ANNUAL REPORT 1989

ANNUAL REPORT
OF THE
ADDICTION RESEARCH CENTER
NATIONAL INSTITUTE ON DRUG ABUSE

P.O. Box 5180
Baltimore, Maryland 21224

NATIONAL INSTITUTES OF HEALTH
NIH LIBRARY

JAN 3 0 2008

BLDG 10, 10 CENTER DR
BETHESDA, MD 20892-1150

U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
Public Health Service
Alcohol, Drug Abuse and Mental Health Administration

TABLE OF CONTENTS

Annual Report of the
Addiction Research Center
National Institute on Drug Abuse
January 1, 1989 - December 31, 1989
Roy W. Pickens, Ph.D., Acting Director

This past year has been a major milestone in the history of the Addiction Research Center (ARC). Jerome H. Jaffe, M.D., who was the Director since July 1984, left the ARC in early September to assume a major leadership role in the National Institute on Drug Abuse's efforts to expand and enhance drug abuse treatment throughout the United States. At that time, Roy W. Pickens, Ph.D., Director of the Institute's Division of Clinical Research, was designated Acting Director, ARC.

Assuring the safety of human subjects in clinical studies has been a major focus during the past year. Plans to establish an Office of the Medical Director that were developed under Dr. Jaffe's leadership have recently been implemented. That Office has been established independent from the research branches with responsibility for ensuring that the protection of subjects is paramount. A medical safety officer who is not a study investigator is designated to provide medical oversight for each clinical study that involves more than minimal risk. Responsibilities for recruitment and counseling of human subjects were assigned to the Office of the Medical Director, and policies and procedures for the recruitment and protection of human subjects are being reviewed and expanded.

A number of other organizational changes have been implemented on an interim basis pending formal approval. Most significant among these are three changes in Branch structure. To strengthen our treatment research program, the Clinical Trials Laboratory was incorporated within the Treatment Branch. The Psychopathology and Immunological Studies Branch was redesignated as the Etiology Branch to increase focus on this area of research. And the Neuroendocrinology/ Immunology Laboratory, which formerly was part of the Psychopathology and Immunological Studies Branch, was transferred to the Clinical Pharmacology Branch.

As evident in this Annual Report, the ARC maintains a highly diverse and productive research program. Major contributions have been made to our knowledge of the causes, consequences, and treatment of drug abuse as indicated by the project summaries and literature citations. These accomplishments are a tribute to the talented and dedicated staff and visiting scientists. Development of

the Annual Report is particularly fitting at this time of transition in leadership, since it provides an opportunity to reflect on past accomplishments, current activities, and future directions. This report reflects the strength and diversity of our neuroscience, preclinical pharmacology, and clinical pharmacology research programs. While the research of our treatment and etiology programs is also of uniformly high quality, an expansion of research in these areas is warranted, and such expansion is a high priority over the next several years. The treatment research program is being expanded primarily in the area of medications development. The etiology research program is being expanded primarily through the development of a large-scale, longitudinal study of drug abusers and their first degree relatives.

To assist in the development of major new research initiatives, a formal planning process has been established. When initial internal ARC review indicates that a proposed initiative is promising and warrants further consideration, outside consultants are involved in the formative planning stages to help us further evaluate the proposal and factors impacting on potential implementation. For example, a group of outside consultants was convened in November 1989 to recommend future etiologic research directions. Further input is obtained from our Board of Scientific Counselors as planning proceeds.

While the Annual Report reflects the direct contributions of the ARC to the scientific body of knowledge in the drug abuse field, an equally important indirect contribution is less evident. This is the contribution of the ARC to the training of current and future drug abuse researchers. During 1989, a large number of scientists received training at the ARC including 38 post-doctoral research fellows and 26 visiting foreign fellows who conducted research under the direction of ARC senior scientists. In addition, 20 high school, undergraduate, and graduate students participated in research projects as part of our Student Fellowship Program sponsored by Merrell Dow Pharmaceuticals, Inc. A number of other foreign and U.S. scientists visited our facility, spending from a few hours to several weeks consulting with staff scientists. Also, the ARC sponsors a weekly seminar series on drug addiction for scientists from other laboratories and students from local training programs, as well as our own staff. During 1989, we initiated planning for another training program, the Medical Staff Fellows program. This program, to be implemented in 1990, will provide physicians interested in clinical research with up to seven years of post-doctoral clinical and research training in the addictions. Clinical training opportunities will be provided in conjunction with other area drug abuse clinical and research facilities.

We wish to acknowledge the contributions of our research staff, staff fellows, visiting scientists, research support staff, and

administrative staff, as well as collaborators from other institutions. Our Board of Scientific Counselors continues to provide advice and guidance that helps assure the quality and balance in our research program. Finally, the encouragement, guidance, and support of Institute and Agency leadership is appreciated.

Clinical Pharmacology Branch

Jack E. Henningfield, Ph.D., Chief

Introduction

The Clinical Pharmacology Branch conducts studies of the effects of drugs in human volunteers to provide the basic information necessary to understanding, treating, and preventing drug addiction. The Branch is comprised of three laboratories: the Biology of Dependence and Abuse Potential Assessment Laboratory, headed by Jack E. Henningfield, Ph.D., the Chemistry and Drug Metabolism Laboratory, headed by Edward J. Cone, Ph.D., and the Neuroendocrinology/Immunology Laboratory, headed by Elizabeth Dax, M.D., Ph.D.

The Branch was reestablished in June, 1989, with the selection of Dr. Henningfield as Branch Chief. It replaces the Clinical Biology Branch described in the three preceeding Annual Reports.

The Clinical Pharmacology Branch emerged from the initial program of basic human research which began in the 1930's at the Addiction Research Center in Lexington, Kentucky. Those pioneering clinical researchers recognized that reconciliation of clinical reports and anecdotal observations with scientific theories of drug addiction could best be accomplished through the careful observation of human volunteers in a controlled research setting. In such a setting, drugs could be administered or withdrawn, and the resulting phenomena objectively documented, while protecting the safety and welfare of the human volunteers. Furthermore, factors likely to be relevant to the course and/or treatment of drug dependence could be manipulated (e.g., treatment with therapeutic medications). This program of research constituted the clinical pharmacology program of the ARC and today is represented by the Clinical Pharmacology Branch along with the Etiology and Treatment Branches.

Much of the present research effort of the Branch represents collaborations among its laboratories as well as with those of other Branches of the ARC. Such multidisciplinary collaborations represent one of the unique strengths of scientific exploration at the ARC. Active current research directions include the following: (1) Established methods are refined, and new methods are being developed, for determining the amount and type of prior drug exposure. (2) In an effort to develop worksite type assessment batteries and more precise research tools, the effects of drug administration and withdrawal over a wide range of behavioral and cognitive performance measures are determined and correlated with physiologic indices such as pupillary function and drug level. (3) The abuse liability testing methodology is used both to quantitate the addiction potential of chemicals as well as to provide information basic to understanding the behavioral and neurochemical mechanisms of drug action. (4) An active program of the development of medications for the treatment of drug dependence involves laboratory assessment of efficacy, toxicity, and abuse

liability of putative medications. (5) Studies are conducted to assess the role of the neuroendocrine system in drug dependence. (6) Studies of the involvement of drug abuse in the transmission and vulnerability to AIDS. (7) The Branch also performs urine toxicology screening as a general support service to the ARC.

The Clinical Pharmacology Branch has also continued in its historically important mission of providing a training ground for the development of researchers and clinicians needed to serve national and international public health needs in the area of drug dependence. This training occurs through the specific hiring of staff fellow and visiting scientists, the occasional participation of students from academic institutions, and through collaborations with other researchers and clinicians. Additionally, over the past four years scientists of the Clinical Pharmacology Branch developed a privately sponsored student fellowship program. This program provides 2-3 month intensive training opportunities for one or more students in each of the laboratories of the Addiction Research Center every year.

1. Biology of Dependence and Abuse Potential Assessment Laboratory - Jack E. Henningfield, Ph.D., Chief

Overview

The Biology of Dependence and Abuse Potential Assessment Laboratory (BDL) is one of three laboratories of the Clinical Pharmacology Branch of the Addiction Research Center (ARC). The purposes of this Laboratory are: first, to assess the biological basis of drug dependence using quantitative experimental procedures of the behavioral and pharmacological disciplines; and, second, to assess the abuse liability and physical dependence potential of selected compounds. These aims are intended to serve the overall mission of the ARC in providing a better foundation for understanding drug dependence and for developing rational approaches for preventing and treating drug dependence.

The BDL evolved out of a tradition of research whose goal was to characterize drug-induced changes in behavior and physiologic function; specifically, phenomena such as drug seeking, tolerance, and physical dependence. The understanding of these phenomena and their interrelations provides much of the pharmacologic and behavioral basis for evolving theories of drug dependence. A practical product of this research was the development of standardized procedures to assess the potential of drugs to produce dependence (i.e., abuse liability and physical dependence potential tests). Early research by Himmelsbach, Frazier, Isbell, Martin, and others, produced fundamental observations upon which much of current theory about the understanding and treatment of drug dependence is based. Specific areas of exploration included the following:

(a) the relationship between drug administration and development of tolerance, physiological dependence and changes in mood and behavior;

(b) the use of drug substitution and antagonist administration procedures to study the biologic basis of drug dependence and to treat addicted people;

(c) the phenomena whereby drug administration could lead to the alleviation of dysphoric mood states and/or the production of euphoric mood states by the presentation of certain drugs; and,

(d) patterns of drug seeking in the presence and absence of pharmacologic pretreatment.

In the course of conduct of these and other basic studies, new strategies of assessment emerged. The methods included the use of observer ratings, pupilometry and cardiovascular assessment, and electroencephalogram (EEG) to provide objective markers of drug administration, as well as the development of new instruments for assessing the effects of drugs on mood, feeling, and behavior. Data obtained using such methods and instruments proved not only to be useful in exploration of the basic phenomena underlying drug dependence, but also led to objective methods of abuse liability assessment. The ability to both quantitatively and qualitatively characterize the clinical pharmacology of substances was also fundamental in development of more selective, safer, and more efficacious agents for the alleviation of human disease and suffering.

Most studies of the BDL are multidisciplinary in nature and involve collaborations with one or more other laboratories of the ARC. With such multidisciplinary efforts it is possible to quantitate the subjective, physiologic, behavioral, electrophysiologic, cognitive, pharmacodynamic, pharmacokinetic, reinforcing, aversive, and other effects of drugs, as well as to assess the biologic generality of phenomena by comparative animal-human research.

In the summary that follows, research is divided into that which is ongoing (Section I) and that in which human testing is completed, but for which follow-up analyses are in progress (Section II).

Special Projects During the Last Year

A. National Cancer Institute Physician Training Program. The Chief, CPB, assisted in the development of a videotaped program to train physicians to treat nicotine dependence using nicotine replacement therapy.

B. Grant Reviews. The Chief, CPB, serves in the NIDA Drug Abuse Clinical and Behavioral Initial Review Group.

C. Student Training Program. We have obtained funding for the fourth consecutive year to provide training fellowships for students. Under the direction of Dr. Heishman, twenty students (high school to postgraduate) were assigned to fourteen laboratories within the ARC. The students assisted in the conduct of basic and clinical research, participated in a formal seminar series, and some co-authored papers submitted for publication.

D. Consulting Services to the Federal Trade Commission (FTC) and the Office on Smoking and Health. The Chief, CPB, provided technical reviews and consultive advice to the Office on Smoking and Health and to the Division of Advertising Practices of the FTC about scientific aspects of a new nicotine delivery systems including a possible non nicotine delivering cigarette.

Summary of Ongoing Research

(Note: The first listed investigator is actively directing the research under the general supervision of the Laboratory Chief; investigators shown in parenthesis have left the ARC but either have contributed, or continue to contribute, to the research.)

A. Assessment of Opioid Agonists and Antagonists: Abuse Potential, Pharmacokinetics, and Pharmacodynamics: Heishman, S.J., Henningfield, J.E., Fudala, P.J, Johnson, R.E., and Cone, E.J.

Subjects with histories of opioid abuse are studied on the Residential Research Unit to determine the possible abuse potential of nalmefene (a new investigational opioid antagonist with relatively few agonist effects). The efficacy of nalmefene in blocking the morphine's effects are also evaluated. Blood samples taken over time are analyzed to provide an assessment of the relationship between the effects of nalmefene and plasma levels of nalmefene and its metabolites. It is hoped that the results of this study will be useful in determining the possible utility of this long-acting opioid antagonist for the treatment of opioid-dependent persons. This study is done in collaboration with the Chemistry and Drug Metabolism Laboratory and the Research Support Branch.

Update: Preliminary subject testing began in the past year.

B. Psychotropic Properties of Stimulants: Stimulus and Sedatives: Discriminative Properties: Heishman, S.J., (Lamb, R.J.), and Henningfield, J.E.

Subjects with histories of stimulant and sedative abuse are studied

on the Residential Research Unit to determine their ability to discriminate between prototypic stimulants and sedatives using both traditional subjective effects measures and behavioral discrimination procedures. An opioid will also be evaluated in some tests involving subjects with histories of opioid abuse. It is hoped that this study will help to improve the accuracy of procedures for assessing abuse liability by quantitatively assessing similarities and differences among drugs based upon studies employing controlled exposure of human volunteers. This study will also generate a base of data obtained from human volunteers that may be compared to the extensive amount of data that has been collected using animal subjects.

Update: Subject testing began in the past year.

C. **Assessment of Mazindol for Abuse Liability: W.B. Pickworth, (Klein, S.A.), Henningfield, J.E., and Kuhar, M.J.**

Subjects with histories of stimulant abuse are studied on the Residential Research Unit to compare the abuse liability of mazindol (an anorectant with some psychomotor stimulant properties) to methylphenidate (a prototypic psychomotor stimulant with a known potential for abuse). One reason for conducting this study is to generate comparative data on the abuse liability of mazindol because the compound has been used in binding studies aimed at isolating the cocaine receptor. In addition, mazindol is a theoretically interesting drug since its mechanism of action, which involves blocking the reuptake of norepinephine and dopamine, would suggest that the compound might be expected to exhibit some abuse liability. Despite this, one previous study and limited clinical experience suggest that actual abuse of the compound is not substantial. Thus, additional characterization of the clinical pharmacology of mazindol could be of importance in analytic efforts directed to dissecting out properties of substances which may be related to abuse potential as well as for considerations about drug development efforts. This study is conducted with the collaboration of the Neuroscience Branch.

Update: Subject testing began in the previous year.

D. **Interaction Between Ethanol and Prostaglandin Synthetase Inhibitors: W.B. Pickworth, (Klein, S.A.), Henningfield, J.E., George, F.R.**

Subjects with histories of moderate alcohol use are studied on the Residential Research Unit to assess the effects of ethanol following pretreatment with either acetaminophen or placebo. Acetaminophen is a prostaglandin synthetase inhibitor that has been shown to reduce several behavioral and physiologic effects of

alcohol in animal studies. Alcohol appears to act, at least in part, by increasing prostaglandin levels. Thus, this drug interaction study makes use of the ARC's standard procedures for assessing abuse potential and performance effects to evalate the possibility of antagonistic effects between acetaminophen and alcohol may be demonstrated in human subjects. This study is conducted in collaboration with the Preclinical Branch.

1988 Update: Subject testing began in the previous year.

E. Passive Tobacco Smoke Exposure: Nicotine Absorption, Subjective Effects and Performance: (Woodson, P.P.), (Roache, J.D.), and Henningfield, J.E.

Three subject groups are being compared in a study of the effects of exposure to ambient tobacco smoke (generated by a cigarette smoking machine) on standard measures of subjective and physiologic effect and performance. The groups are: nondeprived cigarette smokers, 12-hour smoke deprived cigarette smokers, and nonsmokers. It is hoped that the use of the performance battery will provide a quantitative assay by which to determine if ambient levels of tobacco produce effects similar to those previously observed in studies in which nicotine is administered by cigarette smoking or by nicotine gum use.

Update: Subject testing began in 1987 and is continuing as resources permit. Initial research demonstrated the safety and reliability of the procedures for inducing passive tobacco smoke exposure.

F. Effects of Nicotine in Nonsmokers. Heishman, (S.J., Snyder, F.R.), and Henningfield, J.E.

Nonsmokers are exposed to nicotine given in the form of policrilex gum; preliminary testing suggests that this formulation is of low abuse liability and is safe given according to prescribed procedures. Two important experimental questions are addressed in this study. One is a further evaluation of the effects of nicotine polacrilex given to nonsmokers to determine the possible effects of nicotine on cognitive performance in the absence of preexisting nicotine dependence. Notably nicotine enhances performance in deprived smokers but, it remains to be determined if nicotine dependence is a precondition for this effect. The second question is of general import to the understanding of the development of drug dependence. That is, a model of daily repeated voluntary cumulative dosing, we will determine the course of possible development of tolerance to subjective, behavioral and physiologic actions of nicotine. Such data cannot be readily obtained with other drugs of abuse, and probably not with forms of nicotine known to be of high abuse liability (e.g., cigarettes).

Update: An initial study indicated safety of procedures to be used as well as the low toxicity of the polacrilex. No reliable changes in performance were found to be induced by one or two exposures to nicotine. Preliminary subject testing on the present study has began.

G. **Behavioral and Pharmacologic Factors in Nicotine Replacement for Tobacco Dependence: Henningfield, J.E., (Nemeth-Coslett, R.), (Snyder, F.R.), Pickworth, W.B. and Herning, R.I.**

A series of ongoing studies are being conducted to further characterize the pharmacology of nicotine polacrilex gum, and to provide information of practical clinical utility associated with its therapeutic use. These studies include assessing the effects of various doses of nicotine gum dose on performance and mood in tobacco deprived cigarette smokers, nondeprived smokers, and nonsmokers. Other studies have assessed factors that determine the dose of nicotine delivered via this route, e.g., chewing rate. Results from currently completed studies may have practical implications for more efficacious use of the gum to treatment of tobacco dependence as well as for understanding the behavioral pharmacology of nicotine delivered via this route of administration.

Update: New findings from this series of studies were that chewing rate affected the functional dose of nicotine delivered of the gum and that consumption of acidic beverages (i.e., coffee, and soft drinks) substantially reduced absorption of nicotine from the polacrilex preparation.

H. **Opioid Self-Administration: Humans Compared to Animals: Heishman, S.J., (Lamb, R.J.), Henningfield, J.E., Katz, J.L. and Goldberg, S.R.**

Subjects with histories of opioid abuse are given the opportunity to receive an intramuscular injection of morphine on the Residential Research Unit to assess the effects of the schedule of reinforcement and drug paired stimuli on the strength and persistance of the behavior. In the seven initial subjects tested, rates and patterns of responding were similar to those obtained when animals have been tested under analogous conditions, confirming the power of the schedule of reinforcement as well as the cross-species generality of the effect. Interestingly, it appears that the drug-seeking behavior may persist at dose levels which appear to be subthreshold for discrimination by conventional self-report measures.

Update: Subject testing was completed on the first stage of this study; further testing will be done as resources permit.

I. **Effects of Commonly Used Drugs on Behavioral Performance in Normal Subjects (Army Contract-Related Study): (Woodson, P.P.), (Roache, J.D.) and Henningfield, J.E.**

Non-residential subjects are being tested in a study which is of basic interest to the characterization of their clinical pharmacology of commonly used drugs, as well as to partially fulfill contractual obligations to the Army. The study involves the use of performance assessment and other behavioral measures in an examination of the effects of prescription and nonprescription drugs in normal volunteer subjects in the nonresidential paradigm. An additional purpose of these studies is to further development of standardized behavioral performance assessment batteries.

Update: Testing was completed on the alcohol versus chlorpheniramine study and final data analyses are being completed. A new protocol to compare a non-centrally acting antihistamine (terfenadine) to diphenhydramine has been developed and approved, and the study has been initiated. This is the final study in the series of those conducted in collaboration with the Joint Working Group (Army Contract).

J. **Effects of Drugs on Cigarette Smoking and Reponses to Nicotine: (Nemeth-Coslett, R.), Davis, F., Sampson, A., Henningfield, J.E. and (Griffiths, R.R.)**

In an ongoing series of studies, conducted in collaboration with Johns Hopkins School of Medicine, a variety of experimental preparations were used to assess the effects of drugs on rate of cigarette smoking as well as on the self-reported responses to smoking (e.g., satisfaction obtained by smoking). Recently completed studies included evaluations of the effects of marijuana, naloxone, nicotine gum and mecamylamine. Currently being collected and evaluated are data from the Residential Research Unit which have been (and are being) obtained as an adjunct to all Residential studies. These data include information on the rate and pattern of cigarette smoking; they are collected from all subjects using an automated cigarette dispensing and recording system.

Update: Data on cigarette smoking behavior from the Research Unit continue to be collected and evaluated from the automated cigarette dispensing system.

K. **Archival Data Base Project: Haertzen, C.A., Chairman, Data base Committee**

The main purpose of the Database Committee is to combine data from diverse studies and perform analysis on the combined data, building on the extensive screening/testing program initiated the Director

of the ARC at both the recruitment and admission levels. Database activity has been focused on assembling files of scores collected at the two time periods and linking these. This effort has made it possible to compare results from tests collected at the two time periods as well as to relate scores on the various tasks.

The Database project has served several other laboratories of the ARC, and has enabled numerous collaborations on research problems such as the involvement of aggression and personality correlates in drug abuse as well as identification of factors that may be related to treatment outcome. Depending upon the drug, data from about 200 ARCI tests have be entered into the Data base. Morphine data were entered initially; subsequently, data on amphetamine, pentobarbital, alcohol and other drugs have been included.

Update: The database system continues to be refined, data are being collected and diseminated to collaborating laboratories and six manuscripts are in various stages of preparation and/or editorial review. One analysis suggests that hostility, as measured by the Jenkins Composite Hostility Scale, is a general predictor of drug induced effects such as euphoria.

L. Physiologic, Cognitive and Subject Effects of Commonly Abused Drugs: Henningfield, J.E. and Pickworth, W.B.

Subjects with histories of polydrug abuse are being studied to assess the effects of several classes of abused drugs on cognitive, subjective and physiologic measures. The main purpose of the study is to parametrically compare the sensitivity of various testing instruments, including a new pupilometry system, across several classes of drugs, doses and time.

M. Opioid Self-Administration in Humans: Henningfield, J.E. and Heishman, S.T.

The effect of withdrawal states on drug reinforcement and drug self-administration has received little systematic attention from drug abuse researchers. This is a critical omission because it is generally assumed that humans will seek drugs to alleviate unpleasant or relapsing withdrawal symptoms. This research should also be useful in the development of better methods to predict abuse liability of drugs, because it combines the two primary strategies of abuse liability assessment, self-administration and subjective effects testing, in a single study.

N. Nicotine Patch: Effects on Smoking Subjective and Physiologic Function: Henningfield, J.E. and Pickworth, W.B.

A recently developed nicotine patch will be studied in residential research volunteers. The effect of two patches containing 0, 30 and 60 mg will be evaluated on ad lib smoking, subjective effects and physiologic measures. The patch will be tested in subjects with and without histories of drug abuse. The study is of practical importance in the development of a new therapy for smoking cessation.

O. Dopaminergic Lessions and Subjective Effects of Methylphenidate: Uhl, G.R., Kuhar, M.J. and Henningfield, J.E.

The purpose of this research study is to examine whether the effect of the drug "Methylphenidate" that has been used in the therapy of Parkinson's disease is different in patients with Parkinson's disease compared with individuals without this disease. The study will test whether differences in feeling that these drugs can induce in normal individuals may or may not be present in patients with Parkinson's disease. Preliminary testing was initiated in 1989.

Summary of Projects in Which Human Testing is Completed.

A. Triazolam Self-Administration: Effects of Yohimbine Pretreatment: (Roache, J.D.), (Klein, S.A.), (Meisch, R.A.), Henningfield, J.E.,and (Jaffe, J.H.)

Subjects with histories of sedative abuse were studied on the Residential Research Unit to determine the possible effects of an experimental model of anxiety induction (yohimbine pretreatment) on responses to a rapidly-acting benzodiazepine (triazolam). Completion of testing in 3 subjects revealed that: (1) yohimbine pretreatment did produce responses characteristic of anxiety; (2) triazolam self-administration appeared to be increased by yohimbine pretreatment; and (3) triazolam produced deficits on performance and memory tasks to which some tolerance developed. A manuscript is in preparation.

B. Comparative Studies of Intravenous Drug Self-Administration by Monkeys and Human Volunteers: Nicotine and Cocaine. Henningfield, J.E., (Nemeth-Coslett, R.), Katz, J.L. and Schindler, C.W. and Goldberg, S.R.

Volunteers were given access to intravenous nicotine and cocaine delivery in a paradigm similar to that employed to study the reinforcing effects of drugs in animals. Such studies permit comparison of findings obtained with animals and humans and thereby

offer the opportunity to cross-validate human and animal models of drug abuse. In addition, the studies can yield data not possible from studies conducted with either species alone. For instance, the effects of drug-associated stimuli on drug self-administration as well as on the occurrences of subjective effect can be investigated using humans, yet studies with animals permit a much more extensive range of test conditions.

In brief, the studies showed that there was a considerable degree of cross-species generality in the functional effects of variables such as dose and schedule of reinforcement. In addition, an intensive study of the effects of cocaine-paired stimuli showed that these could be important factors involved in the maintenance of drug seeking behavior as well as in the susceptibility to relapse. Manuscripts are in preparation.

C. **Acquisition of Dependence to Cigarettes and Smokeless Tobacco: Henningfield, J.E., Haertzen, C.A. and (Fagerstrom, K.O.), (Nemeth-Coslett, R.), Radzius, A.**

A survey was conducted in collaboration with The Johns Hopkins University School of Medicine to retrospectively assess the patterns of use of cigarettes and smokeless tobacco products. The questionnaire included a scale used to evaluate level of dependence (Fagerstrom Tolerance Questionnaire or FTQ). Preliminary analyses revealed that acquisition of tobacco use is marked by a gradual increase in use over many (8+) years in most tobacco users. Approximately 5% of cigarette smokers remained "chippers" (less than 6 cigarettes per day) for more than two years. There were no clear correlates of dependence development during early exposure to tobacco, however, smoking rates at 6 months were related to smoking rates and levels of dependence 8 years or more later. Another analysis showed that the nicotine yields of different cigarette brands were related in a curvilinear fashion to dependence. That is, smokers of highest nicotine-yielding brands had the highest scores on the FTQ. Manuscripts are in preparation.

D. **Cholinergic Agonists and Antagonists (Army Contract Related): (Roache, J.D.), Henningfield, J.E., Herning R.I.**

Human volunteers without histories of drug abuse, except for cigarette smoking, were tested to assess the possible adverse performance effects of a cholinergic agonist and antagonist, given singley and in combination. The Army Performance Assessment Battery including components of the Triservices Performance Assessment Battery (PAB), was used to evaluate behavioral performance. Manuscript currently in preparation.

E. Factors Influencing Behavioral and Physiological Response to Opioids: Henningfield, J.E., (Higgins, S.T.), (Preston, K.L.), Cone, E.J., and (Jaffe, J.H.)

Postaddicts and nonopiod users have been reported to respond differentially to opioids. This project was designed to experimentally examine such population differences in response to mu and kappa opioids on subjective, behavioral, physiological and neuroendocrine parameters using postaddicts and opiate-naive normal residential volunteers. In the initial study (completed), the effects of single doses of naloxone following either placebo or morphine pretreatment were studied in subjects with histories of opioid dependence. Laboratory testing is complete on the first phase of the study. Initial results suggest that a single dose of morphine produces sufficient physical dependence such that a mild morphine withdrawal-like effect was observed when the opioid antagonist, naloxone, was subsequently administered. A manuscript is currently in preparation.

F. Abuse liability of Smokeless Tobacco Products: Henningfield, J.E., Radzius, A., (Nemeth-Coslett, R.) and Cone, E.J.

Two smokeless nicotine delivery systems were evaluated using standardized procedures to assess the pharmacodynamic variables relevant to their potential liability for abuse, as well as the degree to which effects were similar to those known to be produced by cigarette smoking. One of the systems was a commercially available smokeless tobacco product (snuff pouches) which was held in the mouth to provide buccal nicotine absorption; the other was a smokeless "cigarette" through which air was sucked to inhale vaporized nicotine. Both products produced orderly dose-related effects which were generally similar to nicotine delivered by cigarette smoke. A third smokeless nicotine delivery system, a pleasantly flavored nicotine delivering chewing gum, is currently under review for possible clinical testing. Manuscripts from the first two studies are in preparation.

G. Physiologic Dependence to Tobacco: Cigarette Withdrawal and Nicotine Substitution: Henningfield, J.E., (Nemeth-Coslett, R.), (Snyder, F.R.), Pickworth, W.B., Herning, R.I. and Cone, E.J.

Two intensive multidisciplinary collaborative studies were conducted using variations on the "direct addiction" and "substitution" strategies for assessing withdrawal reactions and cross tolerance. In the first study, cigarette smokers were abruptly withdrawn from tobacco for ten days, and then allowed to resume smoking. In the second study, smokers were tested in repeating cycles of 4 days smoking and 3 days abstinence; during abstinence, they were given either 0, 2 or 4 mg nicotine containing

pieces of gum to chew. A characteristic tobacco withdrawal syndrome was obtained in the first study and on 0 mg gum days in the second study. Of particular interest were certain performance and electrophysiologic data that showed little tendency to recover over the 10 day period of observation. Nicotine gum produced a dose-related blockade of withdrawal responses. Two manuscripts have been submitted for publication and four others are in preparation.

H. Behavioral Performance and Physiologic Effects of Drugs: Atropine and Diazepam (Army Contract Related): (Higgins, S.T.), (Lamb, R.J.), Pickworth, W.B., Herning, R.I. and Henningfield, J.E.

Volunteers without histories of drug abuse, except cigarette smoking, were studied on the Residential Research Unit to assess the effects of atropine or diazepam on mood and performance. These studies were conducted in collaboration with the Joint Triservices Working Group. Both atropine and diazepam produced dose related impairments on some performance measures. The differential sensitivity of the various PAB components provided useful practical informaton for subsequent performance testing at the ARC and elsewhere. One manuscript is in press, four others are in preparation.

I. Why Do Substance Abusers Seek Help? What Are Their Worries About That Help?: Henningfield, J.E., Johnson, R.E., (Brooke, D.)

A survey of ARC research subjects was conducted to investigate the reasons that people seek treatment, and what their worries about that treatment are. We hope that the answers to these questions will enable us to make it easier for people to seek help. Subjects were asked to fill out two questionnaires and a cover sheet on their past experience of seeking help.

Papers Published or Accepted for Publication

Glover, E.D., Schroeder, K.L., Henningfield, J.E., Winn, D.M., Severson, H.H. and Christen, A.G.: A compendium of smokeless tobacco research. Journal of Drug Education. In press.

Goldberg, S.R. and Henningfield, J.E. (Eds.). Meeting Report: Nine papers on progress in understanding the relationship between the pharmacological effects of nicotine and human tobacco dependence. Pharmacol Biochem Behav 30:215-294, 1988.

Henningfield, J.E.: Redefining craving. NIDA Notes 2:9, 1987.

Henningfield, J.E.: Reducing urges to smoke. Chest 92:963, 1988.

Henningfield, J.E.: Tobacco-caused diseases are side effects of nicotine dependence. J Med Soc NJ 85(2):108-112, 1988.

Henningfield, J.E. and Brown, B.S.: Do replacement therapies treat craving? NIDA Notes 2:8-9, 1987.

Henningfield, J.E. and Goldberg, S.R.: Introduction: Progress in understanding the relationship between the pharmacological effects of nicotine and human tobacco dependence. Pharmacol Biochem Behav 30:217-220, 1988.

Henningfield, J.E. and Goldberg, S.R.: Pharmacological determinants of tobacco self-administration by humans. Pharmacol Biochem Behav 30:221-226, 1988.

Goldberg, S.R. and Henningfield, J.E.: Reinforcing effect of nicotine in humans and experimental animals responding under intermittent schedules of i.v. drug injection. Pharmacol Biochem Behav 30:227-234, 1988.

Henningfield, J.E., Goldberg, S.R. and Jasinski, D.R.: Abuse Liability and Dependence Potential of Nicotine. In Martin, W.R., Van Loon, G.R., Iwamoto, E.T., and Davis, D.L. (Eds.) Tobacco Smoke and Nicotine: A Neurobiologic Approach. New York: Plenum Press, 1987 pp. 81-99.

Henningfield, J.E. and Jasinski, D.R.: Pharmacological Basis for Nicotine Replacement. In: Pomerleau, O.F., Pomerleau, C.S., Fagerstrom, K.O., Henningfield, J.E. and Hughes, J.R. (Eds.): Nicotine Replacement: A Critical Evaluation. New York, NY: Alan R. Liss, 1988 pp. 35-61.

Henningfield, J.E., Johnson, R.E. and Jasinski, D.R.: Clinical Procedures for the Assessment of Abuse Potential. In Bozarth, M.A., (Ed.): Methods of Assessing the Reinforcing Properties of Abused Drugs, New York, NY: Springer-Verlag, 1987 pp. 573-590.

Higgins, S.T., Preston, K.L., Cone, E.J., Henningfield, J.E. and Jaffe, J.H.: Behavioral, Physiological, and Hormonal Effects of a Naloxone Challenge following Acute Morphine Pretreatment in Humans. In Harris, L.S. (Ed.): NIDA Research Monograph. Washington, D.C.: U.S. Government Printing Office, 1988 pp. 266-273.

Jarvik, M.E. and Henningfield, J.E.: Pharmaologic treatment of tobacco dependence. Pharmacol Biochem Behav 30:279-294, 1988.

Jasinski, D.R. and Henningfield, J.E.: Conceptual Basis of Replacement Therapies for Chemical Dependence. In: Pomerleau, O.F., Pomerleau, C.S., Fagerstrom, K.O., Henningfield, J.E. and Hughes, J.R., (Eds.): Nicotine Replacement: A Critic1 Evaluation. New York, NY, Alan R. Liss, 1988 pp. 13-34.

Nemeth-Coslett, R., Henningfield, J.E., O'Keeffe, M.K. and Griffiths, R.R. Nicotine Gum: Dose-related effects on cigarette smoking and subjective ratings. Psychopharmacology 92: 424-430, 1987.

Nemeth-Coslett, R., Robinson, N., and Benowitz, N. and Henningfield, J.E.: Nicotine Gum: Chew rate, subjective effects and plasma nicotine. Pharmacol Biochem Behav 29:747-751, 1988.

Pomerleau, O.F., Pomerleau, C.S., Fagerstrom, K.O., Henningfield, J.E. and Hughes, J.R. (Eds.): Nicotine Replacement: A Critical Evaluation. New York, NY, Alan R. Liss, 1988.

Henningfield, J.E. and Nemeth-Coslett, R.: Nicotine dependence: Interface between tobacco and tobacco-related disease. Chest 90:37S-55S, 1988.

Waranch, H.R., Henningfield, J.E. and Edmunds, M.: Letter to the Editor: Elimination of nicotine gum use following successful replacement therapy for cigarette smoking. Lancet January 2-9:49-50, 1988

Pickworth, W.B., Herning, R.I. and Henningfield, J.E.: Mecamylamine reduces some EEG effects of nicotine chewing gum in humans. Pharmacol Biochem Behav 30:149-153, 1988.

Henningfield, J.E.: Improving the diagnosis and treatment of nicotine dependence (Editorial). JAMA 260(11):1613-1614, 1988.

Higgins, S.T., Woodward, B.M. and Henningfield, J.E. Effects of atropine on the repeated acquisition and performance of response sequences in humans. J Exp Anal Behav, In press.

Snyder, F.R. and Henningfield, J.E.: Effects of acute nicotine deprivation and administration: Assessment on computerized performance tasks. Psychopharmacology, in press.

Rose, J.E., Sampson, A., Levin, E.D. and Henningfield, J.E. Mecamylamine increases nicotine preference and attenuates nicotine discrimination. Pharmacol Biochem Behav, in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00026-01 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Assessment of Opioid Agonists and Antagonists

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
S.J. Heishman	Staff Fellow	BDL, ARC, NIDA
E.J. Cone	Chief	CDM, ARC, NIDA
R.E. Johnson	Chief	RSB, ARC, NIDA
P.J. Fudala	Deputy Chief	RSB, ARC, NIDA

COOPERATING UNITS *(if any)*

Chemistry & Drug Metabolism Laboratory
Research Support Branch

LAB/BRANCH
Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
1.10	0.40	0.70

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Subjects with histories of opioid abuse were studied on the Residential Research Unit to investigate the effects of nalmefene, a new investigational opioid antagonist with relatively few agonist effects. This research should be useful in determining the possible utility of this long-acting (several days) opioid antagonist for the treatment of opioid dependent persons. This research is being conducted in collaboration with the Chemistry and Drug Metabolism Laboratory and the Research Support Branch. Two studies were planned, the first will assess the abuse liability of nalmefene and the second will determine the efficacy of nalmefene to block the subjective and physiological effects of morphine. The first study has been completed. Results indicated that nalmefene did not produce typical opiate-like abuse liability, but that side effects, such as feelings of agitation and irritability, muscle tension, headache, and insomnia may limit its use as possible treatment for opioid dependence. The second study has begun with one subject having completed protocol.

Abstract:

Fudala, P.J., Johnson, R.E., Heishman, Cone, E.J. & Henningfield, J.E. Abuse liability assessment of the long-acting opiate antagonist nalmefene. In L.S. Harris (Ed.), Problem of Drug Dependence 1989. NIDA Research Monograph, in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00027-01 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Psychotropic Properties of Stimulants and Sedatives: Discriminative Properties

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
S.J. Heishman	Staff Fellow	BDL, ARC, NIDA
R.J. Lamb	Staff Fellow	BDL, ACR, NIDA
W.R. Lange	Medical Officer	RSB, ARC, NIDA

COOPERATING UNITS *(if any)*

Research Support Branch

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.85	0.25	0.60

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

In these studies the psychotropic effects of the prototypical stimulant, d-amphetamine, were compared to those of another stimulant and to the effects of a sedative and an opioid. These comparisons were conducted in subjects with histories of stimulant and either opioid or sedative abuse, and were carried out using two different types of procedures simultaneously. The first procedure was a drug discrimination task. In the drug discrimination procedure subjects were trained to respond on one lever following administration of the training dose of d-amphetamine and on another lever in the absence of d-amphetamine. Correct responding was reinforced by money. The second procedure was a traditional abuse liability assessment procedure that utilized physiologic and self-report measures.

To date two studies have been conducted. In the first the effects of amphetamine and hydromorphone were compared. In the second the effects of amphetamine, methylphenidate, and diazepam were compared. In both studies amphetamine dose-relatedly occasioned d-amphetamine appropriate responding. Methylphenidate, also, dose-relatedly occasioned d-amphetamine appropriate responding. In contrast neither diazepam nor hydromorphone occasioned d-amphetamine appopriate responding. The subjective effects of d-amphetamine and methylphenidate were similar and covaried with their discriminative effects, while the subjective effects of diazepam were clearly different. In contrast, the only self-report measure that distinguished hydromorphone from d-amphetamine were drug identifications. Thus these studies show that drug discrimination procedures can be drug-class specific in humans, and that while these discriminative effects can covary with the subjective effects of the drug. The discriminative effects of amphetamine under these conditions appear to be controlled in a manner most similar to the identification of a drug.

PHS 6040 (Rev 1/84)

GPO 914-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: ZO1 DA00028-01 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Assessment of Mazindol for Abuse Liability

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
W.B. Pickworth	Pharmacologist	BDL, ARC, NIDA
M.J. Kuhar	Chief	MPL, ARC, NIDA

COOPERATING UNITS *(if any)*
Molecular Pharmacology Laboratory

LAB/BRANCH
Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL	OTHER
0.65	0.30	0.35

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Subjects with histories of stimulant abuse are studied on the Residential Research Unit to determine the abuse liability of mazindol (an anorectant with some psychomotor stimulant properties) to methylphenidate (a prototypic psychomotor stimulant with a known potential for abuse). This study was performed because mazindol has been used in binding studies aimed at isolating the cocaine receptor. The results of those studies indicated that mazindol has high affinity binding at cocaine-sensitive dopamine receptor sites. Mazindol is a theoretically interesting drug since its apparent mechanism of action (blocks reuptake of norepinephine and dopamine) suggests that it would have abuse potential. However, one previous study and limited clinical experience, suggest that it is seldom abused. Therefore additional characterization of the clinical pharmacology of mazindol could be of importance in analytic efforts as well as drug development. This study is conducted in collaboration with the Neuroscience Branch. Subject testing has been completed. Preliminary analyses indicate that mazindol and methylphenidate increased heart rate and diastolic blood pressure and decreased hunger. Mazindol decreased vigor and increased measures of fatigue and tired and elevated scores on the PCAG and LSD scales of the ARCI. Methylphenidate did not cause the sedative-like effects seen after mazindol. Subjects reported disliking for each drug. These data indicate that at doses three times the therapeutic level mazindol poses little abuse potential. On the other hand its dysphoric effects call to question the acceptability of mazindol for the treatment of cocaine dependence.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00029-01 BDL

PERIOD COVERED: January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Interaction Between Ethanol and Prostaglandin Synthetase Inhibitors

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

J.E. Henningfield	Chief	BDL, ARC, NIDA
W.B. Pickworth	Pharmacologist	BDL, ARC, NIDA
F.R. George	Staff Fellow	PPB, ARC, NIDA

COOPERATING UNITS (if any)
Preclinical Pharmacology Branch

LAB/BRANCH
Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.65	0.30	0.35

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

Subjects with histories of moderate alcohol use are studied on the Residential Research Unit to assess the effects of ethanol following pretreatment with either acetaminophen (325, 650, 1300 and 1950 mg) or placebo. Acetaminophen is a prostaglandin synthetase inhibitor that has been shown to reduce several behavioral and physiologic effects of alcohol in animal studies, alcohol appears to act in part by increasing prostaglandin levels. This drug interaction study makes use of our standard procedures for assessing abuse potential and performance to evalate the possibility of such antagonistic effects in human subjects. This study is conducted in collaboration with the Preclinical Branch. Subject testing has been completed. Preliminary analyses indicate that alcohol at this dosage (0.625 gm/kg, taken over 90 mins) caused subjective effects (drunk, feel drug, sober, etc) but did not reliably change physiologic or performance measures. Pretreatment with acetaminophen did not influence the subjective effects. The results are being prepared for publication and another study is being proposed involving higher doses of alcohol and a more effective prostaglandin synthesis inhibitor.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00030-01 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Passive Tobacco Smoke: Nicotine Absorption, Subjective Effects and Performance

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
P.P. Woodson	Staff Fellow	BDL, ARC, NIDA
J.D. Roache	Staff Fellow	BDL, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH
Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER
0.35	0.15	0.20

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Three subject groups are being compared in a study of the effects of exposure to ambient tobacco smoke, generated by a cigarette smoking machine, on standard measures of subjective and physiologic effect as well as on performance. The groups are: nondeprived cigarette smokers, 12-hour smoke deprived cigarette smokers, and nonsmokers. It is hoped that the use of the performance battery included in this study will provide a quantitative assay by which to determine if various ambient levels of tobacco smoke can produce dose-dependent effects on performance and physiology which are comparable to those observed with cigarette smoking. Initial research demonstrated the safety and reliability of the procedures for inducing passive tobacco smoke exposure. Further testing continuing as resources permit.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00031-01 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Effects of Nicotine in Nonsmokers

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
S.J. Heishman	Staff Fellow	BDL, ARC, NIDA
F.R. Snyder	Statistician	NOVA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL.	OTHER.
1.10	0.30	0.80

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Nonsmokers are exposed to nicotine given in the form nicotine polacrilex gum; preliminary testing suggests that this formulation is of low abuse liability and is safe if given according to prescribed procedures. Two important experimental questions are addressed in this study. One concerns the further evaluation of the effects of nicotine polacrilex gum in nonsmokers to determine the possible effects of nicotine on cognitive performance in the absence of pre-existing nicotine dependence. Nicotine enhances performance in deprived smokers; however, it remains to be determined if nicotine dependence is a precondition for this effect. The second question is of general importance to the understanding of the development of drug dependence. Using a model of daily repeated voluntary cumulative dosing, the course of possible development of tolerance to the subjective, behavioral and physiologic actions of nicotine will be determined. Such data cannot be readily obtained with other drugs of abuse, and probably not with forms of nicotine known to be of high abuse liability (e.g., cigarettes), but may be safely collected following the procedures used in this study. To date, eight subjects have completed the protocol and 5-8 more subjects will be tested. Preliminary results indicate that over the course of the study, tolerance to the effects of nicotine developed for some, but not all, measures.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00010-05 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Behavioral and Pharmacologic Factors in Nicotine Replacement for Tobacco Dependence

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R. Nemeth-Coslett	Staff Fellow	BDL, ARC, NIDA
P.P. Woodson	Staff Fellow	BDL, ARC, NIDA
R.I. Herning	Chief	CHP, ARC, NIDA
W.B. Pickworth	Pharmacologist	CHP, ARC, NIDA
F.R. Snyder	Statistician	NOVA

COOPERATING UNITS *(if any)*

Cognitive Studies and Human Performance Laboratory

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.50	0.15	0.35

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Nicotine polacrilex (chewing gum) has been under investigation as a replacement for tobacco-delivered nicotine and also as a convenient drug administration modality which provides a model of more general interest for drug dependence researchers. For example, nicotine gum was employed in our initial studies to examine the capabilities of this Laboratory's recently established performance and electrophysiologic assessment approaches for evaluating drug effects. The course of research conducted using this preparation has been determined by the priorities of the ARC and the Chief of the Biology of Dependence Laboratory. These studies have included the following: (1) Effects of nicotine gum replacement on cigarette smoking and tobacco smoke exposure; (2) Pharmacodynamic effects of nicotine gum compared to other routes of nicotine administration; (3) Abuse liability of nicotine gum; (4) Dose-related effects on subjective, behavioral, and physiologic variables, including studies of the factors which may affect the functional dose, such as chewing and swallowing rates; (5) Effects of nicotine gum administration on learning and performance in non-smokers; and, (6) Role of oral pH in nicotine absorption.

PHS 6040 (Rev 1/84) GPO 914-911

Behavioral and Pharmacologic Factors in Nicotine Replacement for Tobacco Dependence
ZO1 DA00010-05

Publications

Nemeth-Coslett, R., Benowitz, N.L., Robinson, N. and Henningfield, J.E.: Nicotine Gum: Chew rate, subjective effects and plasma nicotine. Pharmacol Biochem Behav 29:747-751, 1988.

Pickworth, W.B., Herning, R.I. and Henningfield, J.E.: Electroencephalographic effects of nicotine gum in humans. Pharmacol Biochem Behav 25:879-882, 1986.

Jasinski, D.R. and Henningfield, J.E.: Conceptual Basis of Replacement Therapies for Chemical Dependence. In Pomerleau, O.F., Pomerleau, C.S., Fagerstrom, K.O., Hennigfield, J.E. and Hughes, J.R. (Eds.): Nicotine Replacement: A Critical Evaluation. New York, NY, Alan R. Liss, 1988, pp. 13-34.

Waranch, H.R., Hennigfield, J.E. and Edmunds, M.: Letter to the Editor: Elimination of nicotine gum use following successful replacement therapy for cigarette smoking. Lancet January 2-9:49-50, 1988.

Snyder, F.R. and Henningfield, J.E.: Effects of acute nicotine deprivation and administration: Assessment on computerized performance tasks. Psychopharmacology. In press.

Pickworth, W.B., Herning, R.I. and Henningfield, J.E.: Mecamylamine reduces some EEG effects of nicotine chewing gum in humans. Pharmacol Biochem Behav 30:149-153, 1988.

Pomerleau, O.F., Pomerleau, C.S., Fagerstrom, K.O., Henningfield, J.E. and Hughes, J.R.: (Eds.): Nicotine Replacement: A Critical Evaluation. New York, NY, Alan R. Liss, 1988

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00024-02 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Opioid Self-Administration: Humans Compared to Animals

PRINCIPAL INVESTIGATOR *(List other professional personnal below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R.J. Lamb	Staff Fellow	BDL, ARC, NIDA
S.R. Goldberg	Chief	BPL, ARC, NIDA
J.L. Katz	Staff Fellow	BPL, ARC, NIDA
C.W. Schindler	Staff Fellow	BPL, ARC, NIDA
R.A. Meisch	Visiting Scientist	U of TX, Houston

COOPERATING UNITS *(if any)*

Behavioral Pharmacology Laboratory

LAB/BRANCH
Clinical Pharmacology Branch

SECTION
Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.15	0.10	0.05

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Studies with animals have shown that stimuli associated with drug delivery can come to function as variables that partially control drug-seeking behavior and the likelihood of resumption (i.e., relapse) to such behavior, even in the absence of the drug. Analogous research strategies are being used to assess the generality of these findings to human subjects. In addition, these procedures provide data on the degree of correspondence between self-reported drug effects and drug seeking behavior. The human studies have produced a number of interesting results. When the consequences of varying the dose of morphine available on self-administration, physiological effects, and self-reported effects were examined, it was found that low doses of morphine (3.75 mg) maintained rates of responding above placebo and constricted pupillary diameter, but did not reliably alter the self-reports of the subjects, indicating a dissociation between the subjective effects of morphine and morphine's reinforcing properties. Another study evaluated the role of a stimuli paired with drug administration on the maintenance of responding. Initial results suggested that the stimuli were of less importance than in an analogous study with animals, as well as in somewhat similar study of cocaine self-administration by humans. The basis for these differences is currently under investigation.

Manuscript submitted for publication.

Lamb, R.J., Preston, K.L., Henningfield, J.E., Schindler, C.W., Meisch, R.A., Davis, F., Katz, J.L. and Goldberg, S.R.: The Reinforcing and Subjective Effects of Morphine in Post-Addicts: A Dose-Response Study. J Pharm Exp Ther. Submitted.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00007-04 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effects of Commonly Used Drugs on Behavioral Performance in Normal Subjects

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
P.P. Woodson	Staff Fellow	BDL, ARC, NIDA
J.D. Roache	Visiting Scientist	U of TX, Houston

COOPERATING UNITS *(if any)*
Clinical Pharmacology Branch

LAB/BRANCH
Biology of Dependence and Abuse Potential Assessment Laboratory

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.45	0.45	1.00

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The possible adverse effects on performance of an antihistamine and alcohol are being evaluated in non-residential subjects without histories of drug abuse, other than cigarette smoking. The study involves the use of strategies recommended by the Joint Triservices Working Group (Army Contract) to assess behavioral (i.e., cognitive) performance. Measures include the standard Army Performance Assessment Battery (PAB), prototypic portions of the Unified Triservices Battery (UTC PAB), critical flicker fusion, and mood, as well as cardiovascular and other basic physiologic variables.

Preliminary analysis of data from the first study suggest that alcohol and chlorpheneramine produced dose-related effects on several self-report measures and mixed effects on performance across measures. These initial results suggest that the PAB is less sensitive compared to the Digit Symbol Substitution Task with respect to the level of performance disruption by alcohol or chlorpheramine.

A new protocol to compare a non-centrally acting antihistamine (terfenadine) to a centrally acting one (diphenhydramine) as well as to the benzodiazepine, triazolam, has been developed and approved, and the study has been initiated. This is the final study in the series of those conducted in collaboration with the Joint Triservices Working Group (Army Contract).

PHS 6040 (Rev 1/84) GPO 814-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00009-05 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effects of Drugs on Cigarette Smoking and Responses to Nicotine

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R. Nemeth-Coslett	Staff Fellow	NIDA
F.C. Davis	Nurse	BDL, ARC, NIDA
A.H. Sampson	Nurse	BDL, ARC, NIDA
R.R. Griffiths	Collaborator	Johns Hopkins
J.E. Rose	Collaborator	VA Medical Center Durham, NC

COOPERATING UNITS *(if any)*

LAB/BRANCH
Clinical Pharmacology Branch

SECTION
Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.23	0.03	0.20*

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Some of these studies were conducted in the physical facilities of the Behavioral Pharmacology Research Unit at Johns Hopkins University Medical School (JHMU) which provides some research support*. For example, multiple measures of cigarette smoking, subjective effect, and physiologic effect were collected during ad libitum smoking sessions in normal volunteers following administration of mecamylamine, naloxone, or marijuana. Mecamylamine had effects opposite to those observed with nicotine on several measures of smoking and subjective response: mecamylamine increased smoking and had some sedating effects. Despite this, both drugs decreased the satisfaction derived from smoking. These findings are not consistent with either the hypothesis that smoking is substantially mediated by endorphin release or the hypothesis that smoking is simply related to the level of positive subjective state.

Presently, basic measures of cigarette smoking are being collected from all subjects on the Clinical Research Unit and data analyses have begun. This database-type of study appears to be providing the opportunity to quantitate the effects of a wide range of variables on cigarette smoking (i.e., atropine administration, cocaine withdrawal, buprenorphine administration, and passive tobacco smoke exposure).

Effects of Drugs on Cigarette Smoking and Response to Nicotine
ZO1 DA00009-05 BDL

Publications

Nemeth-Coslett, R. and Griffiths, R.R.: Naloxone does not affect cigarette smoking. Psychopharmacology, 89:261-264, 1986.

Nemeth-Coslett, R., Henningfield, J.E., O'Keeffe, M.K. and Griffiths, R.R.: Effects of mecamylamine on human cigarette smoking and subjective ratings. Psychopharmacology 88:420-425, 1986.

Nemeth-Coslett, R., Henningfield, J.E., O'Keeffe, M.K. and Griffiths, R.R.: Effects of marijuana on human cigarette smoking and physiologic changes and subjective responses. Pharmacol Biochem Behav 25:659-665, 1986.

Nemeth-Coslett, R., Henningfield, J.E., O'Keeffe, M.K. and Griffiths, R.R.: Nicotine gum: Dose-related effects on cigarette smoking and subjective ratings. Psychopharmacology 92:424-430, 1987.

Rose, J.E., Sampson, A., Levin, E.D. and Henningfield, J.E.: Mecamylamine increases nicotine preference and attenuates nicotine discrimination. Pharmacol Biochem Behav In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00013-04 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Archival Data Base Project

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

C.A. Haertzen	Research Psychologist	BDL, ARC, NIDA
J.E. Henningfield	Chief	BDL, ARC, NIDA
W.R. Lange	Medical Officer	RSB, ARC, NIDA
J.H. Jaffe	Former Director	NIDA
W.E. Weddington	Staff Fellow	NIDA
L. Covi	Visiting Scientist	TEI, ARC, NIDA

COOPERATING UNITS *(if any)*

Research Support Branch; Biology of Vulnerability Laboratory
Cognitive Studies and Human Performance Laboratory
Treatment Laboratory

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER
1.12	0.62	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Currently data obtained by the recruitment staff (i.e., Addiction Severity Index, Symptom Checklist [SCL-90], Shipley IQ, Early Childhood Aggression) and admission test data (i.e., Diagnostic Interview Schedule, Buss-Durkee Hostility, Minnesota Multiphasic Personality Inventory [MMPI], Alcohol Related Behavior Questionnaire, and electroencephalogram) have been combined into a single database. The results of the analyses will be reported by those in the Psychology of Vulnerability Laboratory. One finding of particular interest concerned hostility (i.e., assault), which is related to antisocial personality. For example, a database comprised of 97 opiate addicts given the Addiction Research Center Inventory (ARCI) under no-drug and morphine (20 mg, i.m.) conditions and the MMPI under a non-drug condition was assembled. Interest in this data base was focused on the question of whether a high level of hostility constituted a risk factor for feeling greater morphine effects. Hostility was positively related to four morphine-related scales. Further, those high on hostility had twice the change in elevation on a simulated opiate scale as those who were low. This data base has been extended to include a wide range of other psychoactive drugs. A computerized dictionary of drug associations to heroin, Benzedrine, alcohol, barbiturates, and marijuana has been assembled which covers 8625 words or phrases. Current data base activities emphasize the cocaine treatment oriented studies by Drs. Weddington, Covi, and Kolar.

GPO 914-910

Publications

Haertzen, C.A. and Hickey, J.E.: Addiction Research Center Inventory (ARCI): Measurement of Euphoria and Other Drug Effects. In Bozarth, M.A. (Ed.) Methods of Assessing the Reinforcing Properties of Abused Drugs. New York, NY, Springer-Verlag, 1987, pp. 489-524.

Lange, W.R., Haertzen, C.A., Hickey, J.E., Snyder, F.R., Dax, E.M., and Jaffe, J.H.: Nitrite inhalants: Patterns of abuse in Baltimore and Washington, D.C. Am J Drug Alcohol Abuse 14:29-39, 1988.

Rose, M.R., Brown, B.S., and Haertzen, C.A.: Comparison of the characteristics and functioning of cocaine treatment and cocaine research subjects. Am J Drug Alcohol Abuse 15:251-260, 1989.

Fishbein, D.H., Herning, R.I., Pickworth, W.B., Haertzen, C.A., Hickey, J.E., and Jaffe, J.H.: EEG and Brainstem auditory evoked response potentials in adult male drug abusers with self-reported histories of aggressive behavior. Biological Psychiatry 595-611, 1989.

Muntaner, C., Nagoshi, C., Jaffe, J.H., Walter, D., Haertzen, C., Fishbein, D.H.: Correlates of self-reported early childhood aggression in subjects volunteering for drug studies. Am J Drug Alcohol Abuse 15:383-402, 1989.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Cone, E.J., Dax, E.M., Herning, R.I., Michaelson, B.S.: Changes in mood, craving, and sleep during acute abstinence reported by male cocaine addicts: A controlled, residential study. Archives of General Psychiatry. In press.

Haertzen, C.A., Hickey, J.E., Rose, M.R. and Jaffe, J.H.: The relationship between a diagnosis of antisocial personality and hostility: Development of an Antisocial Hostility Scale. J Clin Psychol. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00036-01 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Physiologic, cognitive and subject effects of commonly abused drugs

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
W.B. Pickworth	Pharmacologist	BDL, ARC, NIDA

COOPERATING UNITS *(if any)*

Biology of Dependence and Abuse Potential Assessment Laboratory

Clinical Pharmacology Branch

LAB/BRANCH

SECTION

Addiction Research Center, NIDA, Baltimore, MD 21224

INSTITUTE AND LOCATION

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER:
0.80	0.3	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Subjects with histories of polydrug abuse are studied on the Residential Research Unit to assess the effects of several classes of abused drugs on cognitive, subjective and physiologic measures. The main purpose of the study is to parametrically compare the sensitivity of various testing instruments across several classes of drugs, doses and time. The results are theoretical important because they will evaluate the sensitivities of methods used in the drug abuse field. The study is of practical importance because in evaluating the utility of dynamic pupillography as a drug detection screen. The protocol has been approved by the IRB and subject recruiting has started.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00034-01 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Opioid Self-Administration in Humans

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
S.J. Heishman	Staff Fellow	BDL, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.5	0.2	0.3

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The effect of withdrawal states on drug reinforcement and drug self-administration has received little systematic attention from drug abuse researchers. This is a critical omission because it is generally assumed that humans will seek drugs to alleviate unpleasant or relapsing withdrawal symptoms, thus maintaining their state of physical dependence or relapsing after a period of abstinence. This research will examine the following issues: (a) the pattern of self-administration when only low doses of opiates are available, (b) the effect of opioid antagonist-precipitated withdrawal on opiate self-administration, and (c) the relationship between self-administration behavior and subjective drug effects. This research should be useful in the development of better methods to predict abuse liability of drugs, because it combines the two primary strategies of abuse liability assessment, self-administration and subjective effects testing, in a single study. In addition to addressing these important pharmacological-behavioral interactions, this research may ultimately result in more effective treatment methods for drug abuse. Pilot testing of residential subjects with a history of opioid abuse has been completed, and the main study should begin soon.

PHS 6040 (Rev 1/84)

GPO 914-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00033-01 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Nicotine Patch: Effects on smoking subjective and physiologic function

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
W.B. Pickworth	Pharmacologist	BDL, ARC, NIDA

COOPERATING UNITS *(if any)*

Biology of Dependence and Abuse Potential Assessment Laboratory

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL.	OTHER
0.80	0.3	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

A recently developed nicotine patch will be studied in residential research volunteers. The effect of two patches containing 0, 30 and 60 mg will be evaluated on ad lib smoking, subjective effects and physiologic measures. The patch will be tested in subjects with and without histories of drug abuse. The study is of practical importance in the development of a new therapy for smoking cessation.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00032-01 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Dopaminergic lessions and subjective effects of methylphenidate

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

G.R. Uhl	Visiting Scientist	MNL, ARC, NIDA
M.J. Kuhar	Chief	NB, ARC, NIDA
J.E. Henningfield	Chief	BDL, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH
Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER
0.30	0.10	0.20

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The purpose of this research study is to examine whether the effect of the drug "Methylphenidate" that has been used in the therapy of Parkinson's disease is different in patients with Parkinson's disease compared with individuals without this disease. The study will test whether differences in feeling that these drugs can induce in normal individuals may or may not be present in patients with Parkinson's disease. Preliminary testing was initiated in 1989.

PHS 6040 (Rev 1/84)

GPO 914-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00006-03 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Triazolam Self-Administration: Effects of Yohimbine Pretreatment

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
J.D. Roache	Staff Fellow	BDL, ARC, NIDA
R.A. Meisch	Visiting Scientist	BDL, ARC, NIDA
S.A. Klein	Staff Fellow	BDL, ARC, NIDA
J.H. Jaffe	Former Director	NIDA

COOPERATING UNITS *(if any)*

Biology of Vulnerability

LAB/BRANCH
Clinical Pharmacology Branch

SECTION
Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.70	2.00	5.00

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The purpose of this study is to examine the effects of yohimbine pretreatment on the self-administration of triazolam in subjects with histories of sedative abuse. Two issues of relevance to the behavioral pharmacology of drug abuse are being addressed: the first involves the development of procedures to measure sedative/anxiolytic drug self-administration; and, the second is to examine the effects of yohimbine pretreatment on triazolam self-administration. It is of basic theoretical, as well as clinical, interest to define methods to detect the effects of one drug on the self-administration of another drug. In addition, yohimbine has been shown to produce neuroendocrine changes and subjective mood states in humans which resemble anxiety. Thus, this study could provide important information related to hypotheses of drug abuse which involve psychiatric vulnerability factors.

Six subjects were given the opportunity to orally self-administer capsules containing placebo or triazolam (0.125 or 0.25 mg) on a signalled FI 10 min schedule of reinforcement in which a maximum of 18 capsules could be self-administered over a 3 hr period under double blind conditions. Forty-five min prior to self-administration sessions, subjects received placebo or yohimbine (7.5 - 60 mg) capsules administered as a pretreatment. The results showed: (1) evidence of yohimbine-induced increases in triazolam self-administration in all subjects; (2) yohimbine-induced increases in anxiety ratings in three of the six subjects.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00004-04 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)

Comparative Self-Administration (Monkeys and Human): Nicotine and Cocaine

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

J.E. Henningfield	Chief	BDL, ARC, NIDA
R. Nemeth-Coslett	Staff Fellow	BDL, ARC, NIDA
R.J. Lamb	Staff Fellow	BDL, ARC, NIDA
S.R. Goldberg	Chief	BPL, ARC, NIDA
C.W. Schindler	Staff Fellow	BPL, ARC, NIDA

COOPERATING UNITS (if any)

Behavioral Pharmacology Laboratory

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.12	0.07	0.05

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

This was a collaborative project with the BPL in which the human research was conducted on the Residential Research Unit and parallel animal studies were conducted in the BPL. The use of the self-administration (SA) study paradigm permited an assessment of the relative contribution of environmental and pharmacologic factors to the self-administration of drugs. Parallel comparative studies in squirrel monkeys and humans in which subjects are given the opportunity to self-administer comparable doses of cocaine and nicotine under similar behavioral schedules and experimental conditions also provide a means to assess the generality of biological variables influencing drug SA. This research has shown that responding is maintained in human subjects in the same manner in which it is maintained in non-human subjects. The stimuli that are associated with injections of cocaine develop conditioned reinforcing effects in the humans in a manner similar to the manner in which these effects develop in squirrel monkeys. These studies have also demonstrated that a research strategy employing drug SA in human subjects can yield all of the important information of single-dose studies, and also, provide information on the direct reinforcing effects of the compound which may be compared to the large base of animal drug SA. These data need only to undergo final analyses before publication.

PHS 6040 (Rev. 1/84)

GPO 914-918

Comparative Studies of Drug Self-Administration in Monkeys and Human Volunteers: Nicotine and Cocaine
ZO1 DA00004-01 BDL

Publications

Goldberg, S.R. and Henningfield, J.E.: Reinforcing effect of nicotine in humans and experimental animals responding under intermittent schedules of i.v. drug injection. Pharmacol Biochem Behav 30:227-234, 1988

Henningfield, J.E., Nemeth-Coslett, R., Katz, J.L. and Goldberg, S.R. Intravenous cocaine self-administration by human volunteers: Second-order schedules of reinforcement. In: Harris, L.S. (ed.): NIDA Research Monograph 76, Washington, D.C.: U.S. Government Printing Office, 1987, pp. 266-273.

Henningfield, J.E. and Goldberg, S.R. Pharmacological determinants of tobacco self-administration by humans. Pharmacol Biochem Behav 30:221-226, 1988.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00025-02 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Acquisition of Dependence to Cigarettes and Smokeless Tobacco

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R. Nemeth-Coslett	Staff Fellow	BDL, ARC, NIDA
E.J. Cone	Chief	CDM, ARC, NIDA
C.A. Haertzen	Psychologist	BDL, ARC, NIDA
F. R. Snyder	Statistician	CHP, ARC, NIDA
A. Radzius	Research Assistant	BDL, ARC, NIDA
K.O. Fagerstrom		Pharmacia, Sweden

COOPERATING UNITS *(if any)*

Dr. K.O. Fagerstrom (Pharmacia LEO Therapeutics AB; Helsingborg, Sweden)

LAB/BRANCH
Clinical Pharmacology Branch

SECTION
Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.50	0.25	0.25

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Questionnaires were given to populations of experienced cigarette and/or smokeless tobacco users (785 responses), and to a population which included persons who had never used tobacco (496 responses). The purpose of the questionnaires was to determine changes in the amount of tobacco products consumed as a function of time and to assess the level of nicotine dependence, as measured by the Fagerstrom Tolerance Questionnaire (FTQ). Findings that have emerged from initial analysis of the first population include the following: (1) Smokeless Tobacco use begins about one year earlier than cigarette use (15.5 vs 16.3); (2) Males begin smoking about one year earlier than females; (3) Tobacco consumption increased over time (i.e., dose graduation); (4) The dose escalation was negatively accelerated with no difference between sexes; (5) Age of starting smoking is negatively correlated with the age of quitting and also with predicted FTQ scores after the same number of years of smoking; (6) Four of 8 questions on FTQ scale are correlated with total FTQ score. Analyses in progress are: (1) Analysis of brands smoked; (2) Prediction of dependence based on the amount of tobacco product consumed at some earlier point in history; and, (3) Analysis of the data from the 496 response population. These data need only to undergo final analyses before publication.

GPO 914-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00014-02 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Cholinergic Agonists and Antagonists (Army Contract Related)

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
J.D. Roache	Staff Fellow	BDL, ARC, NIDA
R.I. Herning	Chief	CHP, ARC, NIDA
W.B. Pickworth	Pharmacologist	CHP, ARC, NIDA

COOPERATING UNITS *(if any)*
Research Support Branch
Cognitive Studies and Human Performance Laboratory

LAB/BRANCH
Clinical Pharmacology Branch

SECTION
Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.70	0.20	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Ten male volunteers without histories of drug abuse, except for cigarette smoking, were tested to assess the possible adverse performance effects of a cholinomimetic and a cholinergic antagonist, each given alone and in combination. A dose run-up procedure was employed in which physostigmine was administered i.v. in an ascending dose series (0.25, 0.5, 1.0, 1.5 and 2.0 mg) first alone, then following pretreatment with 5.0 or 10.0 mg of methscopolamine, a peripherally active antagonist. Methscopolamine was given to assess the degree to which peripheral blockade reduces physiological effects and/or performance impairment. The Army Performance Assessment Battery (PAB), including components of the Triservices PAB, was used to evaluate behavioral performance. Preliminary analyses are ongoing.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00012-05 BDL

PERIOD COVERED
January 1, 1989 to December 31, 1889

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Factors Influencing Behavioral and Physiologic Response to Opioids (Mu Project)

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

J.E. Henningfield	Chief	BDL, ARC, NIDA
S.T. Higgins	Staff Fellow	BDL, ARC, NIDA
E.J. Cone	Chief	CDM, ARC, NIDA
J.H. Jaffe	Former Director	ARC, NIDA

COOPERATING UNITS (if any)

Johns Hopkins University (K.L. Preston); Biology of Vulnerability Chemistry and Drug Metabolism

LAB/BRANCH
Clinical Pharmacology Branch

SECTION
Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.90	0.40	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

Following from the observations that post-addicts and non-opioid users are differentially sensitive to opioids, and perhaps even respond qualitatively differently, and the possibility that such differences either predispose certain persons to opioid abuse and/or contribute to relapse, this study was conducted to experimentally examine such population differences in response to mu and kappa opioids. Prominent measures included discrimination thresholds of behavioral effects, physiologic responses, and neuroendocrine response. Post-addict and opioid-naive subjects were intended to be separately tested for comparison. Testing is completed; however, upon the initial phase involving post-addict volunteers, changes in priorities resulted in the termination of the protocol before opioid-naive subjects were tested. Initial results suggest that a single dose of morphine is sufficient to measure a mild withdrawal-like effect when the opioid antagonist, naloxone, is subsequently administered.

Publication:
Higgins, S.T., Preston, K.L., Cone, E.J., Henningfield, J.E. and Jaffe, J.H.: Behavioral, Physiological, and Hormonal Effects of a Naloxone Challenge Following Acute Morphine Pretreatment in Humans. In Harris, L.S. (Ed.), NIDA Research Monograph 76. Washington, DC, U.S. Government Printing Office, 1987, pp. 266-273.

GPO 814-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00005-04 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Abuse Liability of Smokeless Tobacco Products

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R. Nemeth-Coslett	Staff Fellow	BDL, ARC, NIDA
A. Radzius	Research Assistant	BDL, ARC, NIDA
E.J. Cone	Chief	CDM, ARC, NIDA
N.L. Benowitz	Collaborator	U of California

COOPERATING UNITS *(if any)*

Chemistry and Drug Metabolism Laboratory
Division of Clinical Pharmacology and Experimental Therapeutics
University of California, San Francisco

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL	OTHER
0.37	0.07	0.30

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

In two studies, tobacco users were tested with a commercially available smokeless tobacco product (i.e., pouches of snuff), and with a smokeless cigarette through which air is sucked to inhale vaporized nicotine. Standardized methods of abuse liability assessment were used.

The smokeless tobacco study consisted of two phases. The first evaluated the effects of dose and the possibility that rate of expectoration would alter nicotin extraction and effects. Dose-related changes were found in the magnitude and duration of action of measures such as reduction in urge to smoke and strength of effects observed. The second phase evaluated the relationship of the effects observed to plasma levels of nicotine; these were found to be closely related to the dose administered, thus confirming the reliability of this system of nicotine delivery. The study with smokeless cigarettes indicated similar dose-related effects as those found with the commercial tobacco products; nicotine levels were negligible, suggesting the possibility that this route of nicotine administration may produce effects mediated by its peripheral stimulus properties which resemble those of smoking cigarettes.

Abuse Liability of Smokeless Tobacco Products
ZO1 DA00005-05 BDL

Publications

Henningfield, J.E. How tobacco produces drug dependence. In: J.K. Ockene (ed.), The Proceedings of the World Congress on the Pharmacologic Treatment of Tobacco Dependence. Cambridge, MA: Institute for the Study of Smoking Behavior and Policy, pp. 19-31, 1986.

Cullen, J.W., Blot, W., Henningfield, J.E., Boyd, G., Mecklenberg, R. and Massey, M.M. Health consequences of using smokelss tobacco: Summary of the Advisory Committee's Report to the Surgeon General. Public Health Reports 101:355-373, 1986.

Connolly, G.N., Winn, D.M., Hecht, S.S., Henningfield, J.E., Walker, B. and Hoffman, D. The re-emergence of smokeless tobacco. N Eng Med 314:1020-1027, 1986.

Glover, E.D., Schroeder, K.L., Henningfield, J.E., Winn, D.M., Severson, H.H. and Christen, A.G. A compendium of smokeless tobacco research. J of Drug Educ. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00011-04 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Physiologic Dependence to Tobacco: Cigarette Withdrawal and Nicotine Substitution

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R. Nemeth-Coslett	Staff Fellow	BDL, ARC, NIDA
R.I. Herning	Chief	CHP, ARC, NIDA
W.B. Pickworth	Pharmacologist	CHP, ARC, NIDA
W.R. Lange	Medical Officer	RSB, ARC, NIDA

COOPERATING UNITS *(if any)*

Cognitive Studies and Human Performance Laboratory, Research Support Branch

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER
.80	.55	.25

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Heavy tobacco users, otherwise without drug abuse histories, were studied on the Residential Research Unit. In the withdrawal study, subjects were assessed for nicotine and cotinine, general cardiovascular functioning, passive EEG and evoked cortical potential, and caloric intake, during 10 days of cigarette deprivation and when smoking resumed. In the substitution phase of the study, subjects were tested during alternating cycles of 4 days smoking and 3 days abstinence. In this phase, subjects were similarly assessed as described above, but on days in which they were not permitted to smoke, they were given pieces of gum to chew 12 times per day at one hour intervals: the gum contained either 0, 2 or 4 mg of nicotine. We found that an orderly withdrawal emerged during. It included impaired performance, which did not recover within the ten days of abstinence, but did recover when cigarette smoking resumed. Nicotine gum reversed major signs of tobacco withdrawal, confirming that the withdrawal was nicotine specific. This effect was dose-related, e.g., 4 mg gum restored performance to baseline levels, whereas 2 mg gum only partially restored performance. Placebo gum use was accompanied by withdrawal. Together, these results confirm that nicotine replacement can be a viable mode of alleviation of the tobacco withdrawal syndrome, but is of little benefit in reducing desire to smoke (which appears to be pharmacologically related to abstinence but appears readily elicited by environmental stimuli).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00008-03 BDL

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)

Behavioral Performance and Physiologic Effects of Drugs: Atropine and Diazepam (Army Contract Related)

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

J.E. Henningfield, Ph.D.	Chief	BDL, NIDA, ARC
R.J. Lamb, Ph.D. (Left ARC)	Staff Fellow	BDL, NIDA, ARC
S.T. Higgins, Ph.D. (Left ARC)	Staff Fellow	BDL, NIDA, ARC
R.I. Herning, Ph.D.	Chief	CHP, NIDA, ARC
W.B. Pickworth, Ph.D.	Pharmacologist	CHP, NIDA, ARC
F.R. Snyder, B.S.	Statistician	CHP, NIDA, ARC
W.R. Lange, M.D.	Medical Officer	RSB, NIDA, ARC

COOPERATING UNITS (if any)

Cognitive Studies and Human Performance Laboratory, Research Support Branch

LAB/BRANCH

Clinical Pharmacology Branch

SECTION

Biology of Dependence and Abuse Potential Assessment Laboratory

INSTITUTE AND LOCATION

National Institute on Drug Abuse, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
.10	5	5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

Studies were conducted to assess the effects of the drugs on performance at various tasks. An additional aspect of this research was to identify possible electrophysiological effects. Atropine and diazepam were found to produce dose-related effects on a variety of measures of subjective response as well as performance on the computerized Performance Assessment Battery (PAB).

These studies have been completed and data analyses are underway. Preliminary analysis of the results from the study on diazepam indicate that most measures were affected in an orderly time and dose-related manner. Most measures were surprisingly insensitive, however, and significant effects were often not seen until the administration of the highest dose of diazepam (40 mg). The Army developed measures did not appear to be more sensitive than traditional measures (e.g., DSST).

Publication:

Higgins, S.T., Woodward, B.M. and Henningfield, J.E. Effects of atropine on the repeated acquisition and performance of response sequences in humans. Journal of the Experimental Analysis of Behavior. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00035-01 BDL

PERIOD COVERED: January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*: Why do substance abusers seek help? What are their worries about that help?

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

J.E. Henningfield	Chief	BDL, ARC, NIDA
R.E. Johnson	Chief	RSB, ARC, NIDA
D. Brooke	Visiting Scientist	RSB, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH: Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION: Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
0.20	0.10	0.10

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

A survey of ARC research subjects was conducted to investigate the reasons that people seek treatment, and what their worries about that treatment are. We hope that the answers to these questions will enable us to make it easier for people to seek help. Subjects were asked to fill out two questionnaires and a cover sheet on their past experience of seeking help.

2. Chemistry and Drug Metabolism Laboratory - Edward J. Cone, Ph.D., Chief

Overview

The Laboratory of Chemistry and Drug Metabolism performs chemical, pharmacokinetic, metabolic and pharmacodynamic research with human subjects related to the chemistry of substance abuse. Presently, studies are underway to further delineate the pharmacokinetic and pharmacodynamic profile of marijuana, opiates and methamphetamine. The focus of these studies is the exploration of the relationship between drug levels in various body fluids to behavior, performance and physiological effects. An additional focus is the evaluation of the usefulness of unusual body fluids or tissues, e.g., saliva, nails and hair for drug detection. Our initial findings indicate that each body fluid or tissue provides a unique but slightly different historical record of drug exposure. It is important in the diagnosis, treatment and prevention of drug abuse that we have an understanding of the information provided by drug tests on various body fluids and the relationship of these tests to drug-induced effects. Also, the risk of unknowing drug exposure, i.e., "crack" smoke and methamphetamine smoke, is being evaluated. The Laboratory performs basic research in the area of chemical methodology development; new methods must be developed as new drugs appear in the illicit drug market and as new and important metabolites are identified in metabolic studies. In related studies, the validity of commercial test methods presently used in employment drug testing are being evaluated for precision and accuracy with clinical specimens collected under highly controlled conditions.

Summary of Ongoing Research

Specific research projects which were actively pursued in FY '89 are briefly summarized below. Only those studies for which personnel from this Laboratory were the principal investigators are discussed.

A. The Pharmacokinetics and Pharmacodynamics of Opiate Analgesics: Cone, E.J.; Collaborating Investigators: Mitchell, J. and Paul, B., Navy Screening Laboratory, Norfolk, VA.

The goals of this study include the following: evaluation of the usefulness of a heroin "marker" as a means of detecting heroin addicts in urine and saliva of subjects after heroin abuse; determine the relationship between plasma levels and saliva levels of active drug or metabolite and pharmacologic effects; use of saliva as a screening media for opiates; and validity assessment studies of commercial drug assays for opiates.

B. **Studies on the Validity of Drug Testing Methodology: Cone, E.J.; Collaborating Investigators: Mitchell J. and Paul B., Navy Screening Laboratory, Norfolk, VA.**

The goal of this study is to compare test results of commercial screening assays for drugs of abuse in urine with test results obtained by gas chromatography/mass spectrometry (GC/MS). Standard specimen sets utilized in these studies consist of clinical drug specimens collected under highly controlled conditions following drug administration and "spiked" standards at known concentration. A complete validity assessment of opiate assays was completed utilizing clinical drug specimens collected following heroin, morphine, codeine, hydromorphone, hydrocodone, buprenorphine, oxymorphone and oxycodone administration.

C. **Drug Assay Development Studies on Drugs of Abuse: Cone, E.J.**

The aim of this ongoing project is to develop specific, sensitive and reliable assays for drugs of abuse in a variety of biological media. For example, test methodology was developed for the detection of opiates in hair. This assay was used to study the appearance of morphine and codeine in human facial hair after controlled dosing. This assay provided the first documented evidence of the time period required for an administered opiate to appear in hair. Work also continues on the refinement of an assay for the simultaneous assay of cocaine and metabolites in body fluids. Another assay is currently under development for the determination of buprenorphine in blood, saliva and urine. Buprenorphine is a promising new drug for the treatment of opiate and cocaine addiction. Other assays also are developed for support of ongoing pharmacokinetic and pharmacodynamic studies.

D. **Buprenorphine Pharmacodynamics: Cone, E.J., Johnson, R.E. and Fudala, P.**

Buprenorphine is an opioid partial agonist which shows promise as a treatment agent for heroin and cocaine addiction. Although buprenorphine has limited bioavailability by the oral route of administration, it is effective by the sublingual route. Current studies are underway to determine its bioavailability by the sublingual and buccal route. Concurrent behavioral and physiological effects will be measured for correlation with blood levels. Urine will be tested for buprenorphine and metabolite content. The detection period for buprenorphine in urine and saliva will be determined. In addition, following chronic buprenorphine dosing, the blood levels of drug and metabolite will be determined in order to evaluate the importance of accumulation of drug and active metabolites.

E. Fast Action Dynamics of Marijuana Smoking: Huestis, M.A. and Cone, E.J.

The immediate effects of smoking marijuana on behavior and performance will be evaluated in this study. Behavioral and physiological measures will be collected before, during and after smoking. Blood and saliva samples will be collected concurrently and will be analyzed for tetrahydrocannabinol and metabolite content as well as selected hormones. The study is designed to evaluate the mechanistic and functional effects of smoking marijuana in human subjects. The study will have a unique focus on the early changes that occur in the physiology, behavior and the neuroendocrine system during the smoking of marijuana cigarettes.

F. Passive Inhalation of Drugs of Abuse: Cone, E.J.

When drugs of abuse are smoked, e.g., marijuana, cocaine, heroin, phencyclidine and methamphetamine, some of the volatile material is released into the atmosphere. Depending on the local environment, bystanders may be exposed to small doses of the drug and its pyrolyzed breakdown materials. Laboratory methods are being developed to experimentally simulate an atmosphere of drug smoke and means of withdrawing air samples for chemical analysis. These methods will be used to assess the potential hazards of passive inhalation of drugs of abuse.

G. Methamphetamine Pharmacodynamics: Cone, E.J.

Methamphetamine is a stimulant with effects similar to cocaine. Historically, it has a history of abuse both as a licit and illicit drug. Presently, there is concern that a new form of methamphetamine, "ice", may be abused in the same manner as the smokeable form of cocaine, i.e., "crack". Methods are under development to study the effects of this new form of methamphetamine. The pharmacokinetic profile, abuse liability and chemistry of "ice" will be evaluated.

Publications

Articles

Cone, E.J. , Kumor, K., Thompson, L.K.:Correlation of saliva cocaine levels with plasma levels and with pharmacologic effects after intravenous cocaine administration in human subjects. J. Anal. Toxicol., 12: 200-206, 1988.

Cone, E.J., Menchen, S.L., Paul, B.D., Mell, L.D. and Mitchell, J. Validity testing of commercial cocaine metabolite assays. I. Assay detection times, individual excretion patterns and kinetics after cocaine administration to humans. J. Forensic Sci., 34:15-31, 1989.

Cone, E.J. and Mitchell, J. Validity testing of commercial cocaine metabolite assays. II. Sensitivity, specificity, accuracy and confirmation by gas chromatography/mass spectrometry. J. Forensic Sci., 34:32-45, 1989.

Della Puppa, A. Ford-Rice, F., Snyder, F.R. Cone, E. and London, E.D. Time course of verapamil interaction with morphine effects on physiological parameters in rats. J. Pharm. Pharmacol., 41:617-623, 1988.

Higgins, S.T., Preston, K.L., Cone, E.J., Henningfield, J.E. and Jaffe, J.H. Supersensitivity to naloxone following acute morphine treatment in humans: Horomonal, physiological and behavioral effects., in press, JPET., 1990.

Cone, E.J. and Henningfield, J.E. Premier 'Smokeless Cigarettes' can be used to deliver crack. JAMA, 261:41, 1989.

Johnson, R.E., Cone, E.J., Henningfield, J.E. and Fudala, P.J. Use of buprenorphine in the treatment of opiate addiction. I: Physiologic and behavioral effects during a rapid dose induction. Clin. Pharmacol. Ther., 46:335-343, 1989.

Cone, E.J. Validity testing of commercial cocaine metabolite assays. III. Evaluation of an ELISA assay for detection of cocaine and cocaine metabolite. J. Forensic Sci., 34:991-995, 1989.

Radzius, A., Welch, P., Cone, E.J. and Henningfield, J.E. A portable pupilometer system for measuring pupillary dynamics. Behavior Research Methods, Instruments and Computers, 21:611-618, 1989.

Cone, E.J. Testing human hair for drugs of abuse. I. Individual dose and time profiles of morphine and codeine in plasma, saliva, urine and beard compared to drug-induced effects on pupils and behavior. J. Anal. Toxicol.
in press, 1990.

Pickworth, W.B., Welch, P., Henningfield, J.E. and Cone, E.J. Opiate-induced pupillary effects in humans. Methods and Findings in Clinical Pharmacology, in press, 1990.

Cone, E.J., Hair testing for drugs-Developments in an infant Science, Employment Testing. 3:BWR439-440, 1989.

Cone, E.J. and Huestis, M.A. Urinary excretion of commonly abused drugs following unconventional means of administration. Forensic Sci. Rev., in press, 1990.

Cone, E.J., Yousefnejad, D. and Dickerson, S.L., Validity testing of commercial urine cocaine metabolite assays. IV. Evaluation of the Emit^{R} d.a.u.^{TM} Cocaine Metabolite Assay in a quantitative mode for detection of cocaine metabolite. J. Forensic Sci., in press, 1990.

Weddington, W.W.k Brown, B.S., Haertzen, C.A., Cone, E.J., Dax, E.M., Herning, R.I. and Michaelson, B.S. Changes in mood, craving, and sleep during acute abstinence reported by male cocaine addicts: A controlled, residential study. Arch. Gen. Psychiatry., in press, 1990.

Lange, W.R., Cone, E.J. and Snyder, F.R. The association of hepatitis delta and hepatitis B virus in parental drug abusers, Arch. Intern. Med., in press, 1990.

Kuhar, M.J., Boja, J.W. and Cone E.J. Phencyclidine binding to striatal cocaine receptors, Life Science, in press, 1990.

Lange, W.R., Ball, J.C., Pfieffer, M.B., Snyder, F.R. and Cone, E.J. The Lexington Addicts, 1971-1972: Demographic characteristics, drug use patterns, and selected infectious disease experience. Intern. J. Addict. 24:609-626, 1989.

Cone, E.J. and Menchen, S.L. Stability of cocaine in saliva, Clin. Chem., 34:1508, 1988.

Cone, E.J. and Weddington, W.W., Jr. Prolonged occurrence of cocaine in human saliva and urine after chronic use. J. Anal. Toxicol. 13:65-68.

Chapters

Cone, E.J. and Dickerson, S.L. Analysis of human facial hair for morphine and codeine; Excretion patterns after single doses. In Proceedings of the International Association of Forensic Toxicologists, 1989, in press, 1990.

Cone, E.J. Darwin, W.D. and Dickerson, S.L. Prolonged Excretion of cocaine and metabolites in human urine after chronic use. In Proceedings of the International Association of Forensic Toxicologists, 1989, in press, 1990.

Cone, E.J., Yousefnejad, D., Darwin, W.D. and Menchen, S.L.: Detection of morphine and cocaine in human saliva by Coat-A-CountR radioimmunoassay, TIAFT 88 Proceedings, 1988, pp. 240-248.

Reviews

Cone, E.J. and Huestis, M.A. Urinary excretion of commonly abused drugs following unconventional means of drug administration. Forensic Sci. Rev., in press, 1990.

Abstracts

Yousefnejad, D. and Cone, E.J. Drug assay development. XXIV. Determination of cocaine in air by capillary gas chromatography/mass spectrometry. American Chemical Society Meeting, 24th MARM, May 23-25, 1990.

Yousefnejad, D., Darwin, W.D., Henningfield, J. and Cone, E.J. Drug assay development. XVIII. Determination of cocaine and phencyclidine (PCP) in the smoke of drug-loaded cigarettes by capillary gas chromatography/mass spectrometry (GC/MS) or by GC with a flame ionization detector (FID). ACS, 23th MARM, Cherry Hill, N.J., May 24-26, 1989.

Dickerson, S.L. and Cone, E.J. Drug assay development. XIX. Analysis of opiates in human facial hair. ACS, 23th MARM, Cherry Hill, N.J., May 24-26, 1989.

Darwin, W.D. and Cone, E.J. Drug assay development. XX. Solid phase extraction (SPE) of opiates from human biofluids. ACS, 23th MARM, Cherry Hill, N.J., May 24-26, 1989.

Cone, E.J. and Dickerson, S.L. Urine screening for cocaine use with an enzyme linked immunosorbent assay (ELISA) and confirmation with gas chromatography/mass spectrometry (GC/MS). AACC, Atlanta, GA, July 23-27, 1989.

Cone, E.J., Welch, P., Mitchell, J. and Paul, B. Time course of 6-acetylmorphine in urine after heroin administration. CPDD, Keystone, CO, June 18-22, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00006-03 CDM

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Pharmacokinetics and Pharmacodynamics of Opiate Analgesics

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	D. Darwin	Chemist	ARC, NIDA
	S. Dickerson	Lab Tech	ARC, NIDA
	B. Holicky	Nurse	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL	OTHER
1.25	0.25	1.0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The effects of single doses of intramuscularly administered opiates (heroin, morphine, dilaudid, codeine, oxycodone, oxymorphone) and sublingual buprenorphine are being studied in male human volunteers in order to determine the relationship of blood and saliva levels to pharmacologic effects. Additionally, the study is being performed to determine if a metabolic marker for heroin abuse can be found in urine.

The subjects are healthy males with a history of heroin abuse. Informed consent is is obtained and all procedures are approved by the hospital Institutional Review Board. A total of three test doses (placebo and two active doses) are administered in random order. Test measures are made for 24 hrs and biological fluids are collected for 7 days after each test. The biological fluids will be analyzed for drug and metabolites by chromatographic and immunoassay techniques.

The significance of this study lies in the potential value of saliva as a new test medium for detection of drugs of abuse and the characterization of the time course of excretion of metabolic markers for heroin abuse in urine and saliva.

PHS 6040 (Rev. 1/84)

GPO 914-918

Pharmacokinetics and Pharmacodynamics of Opiate Analgesics- FY - 1989
Z01 DA 00006-03 CDM
Periodicals:

Higgins, S.T., Preston, K.L., Cone, E.J., Henningfield, J.E. and Jaffe, J.H. Supersensitivity to naloxone following acute morphine treatment in humans: Horomonal, physiological and behavioral effects., in press, JPET., 1990.

Radzius, A., Welch, P., Cone, E.J. and Henningfield, J.E. A portable pupilometer system for measuring pupillary dynamics. Behavior Research Methods, Instruments and Computers, 21:611-618, 1989.

Pickworth, W.B., Welch, P., Henningfield, J.E. and Cone, E.J. Opiate-induced pupillary effects in humans. in press, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00002-04 CDM

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Validity Studies of Commercial Drug Screening Assays

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	D. Darwin	Chemist	ARC, NIDA
	D. Yousefnejad	Chemist	ARC, NIDA
	S. dickerson	Lab Tech	ARC, NIDA
	B. Holicky	Nurse	ARC, NIDA

COOPERATING UNITS *(if any)*

Naval Screening Laboratory, Norfolk, VA (J. Mitchell and B. Paul).

LAB/BRANCH

Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
1.125	0.125	1

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Commercial assays for the detection of drugs of abuse in urine change periodically and must be reevaluated for validity of detection. Studies are designed to test the validity of new assays on clinical specimens obtained from drug users under controlled conditions.

Healthy male volunteers with a history of chemical substance abuse participate in these studies. Informed consent is obtained and all procedures are approved by the hospital Institutional Review Board. Commercial assays for detection of drugs of abuse in urine are tested for validity with specimens collected under controlled dosing conditions. A variety of drugs of abuse are studied at various dose levels. The results of the assays are compared to GC/MS analyses.

These studies test the validity of commercial assays on clinical samples instead of "spiked" samples and provide unique and valuable information to the military and industry concerning their time course of detection, specificity and accuracy.

PHS 6040 (Rev 1/84)

GPO 914-918

Validity Studies of Commercial Drug Screening Assays - Publications
Z01- DA 0002-03 CDM

Periodicals

Cone, E.J., Menchen, S.L., Paul, B.D., Mell, L.D. and Mitchell, J.: Validity testing of commercial cocaine metabolite assays.: I. Assay detection times, individual excretion patterns and kinetics after cocaine administration to humans, J. Forensic Sci., 34:15-31, 1989.

Cone, E.J. and Mitchell, J.: Validity testing of commercial cocaine metabolite assays. II. Sensitivity, specificity, accuracy and confirmation by gas chromatography/mass spectrometry. J. Forensic Sci., 34:32-45, 1989.

Cone, E.J. Validity testing of commercial cocaine metabolite assays. III. Evaluation of an ELISA assay for detection of cocaine and cocaine metabolite. J. Forensic Sci., 34:991-995, 1989.

Cone, E.J., Yousefnejad, D. and Dickerson, S.L., Validity testing of commercial urine cocaine metabolite assays. IV. Evaluation of the Emit® d.a.u.™ Cocaine Metabolite Assay in a quantitative mode for detection of cocaine metabolite. J. Forensic Sci., in press, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00003-04 CDM

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)

Detection of Drugs of Abuse in Human Saliva

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	D. Darwin	Chemist	ARC, NIDA
	D. Yousefnejad	Chemist	ARC, NIDA
	S. Dickerson	Lab Tech	ARC, NIDA
	B. Holicky	Nurse	ARC, NIDA

COOPERATING UNITS (if any)

LAB/BRANCH

Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL	OTHER
1.25	0.25	1.0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The presence of drugs of abuse in saliva of human subjects after drug administration was studied to determine the feasibility of drug testing with saliva.

Healthy male subjects with a history of chemical substance abuse volunteered for the studies. Informed consent was obtained and all procedures were approved by the hospital Institutional Review Board. Following the administration of cocaine, marijuana or opiate, saliva and blood samples were collected periodically. Behavioral and physiological measures were made concurrently with collection of biofluids. Samples were analyzed by gas chromatography or RIA. Significant correlations of blood levels with saliva levels were found for cocaine and opiates. Investigations are continuing on marijuana.

These studies provide the scientific basis for development of new non-invasive tests for drug abuse.

Detection of Drugs of Abuse in Human Saliva, Publications - FY - 1989
Z01 DA 00003-03 CDM

Periodicals

Cone, E.J. and Menchen, S.L. Stability of cocaine in saliva, Clin. Chem., in press, 1988.

Cone, E.J., Kumor, K., Thompson, L.K. and Sherer, M.: Correlation of saliva cocaine levels with plasma levels and with pharmacologic effects after intravenous cocaine administration in human subjects. J. Anal. Toxicol. 12:200-206(1988).

Cone, E.J. and Weddington, W.W., Jr. Prolonged occurrence of cocaine in human saliva and urine after chronic use. J. Anal. Toxicol. 13:65-68.

Chapter

Cone, E.J., Yousefnejad, D., Darwin, W.D. and Menchen, S.L.: Detection of morphine and cocaine in human saliva by Coat-a-countr radioimmunoassay, TIAFT 88 Proceedings, 1988, pp. 240-248.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00007-02 CDM

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Pharmacokinetics and Pharmacodynamics of Drugs of Abuse in Hair

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	B. Holicky	Nurse	ARC, NIDA
	S. Dickerson	Lab Tech	ARC, NIDA
	D. Darwin	Chemist	ARC, NIDA
	D. Yousefnjad	Chemist	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
0.75	0.125	0.625

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Drug residues have been detected in human hair specimens by a variety of analytical techniques. These reports have generated substantial interest in using hair as a historical record of drug usage. Studies are designed to determine the presence and time course of drugs of abuse in human hair.

Healthy male volunteers with a history of chemical substance abuse will participate in the study. Informed consent will be obtained and all procedures will be approved by the hospital Institutional Review Board. Subjects will reside on the clinical ward of the ARC. Head and facial hair specimens will be obtained prior to and after administration of drugs of abuse. Blood, saliva and urine specimens also will be obtained. Analyses of tissue and biofluids for drug will be performed by radioimmunoassay and gas chromatography/mass spectrometry.

The studies will provide the scientific basis for determining the usefulness of hair as a "historical record" for substance abuse.

Pharmacokinetics and Pharmacodynamics of Drugs of Abuse in Hair- FY - 1989
Z01 DA 00007-02 CDM

Periodicals

Cone, E.J. Testing human hair for drugs of abuse. I. Individual dose and time profiles of morphine and codeine in plasma, saliva, urine and beard compared to drug-induced effects on pupils and behavior. J. Anal. Toxicol.
in press, 1990.

Cone, E.J., Hair testing for drugs-Develpoments in an infant Science, Employment Testing. 3:BWR439-440, 1989.

Chapter:

Cone, E.J. and Dickerson, S.L. Analysis of human facial hair for morphine and codeine; Excretion patterns after single doses. In Proceedings of the International Association of Forensic Toxicologists, 1989, in press, 1990.

Abstract:

Dickerson, S.L. and Cone, E.J. Drug assay development. XIX. Analysis of opiates in human facial hair. ACS, 23th MARM, Cherry Hill, N.J., May 24-26, 1989 (Abstract).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00009-01 CDM

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Fast Action Dynamics of Marijuana

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	B. Holicky	Nurse	ARC, NIDA
	S. Dickerson	Lab Tech	ARC, NIDA
	D. Darwin	Chemist	ARC, NIDA
	D. Yousefnjad	Chemist	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL.	OTHER:
0.75	0.125	0.625

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Although early changes which occur during the smoking of marijuana are more likely to be indicative of its mode of action, this phase of the use of marijuana has largely been ignored. This study will detail the effects of smoking marijuana cigarettes on a variety of systems including physiologic effects, behavior and hormonal systems. In addition, blood and saliva levels will be determined during and after smoking. Blood and saliva levels will be compared to drug-induced effects and hormonal changes.

This study will provide the most comprehensive assessment of marijuana's effects that occur both during and after smoking and should provide important insight to the mode of action of this widely abused drug.

PHS 6040 (Rev. 1/84) GPO 914-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00010-01 CDM

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Methodological Assessment of the Risk of Passive Inhalation of Drugs of Abuse

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	B. Holicky	Nurse	ARC, NIDA
	S. Dickerson	Lab Tech	ARC, NIDA
	D. Darwin	Chemist	ARC, NIDA
	D. Yousefnjad	Chemist	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.75	0.125	0.625

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

When drugs of abuse are smoked, volatile components and pyrolysis material escape into the atmosphere. Depending on the local environment, bystanders may be exposed to the drug by passive inhalation of the contaminated air. Present studies are underway to develop means of heating drugs of abuse in a controlled environment and measuring air levels of drug in order to evaluate this potential hazard. Initially, free-base cocaine "crack" and methamphetamine "ice" will be evaluated for potential passive inhalation exposure.

Unknowing drug exposure could be dangerous to unsuspecting bystanders, particularly to small children. These studies will establish limits of exposure to volatilized drugs under controlled conditions.

PHS 6040 (Rev. 1/84)

GPO 914-9

Methodological Assessment of the Risk of Passive Inhalation of Drugs of Abuse
FY - 1989
Z01 DA 00010-01 CDM

Abstract

Yousefnejad, D. and Cone, E.J. Drug assay development. XXIV. Determination of cocaine in air by capillary gas chromatography/mass spectormetry. American Chemical Society Meeting, 24th MARM, May 23-25, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00008-01 CDM

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Pharmacokinetics and Pharmacodynamics of Methamphetamine

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	E.J. Cone	Chief	CDM, ARC, NIDA
Others	B. Holicky	Nurse	ARC, NIDA
	S. Dickerson	Lab Tech	ARC, NIDA
	D. Darwin	Chemist	ARC, NIDA
	D. Yousefnjad	Chemist	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH
Laboratory of Chemistry and Drug Metabolism, Clinical Pharmacology Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.75	0.125	0.625

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Methamphetamine is an amphetamine-like stimulant with substantial abuse liability by the intravenous route. Recently, a new form of methamphetamine ("ice") has been reported to be abused via the smoking route. This study will evaluate various chemical forms of methamphetamine in human volunteer subjects. Informed consent will be obtained and all procedures will be approved by the hospital Institutional Review Board. Methamphetamine will be administered by the smoking and intravenous routes. The pharmacokinetic profile will be determined by analysis of blood samples. The bioavailability and abuse liability of the smoked drug will be obtained by comparison to the intravenous route. Urine, saliva, and hair specimens will be collected for drug detection studies on methamphetamine.

These studies will evaluate for the first time, the abuse potential of smoked methamphetamine and will allow drug detection methods to be developed for this new form of methamphetamine.

3. Neuroendocrinology/Immunology Laboratory - Elizabeth M. Dax, M.B., B.S., M.D., Ph.D., Chief

Introduction

The neuroendocrinology/immunology laboratory is investigating mechanisms by which substances of abuse, particularly cocaine, act to alter two of the body's homeostatic mechanisms; the endocrine and immune systems. By utilizing neuroendocrine hormone secretion as CNS markers the effects of physiological and pharmacological manipulations on the CNS may be followed in clinical and basic research settings. The neuroendocrine hormones (prolactin, growth hormone, adrenocorticotrophic hormone, thyroid stimulating hormone, leutinizing hormone) are regulated by releasing factors and neurotransmitters released from the hypothalamus. Principal neurotransmitters are dopamine, serotonin and norepinephrine; each of these may be perturbed by specific drugs. Other regulators include peptide releasing factors, feed back loops, steroid hormones and influences from higher CNS centers. Manipulation of these perturbators or regulators and subsequent measurement of the release of neurohormones may be used to dissect mechanisms of drug actions on the hypothalamo-pituitary system axis. The relevance of the hypothalamo pituitary responses to CNS mechanisms may be established in animal models. Therefore, by examining patterns of secretion and secretory responses to specific manipulations by peptide regulators or drugs with known actions on neurotransmitters, the mechanisms of actions substances of abuse, the altered physiology consequent to their actions and possibly mechanisms of addiction may be investigated. Furthermore, hormones interact with the immune system. Examination of temporal alterations in endocrine function simultaneously with immune function may help to explain altered immune function in substance abusers and mechanisms of action of substances of abuse on the immune system. The more rapid progress to AIDS and death in substance abusers with HIV infection when compared with other groups of HIV infected individuals may, thus, be explained.

Overview

Studies on the role of substances of abuse in perturbing CNS function are being carried out. Neurohormonal secretion is being characterized as a marker of CNS actions. Our initial studies center on the mechanisms by which cocaine disrupts dopamine-mediated secretion of hormones. We are examining hormones with known circadian periodicities in men withdrawing from cocaine and have shown that dopamine-mediated hormonal secretion is altered for up to 3 weeks. The human studies are supported by investigations into the CNS mechanisms and validation of hormones as CNS markers, by producing similar alterations in endocrine function in rats by the administration of cocaine in specific regimens. Secretion of neurohormones is being monitored, during

different regimens of cocaine administration, and subsequently receptor distribution and neurotransmitter content of the CNS being quantitated. To examine secretion of neurotransmitters and hormones from hypothalami and pituitaries without the influence of higher centers, they are superfused alone and in tandem. Hormones and neurotransmitters are measured in the superfusates. Secretion from dispersed anterior pituitary cells also is being examined. Thus, we are disecting the levels of control at which these drugs act on dopaminergic and serotonergic neurosecretory systems with different regimens of drug administration.

Substance abusers constitute a large proportion of those people who have been exposed to the Human Immunodeficiency Virus (HIV) or have contracted the disease of AIDS - Acquired ImmunoDeficiency Syndrome. It is this group of those infected with HIV who will be largely responsible for the continued spread of the virus into the heterosexual community. Furthermore, substances of abuse are known to perturb immune function and therefore may be important cofactors in the development of AIDS in those exposed to HIV. The Laboratory of Neuroendocrinology and Immunology evolved in part from the "AIDS Laboratory" which was established to investigate the prevalence of HIV in substance abusers and the effects of substances of abuse on the immune system. The AIDS laboratory has formed part of NIDA's effort to curb AIDS in the substance-abusing community by carrying out the laboratory component of a multi-city survey of HIV antibody prevalence in known addicts.

The NEI laboratory is carrying out multidisciplinary studies of the effects of substances on clinical immune function in lymphocytes. These studies have been carried out in collaboration with the Immunology Section of the Gerontology Research Center (William Adler, M.D., Chief). The effects of cannabinoids and cocaine on these clinical parameters is being carried out. Volatile nitrites are abused primarily by homosexual men and it is these men who are the only group of AIDS sufferers with a high prevalence of Kaposi's sarcoma. Thus, it is suspected that the nitrites are responsible for a disturbance in immune function which facilitates the development of this disease. Immune function in a group of ARC volunteers has been shown to be depressed in response to the administration of amyl nitrites. This type of study has been extended to examine immune function in volunteers using other substances of abuse. Studies on the effects of Δ^9Tetrahydrocannabinoid (THC) and cocaine on the immune function of volunteers have been commenced. Future studies will assess immune function with the drugs in combination because they are frequently used together. Alterations in prolactin secretion has been observed in both cocaine and heavy THC users. Prolactin has been postulated to maintain the integrity of the immune system. Therefore, the possible role of hormones, particularly prolactin, in the interactions between substances of abuse and altered immune function are being carried out. More detailed mechanistic studies are being carried out in rats.

In addition to its research activities, the NEI laboratory is responsible for carrying out the tests for establishing the sero-status of individuals at risk for exposure to HIV. The laboratory is supporting the NIDA national study of sero-prevalence in substance abusers. Studies originating at ARC have been carried out and further studies examining the HIV status of high risk individuals are in progress.

Monoclonal antibodies against new treatment drugs and substances of abuse that do not hold immediate commercial value, are being raised.

The neuroendocrine laboratory is carrying out the urine toxicology services for the ARC. This has allowed the laboratory to adapt drug assays for measurement of drug levels in plasma and tissue samples as well as urine.

The Goals of the Laboratory are:

a) to investigate the disturbances of neuroendocrine secretion caused by substances of abuse, in order to investigate their mechanisms of action, the altered physiological consequences of substance abuse, and possibly the mechanisms of perpetuation of a substance abuse habit.
b) continue research into basic mechanisms of neuroendocrine secretion.
c) to investigate the possible interactions between substances of abuse and neurohormones and their relationship to altered immune function. Further, the hypothesis that substances of abuse as immunodepressors, may be co-factors in the development of AIDS in HIV infected people will be tested.
d) determine the HIV antibody status of ARC volunteers, research subjects and addicts in the NIDA HIV-antibody prevalence study.
e) to provide quantitation of drug concentrations in body fluids and tissue extracts.

Summary of Ongoing Research

A. Changes in Neurosecretion caused by Substances of Abuse: Dax, E.M., Pilotte, N.S., Contoreggi, C.S., Partilla, J.S., Fishbein, D., Ulrichsen, J., Weddington, W.W., Lange, W.R.

The purpose of these investigations is to define neuroendocrine changes in response to administration of drugs in order to determine mechanisms of action, physiological consequences of using substances of abuse and possibly mechanisms of addiction.

Cocaine may act primarily on dopamine uptake sites to inhibit dopamine reuptake, at the same time directly stimulate dopamine release. Thus, in chronic administration paradigms, cocaine may

cause dopamine depletion. It may have similar effects on serotonergic systems. We have followed neuroendocrine secretion in men acutely withdrawn from cocaine (Project number Z01 DA 00007-03 NEI). By measuring prolactin and cortisol at 2 hr. intervals over 24 hr. up to the 21st day after cocaine cessation, it has been shown that the hormone, prolactin, which is under tonic inhibitory control by dopamine, is higher in those men than non-cocaine abusers. This finding is consistant with the hypothesis that chronic cocaine use results in depletion of hypothalamic dopamine. Cortisol levels (controlled by ACTH with input from serotonergic mechanisms) was not different from controls. Further, the diurnal variation of prolactin secretion was not present in those men, up to 21 days after cocaine cessation. The results show that the altered neuroendocrine effects of cocaine may persist for at least 21 days after withdrawal. Men withdrawing from cocaine are being subjected to provocative endocrine tests to establish whether control mechanisms are hypersensitive at the level of the hypothalamus or at anterior pituitary cells. Future studies will examine the sensitivity of the hypothalamus to medications which perturb the neurotransmitters, dopamine and serotonin. The aims of these studies is to decipher mechanisms of cocaine's action and to provide objective assessment parameters in treatment strategies.

The mechanisms of the neuroendocrine alterations human studies are being followed up by studies in rats. (Project number Z01 DA 00008-03 NEI). The neuroendocrinology of administering cocaine in different regimens is being examined in order to investigate the progression of changes that occurs as cocaine is administered in increasing amounts and to investigate how long these alterations persist when cocaine is withdrawn. Neurotransmitter content and receptor parameters are being quantified. A regime of cocaine administration in rats and that mimics the endocrine changes seen in humans, has been established as a model of chronic cocaine abuse. Alterations in appropriate CNS receptors are being examined in this model. In the same rats cardiac manifestations of this administration regime will be investigated, since cardiac complications of cocaine are the most common cause of acute deaths in cocaine users.

Since there are many influences on the hypothalamus of the intact brain, rat studies where hypothalami, anterior pituitary glands or hypothalami and pituitaries in tandem, are perifused, will examine direct effects of cocaine (and other substances of abuse) on the hypothalamus or anterior pituitary. This will be examined in tissues naive to the drugs and those from rats which have been exposed to cocaine and other substances of abuse <u>in vivo</u>. Again, the release of neurohormones and neurotransmitters will be measured. Using reverse hemolytic plaque assays, secretion from single anterior pituitary cells has been quantitated (Project number Z01 DA 00014-01 NEI). Cocaine has no direct effect on prolactin secretion in anterior pituitary cells from rats.

However, the dynamics of anterior pituitary cell responses to dopamine are altered markedly by cocaine treatment. We will investigate the possibility of examining secretion of releasing factors from hypothalamic cells, by the same method. To examine release of neurotransmitters in rats in vivo, microdialysis techniques are being established in the laboratory.

Alterations in neuroendocrine secretion has also been examined in men taking Δ^9tetrahydrocannabinol (THC) (Project number Z01 DA 00006-03 NEI). This substance has been postulated to have steroid-like properties. Therefore THC may modify neuroendocrine hormone secretion because steroids are important regulators of the endocrine system. By studying endocrine and immune effects simultaneously, some important information on endocrine-immune system interrelationships may be elucidated. Particularly, the role of THC in altering immune function via alterations in neuroendocrine modulation is being investigated. Some of these studies will be extended in rats to further investigate mechanisms of the action of THC. Receptor mechanisms will be investigated. Particular attention will be paid to the possible role of THC in altering the modulating immune function by prolactin because we have found alterations in prolactin secretion by THC.

It has been established that in ARC volunteers, divided into groups according to aggression and impulsivity scales, that endocrine responses are attenuated in the more aggressive/impulsive men to a challenge by a drug with predominantly serontonergic effects (Project number Z01 DA 00013-02 NEI). These studies are being extended by examing responses to a more specific serotonergic drug, metachlorophenylpiperazine (mCPP). Protocols to administer mCPP in more chronic regimes are approved. We will examine the treatment possibilities of mCPP in aggressive/impulsive behavior and concommitantly measure neuroendocrine parameters in order to explain the mechanisms of the drug's actions. Again, studies in rats will be used to investigate the mechanism of action of this potential therapeutic agent (Project number Z01 DA 00018-01 NEI).

B. Mechanisms of Neuroendocrine Secretion: Dax, E.M., Pilotte, N.S.

The above-mentioned studies all include investigations to extend our understanding of basic mechanisms of neurosecretion. Findings that are being explored further, are the role of individual hypothalamic neurotransmitters in mediating diurnal rhythms. In the men withdrawing from cocaine we saw a disruption of the prolactin diurnal rhythm (Project number Z01 DA 00007-03 NEI) while cortisol diurnal rhythms remained intact. We have subsequently examined ACTH rhythms in these men. In rat studies receptor studies are being carried out to investigate the neurotransmitter changes which may account for the differential effects of cocaine on these secretory systems.

Hormones and neurotransmitters are sequestered in 2 or more intracellular pools. The significance of these pools in cell function is unknown (Project number Z01 DA 00009-03 NEI). In order to examine release from the intracellular pools, investigations using direct visualization of the secretion of prolactin from single anterior pituitary cells by use of the reverse hemolytic plaque assay (RHPA), will be carried out. Other experiments will examine secretion of newly synthesized hormone by combining the RHPA with autoradiographic techniques.

C. **Neuroendocrine-Immune System Interactions: Dax, E.M., Adler, W.H., Nagel, J.E., Partilla, J.S., Dersch, C., Lange, W.R.**

There is extensive evidence that substances of abuse induce changes in immune function. Few studies have been carried out where the drug administration and the immune function of lymphocytes have been temporally related. A study of immune function in lymphocytes of men taking volatile nitrites (the use of which has been linked with the development of Kaposi's sarcoma in AIDS), has been carried out. During the period when the volunteers were exposed to amyl nitrite immune function alterations occurred. Depressed natural killer cell activity and T-lymphocyte cell function were observed (Project number Z01 DA 00004-03 NEI). A similar protocol studying the effects of Δ^9THC on lymphocyte function has been carried out (Project number Z01 DA 00006-03 NEI). However, the THC study also includes detailed endocrinological studies, in order to examine any temporal relationships between the alterations in neuroendocrine and immune function.

The hypothesis that substances of abuse may be cofactors in the development of AIDS in HIV infected people is being tested. Data from men with documented substance abuse and medical histories, and whose HIV status is known are being collected. The relationship between HIV status and clinical status in relationship to substance abuse will be correlated.

D. **HIV - Status of Risk Groups: Dax, E.M., Lange, W.R.**

The HIV serology laboratory processes approximately 200 samples per week. A major portion of these samples are supporting the multicity survey being conducted by NIDA, in which the HIV status in substance abusers is being determined. The origin of the HIV virus in its present form of efficacy and ability to produce the immunodeficiency syndrome, is unknown. We have examined several thousand plasmas from missionaries travelling between Africa and other countries and the U.S. between 1968 and 1983. Although this group of people is not high risk, they are a group with a high

level of casual contact with people in whom the virus is endemic. Selected plasmas have been examined for the presence of related retrovirus (Project number Z01 DA 00012-02 NEI). None of these plasmas was found to be positive for HIV antibodies by Western Blot despite about 200 being ELISA positive. The ELISA positive samples and twice the number of as closely matched controls as possible, were further screened.

In 1129 plasmas from people admitted to Lexington, KY, between 1971 and 1972, 29 were found to be ELISA positive (Project number Z01 DA 00002-03 NEI, in 1987 ARC Annual Report). However, follow up revealed no excess mortality compared with matched controls among the seronegative group. It is of interest that unexpectedly high proportions of these groups were ELISA positive.

E. Measurements of Drugs and Their Metabolites in Body Fluids and Tissues: Dax, E.M., Pilotte, N.S., Partilla, J.S.

The urine toxicology radioimmunoassays are being adapted to quantify drug concentrations in other body fluids besides urine and in tissue extracts. The ability to quantify drugs in tissues will be of value when drug effects are being determined because tissue and plasma concentrations as well as doses may be related to responses.

Publications

Dax, E.M., Lange, W.R. and Jaffe, J.H. (1989) Allergic reactions to amyl nitrite inhalation. American J. Medicine, 86, 732.

Lange, W.R., Dax, E.M., Kovacs, R., Kreider, S., and Frame, J. (1989) Are missionaries at risk of AIDS? Southern Medical Journal, 82, 1075-1078.

Dax, E.M. (1989) Endocrine alterations associated with the abuse of cocaine. T.E.M.S., 1, 55-56.

Dax, E.M., Pilotte, N.S., Adler, W.H., Nagel, J.E., Lange. W.R. (1990) The effects of 9-ene-tetrahydrocannabinol on hormone release and immune function. J. Steroid Biochem., 34, 263-268.

Dax, E.M., Ingram, D.I., Partilla, J.S. and Gregerman, R.I. (1989) Food restriction prevents an age-associated increase in liver beta-adrenergic receptors. J. Gerontology, 44, B72-B76.

Buckenmeyer, P.J., Goldfarb, A.H., Partilla. J.S., Piñeyro, M.A., Dax, E.M. (1990) Beta-adrenergic receptor;adenylate cyclase complex in rat skeletal muscle: responses to endurance training and acute exercise. Am. J. Physiology: Endocrinol. Metab., 258, E71-E77.

Pilotte, N.S., Sharpe, L.G., and Dax, E.M. Multiple, but not acute, infusions of cocaine alter the release of prolactin in male rats. Brain Res. In press.

Lange, W.R., Fudala, P.J., Dax, E.M., Johnson, R.E. Safety and side-effects of buprenorphine in the clinical management of heroin addiction. J. Alcohol and Drug Abuse. In press.

Dax, E.M., Adler, W.H, Nagel, J.E., Lange, W.R. and Jaffe, J.H. Inhalation of volatile nitrites induces changes in *in vitro* immune function. In press.

Litow, R., Robinson, N., Herning, R., Jaffe, J.H. and Dax, E.M. Cognitive function and EEG changes with the inhalation of amyl-nitrite. In press.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Cone, E.J., Dax, E.M., Herning, R.I., Michaelson, B.S. Changes in mood, craving, and sleep reported by male cocaine addicts during acute abstinence: A controlled, residential study. In press.

Fudala, P.J., Jaffe, J.H., Dax, E.M., Johnson, R.E. Use of buprenorphine in the treatment of opioid addiction II: Physiological and behavioral effects of daily and alternate day administration and abrupt withdrawal. In press.

Dax, E.M. Drug dependence in the differential diagnosis of allergic respiratory disease. Annal Allergy. In press.

Dax, E.M., Partilla, J.S., Piñeyro, M.A., and Gregerman, R.I. Hormone receptor:adenylyl cyclase interactions are affected by dietary fatty acid manipulations: Effects in rat liver. In press.

Robinson, N., Lange, W.R., Dax, E.M. EKG alterations following Δ^9tetrahydrocannabinol. (Submitted).

Lange, W.R., Ball, J.C., Dax, E.M. et al. A follow-up study of a parent HIV seropositivity among parenteral drug abusers in 1971-72. (Submitted).

Dax, E.M. (1989) Mechanisms of altered target cell response. In Endocrinology, Hormones and Aging. Timiras, P.S. and Quay, W.B. (Eds) Prentice Hall, NY. (Submitted).

Abstracts

Pilotte, N.S., Johnson, R.L., and Dax, E.M. (1989) Chronic cocaine in vivo modifies prolactin release in the presence of dopamine in vitro. Proceedings 19th Annual Society for Neuroscience Meetings, Toronto, Canada.

Dax, E.M., Adler, W.H., Nagel, J.E., Lange, W.R., and Jaffe, J.H. (1989) The effects of Δ^9tetrahydrocannabinol on immune function and hormone release. 9th International Symposium of the Journal of Steroid Biochemistry, Las Palmas, Canary Islands, Spain.

Lange, W.R., Dax, E.M., Kovacs, R., Kreider, S., and Frame, J. (1989) Are missionaries at risk of AIDS? V International Conference on AIDS, Montreal, Canada.

Dax, E.M. (1989) Adrenergic Receptor Systems in Aging. 42nd Annual Scientific Meeting of the Gerontological Society of America, Minneapolis, Minnesota.

Dax, E.M., Pilotte, N.S., Sharpe, L.G. and Weddington, W.W. (1989) Withdrawal from chronic cocaine alters neuroendocrine function in humans and rats. Johns Hopkins Hospital Department of Medicine Symposium.

Dax, E.M., Weddington, W.W., Pilotte, N.S. and Jaffe, J.H. (1989) Changes in hypothalamo-pituitary-adrenal axis hormone release following abrupt cocaine cessation. 71st Annual Meeting of the Endocrine Society, Seattle, Washington.

Dax, E.M., Pilotte, N.S., Adler, W.H., Nagel, J.E. and Lange, W.R. (1989) Neuroendocrine immune effects of short term and long term intake of Δ^9tetrahydrocannabinol. Drugs of Abuse, Immunity and Immunodeficiency, Tampa, Florida.

Dax, E.M., Pilotte, N.S., Adler, W.H., Nagel, J.E. and Lange, W.R. (1990) Short-term Δ^9tetrahydrocannabinol (THC) does not affect neuroendocrine or immune parameters. CPDD 52nd Annual Scientific Meeting, Richmond, Virginia.

Newlin, D.B., Pretorius, M.B., Wong, C. and Dax, E.M. Acute marijuana smoking reduces vagal tone. (Submitted).

Pilotte, N.S., Mitchell, W.M., Sharpe, L.G., DeSouza, E.B. and Dax, E.M. Chronic cocaine administration and withdrawal from cocaine modify central neurotensin receptors in rats. (Submitted).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00004-03 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Inhalable Nitrites - Immune Function and Abuse Potential

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI,ARC,NIDA
Others:	J.H. Jaffe		NIDA
	W.R. Lange	Medical Director	ARC, NIDA
	R. Herning	Laboratory Chief	CHP, ARC, NIDA
	R.M. Litow	Research Technologist	NEI, ARC, NIDA
	N. Robinson	Registered Nurse	NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

W.H. Adler	Clin. Immunology Section	GRC, NIA
J.A. Nagel	Clin. Immunology Section	GRC, NIA

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1	0.25	0.75

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The intake and frequency of inhalation of volatile nitrites has been associated with the incidence of Kaposi's sarcoma in people suffering from AIDS. Animal and in vitro lymphocyte studies have shown that immune cell function can be altered by these agents. However, no study has related directly the effects of nitrites administered in vivo to disturbances of immune function in humans. Thus, a study has been conducted in healthy, HIV negative volunteers. An inhlation protocol in which the subject inhaled 3 doses of amyl nitrite for 3 days and 1 dose on the fourth day has been conducted. In an extended protocol a second group of volunteers were administered subsequent, single inhalations of nitrite 3-4 days apart, to a total of 13 inhalations over 3 weeks. A battery of immune function tests in the subjects' lyphocytes, was carried out on 2 occasions prior to the inhalation protocol, immediately following the last dose, and at 1, 4, and 7 days after the last dose. Results showed a decrease in natural killer cell activity, the lymphocyte function reputedly responsible for tumor cell scavenging. The single doses of nitrite administered at 3-4 day intervals continued to suppress this activity. Lymphocyte numbers and subsets were not altered during the inhalation protocols, but showed a non-specific rise on cessation of the drug. Discrepencies between mitogen stimulated [^{3}H]thymidine incorporation, a measure of the activity potential of lymphocytes, and antibody production by the T lymphocyte-dependent, B-cells indicated a deficit in T-cell function during nitrite exposure.

The nitrites were demonstrated to have minimal abuse potential.

Inhalable Nitrites - Immune Function and Abuse Potential
Z01 DA00004-03

Publications

Litow, R., Robinson, N., Herning, R., Jaffe, J.H. and Dax, E.M. Cognit function and EEG changes with the inhalation of amyl-nitri Psychopharmacology. In press.

Dax, E.M., Lange, W.R. and Jaffe, J.H. (1989) Allergic reactions to amyl nitr inhalation. American J. Medicine 86, 732.

Dax, E.M., Adler, W.H, Nagel, J.E., Lange, W.R. and Jaffe, J.H. Inhalation volatile nitrites induces changes in in vitro immune function. Clin. and E Immunol. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00005-03 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

HIV Prevalence: In Depth Survey of Baltimore

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
Others:	W.R. Lange	Medical Director	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.0	0.25	0.75

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The seroprevalence of HIV antibodies in surveyed intravenous substance users (IVSUs) who were either recently enrolled into treatment or were on a waiting list for enrollment was 29%. The rate among ARC research subjects with parenteral drug use histories has averaged 24%, and among area prostitutes with heavy drug use histories, 34%. In Baltimore, 94% of IVSUs had shared needles, and even though HIV seropositivity was not assoicated with a needle-sharing history, there was an association between the intensity of sharing and the probability of being seropositive. A much stronger association was observed between seropositivity and "shooting gallery" visitation, suggesting that this milieu of sharing, rather than other environments, is the real risk factor.

Very distinct ethnic group differences in HIV infection were observed, with Blacks being much more likely to be seropositive than Whites (odds ratio = 8.18, 95% CI 3.35-19.97). There was no significant difference in HIV infection between Blacks in Baltimore and in New York City. Shooting gallery visitation appears to be much more a phenomenon among Black IVSUs than it is in White (X^2 = 8.23, $p<0.01$). HIV infection has appreciably penetrated Baltimore's addict community. The overall seroprevalence rate in Baltimore in 1986 (29%) approximated that of New York in 1979 (27%) where the rate subsequently jumped to 58% in some areas by 1984 and has increased to 60% in 1987.

Hepatitis antigen and antibody status of these subjects has been assessed. There was no concordance of Hepatitis B infection and HIV infection. Other data concerning Hetatitis D is being analysed.

The second wave of this study is being initiated.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00006-03 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Cannabinoids and Their Effects on the Immune System and Cognitive Function

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
Others:	W.R. Lange	Medical Director	ARC, NIDA
	N.S. Pilotte	Staff Fellow	NEI, ARC, NIDA
	R.M. Litow	Research Technologist	NEI, ARC, NIDA
	J.S. Partilla	Chemist	NEI, ARC, NIDA
	N. Robinson	Registered Nurse	NEI, ARC, NIDA
	J.R. Mahaffy	Registered Nurse	NEI, ARC, NIDA

COOPERATING UNITS *(If any)*

W.H. Adler	Clin. Immunology Section	GRC, NIA
J.A. Nagel	Clin. Immunology Section	GRC, NIA

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2	0.5	1.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Delta-9-tetrahydrocannabinol (THC) has been hypothesized to influence immune function. However, this has not been investigated in a comprehensive fashion in humans. The purpose of this study is to measure and study the effects of THC on immune function. To investigate immune-endocrine correlations, hormone parameters defining the activity of the hypothalamo-pituitary-adrenal axis have been measured during THC administration. (The effects of THC on cognitive function will also be investigated.) Experienced THC users have been recruited for study. Immune function of lymphocytes in vitro has been investigated during orally administered and inhaled THC and during the washout phase.

Cannabinoids and Their Effects on the Immune System and Cognitive Function
Z01 DA00006-03

Publications

Dax, E.M., Pilotte, N.S., Adler, W.H., Nagel, J.E., Lange. W.R. The effects of 9-ene-tetrahydrocannabinol on hormone release and immune function. J. Steroid Biochem. In press.

Dax, E.M., Adler, W.H., Nagel, J.E., Lange, W.R., and Jaffe, J.H. (1989) The effects of Δ^9tetrahydrocannabinol on immune function and hormone release. 9th International Symposium of the Journal of Steroid Biochemistry, Las Palmas, Canary Islands, Spain.

Dax, E.M., Pilotte, N.S., Adler, W.H., Nagel, J.E. and Lange, W.R. (1989) Neuroendocrine immune effects of short term and long term intake of Δ^9tetrahydrocannabinol. Drugs of Abuse, Immunity and Immunodeficiency, Tampa, Florida.

Dax, E.M., Pilotte, N.S., Adler, W.H., Nagel, J.E. and Lange, W.R. Short-term Δ^9tetrahydrocannabinol (THC) does not affect neuroendocrine or immune parameters. (Submitted).

Newlin, D.B., Pretorius, M.B., Wong, C. and Dax, E.M. Acute marijuana smoking reduces vagal tone. (Submitted).

Robinson, N., Lange, W.R., Dax, E.M. EKG alterations following Δ^9tetrahydrocannabinol. (Submitted).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00007-03 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Neuroendocrine Secretion During Cocaine Withdrawal

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	Elizabeth Dax, M.D. Ph.D.	Laboratory Chief	NEI, ARC, NIDA
Others:	W. Weddington, M.D.		TEI NIDA
	Nancy Pilotte, Ph.D.	Staff Fellow	NEI, ARC, NIDA
	Edrich Anderson, R.N.	Registered Nurse	NEI, ARC, NIDA
	Teri Gendron	Research Technologist	NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3	1	2

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Cocaine withdrawal (and withdrawal from other drugs) is associated with CNS disturbances which are reflected in altered neurohormonal secretion, secondary to CNS neurotransmitter alterations. In men known to be cocaine abusers, the secretion of neurohormones has been examined during cocaine withdrawal. Prolactin secretion is under tonic inhibition by dopamine from the hypothalamus. The neurotransmitter most closely associated with cortisol release is serotonin. In the volunteers withdrawn from cocaine, prolactin levels were higher than in men who had not ever taken cocaine, and the diurnal rhythms in prolactin secretion were disturbed. The men were followed for up to 21 days with little change in the profiles of prolactin release. Cortisol levels and rhythms were similar to controls over this withdrawal period. These results suggest that chronic cocaine abuse results in dysfunction of dopamine mediated mechanisms of neurosecretion.

The alterations in the hypothalamo-pituitary-adrenal axis resulting from chronic cocaine abuse, is being further defined. This will enable study in volunteers whose serotonergic and dopaminergic functions are predictably manipulated, with tests that perturb the hypothalamic-pituitary-adrenal axis at a known level. Standard endocrine diagnostic tests (TRH, CRF stimulation and L-dopa suppression) in conjunction with drugs that perturb dopaminergic and serotonergic function have been carried out. The study will provide further information on dopaminergic control of hormonal secretion and its role in maintaining diurnal rhythms of hormones. It may provide an important means of assessing the efficacy of treatment protocols. Mechanisms of these changes is being investigated in a rat model of cocaine withdrawal.

Neuroendocrine Secretion During Cocaine Withdrawal
Z01 DA00007-03

Publications

Dax, E.M., Weddington, W.W., Pilotte, N.S. and Jaffe, J.H. (1989) Changes in hypothalamo-pituitary-adrenal axis hormone release following abrupt cocaine cessation. 71st Annual Meeting of the Endocrine Society, Seattle, Washington.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Cone, E.J., Dax, E.M., Herning, R.I., Michaelson, B.S. Changes in mood, craving, and sleep reported by male cocaine addicts during acute abstinence: A controlled, residential study. Biol. Psychiat. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00008-03 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

The Effects of Cocaine on Hormone Secretion from the Anterior Pituitary

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
	N.S. Pilotte	Staff Fellow	NEI, ARC, NIDA
Others:	C.S. Contoreggi	Staff Fellow	NEI, ARC, NIDA
	L.G. Sharpe	Research Psychologist	BVL, ARC, NIDA
	J.S. Partilla	Research Chemist	NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2	1	1

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The physiologic effects of cocaine, at least in part, are dopamine mediated. Prolactin, an anterior pituitary hormone, is regulated primarily by dopamine which is released from a discrete population of neurons within the medial basal hypothalamus. Thus, prolactin release is an indirect measure of dopamine release.

Secretion mechanisms will be examined during different cocaine administration regimens, using several techniques now established in this laboratory. Pituitary hormones measured in the blood of catheterized rats have demonstrated that adrenocorticotrophin (ACTH) is released in response to acute cocaine administration. Direct secretion of dopamine has been examined in the hypothalamo-portal blood of live, anesthetized animals, before and after acute cocaine treatment and, subsequently, in rats treated with cocaine for varying periods of time. In isolated pituitaries and hypothalami, perifused alone or in tandem, the output of dopamine and neuropeptides are being examined concomittantly with the release of prolactin in order to examine release which is not under the influence of higher centers. Neurotransmitter release will be measured by microdialysis where microdialysis probes are inserted stereotoxically through fixed cannulae which have been previously inserted under anesthesia. Thus, secretion is being studied in particular brain areas of unanesthetized rats to examine neurosecretion in vivo. Finally, prolactin release will be examined in dispersed anterior pituitary cells to assess cocaine's effects on secretion at the level of anterior pituitary cell.

Thus, the role of the dopaminergic system in acute and chronic administration of cocaine of assessing the use of prolactin as an accurate marker for dopaminergic function in cocaine users is being assessed.

The Effects of Cocaine on Hormone Secretion from the Anterior Pituitary
Z01 DA00008-03

Publication

Pilotte, N.S., Sharpe, L. G. and Dax, E.M. Multiple, but not acute, infusions of cocaine alter the release of prolactin in male rats. Brain Res. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00009-03 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Mobilization of Pools of Peptide Hormone as a Function of Drug Environment

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: N.S. Pilotte Staff Fellow NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.0	1.0	1.0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

One physiological consequence of the administration of drugs of abuse such as cocaine or opiates or amphetamine is increased release of prolactin from the anterior pituitary gland. Prolactin, like other hormones and neurotransmitters, appears to be sequestered in 2 or more intracellular pools. The release of hormone from each pool is governed normally by the actions of releasing or release-inhibiting factors. However, the administration of cocaine, opiates, or amphetamine may alter the secretion of prolactin by a direct action on the lactotrope (prolactin-secreting cell). These agents may modify the ability of the cell to produce and release new hormone or may affect the release of the older, stored pool. These possibilities will be assessed by visualizing directly the secretion of prolactin from single cells challenged with the drugs in combination with known secretagogues through the use of a reverse hemolytic plaque assay. This technique will be modified to permit the production of radioisotopically-labelled new hormone. The secretion and sequestration of the labelled hormone will be monitored by utilizing the combination of hemolytic plaque formation with autoradiography.

It is expected that these experiments, using the lactotrope as a model system, will permit the identification of the possible mechanism(s) by which drugs of abuse alter the endocrine regulatory systems and provide insight into similar processes which occur in neuronal systems.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00010-02 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Development of Monoclonal Antibodies to Drugs and Hormones

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
Others:	C. Dersh	Chemist	NEI, ARC, NIDA
	R. Zaczek	Staff Fellow	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER:
2	.25	1.75

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Usually antibodies to treatment drugs, substances of abuse or hormones are not commercially available unless there is wide market. Treatment drugs, such as buprenorphine, are unlikely to have antibodies developed until the efficacy of the drug is established. Therefore, this laboratory is developing antibodies to buprenorphine in order to establish a radioimmunoassay. Mice have been immunized with buprenorphine and demonstrate the presence of antibodies on initial tests. Spleen cells will be harvested and fused to myeloma cells to produce cell lines. Testing for production of antibodies in the cell line will be carried out and any clones producing highly specific and high affinity antibodies will be isolated. Buprenorphine antibodies will be used to establish a radioimmunoassay so that the drug may be quantitated in urine, plasma and tissue extracts. Other drugs that antibodies may be raised against include amphetamine and metamphetamine, cocaine and cocaine metabolites, and metachloropheylpiperazine (mCPP). Antibodies are also presently being raised against vasopressin and rat prolactin, for which commercially available antibodies are limited.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00011-02 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Neuroendocrine Correlates of HIV Infection and the Development of ARC and AIDS

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
	W.R. Lange	Medical Director	ARC, NIDA

COOPERATING UNITS *(If any)*

Lawrence Brown, M.D., M.P.H., Vice President for Research and Medical Affairs, Addiction Research and Treatment Inc., Brooklyn, N.Y.

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1	0.5	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The human immunodeficiency virus infects the central nervous system resulting in a wide range of neurological deficits. One problem in management of HIV-infected people is predicting the disease's prognosis and course. The control of the neuroendocrine system and many feed back loops both endocrine and immunological are a property of the CNS, particularly of the hypothalamus. Several studies have shown disruption of neuroendocrine function. In a large group of drug abusers the neuroendocrine/endocrine status will be correlated with HIV status, clinical history, drug history, and presence of opportunistic infections. To date, clinical data from 800 patients have been collected. Their HIV antibody status and hormonal measurements are being assessed.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00012-02 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

HIV Sero-status in Missionaries From Africa, 1968-1983

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	W.R. Lange	Medical Director	ARC, NIDA
	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1	0.5	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The origins and timing of HIV spread into the U.S. remain in question. One possibility is that alterations in the virus' genetic makeup led to its current pathogenic properties, but a similar virus may have been transported from Africa where HIV is endemic. We have screened approximately 6000 plasmas from missionaries travelling between Africa and the U.S. between 1968 and 1983. Although the group may be considered low risk for sexually transmitted diseases, they are a group with high casual contact with the African people. Approximately 200 plasmas were found to be positive on ELISA screening but none was found to have HIV specific proteins detected by Western Blot. Further analysis of the HIV proteins are being carried out. Selected plasmas are being screened for related virus, including HTLV1.

HIV Sero-status in Missionaries From Africa, 1968-1983
Z01 DA00012-02

Publications

Lange, W.R., Dax, E.M., Kovacs, R., Kreider, S., and Frame, J. (1989) Are missionaries at risk of AIDS? Southern Medical Journal 82, 1075-1078.

Lange, W.R., Dax, E.M., Kovacs, R., Kreider, S., and Frame, J. (1989) Are missionaries at risk of AIDS? V International Conference on AIDS, Montreal, Canada.

Lange, W.R., Ball, J.C., Dax, E.M. et al. A follow-up study of a parent HIV seropositivity among parenteral drug abusers in 1971-72. (Submitted).

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00013-02 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Neuroendocrine Correlates of Aggressive/Impulsive Behavior

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
Others:	C.S. Contoreggi	Staff Fellow	NEI, ARC, NIDA
	D. Fishbein	Guest Scientist	ARC, NIDA
	J.H. Jaffe		NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA, Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
4.0	1.5	2.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

When men grouped according to their aggressive/impulsive scores on standard psychological tests, are challenged with a serotonergic stimulator, such as fenfluramine, the neuroendocrine response is attenuated in the more aggressive, more impulsive men, suggestive of alterations in central serotonergic mechanisms. Further, results suggest that hostility ratings decrease with fenfluramine administration suggesting possible treatment rationales for the study. Aggression and impusivity may be important personality characteristics in initiating and perpetuating addictive behavior. In order to investigate mechanisms of this behavior, establish neuroendocrine markers, suggest treatment possibilities and assess the efficacy of treatment paradigms, we are extending these studies with the more specific serotonergic agonist, metachlorophenylpiperazine (mCPP). We are examining whether mCPP administration gives similar results to fenfluramine. Subsequent studies will examine serotonergic as well as dopaminergic secretion in greater detail. Using neurohormones as markers of these responses, secretion will be examined in the presence of either a serotonergic or dopaminergic agonist (mCPP or bromocryptine, respectively). Neuroendocrine provocation tests will be used to further define alterations of function in the aggressive men.

Publications

Fishbein, D.H., Lozovsky, D., and Jaffe, J.H. (1989) Impulsivity, aggression, and neuroendocrine responses to serotonergic stimulatin in substance abusers. Biological Psychiatry, 25:1049-1066.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00014-01 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

The effects of cocaine on prolactin secretion from single cells of the anterior pituitary gland

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	N.S. Pilotte	Staff Fellow	NEI, ARC, NIDA
Others:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA/Addiction Research Center P.O. Box 5180 Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1	.5	.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

In a recently completed study, we found that in rats that received programmed infusions of 1 mg/kg cocaine every 12 min for 2 hr over 10 days, the pre-infusion concentrations of prolactin (PRL) increased in a time-dependent manner whereas post-infusion levels of PRL were decreased by cocaine. These changes could involve modification of adenohypophysial dopamine D2 receptors. Dispersed anterior pituitary cells were obtained from rats treated chronically with cocaine or saline and were subjected to a reverse hemolytic plaque assay that permitted direct visualization of PRL release from single lactotropes. The cells were incubated with media, cocaine, thyrotropin releasing hormone (TRH), or dopamine (DA) in vitro. There were 4 major findings. 1) Basal PRL release is greater in rats treated with cocaine: more cells secreted PRL and the individual cells secreted more PRL. 2) Cocaine in vitro did not affect PRL release. 3) TRH stimulated PRL release similarly from lactotropes of cocaine- or saline-treated rats. 4) DA in vitro inhibited PRL release dose-dependently from both cocaine- and saline-treated rats when the concentration of DA met or exceeded that observed in hpothalamo-hypophysial portal blood. However, lactotropes from cocaine-treated rats were more sensitive to the inhibition by DA. Paradoxically, very low concentrations of DA (<10-9M) enhanced PRL release from cells from cocaine-treated rats. These data confirm the findings of other that DA-deprived lactotropes release more PRL when challenged with low concentrations of DA and suggest that one consequence of chronic use of cocaine is a diminished release of DA in the absence of cocaine.

The Effects of Cocaine on Prolactin Secretion from Single Cells of the Anterior Pituitary Gland
Z01 DA00014-01

Publications

Pilotte, NS, Johnson, RL, Dax, EM. Chronic cocaine *in vivo* modifies prolactin release in the presence of dopamine *in vitro*. Presented at 19th Ann. Mtg. of Soc. for Neuroscience, Phoenix, AZ, Abst. #322.9.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00015-01 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

The effects of cocaine on dopamine release from hypothalamic neurons.

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	N.S. Pilotte	Staff Fellow	NEI, ARC, NIDA
Others:	L.G. Sharpe	Research Psychologist	ARC, NIDA
	I.M. Mefford	Special Expert	CP, NIMH
	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA

COOPERATING UNITS *(If any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA/Addiction Research Center P.O. Box 5180 Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL.	OTHER.
1	.75	.25

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

In a recently completed study, we found that in rats that received programmed infusions of 1 mg/kg cocaine every 12 min for 2 hr over 10 days, the pre-infusion concentrations of prolactin (PRL) increased in a time-dependent manner whereas post-infusion levels of PRL were decreased by cocaine. Because doapmine (DA) and PRL are reciproaclly related in male rats, these changes could involve modification of the release of DA from hypothalamic tuberoinfundibular neurons. We tested this hypothesis in rats treated as described above for 9 days with cocaine or saline. On the 10th day, the hypothalamo-hypophysial portal blood for 30 min before the initiation of passive infusions of cocaine or saline, during 60 min of intermittent infusion, and for 30 min following a challenge of amphetamine. Arterial blood was collected concurrently. These aliquots are currently being assayed in Dr. Mefford's laboratory using microbore high performance liquid chromatography. If there are differences between cocaine- and saline-treated animals, another series will be performed with lidocaine as the infusate as a control for the local anesthetic effects of cocaine. Neurochemical correlates of sensitization of the tuberoinfundibular DA neurons will be assessed as well by treating another group of rats with cocaine or saline for 10 days followed by a 10-day withdrawal period. Animals will be surgically prepared on the 11th day after cocaine cessation and portal blood will be collected as described above. Together, these experiments will provide the first evidence of cocaine-induced modifications of functional DA release coupled to a physiological relevant event, the release of PRL, and can serve as a model of the action of cocaine on other central DA systems.

The Effects of Cocaine on dopamine Release from Hypothalamic Neurons
Z01 DA00015-01

Publications

Pilotte, N.S., Sharpe, L.G., and Dax, E.M. Multiple, but not acute, infusions of cocaine later the release of prolactin in male rats. Brain Res. In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00016-01 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Effects of cocaine and withdrawal from cocaine on central receptors for peptides, catecholamines, and catecholamine uptake markers.

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	N.S. Pilotte	Staff Fellow	NEI, ARC, NIDA
Others:	W.M. Mitchell	Lab Manager	NBL, ARC, NIDA
	L.G. Sharpe	Research Psychologist	PNP, ARC, NIDA
	E.B. de Souza	Laboratory Chief	NBL, ARC, NIDA
	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA

COOPERATING UNITS *(If any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA/Addiction Research Center P.O. Box 5180 Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
3	1.5	1.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Cocaine is thought to produce many of its effects through an interaction with dopaminergic neuronal systems. Cocaine's neurochemical effects may include modifications in the numbers of receptors or uptake sites for dopamine (DA) or other regulatory peptides colocalized with DA, such as neurotensin (NT). If such changes occur, it is not known if they are permanent. Thus, we treated rats with programmed infusions of isotonic saline or 1 mg/kg cocaine every 12 min for 2 hr over 10 days and killed them within 15 min of the last infusion. Other rats were treated identically, but were killed 10 days later. Brains were removed and immediately frozen. Ten micron sections were taken through areas known to contain DA perikarya or terminals and binding experiments were conducted on the slices to determine the loci and number of binding sites for NT, mazindol, and DA receptors of the D1 and D2 classes. Additional sections were taken for anlaysis of binding sites for paroxetine, corticotropin releasing hormone, and the mu, kappa and delta opiate ligands. Analysis for NT sites is complete at this time. We found that cocaine reduces the number of NT binding sites in ventral tegmental area, substantia nigra and pars lateralis and that this observed reduction is reversed 10 days later.

Effects of Cocaine and Withdrawal from Cocaine on Central Receptors for Peptides, Catecholamines, and Catecholamine Uptake Markers
Z01 DA00016-01

Publications

Pilotte, NS, Mitchell, WM, Sharpe, LG, de Souza, EB, Dax, EM. Cocaine-induced reduction in neurotensin binding in midbrain is reversed during withdrawal from cocaine. Submitted to CPDD, June, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00017-01 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Cardiac effects of I.V. cocaine administration as measured by radionucleotide scanning, echocardiography and holter monitoring.

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
	W.R. Lange	Medical Officer	ARC, NIDA
	N. Chandra	Cardiologist	FSKMC
Others:	C.S. Contoreggi	Assistant Medical Officer	ARC, NIDA
	J. Fralich	Physician's Assistant	ARC, NIDA
	F. Levin	Staff Fellow	ARC, NIDA

COOPERATING UNITS *(If any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA/Addiction Research Center P.O. Box 5180 Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
2	1.5	.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Cocaine use is associated with sudden death which is often due to cardiac complications. The mechanism of cocaine's effect on cardiac function is not understood. In healthy volunteers who use cocaine, cardiac function will be monitored in the absence and presence of cocaine by holter monitoring, echocardiography and radionucleotide (Thallium) scanning. In addition, physiological, neuroendocrinological and peripheral nervous system data will be obtained.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA00018-01 NEI

PERIOD COVERED

January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

The effect of chronically administered metachlorophenylpiperazine on rat brain receptors, neurotransmitters and neuroendocrine hormone secretion.

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.M. Dax	Laboratory Chief	NEI, ARC, NIDA
Others:	J. Ulrichsen	Foreign Fellow	NEI, ARC, NIDA
	N.S. Pilotte	Staff Fellow	NEI, ARC, NIDA
	J.S. Partilla	Research Chemist	NEI, ARC, NIDA
	T. Richardson	Lab Technician	NEI, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

NEI

SECTION

Clinical Pharmacology Branch

INSTITUTE AND LOCATION

NIDA/Addiction Research Center P.O. Box 5180 Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2	1	1

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Metachlorophenylpiperazine (mCPP) is a serotonergic agonist/antagonist which may be useful in treatment of disorders such as depression and aggression which are associated with substance abuse. mCPP may have therapeutic value for cocaine abuse. Although neuroendocrine and other responses to acute treatment with mCPP have been studied, no studies of alterations with chronic administration have been made. We will conduct these studies in rats chronically treated with mCPP. Appropriate receptors and neurotransmitters along with neuroendocrine responses and behavioral parameters will be quantitated. The interaction of mCPP with cocaine effects will be investigated.

Etiology Branch

David B. Newlin, Ph.D., Acting Chief

Introduction

The Etiology Branch was formed in the 1989 fiscal year with scientists and staff from the Psychology of Vulnerability and Cognitive and Electrophysiology Laboratories. The Etiology Branch has at the present time one laboratory, the Vulnerability Laboratory. The domain of this branch is the study of causes and models of drug abuse and relapse, as well as residual effects of abused drugs. This research is conducted with individuals at risk for drug abuse, established drug users, drug addicts in periods of withdrawal from drugs, and normal individuals. The Etiology Branch has maintained close research collaborations with other branches, including Neuroscience, Preclinical Pharmacology, and Clinical Pharmacology.

The primary goals of this Branch are to determine psychobiological mechanisms involved in risk for drug abuse, develop theoretical models of addictive behavior and relapse to drug abuse, and study residual drug effects. Much research was carried out in support of these objectives during the 1989 Fiscal Year, which is summarized below.

Vulnerability Laboratory - David B. Newlin, Ph.D., Acting Chief

Overview

The Vulnerability Laboratory has conducted several investigations concerning the etiology of drug abuse. More importantly, this research has been guided by theoretical models developed at the ARC which dictate specific empirical studies.

One such model concerns cardiovascular responses to abused drugs. This model is based on the observation that heavily abused drugs such as cocaine, marijuana, alcohol, amphetamine, inhalants, and nicotine produce relatively robust increases in heart rate when self-administered. This observation can be theoretically linked to the psychomotor stimulant theory of addiction, which proposes a special status for psychostimulant effects of abused drugs. The heart rate responses may provide a general measure of the psychomotor stimulant properties of abused drugs, and may therefore have direct relevance to mechanisms of reward and abuse liability. These studies have been conducted in collaboration with the Clinical Pharmacology and Neuroscience Branches.

Therefore, studies have been conducted in which cardiovascular responses, including heart rate, blood pressure, and vagal tone (a noninvasive measure of parasympathetic influences on the heart) have been measured to a variety of drugs of abuse. Initial results indicate that several different abused drugs, including alcohol, marijuana, cocaine, and morphine tend to increase heart rate and decrease vagal tone. This is preliminary evidence in favor of the theoretical model. Further theoretical and empirical work is anticipated in this area.

Other theoretical models concerning the addictive process have focused on Pavlovian conditioning mechanisms of drug tolerance and dependence, as well as gene neuroscience applications to the problems of peptide regulation in morphine tolerance and withdrawal. It was proposed that conditioning mechanisms may be better understood with consideration of the excitatory vs. inhibitory nature of specific drug effects. Excitatory drug effects tend to exhibit conditioned sensitization, and inhibitory drug effects tend to show conditioned tolerance. A review of the Pavlovian drug conditioning literature also found that the conditioned response to abused drugs was always excitatory, except for 24 hr cues for these drugs. These observations were also related conceptually to the psychomotor stimulant theory of addiction (Wise & Bozarth, 1987) and to reward mechanisms of these drugs.

In collaboration with Dr. Uhl of the Neuroscience Branch, studies have been conducted concerning Dr. Uhl's model of addiction based on gene regulation of peptide production. Outpatients were given analgesia tests before and after longterm blockade of the opiate receptors. Preliminary results support Dr. Uhl's suggestion that gene regulation may provide a "memory store" for drug history, although alternative explanations of the results must be ruled out by further testing. Further studies will be conducted in this area.

Other research on mechanisms of the addictive process focus on "priming" of cocaine craving by alcohol intoxication. The concept of drug craving has received a great deal of attention recently, but is still difficult to define and measure. The Etiology Branch has been actively involved in research on drug craving, and will continue to support such research. Studies in the Etiology Branch use both self-report (on a joystick device) and physiological measures of autonomic and cortical function as possible correlates of craving processes. Craving is induced by tapes developed at the ARC in which individuals self-administer drugs, and by drug paraphernalia that are shown to the subjects. The purpose of this research is to determine whether concurrent use of alcohol may enhance cocaine craving in cocaine abusers who are also heavy users of alcohol. Preliminary results indicate that alcohol intoxication does increase cocaine craving.

Studies begun in previous years were continued during this year. We focused our resources on a study which monitors psychological, neurophysiological and cognitive alterations in cocaine abusers as they abstained from cocaine on our inpatient ward. Other collaborative

studies include (1) the electrophysiologic effects of cocaine and drugs which might block the central nervous effects of cocaine, (2) the relationship between the electrophysiologic and subjective effects of cocaine and morphine and (3) the effects of cholinergic agents on the behavioral and neurophysiologic indices of cognitive information processing.

Topographic mapping of multichannel EEG and evoked potential measures were expanded to include brain electrical mapping during complex cognitive tasks. These color maps of electrical activity from the human brain during resting and cognitive processing will aid in localization of drug effects in the brain. These new technical advances will also aid in the quantification of drug-produced alterations in cognition and performance, the characterization of cognitive and performance deficits observed during drug withdrawal, the evaluation of sensory and cognitive information processing abilities in populations at risk for drug abuse, and the investigation of drug effects on the brain electrical activity as both a correlate and as a probe to delineate drug-related activity.

Summary of Ongoing Research

A. Alcohol-Induced Craving: Newlin, D.B., Pretorius, M.B., and Wong, C.

This study concerned the effect of alcohol and intoxication on craving for cocaine. In addition to high and low doses of alcohol, subjects receive both placebo and water administrations as control conditions for the effect of alcohol; this will allow determination of the effect of placebo on craving for cocaine. Subjective report of craving is measured on a joystick that provides a continuous report throughout the experiment. Autonomic and electrocortical responses to cocaine stimuli and a videotape of an individual self-administering cocaine are used to evoke craving responses on both the rising and falling blood alcohol limbs. It is anticipated that this research will provide important information concerning the pharmacological and psychological mechanisms that encourage cocaine self-administration, and provide a model for relapse following periods of cocaine abstinence.

B. Cardiovascular Response to Drugs: Newlin, D.B., Pretorius, M.B., and Wong, C.

These studies concern cardiovascular and subjective responses to a number of different abused drugs, including oral alcohol, chewed nicotine gum, i.v. nicotine, i.v. cocaine, i.v. morphine, oral amphetamine, oral phenobarbitol, and oral hydromorphone. Preliminary results indicate that these drugs, other than phenobarbitol, increase heart rate and decrease vagal tone. Thus, these preliminary results support the theoretical linkage between brain reward mechanisms and vagal influences on the heart. This research has implications for abuse liability, and has been conducted in collaboration with the Clinical Pharmacology and Neuroscience Branches.

C. **Review of Drug Conditioning: Newlin, D.B.**

This research summarizes the available literature on the conditioning mechanisms that are theoretically related to drug craving and reward. It was felt that better understanding of the animal literature would lead to more effective clinical research on drug craving and relapse. This research underscores the importance of considering the excitatory vs. inhibitory nature of response systems in the analysis of conditioning mechanisms. It also emphasizes commonalities in conditioned responses across drugs of abuse.

D. **Gene Regulation in Humans: Uhl, G., Newlin, D.B., Pretorius, M.B., Park, J., and Cone, E.**

This research, in collaboration with the Neuroscience and Clinical Pharmacology Branches, is in the process of providing evidence for exogenous control of the regulation of endogenous opiate genes in humans. Normal individuals given an opiate antagonist for a week show greater antinociceptive effects after the antagonist leaves the system than individuals given placebo. This research is currently being replicated and extended to include a condition to rule out receptor up-regulation as a possible mechanism for the effect.

E. **Calcium Channel Blocker/Cocaine Studies: Herning, R.and Lange, R.**

Earlier studies found cocaine increased EEG beta, blood pressure, pulse and subjective feelings of "rush." Pretreatment by an appropriate blocking drug might either block all of cocaine effects or selectively alter the time course and magnitude of some of these effects. However, in either case the craving for the drug might be reduced and cocaine's mechanism of action clarified.

Single doses of 10 mg nifedipine, a calcium channel blocker was used in an attempt to antagonize the effects of cocaine. The EEG beta increase, which is observed with cocaine, appears to be blocked by a single dose of nifedipine. The results of this study will be of theoretical and practical significance.

F. **Cognitive Neurophysiologic Signs of Cocaine Abstinance: Herning, R. I., Dax, E.M., Levin, F., and Glover, B.J.**

Neurophysiologic signs and cognitive performance have not been studied during cocaine withdrawal. Cognitive deficits are being studied in patients withdrawing from cocaine. The present study evaluates cognitive information processing in subjects on a clinical ward withdrawing from cocaine with a battery of cognitive tasks. Sleep quality and duration is monitored by a subjective

questionnaire. Twenty subjects including controls have been tested in this study over a one-month withdrawal period. Stimulus evaluation and memory deficits were observed in the cocaine addicts. The memory deficits persisted for over three weeks. Additional subjects are being tested on a more extensive electrophysiological battery for six to eight weeks to determine whether the deficits observed in the original study will persist past three weeks. Clarification of the nature of the cognitive deficits and of sleep loss will lead to more effective treatment strategies for cocaine abuse.

G. **Mapping the Effects of Cocaine by EEG: Herning, R.I., Glover, B.J., London, E. and Stapleton, J.**

The effects of cocaine on scalp EEG are being studied to determine the brain areas involved in the cocaine-induced euphoria. In previous studies, cocaine increased EEG beta power. The distribution of cortical areas responsible for the EEG beta increase and the time course of the beta increase have not as yet been determined. The present study was designed to answer these two questions.

H. **Vulnerability Studies: Herning, R.I., Fishbein, D. and Dax, E.M.**

Many factors appear to be important in the etiology of drug abuse. Both antisocial behavior and early aggression are risk factors for later drug use. Both sensory and cognitive information processing deficits were observed in aggressive adolescents and adults. Data from two studies were published during the current year. Direct measures of aggression were more strongly related to electrophysiological measures of sensory and cognitive impairment than the DSM-IIIR diagnosis of antisocial behavior.

In another study, electrophysiological markers of impulsivity and aggression were modulated by serotinergic agents. Thus far the data from 46 subjects have been analyzed. Psychometric measures of aggression were related to different electrophysiological indices than were measures of impulsitivity. A serotininergic agent altered these relationships in a complex manner.

I. **Acute Abstinance from Tobacco: Electrophysiological and Cognitive Signs: Herning, R.I., Henningfield, J.E. and Glover, B.J.**

The EEG, cognitive, and cognitive process was monitored during a ten-day period of tobacco withdrawal in heavy smokers. Some of the changes persisted over the entire ten-day deprivation period. These measures included EEG alpha frequency, theta power, performance on selected cognitive tasks (especially a rapid arithmetic task) and a cognitive event related potential measure (N100 amplitude). Stimulus

evaluation time, as measured by P300 latency, and the depth of stimulus evaluation battery were affected early during the tobacco deprivation period, but returned to smoking levels later during the deprivation period. Thus, the cognitive deficits are clearly apparent during abstinance from tobacco and contribute to relapse during treatment. The treatment of these cognitive deficits with nicotine gum is also being studied.

J. **Mapping the Effects of Opioid Agonists by EEG: Herning, R.I., London, E., Phillips, R., and Glover, B.J.**

Effects of morphine on the scalp EEG are being studied to determine the brain areas invoked in euphoria. Etiology collected in past years and is now analyzing the EEG data from 20 scalp locations from post addicts receiving placebo, 15 and 30 mg injections of morphine. These subjects subsequently received FDG PET scans while receiving placebo and 30 mg of morphine. The PET scans are performed by our collaborators. The EEG data by itself provides insight into time course of electrophysiologic effects of a mu agonist in humans and the cortical distribution of mu effects.

K. **Cholinergic Pharmacology: Cognitive and Neurophysiologic Screen: Herning, R.I., Glover, B.J., and Koeppl, B.**

The purpose of the study is to better understand the effects of cholinergic agents or sensory, motor and cognitive performance at the neurophysiological level. Cholinesterase inhibitors are commonly used in biological warfare agents. Techniques for determining the cognitive impairments prroduced by anticholinergics and safe models for inducing cholinergic stimulation are important steps in developing useful and effective antidotes to cholinesterase inhibitors.

L. **Effects of Atropine on Cognitive Information Processing: Herning, R.I., Glover, B.J., and Koeppl, B.**

The purpose of this effort is to better understand the effects of cholinergic agents on cognition and performance; in particular, where in the information processing sequence atropine exerts its effects. The EEG and evoked response data have been reported in military and scientific journals. Atrophine at doses 4 mg or greater increase EEG slowing and reduces cognitive evoked potentials and performance.

M. **Effects of Benzodiazepines on Cognitive Information Processing: Herning, R.I., Glover, B.J., and Koeppl, B.**

The purpose of the study is to determine where in the information processing sequence the benzodiazepines exert their effects. Memory deficits have been previously noted, but it is yet unclear whether the deficit is due to poor encoding of the information or loss of the newly formed memory trace. The study is important in understanding the ways in which drugs of this class impair cognition and performance.

Publications

Newlin, D.B., & Thomson, J.B. (1989). Alcohol challenge studies with sons of alcoholics: A critical review and analysis. Psychological Bulletin, in press.

Herning, R.I., Glover, B.J., Weddington, W.W., Koeppl, B.S., and Jaffe, J.H. Cognitive Decrements during cocaine abstinence were not related to depression. Biological Psychiatry. Submitted, January, 1990.

Weddington, W.W., Brown, B.S., Haertzen, C.H., Cone, E.J., Dax, E.M., Herning, R.I., and Michaelson, B.S. Changes in mood, craving and sleep during acute abstinence reported by male cocaine addicts: A controlled residential study. Archives of General Psychiatry, In press, 1990.

Litow, R.M., Herning, R.I., Robinson, N., Jaffe, J.H., and Dax, E.M. Cognitive function and EEG testing in volunteer men inhaling volatile nitrites. Submitted Psychopharmacology June, 1989.

London, E.D., Cascella, N.G., Wong, D.F., Phillips, R.L., Dannels, R.F., Links, J.M., Herning, R.I., Grayson, R., Jaffe, J.H., and Wagner, H.N. Cocaine-induced reduction of glucose utilization in human brain: A study using Positron Emission Tomography and FDG. Archives of General Psychiatry, Submitted, August, 1989.

Herning, R.I., Glover, B.J., Koeppl, B., Weddington, W., and Jaffe, J.H. Cognitive deficits in abstaining cocaine abusers. In: Residual Effects of Abused Drugs (J. Spenser and J.J. Boren Eds.) National Institute on Drug Abuse Monograph Series, in press, 1989.

Pickworth, W.B., Herning, R.I., and Henningfield, J.E. Spontaneous EEG changes during abstinence and nicotine substitution. Journal of Pharmacology and Experimental Therapeutics, In press, 1989.

Muntaner, C., Cascella, N.G., Kumor, K.M., Herning, R.I., and Jaffe, J. Placebo Response to cocaine administration in humans: Effects of prior verbal instructions. Psychopharmacology, 1989, 99: 282-286.

London, E.D., Broussole, E.P.M., Links, J.M., Wong, D.F., Cascella, N.G., Dannels, R.F., Sono, M., Herning, R., Snyder, F.R., Rippetoe, L.R., Toung, T.J.K., Jaffe, J.H., Wagner, H.N. Morphine-induced metabolic changes in the brain: studies with Positron Emission Tomography and FDG. Archives of General Psychiatry, In press, 1989.

Herning, R.I., Glover, B.J. and Henningfield, J.E. Attention deficits during nicotine abstinence. Psychopharmacology, Submitted Jan., 1989.

Pickworth, W.B., Herning, R.I., Koeppl, B. and Henningfield, J.E. Atropine-induced changes in spontaneous electroencephalogram in human volunteers. Military Medicine. In press, 1989.

Herning, R.I., Hickey, J., Pickworth, W. and Jaffe, J.H. Auditory event-related potentials in adolescents at risk for drug abuse. Biological Psychiatry, 1989, 25: 598-609

Fishbein, D., Herning, R.I., Pickworth, W.B., Haertzen, C.A. and Jaffe, J.H. Brainstem evoked response potentials in adult male drug abusers with self-reported histories of aggressive behavior. Biological Psychiatry, 1989, 26: 595-611.

Newlin, D.B. (1989). Placebo responding in the same direction as alcohol in women. Alcoholism: Clinical and Experimental Research, 13, 36-39.

Newlin, D.B., Hotchkiss, B., Cox, W.M., Rauscher, R., & Li, T.-K. (1989). Autonomic and subjective responses to alcohol stimuli with appropriate control stimuli. Addictive Behaviors, 14, 625-630.

Newlin, D.B. (1989). The skin flushing response: Autonomic, subjective, and conditioned responses to repeated administrations of alcohol in Asian men. Journal of Abnormal Psychology, 98, 421-425.

Newlin, D.B., Byrne, E.A., & Porges, S.W. (1989). Vagal mediation of the effect of alcohol on heart rate. Alcoholism: Clinical and Experimental Research, in press.

Newlin, D.B., Pretorius, M.B., & Jaffe, J.H. (1989). Pavlovian conditioning to morphine in opiate abusers. NIDA Research Monograph, Problems of Drug Dependence, 1989, Proceedings of the 51st annual scientific meeting, Committee on Problems of Drug Dependence. Washington, DC: U.S. Government Printing Office.

Thomson, J.B., & Newlin, D.B. (1989). Pavlovian conditioning history vs. placebo expectance effects to alcohol in humans. Alcoholism: Clinical and Experimental Research, 10.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 07501-02 CPH

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Acute Marijuana Smoking Reduces Vagal Tone

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI:	D.B. Newlin	Acting Chief	Etiology, ARC, NIDA
Others:	E. Dax	Chief	NEIL, ARC, NIDA
	M.B. Pretorius	Research Asst.	Etiology, ARC, NIDA
	C. Wong	Guest Worker	Etiology, ARC, NIDA

COOPERATING UNITS (If any)

Neuroendocrinology and Immunology Laboratory, Clinical Pharmacology Branch

LAB/BRANCH
Etiology Branch

SECTION
Vulnerability Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
2.00	1.00	1.00

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

It has been assumed that the pronounced tachycardia from marijuana smoking is sympathetic in origin (Goodman and Gilman, 1985). It is difficult to draw inferences from parasympathetic blockade during marijuana smoking because vagal blockade has dramatic effects on baseline cardiovascular values. Therefore, we sought to evaluate parasympathetic components of THC-induced tachycardia using a noninvasive measure of vagal influences. This vagal tone index (V) quantified rhythmicity in heart rate at the respiratory frequency (i.e., respiratory sinus arrhythmia) and at a lower frequency associated with blood pressure homeostasis. 19 male marijuana smokers who had been deprived of THC for at least one week on an inpatient research unit were given either oral placebo, 10 mg THC orally, or they smoked 2.7% marijuana cigarettes. We found that particularly marijuana smoking produced increases in static ataxia, and decreases in vagal tone index (V) and heart rate rhythmicity at lower frequencies. There was a pronounced flattening of heart rate variability and attenuation of respiratory sinus arrhythmia with the higher THC delivery associated with marijuana smoking. Oral THC at this dosage had no clear effects on heart rate, vagal tone index (V), or the lower cardiovascular rhythm. This preliminary data indicated that marijuana produces significant withdrawal of vagal inhibition that elicits tachycardia and increased blood pressure, and that the cardiovascular effects of marijuana are not purely sympathetic.

Z01 DA 07501-02 CPH

Acute Marijuana Smoking Reduces Vagal Tone

Publications

Newlin, D.B., Pretorius, M.B., Wong, C.J., and Dax, E.M. (1990). Acute marijuana smoking reduces vagal tone. Paper to be presented at the Annual Meeting of the Committee on Problems of Drug Dependence, Richmond, VA, June, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 07001-02 ETL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Cardiovascular Components of the Response to Morphine

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI: D.B. Newlin Acting Chief Etiology, ARC, NIDA

Others: M.B. Pretorius Research Asst. Etiology, ARC, NIDA
0 C. Wong Guest Worker Etiology, ARC, NIDA

COOPERATING UNITS (If any)

none

LAB/BRANCH
Etiology Branch

SECTION
Vulnerability Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.5	1.00	1.00

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The cardiovascular response to morphine is biphasic in humans and rats. We used noninvasive measures based on EKG monitoring to study different components of the cardiovascular response to morphine. Vagal tone index (V) quantifies heart rate (HR) variability in the respiratory frequency band of approx. 0.33 Hz (i.e., respiratory sinus arrhythmia), and the THM band measures HR variability at a frequency (approx. 0.10 Hz) associated with blood pressure homeostasis. Twelve opiate abusers on a research residential unit received morphine (i.m., 20 mg) in five separate sessions, compared to a placebo injection session and a no injection control session. HR was monitored continuously in each session. The initial HR increase to morphine appeared to be due to withdrawal of vagal inhibition (significant decreases in V) and changes in the baroreceptor feedback system (significant decreases in THM). Initial changes in V were negatively correlated ($r = -0.43$) with MBG (ARCI) morphine-induced euphoria. Only THM was significantly decreased to morphine over the 100 min recording period, indicating that the longer duration effects of the drug may be related to the baroreceptor system. The placebo effect (compared to no injection) consisted of a decrease in HR and THM. These results suggest that the initial tachycardia to morphine may be physiologically and psychologically distinct from subsequent cardiovascular changes.

Z01 DA 07001-02 ETL

Cardiovascular Components of the Response to Morphine

Publications

Pretorius, M.B., Wong, C.J., and Newlin, D.B. (1990). Cardiovascular components of the response to morphine. Paper to be presented at the Annual Meeting of the Committee on Problems of Drug Dependence, Richmond, VA, June, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 07701-02 ETL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Excitatory & Inhibitory Drug Effects in Pavlovian Drug Conditioning

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: D.B. Newlin Acting Chief Etiology, ARC, NIDA

COOPERATING UNITS *(if any)*

none

LAB/BRANCH
Etiology Branch

SECTION
Vulnerability Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS: 1.00	PROFESSIONAL: 1.00	OTHER: 0.00

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This paper reviews Pavlovian drug conditioning studies in terms of excitatory and inhibitory drug effects. Conditioned tolerance was found with inhibitory drug effects, and conditioned sensitization with excitatory drug effects. This did not depend on the class of drug (i.e., stimulant or depressant), but only on the response measure (i.e., excitatory or inhibitory). There was always a trend toward increased excitation over repeated administrations of these drugs. The conditioned response (CR) to environmental cues for these drugs was always excitatory. Inhibitory CRs were found only with 24 hr cues for drugs. Consideration of these patterns led to theoretical conclusions concerning adaptation to abused drugs and conditional responding. A model of motivational aspects of conditioned tolerance, sensitization, and conditioned responses was proposed.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 09601-02 E

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Antagonist-Withdrawal Up-Regulation of Endogenous Opiate

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	G. Uhl	Chief	MNL, ARC NIDA
Others:	D.B. Newlin	Acting Chief	Etiology, ARC, NIDA
	M.B. Pretorius	Research Asst.	Etiology, ARC, NIDA

COOPERATING UNITS *(If any)*
Molecular Neurobiology Laboratory, Neuroscience Branch

LAB/BRANCH
Etiology Branch

SECTION
Vulnerability Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3.00	2.00	1.00

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Changes in expression of the principle opioid peptide gene, preproenkephalin, follow stimulation or inhibition of inputs to enkephalinergic neurons, and administration of opiate agonist and antagonist drugs. The duration of proenkephalin gene up- or down-regulation can outlast the duration of the stimulus, suggesting that regulated gene expression could provide one substrate for storing information about previous stimulus or drug exposure. This would have direct implications for possible mechanisms of opiate tolerance and withdrawal. We have thus sought evidence for functional up-regulation of enkephalinergic gene regulation systems in humans. The subjects were 19 males with no reported histories of opiate abuse. Pain self-report to a 5 min ice-water immersion and cold-stress-induced analgesia were tested before 8 days of antagonist treatment, 2 days after the last dose of antagonist (when excretion of antagonist metabolites was finished) or placebo, and finally 30 min after an acute oral dose of naloxone. Self-report of pain to the initial stages of the ice-water immersion were significantly reduced in subjects 2 days after opiate antagonist treatment. Initial results of the study provide evidence for increased function in endogenous opioid systems after antagonist washout. Current studies aim to separate pre- and post-synaptic components to this effect.

Z01 DA 09601-02 ETL

Antagonist-Withdrawal Up-Regulation of Endogenous Opiate

Publications

Uhl, G.R., Newlin, D.B., Pretorius, M.B., Park, J., and Cone, E. (1990). Antagonist-withdrawal up-regulation of endogenous opiate antinociceptive systems. Paper to be presented at the Annual Meeting of the Committee on Problems of Drug Dependence, Richmond, VA, June, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 06801-03 E

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Cognitive Neurophysiologic Signs of Cocaine Abstience

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: R.I. Herning Visiting Scientist ETL, NIDA

COOPERATING UNITS *(if any)*

Immunology Lab (Elizabeth Dax, MD) Treatment Branch (Frances Levin, MD)

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.90	.70	1.20

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Cognitive impairments and sleep disruption have been reported in patients withdrawing from cocaine. The nature of these disorders have yet to be documented in clinical laboratory studies. The present study evaluates cognitive information processing in subjects on a clinical ward withdrawing from cocaine with a battery of tasks (auditory rare event monitoring, complex visual rare event monitoring, Sternburg memory, visual motor tracking). Sleep quality and duration is monitored by a subjective questionnaire. Twenty subjects including controls have been tested in this study over a one month withdrawal period. Stimulus evaluation and memory deficits were observed in the cocaine addicts. The memory deficits persited for over three weeks. Additional subjects are being tested on a more extensive electrophysiological battery for six to eight weeks to determine whether the deficits observed in the orginal study will persist past three weeks.

Clarification of the nature of the cognitive deficits and of sleep loss will lead to more effective treatment strategies for cocaine withdrawal.

Z01 DA06801-03 ETL

Cognitive Neurophysiologic Signs of Cocaine Abstinence

Publications:

Herning, R.I., Glover, B.J., Weddington, W.W., Koeppl, B.S., and Jaffe, J.H. Cognitive Decrements during cocaine abstinence were not related to depression. Biological Psychiatry Submitted, January, 1990.

Herning, R.I., Glover, B.J., Koeppl, B., Weddington, W., and Jaffe, J.H. Cognitive deficits in abstaining cocaine abusers. In: Residual Effects of Abused Drugs (J. Spenser and J.J. Boren Eds.) National Institute on Drug Abuse Monograph Series, in press, 1989.

Weddington, W.W., Brown, B.S., Haertzen, C.H., Cone, E.J., Dax, E.M., Herning, R.I., and Michaelson, B.S. Changes in mood, craving and sleep during acute abstinence reported by male cocaine addicts: A controlled residential study. Archives of General Psychiatry, In press, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 05801-03 E

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Mapping the Effects of Cocaine by EEG

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: R.I. Herning Visiting Scientent ETL, NIDA

COOPERATING UNITS *(if any)*

Neuropharmacology Lab (E. London)
Johns Hopkins Hospital (D. Wong)

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.35	.15	.20

CHECK APPROPRIATE BOX(ES)

- [x] (a) Human subjects
  - [ ] (a1) Minors
  - [ ] (a2) Interviews
- [ ] (b) Human tissues
- [ ] (c) Neither

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The effects of cocaine on scalp EEG and FDG PET scans are being compared to determine the brain areas involved in the cocaine-induced euphoria. In previous studies, cocaine increased EEG beta power. The distribution of cortical areas responsible for the EEG beta increase and the time course of the beta increase hav not as yet been determined. The present study was designed to answer these two questions.

The complimentary nature of the EEG and PET data will delineate the anatomical and electrophysiologic mechanisms involved in cocaine induced euphoria.

Six subjects were tested using EEG measures with placebo, 20mg and 40mg of cocaine in double blind order in previous years and seven additional subjects were tested during the current year. EEG beta increased in dose dependent manner starting immediately after the injection and continuing for twenty minutes. The increase i EEG beta was maximal in frontal, temporal and partial cortical areas. The relationship between the increase in beta and subjective state is currently being investigated.

Z01 DA05801-03 ETL

Mapping the Effects of Cocaine by EEG

Publications:

London, E.D., Cascella, N.G., Wong, D.F., Phillips, R.L., Dannels, R.F., Links, J.M., Herning, R.I., Grayson, R., Jaffe, J.H., and Wagner, H.N. Cocaine-induced reduction of glucose utilization in human brain: A study using Positron Emission Tomography and FDG. Archives of General Psychiatry, Submitted, August, 1989.

Muntaner, C., Cascella, N.G., Kumor, K.M., Herning, R.I., and Jaffe, J. Placebo response to cocaine administration in humans: Effects of prior verbal instructions. Psychopharmacology, 1989, 99: 282-286.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 03301-03 CI

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Electrophysiologic Measures of Conduct Disorder (Aggressive) in Adolescents

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	R.I. Herning	Visiting Scientist	ETL, NIDA

COOPERATING UNITS *(if any)*

Treatmemt Branch (J. Hickey)

LAB/BRANCH

Etiology

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER:
.225	.025	.200

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☒ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Adolescents with a history of violence are likely to be at risk for drug abuse. This project studies a group of antisocial adolescents and a group of age, IQ, race, and neighborhood matched adolescents who participated in a study which included psychometric and electrophysiological testing. Etiology is primarily concerned with the electrophysiological measures. The more aggressive group differed from the control group on many electrophysiological measures. This data was published and a new study which will look at similar measures in preadolescene males.

Z01 DA03301-03 CHP

Electrophysiologic Measures of Conduct Disorder (Aggressive) in Adolescents

Publications:

Herning, R.I., Hickey, J., Pickworth, W. and Jaffe, J.H. Auditory event-related potentials in adolescents at risk for drug abuse. Biological Psychiatry, 1989, 25: 598-609

Fishbein, D., Herning, R.I., Pickworth, W.B., Haertzen, C.A. and Jaffe, J.H. Brainstem evoked response potentials in adult male drug abusers with self-reported histories of aggressive behavior. Biological Psychiatry, 1989, 26: 595-611.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 03111-04

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effects of Benzodiazepines on Cognitive Information Processing

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: R.I. Herning Visiting Scientist ETL, NIDA

COOPERATING UNITS *(if any)*

Biology of Dependence Lab (J. Henningfield, W. B. Pickworth, J. Roache, R. Lamb)

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.075	.025	.050

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

An extensive battery of sensory and cognitive electrophysiological tasks was used to assess sensory, cognitive and performance deficits produced by diazepam. The tasks include eyes open and eyes closed EEG, brainstem auditory evoked response, pattern reversal visual evoked response, auditory rare event monitoring task, the auditory continuous performance task and the Sternberg memory task (both immediat and delayed). Six doses (0, 2.5, 5.0, 10.0, 20.0 and 40.0 mg) of diazepam were used. Nine subjects were tested.

The purpose of the study was to determine where in the information processing sequence the benzodiazepines exert their effects. Memory deficits have been previously noted, but it is yet unclear whether the deficit is due to poor encodi of the information or loss of the newly formed memory trace. The study is important in understanding the ways in which drugs of this class impair functioni Evoke potential analysis was begun over the last year.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 02101-04 ETL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Acute Abstinence From Tobacco: Electrophysiological and Cognitive Signs

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI	R. I. Herning	Visiting Scientist	ETL, NIDA

COOPERATING UNITS *(if any)*

Biology of Dependence Lab (J. Henningfield, W B. Pickworth

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.025	.025	0.0

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

- ☐ (a1) Minors
- ☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The laboratory's efforts were directed toward the quantification of the cognitive and performance deficits during nicotine withdrawal and the treatment of these deficits with nicotine chewing gum. The EEG, cognitive, and cognitive process was monitored during a ten day period of tobacco withdrawal in heavy smokers. Some of the changes persisted over the entire ten day deprivation period. These measures included EEG alpha frequency, theta power, performance on selected cognitive tasks (especially a rapid arithmetic task) and a cognitive event related potential measure (N100 amplitude). Stimulus evaluation time, as measured by P300 latency, and the depth of stimulus evaluation battery were affected early during the tobacco deprivation period, but returned to smoking levels later during the deprivation period. Thus, the cognitive deficits are clearly apparent during abstinence from tobacco and contribute to relapse during treatment. The deficits during withdrawal have at least two different components-one affecting stimulus evaluation which dissipates after 5 to 7 days of abstinence and one affecting attention accompanied by lower arousal which persists ten days or longer. During the year this data was analyzed and two papers were submitted for publication.

Z01 DA02101-04 ETL

Acute Abstinence from Tobacco: Electrophysiological and Cognitive Signs

Publications:

Pickworth, W.B., Herning, R.I., and Henningfield, J.E. Spontaneous EEG changes during abstinence and nicotine substitution. Journal of Pharmacology and Experimental Therapeutics, In press, 1989.

Herning, R.I., Glover, B.J. and Henningfield, J.E. Attention deficits during nicotine abstinence. Psychopharmacology, Submitted Jan., 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 02001-04 ETL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Mapping the Effects of Opioid Agonists by EEG

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: R.I. Herning Visiting Scientist ETL, NIDA

COOPERATING UNITS *(If any)*

Neuropharmacology Lab (E. London)
Johns Hopkins Hospital (D. Wong)

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
.075	.025	.050

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Effects of morphine on the scalp EEG and FDG PET scans are being compared to determine the brain areas invoked in euphoria. Etiology collected in past years and is now analyzing the EEG data from 20 scalp locations from post addicts receiving placebo, 15 and 30 mg injections of morphine. These subjects subsequently received FDG PET scans while receiving placebo and 30 mg of morphine. The PET scans are performed by our collaborators. The EEG data by itself provides insight into time course of electrophysiologic effects of a mu agonist in humans and the cortical distribution of mu effects. PET techniques do not by themselves provide information about the time course of the mu effects. In the preliminary analysis, twelve subjects had increased EEG delta and the theta power begining 15 minutes and persisting until 45 minutes after the intramuscular injection. Changes in artifact dectection were being investigated so that the relationship between these EEG changes and subjective effects of morphine can be being investigated.

Z01 DA02001-04 ETL

Mapping the Effects of Opioid Agonists by EEG

Publications:

London, E.D., Broussole, E.P.M., Links, J.M., Wong, D.F., Cascella, N.G., Dannels, R.F., Sono, M., Herning, R., Snyder, F.R., Rippetoe, L.R., Toung, T.J.K., Jaffe, J.H., Wagner, H.N. Morphine-induced metabolic changes in the brain: studies with Positron Emission Tomography and FDG. Archives of General Psychiatry, In press, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 03101-04 ETL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effects of Atropine on Cognitive Information Processing

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: R. I. Herning Visiting Scientist ETL, NIDA

COOPERATING UNITS *(if any)*

Biology of Dependence Lab (J. Henningfield, W. B. Pickworth)

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.075	.025	.050

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

An extensive battery of sensory and cognitive electrophysiological tasks is used to assess sensory, cognitive and performance deficits produced by atropine. The tasks include eyes open and eyes closed EEG, brainstem auditory evoked response, pattern reversal visual evoked response, the auditory rare event monitoring task, auditory continuous performance task and Sternberg auditory memory task (both immediate and delayed). Each of four doses of atropine (0, 2, 4 and 6 mg/70 kg) is investigated on two occasions. Eight subjects have been tested on these procedures.

The purpose of the study is to better understand the effects of cholinergic agents on cognition and performance; in particular, where in the information processing sequence atropine exerts its effects. The EEG and evoked response data have been reported in military and scientific journals. Atrophine at doses 4mg or greater increase EEG slowing and reduces cognitive evoked potentials and performance. The EEG results were published and evoked potential analysis was begun over the last year.

Z01 DA03101-04 ETL

Effects of Atropine on Cognitive Informatin Processing

Publications:

Pickworth, W.B., Herning, R.I., Koeppl, B. and Henningfield, J.E. Atropine-induced changes in spontaneous electroencephalogram in human volunteers. Military Medicine. In press, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 05901-03 ETL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Cholinergic Pharmacology: Cognitive and Neurophysiologic Screen

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: R.I. Herning Visiting Scientist ETL, NIDA

COOPERATING UNITS *(if any)*

Biology of Dependence Lab (J. Henningfield, J. Roache)

LAB/BRANCH
Etiology

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
.075	.025	.050

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

A battery of tasks is being used to assess sensory, cognitive, and motor deficits produced by physostigmine. The tasks (eyes open EEG, physiological tremor, pattern reversal visual evoked response, self paced motor potential, rare event monitoring, and Steinburg tasks) were designed to test neurophysiological indices of brain processing as well as behavioral performance. Sensory and cognitive performance was tested both after placebo or methscopolamine pretreatment. The pretreatment with methscopolamine tests whether or not the performance deficit were due perpherial effect of physostigmine.

The purpose of the study is to better understand the effects of cholinergic agents or sensory, motor and cognitive performance at the neurophysiological level. Cholinesterose inhibitors are commonly used biological warfare agents. Techniques for determining the cognitive impairments produced by anticholinergics and safe models for inducing cholinergic stimulation are important steps in developing useful and effective antidotes to cholinesterose inhibitors. Ten subjects were tested in previos years. During the last year, a preliminary data was reported at a military meeting.

Z01 DA05901-03 ETL

Cholinergic Pharmacology: Cognitive and Neurophysiologic Screen

Publications:

Herning, R.I., Glover, B.J., and Reddish, R. Information Processing Effects of Physostigmine. Military Medicine, 1989, 873-876.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 10701-01 ETL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effect of Alcohol on Cocaine Craving

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	D.B. Newlin	Acting Chief	Etiology, ARC, NIDA
Others:	C. Muntaner	Visiting Fellow	Etiology, ARC, NIDA
	M.B. Pretorius	Research Asst.	Etiology, ARC, NIDA
	C. Wong	Guest Worker	Etiology, ARC, NIDA

COOPERATING UNITS *(if any)*
none

LAB/BRANCH
Etiology Branch

SECTION
Vulnerability Laboratory

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
4.00	2.00	2.00

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This project concerned the elicitation of cocaine craving from ingestion of alcohol. The hypothesis that alcohol intoxication increases cocaine craving is derived from anecdotal reports from cocaine abusers that they cannot abstain from cocaine when intoxicated from alcohol, and from evidence of some generality in the ability of passive injection of drugs to "prime" self-administration of other drugs in animals. We investigated this "priming" effect in cocaine abusers in the laboratory using self-report and noninvasive physiological measures. Residential cocaine abusers with histories of relatively heavy alcohol use were given, on separate days, water (as a control), placebo, 0.64 g/kg alcohol, and 1.1 g/kg alcohol. They continuously rated their craving for cocaine on a joystick for up to two hrs before and after receiving alcohol. On each day, they were exposed to a control tape and a cocaine self-administration tape with cocaine paraphernalia during the rising and falling limbs of the blood alcohol curve. The dependent measures included self-report of cocaine craving and mood, and heart rate, vagal tone, finger and cheek temperature, and right and left frontal electroencephalographic (EEG) measures recorded before and after drinking, and before and after the craving induction procedures. We have examined seven subjects to date, and plan to run several more. Preliminary data analyses showed that alcohol intoxication increased self-reported cocaine craving. Analyses of the psychophysiological effects of the cocaine tape and paraphernalia vs. the control tape, and the rising vs. falling blood alcohol curves await completion of further subjects.

Neuroscience Branch

Michael J. Kuhar, Ph.D., Chief

Introduction

The Neuroscience Branch carries out interdisciplinary research aimed at elucidating the mechanisms of action and the effects of abused drugs on biological systems. Also a goal is to identify and to develop new treatment and prevention strategies and new medications for drug abusers. Research areas include brain imaging, drug and neurotransmitter receptors, neurobiology and neuroanatomy of reinforcement and molecular neurobiology.

The Neuroscience Branch developed a new laboratory over the past year. Reflecting major advances in molecular biology and genetics in recent years, the molecular biology unit within the Molecular Pharmacology Laboratory was elevated to the status of a laboratory with Dr. George Uhl as Chief. The Molecular Neurobiology Laboratory focuses on the regulated expression of genes related to drug abuse and on cloning genes for drug receptors.

The Branch has been productive in a variety of areas. For example, chemical and molecular biologic probes for the cocaine receptor/dopamine transporter have been developed which will aid in its isolation and purification; brain imaging studies have shown that morphine and cocaine cause a reduction in glucose metabolic rate in cerebral cortex, suggesting the involvement of this region in drug-induced euphoria; sigma receptors were identified in immune tissues suggesting that PCP exerts its immunosuppressive effects via these receptors; biochemical "transcription factors" regulating neuronal expression of morphine-like peptide genes and novel means for cloning drug receptors have been identified. These and many more findings are presented in more detail below.

The research of the Neuroscience Branch and its investigators have received national and international recognition and attention. Dr. Kuhar received the ADAMHA Administrator's Award for Meritorious Achievement, and was asked to present the Upjohn Lecture at the Uniformed Services University and the Thomas L. O'Donohue Memorial Lecture at Howard University. Dr. De Souza was given the Joseph Cochin Award by the Committee on Problems of Drug Dependence; the award is given to the outstanding young investigator in drug addiction. Dr. Dimitri Grigoriadis, a staff fellow under Dr. De Souza, was given the ARC Staff Fellow Research Prize.

1. Molecular Pharmacology Laboratory - Michael J. Kuhar, Ph.D., Chief

Overview

The Molecular Pharmacology Laboratory focuses on the molecular

mechanisms of action and the molecular effects of drugs of abuse. As this year began, the laboratory included groups working on receptor binding and visualization on molecular biological approaches towards drug receptors and on molecular mechanisms of action. Over the past year, because of the advances in molecular biology and molecular genetics, and because of the importance of these techniques in neuroscience, it was decided to elevate the molecular biology unit to the status of a laboratory. This emphasizes the committment to sustain a research effort of the highest quality in this area. Accordingly, the Laboratory of Molecular Neurobiology was formed and a chief, Dr. George Uhl, was established as its head. The progress of this new laboratory is indicated separately below.

An area of major research emphasis in the laboratory is the mechanism of action of cocaine. In 1987, our laboratory published data suggesting that the cocaine binding site associated with the dopamine transporter is the physiological receptor related to the self-administration of cocaine in primates and humans. Accordingly, we have been working at characterizing the dopamine transporter in molecular terms. A photoaffinity label, ^{125}I-DEEP, has been developed. Studies with this ligand have indicated that the dopamine transporter is a glycoprotein with a molecular weight of about 58,000 Da. The carbohydrate moiety appears to be rich in sialic acid residues.

In another series of studies, we are collaborating with chemists who are synthesizing analogues of cocaine. With these analogues were are carrying out extensive structure-activity studies so as to delineate structural characteristics of the binding site at the dopamine transporter. Another goal is to identify novel ligands for studying the cocaine receptor and dopamine transporter. For example, novel isothiocyanate derivatives of cocaine have been found which irreversibly inhibit cocaine binding. These compounds may be useful in the purification and elucidation of the structure and properties of the cocaine receptor.

In collaborative studies with the Laboratory of Neuropharmacology and with workers at Johns Hopkins, we are undertaking several PET scanning studies involving the action of cocaine in the brain. The goal of these studies is to elucidate the sites in brain related to the rush and high that follow cocaine ingestion.

Another goal is to identify useful ligands for in vivo receptor binding assays for the cocaine receptor. In vitro binding assays are reproducible and are well established. However, in vitro binding assays may give results that are dependent on buffer conditions, temperature as well as other in vitro assay conditions. A critical question is what is the behavior of the receptor in vivo and what are the properties of drug interactions with this receptor in vivo. According, we have been developing an in vivo receptor binding assay using analogues of cocaine that have been radiolabeled. Our results suggest, at least preliminarily, that an effective in vivo receptor

binding assay for the cocaine receptor has been achieved.

There are many other studies in the laboratory as well. For example, we are continuing our research for dopamine receptor ligands which might be useful for studying the secondary effects of cocaine. For example, we have identified a novel dopaminergic ligand, Spectramide, which hopefully will bind with a low affinity in vivo so that endogenous dopamine will compete for the binding successfully. Hence, administration of cocaine which prevents the inactivation of dopamine and enhances dopaminergic neurotransmission might cause an in vivo reduction in Spectramide binding. This would allow us a measure of the secondary action of cocaine at dopamine receptors.

Thus, the Molecular Pharmacology Laboratory utilizes a variety of molecular and other techniques to elucidate the mechanism of action of drugs of abuse. Over the past year, the emphasis, in accordance with the priority of the Institute, has been on cocaine. Substantial advances have been achieved. These include the identification of new ligands for in vitro and in vivo labeling, as well as the development of molecular probes to further characterize the molecular nature of the cocaine receptor.

Summary of Ongoing Research

A. The Cocaine Receptor: Kuhar, M.J., Lew, R. and Simantov, R.

Because of the evidence that cocaine binding sites at the dopamine transporter are the receptors that mediate drug self-administration, efforts have been focused at characterizing the dopamine transporter. Current efforts include structure-activity studies which will clarify the binding properties and perhaps the spacial structure of the cocaine receptor. Other efforts include the solubilization, purification and characterization of the transporter; this involves traditional purification techniques with studies aimed at characterizing the molecular weight, the carbohydrate moiety, and other molecular properties of the transporter. Ultimately, it is hoped that the gene for the dopamine transporter will be identified.

B. Drug Receptors, Neurotransmitters and Addiction: Kuhar, M.J., Ritz, M., Boja, J.W. and Lew, R.

This multifaceted project continues to be directed toward studying the interation of drugs of abuse with various brain neurochemicals. Current studies involve the exploration of the involvement of the PCP receptor in the action of cocaine. Other studies include an assessment of the various neurotransmitter systems that are affected by cocaine and the relative degree of selectivity or nonselectivity of cocaine and its various

analogues on these systems. Also, a screening effort is underway to determine if various drugs that are potential medications for cocaine addiction interact with the cocaine receptor.

C. **Measuring Drug Receptors In Vivo: Kuhar, M.J. and Boja, J.W.**

Efforts are being directed at developing new ligands for the cocaine receptor some identified ligands have properties that are more favorable than those of cocaine. At least two ligands have been identified which are potentially suitable for PET scanning studies. These compounds are more potent than cocaine in in vitro binding studies and are comparatively resistant to metabolic degradation. Preliminary in vivo screening studies in mice indicate that they have useful characteristics. Preliminary studies involving PET scanning with these compounds will be undertaken.

Publications

Ritz, M.C., R.J. Lamb, S.R. Goldberg and M.J. Kuhar. Cocaine Self-Administration Appears to Be Mediated by Dopamine Uptake Inhibition. Prog. Neuro-Psychopharmacol. & Biol. Psychiat. 12, 233-239, 1988.

O'Hearn, E., G. Battaglia, E.B. De Souza, M.J. Kuhar and M.E. Molliver. Methylenedioxyamphetamine (MDA) and Methylenedioxy-methamphetamine (MDMA) Cause Selective Ablation of Serotonergic Axon Terminals in Forebrain: Immunocytochemical Evidence for Neurotoxicity. J. Neurosci. 8 (8), 2788-2803, 1988.

Sharkey, J., M.C. Ritz, J.A. Schenden, R.C. Hanson and M.J. Kuhar. Cocaine Inhibits Muscarinic Cholinergic Receptors in Heart and Brain. J. Pharmacol. Exp. Ther. 246 (3), 1048-1052, 1988.

Kuhar, M.J. Overview. In: Receptor Localization: Ligand Autoradiography. F.M. Leslie and C.A. Altar (Eds.), Alan R. Liss, New York, pp 1-7, 1988.

Kuhar, M.J., Ritz, M.C. and Sharkey, J. Cocaine receptors on dopamine transporters mediate cocaine-reinforced behavior. In: Mechanisms of Cocaine Abuse and Toxicity. D. Clouet, K. Asglan and R. Brown (Eds.), NIDA Research Monograph No. 88, U.S. Gov't. Printing Office, Washington, D.C., pp 14-22, 1988.

Wagner, H.N., Jr., Weinberger, D.R., Kleinman, J.E., Casanova, M.F., Gibbs, C.J., Jr., Gur, R.E., Hornykiewicz, O., Kuhar, M.J., Pettegrew, J.W. and Seeman, P. Neuroimaging and Neuropathology. Schizophrenia Bulletin 14 (3), 383-397, 1988.

Wong, D.F. and Kuhar, M.J. In Vivo PET and SPECT Receptor Imaging: New Technology and Tactics for Receptor Measurement. In Neuroreceptors and Signal Transduction. S. Kito, T. Segawa, K. Kuriyama, M. Tohyama and R.W. Olsen (Eds.), Plenum Press, New York, pp. 181-193, 1988.

Lo, M.M.S., Conrad, M.K., Mamalaki, C. and Kadan, M.K. Retroviral mediated gene transfer: Applications in Neurobiology. Mol. Neurobiol. Rev. 2:1-29, 1988.

Ritz, M.C. and Kuhar, M.J. Relationship Between Self-administration of Amphetamine and Monoamine Receptors in Brain: Comparison With Cocaine. J. Pharmacol. Exp. Ther. 248, 1010-1017, 1989.

Kuhar, M.J. Perspectives. In Brain Imaging Techniques and Applications. N.A. Sharif and M.E. Lewis (Eds.), John Wiley & Sons, New York, pp. 13-17, 1989.

Lew, R., Grigoriadis, D.E., Sharkey, J. and Kuhar, M.J. Dopamine Transporter: Solubilization From Dog Caudate Nucleus. Synapse 3, 372-375, 1989.

Grigoriadis, D.E., Wilson, A.A., Lew, R., Sharkey, J.S. and Kuhar, M.J. Dopamine Transport Sites Selectively Labeled by a Novel Photoaffinity Probe: ^{125}I-DEEP. J. Neurosci. 9(8), 2664-2670, 1989.

Kuhar, M.J. and De Souza, E.B. Autoradiographic Imaging: Localization of Binding Sites other than Neurotransmitter Receptors. In: Visualizations of Brain Functions. D. Ottoson and W. Rostene (Eds.), M Stockton Press, pp. 57-66, 1989.

Porrino, L.J., Ritz, M.C., Goodman, N.L., Sharpe, L.G., Kuhar, M.J. and Goldberg, S.R. Differential Effects of the Pharmacological Manipulation of Serotonin Systems on Cocaine and Amphetamine Self-Administration in Rats. Life Sciences 45, 1529-1535, 1989.

Sanchez-Roa, P.M., Grigoriadis, D.E., Wilson, A.A., Sharkey, J., Dannals, R.F., Villemagne, V.L., Wong, D.F., Wagner, H.N. and Kuhar, M.J. [^{125}]I-Spectramide: A Novel Benzamide Displaying Potent and Selective Effects at the D_2 Dopamine Receptor. Life Sciences 45, 1821-1829, 1989.

Scheffel, U. J.W. Boja and M.J. Kuhar. Cocaine Receptors: In Vivo Labeling With ^{3}H-(-)Cocaine, ^{3}H-WIN 35,065-2 and ^{3}H-WIN 35,428. Synapse, 4, 390-392, 1989.

Witkin, J.M., S.R. Goldberg, J.L. Katz and M.J. Kuhar. Modulation of the Lethal Effects of Cocaine by Cholinomimetics. Life Sciences 45, 2295-2301, 1989.

Conrad, M.K. and M.M.S. Lo. B-cell hybridoma production by avidin-biotin mediated electrofusion. In: Electromanipulation in Hybridoma Technology (C. Borrebaeck and I. Hagen, Eds.), Stockton Press, New York. 1989.

Conrad, M.K., M.M.S. Lo and M.J. Kadan. Neurotoxicity of dopamine selective drugs. Alternative Methods in Toxicology, Vol. 7, 1989.

Conrad, M.K. and M.M.S. Lo. Facilitated Cell Fusion for Hybridoma Production. Methods in Enzymology, 184, pp. 641-643, Academic Press, New York, 1989.

Wilson, A.A., D.E. Grigoriadis, R.F. Dannals, H.T. Ravert and H.N. Wagner, Jr. A One-Pot Radiosynthesis of [^{125}I]Iodoazido Photoaffinity Labels. J. Labelled Compounds and Radiopharmaceuticals, 27(11), 1299-1305, 1989.

Titeler, M., R.A. Lyon, M.J. Kuhar, J.F. Frost, R.F. Dannals, S. Leonhardt, A. Bullock, L.T. Rydelek, D.L. Price and R.G. Struble. Mu Opiate Receptors are Selectively Labelled by ^{3}H-Carfentanil in Human and Rat Brain. Eur. J. Pharm., 167, 221-228, 1989.

Articles in Press

Boja, J.W. and M.J. Kuhar. ^{3}H-Cocaine Binding and Inhibition of [^{3}H]-Dopamine Uptake is Similar in Both the Rat Stritum and Nucleus Accumbens. Eur. J. Pharmacol., in press.

Abstracts Published

Lyon, R.A., M. Titeler, L.T. Rydelek, A.E. Bullock, J.J. Frost, R. F. Dannals and M.J. Kuhar. Properties of ^{3}H-Carfentanil Binding to Human and Rat Brain Opiate Receptors In Vitro. Soc. Neurosci. 42 (12), 105, 1988.

Sanchez-Roa, P.M., J. Sharkey, A.A. Wilson, D.E. Grigoriadis, R.F. Dannals, D.F. Wong and M.J. Kuhar. [^{125}I]-Spectramide: A Novel Benzamide Ligand Displaying Potent and Selective Effects at the D2 Dopamine Receptor. Soc. Neurosci. 165 (18), 411, 1988.

Battaglia, G., J. Sharkey, M.J. Kuhar and E.B. De Souza. Neuroanatomical Specificity of MDA- and MDMA-Induced Degeneration of Serotonin Neurons in Rat Brain. Soc. Neurosci. 222 (5), 557, 1988.

Johannessen, J.N., T.R. Insel, G. Battaglia, M.J. Kuhar and E.B. De Souza. MDMA Selectively Destroys Brain Serotonin Terminals in Rhesus Monkeys. Soc. Neurosci. 222 (6), 557, 1988.

Kuhar, M.J., J. Sharkey and D.E. Grigoriadis. Solubilization of the Dopamine Transporter. Soc. Neurosci. 376 (12), 930, 1988.

Ritz, M.C., E.J. Cone, J. Sharkey and M.J. Kuhar. Structure Activity Relationship of Cocaine and Related Compounds in Binding to Dopamine Transporters. Soc. Neurosci. 388 (17), 963, 1988.

Wilson, A.A., Grigoriadis, D.E., Lew, R. and Kuhar, M.J. Dopamine Transporters/Cocaine Receptors Selectively Labeled by a Novel Photoaffinity Probe: [^{125}I]DEEP. Soc. Neurosci. 15, 107, 1989.

Lew, R., Grigoriadis, D.E., Wilson, A.A. and Kuhar, M.J. Dopamine Transporter in Rat Caudate-Putamen is Glycosylated. Soc. Neurosci. 15(1), 107, 1989.

Goldberg, S.R., Kuhar, M.J., Katz, J.L. and Witkin, J.M. Modulation of the Lethal Effects of Cocaine by Cholinomimetics. Soc. Neurosci. 15(1), 252, 1989.

Boja, J.W. and Kuhar, M.J. ^{3}H-WIN 35,065-2 (^{3}H-WIN) Binding is Similar in Various Brain Regions of the Rat. Soc. Neurosci. 15(2), 1092, 1989.

Ritz, M.C., Boja, J.W., Carroll, F.I., Lewin, A.H. and Kuhar, M.J. ^{3}H-WIN 35,065-2 (^{3}H-WIN): A Ligand for Cocaine Receptors in Rat Striatum. Soc. Neurosci. 15(2), 1092, 1989.

Wong, D.F., Young, D., Young, L.T., Tune, L.E., Pearlson, G., Minkin, E., Meltzer, C. Cidis, Midha, K., Dannals, R.F., Ravert, H.T., Wilson, A.A., London, E.D., Wagner, Jr., H.N., Casanova, M., Klineman, J., Kuhar, M.J. and Gjedde, A. Validation of PET D_2 Dopamine Receptor Quantification Using [C-11] NMSP and [F-18] Haloperidol. Soc. Neurosci. 15(2), 1132, 1989.

Carson, S.G., M.J. Kadan, R.C. Douglas, K.A. Marcus and M.M.S. Lo. Tissue specific expression of a gene which is mutated in a MPP^{+}-selected PC12 mutant. Soc. Neurosci. 15, 958, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00108-04 MPL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

The Cocaine Receptor

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

P.I.	M.J. Kuhar	Chief, Neuroscience Branch, ARC
Others:	Boja, J.W.	Staff Fellow, ARC
	Lew, R.	Visiting Fellow, ARC
	Simantov, R.	Visiting Scientist, ARC

COOPERATING UNITS *(If any)*

None

LAB/BRANCH

Laboratory of Molecular Pharmacology, Neuroscience Branch

SECTION

None

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3	1 1/4	3/4

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

It was previously shown that cocaine binding to the dopamine transporter could be associated with the phenomenon of cocaine self-administration. Since cocaine and amphetamine are pharmacologically similar, the relationship between binding sites and self-administration of amphetamine was examined, and the data were compared with those for cocaine. While the self-administration of cocaine could be related to inhibition at the dopamine transporter, this could not be accomplished for amphetamine. This suggests that the mechanism of action of amphetamine is different from that for cocaine in that the amphetamine receptor is presumably not the same as the cocaine receptor.

Since binding studies are typically done with tissue from the caudate and putamen because they are easy to dissect, and since the brain regions associated with drug self-administration are limbic, we have compared the cocaine receptor and dopamine transporter in both these regions. Our data indicate that the cocaine binding sites and the dopamine transporter in the nucleus accumbens and striatum of the rat are identical.

An important goal is to understand the molecular nature of the cocaine receptor. Hence, we have undertaken several projects whose goal is to ultimately purify and characterize the dopamine transporter. We have succeeded in solubilizing the transporter from membranes and detecting it in its soluble state. We also have identified a photoaffinity probe which irreversibly binds to the transporter and allows a detailed characterization of its molecular weight, carbohydrate moiety, as well as other features. It is anticipated that a substantial effort in the molecular area will be continued in the future.

PUBLICATIONS
Z01 DA 00108-4 MPL

The Cocaine Receptor

Ritz, M.C., R.J. Lamb, S.R. Goldberg and M.J. Kuhar. Cocaine Self-Administration Appears to Be Mediated by Dopamine Uptake Inhibition. Prog. Neuro-Psychopharmacol. & Biol. Psychiat. 12, 233-239, 1988.

Kuhar, M.J., Ritz, M.C. and Sharkey, J. Cocaine receptors on dopamine transporters mediate cocaine-reinforced behavior. In: Mechanisms of Cocaine Abuse and Toxicity. D. Clouet, K. Asglan and R. Brown (Eds.), NIDA Research Monograph No. 88, U.S. Gov't. Printing Office, Washington, D.C., pp 14-22, 1988.

Ritz, M.C. and Kuhar, M.J. Relationship Between Self-administration of Amphetamine and Monoamine Receptors in Brain: Comparison With Cocaine. J. Pharmacol. Exp. Ther. 248, 1010-1017, 1989.

Lew, R., Grigoriadis, D.E., Sharkey, J. and Kuhar, M.J. Dopamine Transporter: Solubilization From Dog Caudate Nucleus. Synapse 3, 372-375, 1989.

Grigoriadis, D.E., Wilson, A.A., Lew, R., Sharkey, J.S. and Kuhar, M.J. Dopamine Transport Sites Selectively Labeled by a Novel Photoaffinity Probe: ^{125}I-DEEP. J. Neurosci. 9(8), 2664-2670, 1989.

Boja, J.W. and M.J. Kuhar. ^{3}H-Cocaine Binding and Inhibition of ^{3}H-dopamine Uptake is Similar in Both the Rat Striatum and Nucleus Accumbens. Eur. J. Pharmacol., in press.

ABSTRACTS

Kuhar, M.J., J. Sharkey and D.E. Grigoriadis. Solubilization of the Dopamine Transporter. Soc. Neurosci. 376 (12), 930, 1988.

Ritz, M.C., E.J. Cone, J. Sharkey and M.J. Kuhar. Structure Activity Relationship of Cocaine and Related Compounds in Binding to Dopamine Transporters. Soc. Neurosci. 388 (17), 963, 1988.

Wilson, A.A., Grigoriadis, D.E., Lew, R. and Kuhar, M.J. Dopamine Transporters/Cocaine Receptors Selectively Labeled by a Novel Photoaffinity Probe: [^{125}I]DEEP. Soc. Neurosci. 15, 107, 1989.

Lew, R., Grigoriadis, D.E., Wilson, A.A. and Kuhar, M.J. Dopamine Transporter in Rat Caudate-Putamen is Glycosylated. Soc. Neurosci. 15(1), 107, 1989.

The Cocaine Receptor

Boja, J.W. and Kuhar, M.J. ^{3}H-WIN 35,065-2 (^{3}H-WIN) Binding is Similar in Various Brain Regions of the Rat. Soc. Neurosci. 15(2), 1092, 1989.

Ritz, M.C., Boja, J.W., Carroll, F.I., Lewin, A.H. and Kuhar, M.J. ^{3}H-WIN 35,065-2 (^{3}H-WIN): A Ligand for Cocaine Receptors in Rat Striatum. Soc. Neurosci. 15(2), 1092, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00112-03 MPL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Drug Receptors, Neurotransmitters and Addiction

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	Kuhar, M.J.	Chief, Neuroscience Branch, ARC
Others:	Goldberg, S.	Chief, Preclinical Research Branch
	Ritz, M.C.	Staff Fellow, Preclinical Research Branch
	De Souza, E.B.	Chief, Neurobiology Laboratory, Neuroscience Branch
	Sharkey, J.	Department of Clinical Neurosciences, Western General Hospital
	Porrino, L.	NINCDS

COOPERATING UNITS *(if any)*

None

LAB/BRANCH

Laboratory of Molecular Pharmacology, Neuroscience Branch

SECTION

None

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3	2 1/4	3/4

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This project has several facets and involves the study of several neurotransmitters and receptors that are involved in drugs of abuse and addiction. In one of our studies, we examined the effect of substituted amphetamines on sertonergic axon terminals in forebrain. Preliminary biochemical studies had shown that these amphetamines cause a reduction of sertonergic markers in the brain suggesting a degeneration of serotonergic nerve terminals. The immunocytochemical study that was carried out presented clear evidence for degeneration of nerve terminals after higher doses of these drugs.

Another interesting area involved the cholinergic muscarinic properties of cocaine. In a detailed study, we showed that cocaine was a weak anticholinergic drug. The evidence was derived from both binding and physiological studies. One question was whether or not this anticholinergic property of cocaine was involved in the toxicity of cocaine, particularly at the higher doses of cocaine where muscarinic inhibition could be obtained. However, in toxicological studies it was found that pretreatment of animals with anticholinergic drugs did not decrease the LD50 values for cocaine. These latter findings suggest that the anticholinergic properties of cocaine are not involved in its lethality.

In another study, it was shown that sertonergic drugs can modify the self administration of amphetamine in rats while having no effect on the self administration of cocaine. These data point out the possibility of the involvement of sertonergic mechanisms in self administration and, in particular, the involvement of sertonergic mechanisms in the antagonism of the self administration of amphetamine.

PUBLICATIONS
Z01 DA 00112-03 MPL

Drug Receptors, Neurotransmitters and Addiction

O'Hearn, E., G. Battaglia, E.B. De Souza, M.J. Kuhar and M.E. Molliver. Methylenedioxyamphetamine (MDA) and Methylenedioxymethamphetamine (MDMA) Cause Selective Ablation of Serotonergic Axon Terminals in Forebrain: Immunocytochemical Evidence for Neurotoxicity. J. Neurosci. 8 (8), 2788-2803, 1988.

Sharkey, J., M.C. Ritz, J.A. Schenden, R.C. Hanson and M.J. Kuhar. Cocaine Inhibits Muscarinic Cholinergic Receptors in Heart and Brain. J. Pharmacol. Exp. Ther. 246 (3), 1048-1052, 1988.

Kuhar, M.J. Overview. In: Receptor Localization: Ligand Autoradiography. F.M. Leslie and C.A. Altar (Eds.), Alan R. Liss, New York, pp 1-7, 1988.

Kuhar, M.J. and De Souza, E.B. Autoradiographic Imaging: Localization of Binding Sites other than Neurotransmitter Receptors. In: Visualizations of Brain Functions. D. Ottoson and W. Rostene (Eds.), M Stockton Press, pp. 57-66, 1989.

Porrino, L.J., Ritz, M.C., Goodman, N.L., Sharpe, L.G., Kuhar, M.J. and Goldberg, S.R. Differential Effects of the Pharmacological Manipulation of Serotonin Systems on Cocaine and Amphetamine Self-Administration in Rats. Life Sciences 45, 1529-1535, 1989.

Witkin, J.M., S.R. Goldberg, J.L. Katz and M.J. Kuhar. Modulation of the Lethal Effects of Cocaine by Cholinomimetics. Life Sci. 45, 2295-2301, 1989.

ABSTRACTS

Lyon, R.A., M. Titeler, L.T. Rydelek, A.E. Bullock, J.J. Frost, R. F. Dannals and M.J. Kuhar. Properties of ^{3}H-Carfentanil Binding to Human and Rat Brain Opiate Receptors In Vitro. Soc. Neurosci. 42 (12), 105, 1988.

Battaglia, G., J. Sharkey, M.J. Kuhar and E.B. De Souza. Neuroanatomical Specificity of MDA- and MDMA-Induced Degeneration of Serotonin Neurons in Rat Brain. Soc. Neurosci. 222 (5), 557, 1988.

Johannessen, J.N., T.R. Insel, G. Battaglia, M.J. Kuhar and E.B. De Souza. MDMA Selectively Destroys Brain Serotonin Terminals in Rhesus Monkeys. Soc. Neurosci. 222 (6), 557, 1988.

Goldberg, S.R., Kuhar, M.J., Katz, J.L. and Witkin, J.M. Modulation of the Lethal Effects of Cocaine by Cholinomimetics. Soc. Neurosci. 15(1), 252, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00107-04 MPL

PERIOD COVERED

Measuring Drug Receptors In Vivo

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

October 1, 1988 to December 31, 1989

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

Investigators:

PI:	Kuhar, M.J.	Chief, Neuroscience Branch, ARC
Others:	Wong, Dean	Division of Nuclear Medicine, JHU
	Grigoriadis, Dimitri	Neurobiology Laboratory, ARC
	Wagner, H.N.	Division of Nuclear Medicine, JHU
	Sanchez-Roa, Patricia	

COOPERATING UNITS *(If any)*

Division of Nuclear Medicine, Johns Hopkins University School of Medicine

LAB/BRANCH

Laboratory of Molecular Pharmacology, ARC

SECTION

None

INSTITUTE AND LOCATION

NIDA Addiction Research Center, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3	2 1/4	3/4

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Because of the rather dramatic advances in brain imaging, and because of the possibility of measuring brain receptors in vivo by PET scanning, this project has important potential for studying drug receptors in drug abusing populations of human beings. One of the ongoing studies is the development of new ligands for dopamine receptors. Since cocaine is thought to exert its action by potentiating dopaminergic neurotransmission, ligands for measuring dopamine receptors are clearly important in the field of drug abuse. While several ligands for these receptors exists, ligands of lower affinity which would show competition with endogenous dopamine are currently being sought. We have identified a compound, spectramide, which is a novel benzamide that is potent and selective for the dopamine D2 receptor. This compound has an affinity for the receptor such that competition with endogenous ligands might be observed. If this were possible, we could detect the secondary effects of the presence of cocaine acting at its receptor.

Also, we have been developing new ligands for the cocaine receptor. Cocaine can be used for specific in vivo binding and therefore for PET scanning studies. However, its in vivo specific binding is low and its duration of action is short. Thus, we have examined the in vivo labeling of two analogues of cocaine that are more potent at the receptor and lack an ester group which is a metabolically vulnerable. These compounds, WIN 35,065-2 and WIN 35,428 are both resistent to metabolic degradation and in fact show much higher specific binding in vivo and a longer duration of action at the receptors. Hence, both of these compounds are suitable candidates for PET scanning ligands for the cocaine receptor and both compounds appear to be superior to cocaine as PET scanning tools.

PUBLICATIONS
Z01 DA 00107-04 MPL

Measuring Drug Receptors In Vivo

Wagner, H.N., Jr., Weinberger, D.R., Kleinman, J.E., Casanova, M.F., Gibbs, C.J., Jr., Gur, R.E., Hornykiewicz, O., Kuhar, M.J., Pettegrew, J.W. and Seeman, P. Neuroimaging and Neuropathology. Schizophrenia Bulletin 14 (3), 383-397, 1988.

Wong, D.F. and Kuhar, M.J. In Vivo PET and SPECT Receptor Imaging: New Technology and Tactics for Receptor Measurement. In Neuroreceptors and Signal Transduction. S. Kito, T. Segawa, K. Kuriyama, M. Tohyama and R.W. Olsen (Eds.), Plenum Press, New York, pp. 181-193, 1988.

Kuhar, M.J. Perspectives. In Brain Imaging Techniques and Applications. N.A. Sharif and M.E. Lewis (Eds.), John Wiley & Sons, New York, pp. 13-17, 1989.

Sanchez-Roa, P.M., Grigoriadis, D.E., Wilson, A.A., Sharkey, J., Dannals, R.F., Villemagne, V.L., Wong, D.F., Wagner, H.N. and Kuhar, M.J. [^{125}]I-Spectramide: A Novel Benzamide Displaying Potent and Selective Effects at the D_2 Dopamine Receptor. Life Sciences 45, 1821-1829, 1989.

Scheffel, U. J.W. Boja and M.J. Kuhar. Cocaine Receptors: In Vivo Labeling With ^{3}H-(-)Cocaine, ^{3}H-WIN 35,065-2 and ^{3}H-WIN 35,428. Synapse 4, 390-392, 1989.

ABSTRACTS

Sanchez-Roa, P.M., J. Sharkey, A.A. Wilson, D.E. Grigoriadis, R.F. Dannals, D.F. Wong and M.J. Kuhar. [^{125}I]-Spectramide: A Novel Benzamide Ligand Displaying Potent and Selective Effects at the D2 Dopamine Receptor. Soc. Neurosci. 165 (18), 411, 1988.

Wong, D.F., Young, D., Young, L.T., Tune, L.E., Pearlson, G., Minkin, E., Meltzer, C. Cidis, Midha, K., Dannals, R.F., Ravert, H.T., Wilson, A.A., London, E.D., Wagner, Jr., H.N., Casanova, M., Klineman, J., Kuhar, M.J. and Gjedde, A. Validation of PET D_2 Dopamine Receptor Quantification Using [C-11] NMSP and [F-18] Haloperidol. Soc. Neurosci. 15(2), 1132, 1989.

2. Neuropharmacology Laboratory - Edythe D. London, Ph.D., Chief

Overview

The Neuropharmacology Laboratory conducts studies designed to elucidate neurochemical and electrophysiological mechanisms and to identify brain loci that mediate the behavioral and physiological effects of abused drugs. Experiments focus on acute and chronic effects of abused drugs in biochemical and physiological systems *in vitro* as well as in intact organisms. Major areas of concentration include the biological effects of psychomotor stimulants, opioids, and nicotine. Efforts are directed at elucidating molecular mechanisms, involving neurotransmitter systems, that may be targets of abused drugs. Investigation of endocrine or nutritional factors which might modify transmission in neuronal pathways which mediate the effects of abused drugs is a major directive. It is anticipated that the information obtained will be helpful in designing new treatment strategies, including the development of medications, for substance abuse.

The Laboratory is comprised of the following cooperating units: Brain Imaging (E.D. London, Chief), Neurochemistry (T.-P. Su, Chief), and Neurophysiology (J. Bell, Chief). A wide variety of approaches is used in relating neurochemical findings to physiological measures *in vitro* and physiological and behavioral parameters in intact animals and humans. Techniques used include the following: *in vivo* and *in vitro* receptor binding, purification and identification of endogenous neuroactive substances, electrophysiological studies of single neurons and neuronal circuits, isolated tissue bath preparations, and cerebral metabolic mapping, using positron emission tomography (PET) in humans and quantitative autoradiography in laboratory animals.

Collaborative studies during FY89 involved the Neurobiology Laboratory, ARC; the Psychology of Vulnerability and Cognitive Studies Laboratory, ARC; the Preclinical Pharmacology Branch, ARC; the Division of Nuclear Medicine, the Johns Hopkins Medical Institutions; the Department of Pharmacology and Toxicology, University of Maryland School of Pharmacy; Stanford Research Institute; New Jersey Institute of Technology; and University of Kentucky.

Summary of Ongoing Research

A. Cerebral Effects of Abused Drugs: Brain Imaging in Humans and Laboratory Animals

The objective of these studies is to identify brain areas that show alterations in function due to the administration of abused drugs. Information obtained would lead to a better understanding of the mechanisms involved in drug self-administration, tolerance

and dependence. The approach that has been used is mapping and quantitation of regional cerebral metabolic rates for glucose (rCMRglc) using positron emission tomography (PET) in human volunteers and ex *vivo* quantitative autoradiography in rats.

Using the PET [^{18}F]fluorodeoxyglucose (FDG) method in human volunteers, and a double-blind crossover design, we showed that a dose of morphine which was rated as euphorigenic in subjective self-reports significantly reduced rCMRglc in some telencephalic areas with no significant effect in lower brain areas.

Similar findings were obtained with intravenous cocaine. The results of these studies, taken together with previous reports demonstrating that other abused drugs (barbiturates, benzodiazepines, and amphetamine) also reduce telencephalic rCMRglc, suggest that the reduction of telencephalic function, as indicated by rCMRglc, is associated with drug-induced reward/reinforcement.

Studies in rats have afforded greater anatomical resolution than that obtained with PET, but have been limited by the inability to relate cerebral metabolic findings directly to simultaneous effects on mood and feeling state. Responses to abused drugs from several drug classes have been studied. Particular attention has been given to nicotine, cocaine, and other psychomotor stimulants. Acute nicotine stimulated cerebral glucose utilization in a pattern that closely followed the distribution of receptors for radiolabelled nicotine *in vitro*. Studies of chronic nicotine effects on rCMRglc have provided important clues about tolerance to the action of this compound. Cocaine and 3,4-methylenedioxymethamphetamine (MDMA) stimulated rCMRglc in components of the extrapyramidal motor system and reduced rCMRglc in the lateral habenula. A greater sensitivity to cocaine was observed in the responses of Lewis rats as compared with Fischer rats, indicating a genetic difference. MDMA also produced activation in thalamic nuclei and in the visual cortex. The findings were consistent both with a psychomotor stimulant action of MDMA, similar to that of cocaine and amphetamine, and with the production of visual hallucinations by MDMA.

Projected work in this area will assess effects of cocaine on regional cerebral blood flow (rCBF) in humans with techniques that potentially would allow greater time resolution and anatomical localization than that obtained with FDG. The objective is to relate effects of cocaine on specific feeling states (e.g., rush, feeling good, craving) to changes in rCBF. In addition, effects of morphine and cocaine on EEG parameters will be related to subjective effects as well as PET findings.

B. Physiological Effects of Opioids and the Opioid Abstinence Syndrome

This project is designed to help elucidate the physiological effects of morphine. Other objectives include delineation of the anatomical systems in rat brain and spinal cord that mediate the acute and chronic effects of opioid agonists and antagonists, and that contribute to the opioid abstinence syndrome.

Several lines of evidence indicate that the physiological effects of opioids on cellular membranes involve the Ca^{2+} ion, and an understanding of the role of calcium in opioid effects is fundamental to elucidating mechanisms of the acute effects of opioids, as well as tolerance and physical dependence. An extension of earlier work in this laboratory, demonstrating antagonism of opioid-induced respiratory depression and tachycardia by calcium channel blockers, revealed that this antagonism was not due to altered morphine pharmacokinetics. Antagonism of respiratory depressant effects of opioids by Ca^{2+} antagonists, with simultaneous facilitation of analgesia, could lead to drug combinations with fewer opioid side effects and less potential for tolerance and physical dependence. Projected studies will focus on interactions between opioids and calcium channel antagonists as they relate to opioid euphoria and other parameters in human volunteers.

Studies of the opioid abstinence syndrome have provided valuable insight into mechanisms of opioid tolerance and withdrawal, and are relevant to the clinical management of opioid abuse. *In vivo* studies in rats demonstrated complete tolerance to the effects of subchronic morphine treatment on cerebral metabolism, but marked hypermetabolism induced during naloxone-precipitated morphine withdrawal. Cerebral and spinal hypermetabolism during morphine withdrawal as well as its antagonism by clonidine showed a wide distribution in the rat brain, with involvement of various limbic and hypothalamic areas not previously implicated in these phenomena. Studies on the isolated spinal cord of the neonatal rat provided electrophysiological evidence that increased release of substance P from primary afferents contributes to the opioid withdrawal syndrome. Furthermore, evidence from electrophysiological and autoradiographic studies suggested that corticotropin releasing factor (CRF) is a primary afferent neurotransmitter in the neonatal rat spinal cord.

We have previously shown that presynaptic changes contribute to opioid withdrawal in the isolated spinal cord of the neonatal rat. The role of postsynaptic change in spinal withdrawal is unknown. Because of the complexity of intact systems such as the isolated spinal cord, we have developed techniques for studying primary cultures of rat spinal cord. We plan to use electrophysiological techniques to determine the potential contribution of postsynaptic changes in dorsal horn neurons to the opioid abstinence syndrome.

C. Multiple Opioid Receptor Subtypes

Recent studies indicate that opioid receptors may be involved in behavioral conditioning. Weekly injections of naltrexone in rats for eight weeks induced hypersensitivity to the behavior-suppressant effect of naltrexone and produced alterations in kappa and delta opioid receptors in various brain areas. Mu opioid receptors, however, were unaffected. These results suggest that conditioning processes may involve kappa and delta opioid receptors and/or endogenous opioid peptides.

A protein extracted from plasma of hibernating woodchucks (hibernation induction trigger, HIT) induces hibernation, an opioid effect which is blocked by naloxone. Studies were conducted to determine if HIT could also extend organ survival time. HIT dramatically extended preservation time in an autoperfusion multi-organ preparation [from 14 h to 44 h]. The mechanism, which may involve facilitation of microcirculation, may also play a role in the ability of HIT to induce hibernation.

D. *Kappa* and *Sigma* Properties of Antinociceptive Drugs in the Dog

The mechanisms of action of several antinociceptive drugs, including d- and l-ketocyclazocine, flupirtine, and a combination of pentazocine and tripelennamine, were studied in chronic spinal dogs. The observable pharmacological activity of ketocyclazocine probably resides in the l- isomer as no demonstrable activity was associated with the d- isomer. Analgesia induced by flupirtine may not involve opioid mechanism and may occur primarily at supraspinal sites. Although the combination of tripelennamine and pentazocine was the form preferred by the abusers, no consistent pattern among the interactions could be observed in the chronic spinal dog. Actions of tripelennamine may not be opiate-like. Tripelennamine also did not appear to antagonize the apparent σ activity produced by SKF-10047, contrasting previously reported results that tripelennamine partially antagonized the discriminative stimulus produced by SKF-10047 in rat.

E. *Sigma* and Phencyclidine Receptors

A major effort has been directed at discovering the biological role of σ binding sites, which may represent a link between the brain, the endocrine system and the immune system. Efforts to purify and identify an endogenous ligand for σ binding sites have continued. In addition, experiments were directed at functional effects, which could be attributed to σ receptor interactions. A cultured cell line (NCB 20) that expresses σ receptors is being utilized for electrophysiological studies. Electrical effects produced by σ ligands were characterized by voltage clamp and patch clamp techniques. Preliminary evidence identified the K^+ leak conductance ion channel as a potential target of σ action.

Structure-activity relationship studies using molecular modeling techniques revealed that σ ligands possess the following structural features: a primary, secondary or tertiary but not quaternary amine; a hydrophobic cluster composed of phenyl rings or other lipophilic substituents; and an intermediate chain. Electrostatic potential calculations on the van der Waals surfaces of σ ligands suggested that unprotonated forms of the ligands represent the active species for σ receptor interactions. The results explained why progesterone, which lacks nitrogen, interacts with σ receptors. Our findings suggest that the β ring of progesterone represents the hydrophobic moiety for σ receptor interaction, and that the 20-carbonyl oxygen may mimic nitrogen in other σ ligands.

The guinea-pig vas deferens was found to contain σ but not PCP receptors. In NCB-20 cells, two types of binding sites were labelled by the prototypic σ ligand, [^{3}H]N-allylnormetazocine. Whereas the high affinity site was the σ receptor, as described in the rodent brain, the low affinity site appeared to be linked to tonic K^+ channels. The finding may provide an approach for studying such channels. Subcellular fractionation studies indicated that σ receptors reside in non-synaptic components of neurons.

The NMDA receptor complex, which is activated by excitatory neurotransmitters, is a target for the commonly abused psychotomimetic drug, phencyclidine (PCP). This receptor participates in fundamental CNS functions such as neuronal plasticity and memory, and its excessive activity has been implicated in pathologies associated with seizures or anoxia-ischemia. It is regulated by numerous endogenous factors, including ions, polyamines and glycine. In our laboratory we study endogenous mechanisms and nutritional factors involved in regulation of NMDA/PCP receptors, which may partially determine individual sensitivity to PCP. We have observed that function of the NMDA receptor complex is regulated by redox status.

Future directions for this area of research include the following: 1) final purification and characterization of sigmaphin, 2) studies of subcellular localization and biochemical characterization of σ binding sites in guinea-pig brain, 3) development of new σ ligands, 4) further elucidation, by electrophysiological techniques, of functional consequences of σ receptor interactions, and 5) imaging σ and PCP receptors using PET.

F. Nicotinic Receptors: Imaging, Regulation, and Structure-Activity Relations

This project is directed at elucidating the actions of nicotine at various levels of organization, from the receptor to the intact animal. The importance of this project stems from the

following: 1) nicotine is a prototypic drug of abuse, 2) receptor and ion channel interactions which mediate the action of nicotine are fundamental to processes involving acetylcholine, a major neurotransmitter in the brain and periphery, and 3) interactions at nicotinic synapses and related ion channels represent a model for coupling of receptor mediated events to biological processes.

In vivo mapping of the nicotinic receptor with radiolabelled nicotine have provided important clues about the neuroanatomical substrates of the action of this compound. These studies lay the groundwork for the development of probes to study nicotinic receptors in the human brain with PET.

To understand the molecular basis of nicotine addiction, we study endogenous mechanisms regulating the function of brain nicotinic cholinergic receptors. We determined the patterns of interaction of competitive and allosteric ligands binding to various domains of the receptor complex using receptor binding techniques. Binding of noncompetitive blockers for the nicotinic receptor, mecamylamine and chlorpromazine, inside the cationic channel operated by this receptor, is a biochemical marker of receptor activity. We use this assay to determine functional alterations of the nicotinic receptor by various endogenous modulators. Future studies will assess functional relations of nicotinic receptors with dopaminergic systems, which may contribute to nicotine dependence.

G. Regulators of the $GABA_A$ Receptor Complex: Anxiolytics and Endogenous Steroids

The ionotropic $GABA_A$ receptor is a target of anxiolytic/hypnotic drugs, including benzodiazepines (BZ's). Ongoing studies focus on various binding sites and endogenous substances which regulate the function of this complex. *In vivo* studies using the deoxyglucose technique in rats demonstrated that diazepam, which does not differentiate between benzodiazepine (BZ) receptor subtypes *in vitro*, reduced rCMRglc primarily in regions rich in BZ type I receptors. Similar findings were obtained with CL 218,872, a drug which shows selectivity for type I BZ receptors. These findings support a functional distinction between BZ receptor subtypes.

Endogenous steroids are powerful modulators of $GABA_A$ receptors. During pregnancy and the postpartum period, ligand binding to $GABA_A$ receptors in the brain and uterus was altered, suggesting steroid regulation. Such phenomena may influence the physiological changes accompanying pregnancy and the puerperium, and may explain the altered sensitivity to anxiolytics and hypnotics during pregnancy and the puerperium.

Pregnenolone sulfate behaves as an allosteric antagonist of the

$GABA_A$ receptor *in vitro*. We observed that this steroid also shows $GABA_A$ antagonistic actions *in vivo*, as it reduced barbiturate sleep time, suggesting that it contributes to alterations of neuronal excitability and CNS arousal. We examined [^{3}H]pregnenolone sulfate binding in the rat brain, noting that it appears to interact at the interface of the receptor with associated phospholipids. Ongoing biochemical and electrophysiological studies focus on interactions of other steroids with $GABA_A$ receptors.

H. HIV Infection and Drug Abuse Alter Expression/Function of Neuroimmune Receptors

As drug abusers are at high risk for infection by human immunodeficiency virus (HIV), several studies relating immunopharmacology and treatments for CNS infection by HIV were performed. It was of interest to determine if abused drugs, especially those commonly abused by intravenous injection, can affect immune function. Ongoing studies demonstrated a dose-dependent reduction of circulating T-lymphocytes associated with chronic morphine treatment in the mouse, indicating that immunocompetency is compromised by use of this drug. The extension of this study will focus on specificity of the immunological effect, attempting to identify specific neurohumoral factors or receptors which might mediate immunosuppressive effects of opioids and other abused drugs. Other studies attempt to identify the sites and mechanisms of action of peptide T and GP-120, a potential therapeutic agent and a glycoprotein component of the HIV viral envelope, respectively.

Publications

London, E.D., R.J. Connolly, M. Szikszay, J.K. Wamsley and Dam M.: Effects of nicotine on local cerebral glucose utilization in the rat.
J. Neurosci. 8: 3920-3928, 1988.

Weissman, A.D., Su, T.-P., Hedreen, J.C. and London, E.D.: *Sigma* receptors in postmortem human brains. J. Pharmacol. Exp. Ther. 247: 29-33, 1988.

McManus, O.B., Weiss, D.S., Spivak, C.E., Blatz, A.L. and Magleby, K.L.: Fractal models are inadequate for the kinetics of four different ion channels. Biophys. J. 54: 859-870, 1988.

Oeltgen, P.R., Nuchols, P.A., Nilekani, S.P., Spurrier, W.A., and Su, T.-P.: Further studies on opioids and hibernation. Life Sci. 43: 1565-1574, 1988.

Majewska, M.D.: Interaction of ethanol with the $GABA_A$ receptor in the rat brain: Possible involvement of endogenous steroids. Alcohol 5: 269-273, 1988.

Majewska, M.D., Mienville, T.M., and Vicini, S.: Neurosteroid pregnenolone sulfate antagonizes electrophysiological responses to GABA in neurons. Neurosci. Lett. 90: 279-284, 1988.

Tabata, H., Kimes, A.S., Robinson, P., and Rapoport, S.I.: Stability of brain incorporation of plasma palmitate in unanesthetized rats of different ages. Exp. Neurol. 102: 221-229, 1988.

Kimes, A.S., and London, E.D.: Glucose utilization in the rat brain during chronic morphine treatment and naloxone-precipitated morphine withdrawal. J. Pharmacol. Exp. Ther. 248: 538-545, 1989

Fanelli, R.J., Walovitch, R.C., Jasinski, D.R., and London, E.D.: Naloxone fails to alter local cerebral glucose utilization in the rat. Pharmacol. Biochem. Behav. 31: 481-485, 1989.

London, E.D.: Effects of abused drugs on cerebral glucose metabolism. J. Neuropsychiat. Clin. Neurosci. 1, Suppl. 1: S30-S36, 1989.

Majewska, M.D., Ford-Rice, F., and Falkay, G.: Pregnancy-induced alterations of GABA-A receptor sensitivity in maternal brain: an antecedent of post-partum blues? Brain Res. 482: 397-401, 1989.

Bell, J.A., and De Souza, E.B.: Functional corticotropin-releasing factor (CRF) receptors in the neonatal rat spinal cord. Peptides 9: 1317, 1989.

Broussolle, E.P., Wong, D.F., Fanelli, R.J., and London, E.D.: *In vivo* specific binding of [^{3}H]l-nicotine in the mouse brain. Life Sci. 44: 1123-1132, 1989.

Spivak, C.E., Yadav, J.S., Shang, W.C., Hermsmeier, M., and Gund, T.M.: Carbamyl analogues of potent nicotinic agonists: Pharmacology and computer-assisted molecular modeling study. J. Med. Chem. 32: 305-309, 1989.

Cadet, J.L., Della-Puppa, A., and London, E.D.: Involvement of nigrotectal-reticulospinal pathways in the iminodipropionitrile (IDPN) model of spasmodic dyskinesias: A 2-deoxy-D-[1-^{14}C]glucose study in the rat. Brain Res. 484: 57-64, 1989.

Ori, C., Ford-Rice, F.Y., and London, E.D.: Effects of nitrous oxide and halothane on *mu* and *kappa* opioid receptors in guinea-pig brain. Anesthesiology 70: 541-544, 1989.

Vaupel, D.B., Nickel, B., and Becketts, K.: Flupirtine antinociception in the dog is primarily mediated by nonopioid supraspinal mechanisms. Eur. J. Pharmacol. 162: 447-456, 1989.

Kimes, A.S., Kumor, K., McCullough, K., Holtzclaw, D., Teller, D., Dobyan, D. and Spector, D.: Effects of acute and chronic acyclovir on canine renal function. J. Pharmacol. Exp. Ther. 249(2): 483-491, 1989.

Majewska, M.D., Parameswaran, S., Vu, T., and London, E.D.: Divergent ontogeny of *sigma* and phencyclidine binding sites in the rat brain. Dev. Brain Res. 47: 13-18, 1989.

Weissman, A.D., Marquis, K.L., Moreton, J.E., and London, E.D.: Effects of self-administered phencyclidine on regional brain uptake of 2-deoxy-D-[1-^{14}C]glucose. Neuropharmacology 28: 575-583, 1989.

Waller, S.B., and London, E.D.: Choline acetyltransferase activity and muscarinic binding in brain regions of aging Fischer-344 rats. Neurochem. Int. 14: 483-490, 1989.

Moroi, S.E., Mervis, R.F., London, E.D., and Ingram, D.K.: dietary restriction suppresses age-related changes in dendritic spines. Neurobiol. Aging 10: 317-322, 1989.

Vaupel, D.B.: Tripelennamine interactions with the psychotomimetic *sigma* agonist N-allylnormetazocine. Pharmacol. Biochem. Behav. 33: 717-720, 1989.

Spivak, C.E., Waters, J.A. and Aronstam, R.S.: Binding of semirigid agonists to nicotinic and muscarinic receptors. Mol. Pharmacol. 36: 177-184, 1989.

London, E.D., Ball, M.J, and Waller, S.B.: Nicotinic binding sites in cerebral cortex and hippocampus in Alzheimer's dementia. Neurochem. Res. 14: 745-750, 1989.

London, E.D., Margolin, R.A., Wong, D.F., Links, J.M., LaFrance, N.D., Cascella, N.G., Broussolle, E.P.M., Wagner, H.N., Jr., Snyder, F.R., and Jasinski, D.R.: Cerebral glucose utilization in human heroin addicts: Case reports from a positron emission tomographic study. Res. Comm. Substances of Abuse 10: 141-144, 1989.

Majewska, M.D., Bluet-Payot, M.T., Robel, P.: Pregnenolone-sulfate: antagonists barbiturate-induced sleep. Pharmacol. Biochem. Behav. 33: 701-703, 1989.

Della-Puppa, A., Ford-Rice, F.Y., Snyder, F.R., Cone, E., and London, E.D.: Time course of verapamil interaction with morphine effects on physiological parameters in rats. J. Pharm. Pharmacol. 41: 617-623, 1989.

Vaupel, D.B.: Interactions between pentazocine and tripelennamine on autonomic and nociceptive measures in the dog. Pharmacol. Biochem. Behav. 33: 245-251, 1989.

Wilkerson, G. and London, E.D.: Effects of methylenedioxymethamphetamine 'Ecstasy' on local cerebral glucose utilization in the rat. Neuropharmacology 28: 1124-1138, 1989.

McManus, O.B., Spivak, C.E., Blatz, A.L., Weiss, D.S., and Magleby, K.L.: Fractal models, Markov models, and channel kinetics. Biophys. J. 55: 383-385, 1989.

Bell, J.A.: Naloxone-induced facilitation of C-fiber reflexes is reduced by chronic morphine. Eur. J. Pharmacol. 168: 101-105, 1989

Oeltgen, P. R., Nilekani, S. P., Nuchols, P. A., Spurrier, W. A., Su, T.-P, Chien, S., Proffitt, G. E. and Mahony, C.: Identification of opioid receptor ligand(s) involved in summer-induced and natural winter hibernation. In: Living in the Cold, A. Malan and B. Canguihem, eds., pp. 97-104, Lolloque INSERM/John Libbey Eurotext Ltd., France, 1989.

Articles in Press

Vaupel, D.B., Cone, E.J., Johnson, R.E., and Su T.-P.: *Kappa* opioid partial agonist activity of the enkephalin-like pentapeptide BW942C based on urination and *in vitro* studies in humans and animals. J. Pharmacol. Exp. Ther.

London, E.D., Casella, N.G., Wong, D.F., Phillips, R.L., Dannals, R.F., Links, J.M., Herning, R., Grayson, R., Jaffe, J.H., and Wagner, H.N.: Cocaine-induced reduction of glucose utilization in human brain: a study using positron emission tomography and [fluorine^{18}]fluorodeoxyglucose. Arch. Gen. Psychiat.

London, E.D., Broussolle, E.P.M., Links, J.M., Wong, D.F., Cascella, N.G., Dannals, R.F., Sano, M., Herning, R., Snyder, F.R., Rippetoe, L.R., Toung, T.J.R., Jaffe, J.H., and Wagner, H.N., Jr.: Morphine-induced metabolic changes in human brain - studies with positron emission tomography and 2-[^{18}F]fluoro-2-deoxy-D-glucose. Arch. Gen. Psychiat.

Della Puppa, A. and London, E.D.: Cerebral metabolic effects of *sigma* ligands in the rat. Brain Res.

Vu, T.H., Weissman, A.D., and London, E.D.: Pharmacological characteristics and distributions of *sigma* and phencyclidine binding sites in the animal kingdom. J. Neurochem.

Takayama, H., Majewska, M.D. and London, E.D.: Interactions of noncompetitive inhibitors with nicotinic receptors in the rat brain. J. Pharmacol. Exp. Ther.

Majewska, M.D., Falkay, G., and Baulieu, E.E.: Modulation of uterine $GABA_A$ receptors during gestational age and by tetrahydroprogesterone. Eur. J. Pharmacol.

Kimes, A.S., Bell, J.A., and London, E.D.: Clonidine antagonizes increased glucose metabolism during naloxone-precipitated morphine withdrawal. Neuroscience.

Schindler, C.W., Wu, X.-Z., Su, T.-P., Goldberg, S.R. and Katz, J.L.: Enhanced sensitivity to the behavioral effects of naltrexone in rats: A conditioning phenomenon associated with opioid receptor changes. J. Pharmacol. Exp. Ther.

London, E.D.: Studies of S receptors and metabolic responses to S ligands in the brain. In: *Sigma*, PCP and NMDA Receptor Systems, E.B. DeSouza, E.D. London, and D. Clouet, eds. NIDA Research Monographs.

London, E.D.: Effects of nicotine on cerebral metabolism. In: Ciba Foundation Symposium Biology of Nicotine Dependence, J. Marsh, ed.

London, E.D., Wilkerson, G.W., Ori, C. and Kimes, A.S.: Central action of psychomotor stimulants on glucose utilization in extrapyramidal motor areas of the rat brain. Brain Res.

London, E.D., Fanelli, R.J., Kimes, A.S., and Moses, R.L.: Effects of chronic nicotine on cerebral glucose utilization in the rat. Brain Res.

Majewska, M.D. and Vaupel, B.: Steroid control of uterine motility via $GABA_A$ receptor: a model mechanism. XXXI Intern. Congress of Physiology, Helsinki, 1989.

Su, T.-P., London, E.D., and Jaffe, J.H.: Steroid binding at S "opioid" receptor Response to a technical comment, Science, 246:1637-1638, 1989.

Su, T.-P. and Wu, X.-A.: Guinea-pig vas deferens contains σ but not phencyclidine receptors. Neurosci. Lett., 1990.

Su, T.-P.: Pharmacological characterization of σ receptors. In: *Sigma*, PCP and NMDA Receptor Systems, E.B. De Souza, E.D. London, and D. Clouet, eds. NIDA Research Monographs, 1990.

Su, T.-P., Shuklar, K. and Gund, T.: Steroid binding at σ receptors: CNS and immunological implications. In: Steroids and Neuronal Activity (Ciba Found. Symposium 153), D. J. Chadwick, ed., Wiley, Chicester, UK 1990.

Su, T.-P., Chien, S. F. and Oeltgen, P. R.: Hibernation induction trigger (HIT) extends preservation time in an autoperfusion multiorgan preparation. Clin. Pharmacol. Ther., 1990.

Abstracts Published

Della Puppa, A., Kimes, A.S., Vu, T. and London, E.D.: Cerebral metabolic mapping of the effects of agonists and antagonists for σ binding sites. Brain 89. XIVth International Symposium Cerebral Blood Flow and Metabolism. Bologna, Italy, May, 1989.

Ford-Rice, F.Y., Majewska, M.D. and London, E.D.: Effects of *in vivo* steroid hormone treatment on *sigma* receptor binding in the female rat brain. Soc. Neurosci. Abstr. 15: 857, 1989.

Hill, G.R., Della Puppa, A. and London, E.D.: Effects of high-dose *d*-SKF 10047 on cerebral metabolism in rats. Soc. Neurosci. Abstr., 15: 858,1989.

Wong, D.F., Young, D., Young, L.T., Tune, L.E., Pearlson, G., Minkin, E., Cidis Meltzer, C., Midha, K., Dannals, R.F., Ravert, H.T., Wilson, A.A. London, E.D. Wagner, H.N. Jr., Casanova, M., Klineman, J., Kuhar, M.J. and Gjedde, A.: Validation of PET D_2 dopamine receptor quantification using [C-11] NMSP and [F-18] haloperidol. Soc. Neurosci. Abstr. 15: 1123, 1989.

Demirgören, S., Majewska, M.D., Wagner, H.N. Jr. and London, E.D.: Binding of pregnenolone sulfate to rat brain membranes: interaction with the $GABA_A$ receptor complex. Soc. Neurosci. Abstr. 15: 994, 1989.

Kimes, A.S., Szabo, G., London, E.D., Raymon L. and Tabakoff, B.: Metabolic effects of GP120 in brain regions rich in VIP receptors. Soc. Neurosci. Abstr. 15: 381, 1989.

London, E.D. and Majewska M.D.: Binding of [^{3}H]mecamylamine to the nicotinic receptor complex in the rat brain: modulation by ATP. Soc. Neurosci. Abstr. 15: 64, 1989.

Takayama, H., Majewska, M.D. and London, E.D.: ATP: An allosteric modulator of the nicotinic receptor in brain. Soc. Neurosci. Abstr. 15: 64, 1989.

Majewska, M.D., ffrench-Mullen, J.M.H. and London, E.D.: Ascorbic acid and glutathione are antagonists of the NMDA receptor. Soc. Neurosci. Abstr. 15: 1167, 1989.

Della Puppa, A. and London, E.D.: Cerebral metabolic effects of *sigma* ligands in the rat. Soc. Neurosci. Abstr. 15: 857, 1989.

O'Hara, B.F., DiGiorgianni, J.M., Shimada, S., Kushner, L., Spivak, C.E., Lerma, J., Zukin, R.S., Bennett, M.V.L. and Uhl, G.R.: Expression of the neurotensin receptor in *Xenopus* oocytes with RNA transcribed from λ and pCDM8 libraries. Soc. Neurosci. Abstr. 15: 672, 1989.

Cascella, N.G, London, E.D., Sano, M., Wong, D.F., Herning, R.I., Links, J.M.: Acute cocaine reduces brain glucose metabolism in drug users. Am. Psychiatric Assoc. 142nd Annual Meeting, San Francisco, CA, May 6-12, 1989.

London, E.D., Takayama, H., Majewska, M.D.: [^{3}H]Mecamylamine binding to the nicotinic receptor complex in the rat brain: Modulation by ATP. ACNP Annual Meeting, Maui, Hawaii, December 9-16, 1989.

Majewska, M.D., ffrench-Mullen, J.R. and London, E.D.: Ascorbic acid and glutathione are allosteric antagonists of the NMDA receptor. 1989 ACNP Annual Meeting, Maui, Hawaii, December 9-16, 1989.

London, E.D., Cascella, N.G., Wong, D.F., Dannals, R.F., Jaffe, J.H. and Wagner, H.N. Jr.: Decrease in brain glucose utilization by cocaine. Am. Psychiatric Assoc. 143rd Annual Meeting, New York City, New York, May 12-17, 1990.

London, E.D., Broussolle, E.P., Links, J.M., Wong, D.F., Wagner, H.N. Jr. and Jaffe, J.H.: Decreased brain glucose utilization by morphine. Am. Psychiatric Assoc. 143rd Annual Meeting, New York City, New York, May 12-17, 1990.

Majewska, M.D. and Vaupel, D.B.: Steroids regulate uterine motility via $GABA_A$ receptors. XXXI Intern. Cong. Physiology, Helsinki, 1989.

Majewska, M.D.: Steroids regulation of $GABA_A$ receptor: The brain and body dialogue. XXXI Intern. Cong. Physiology, Helsinki, 1989.

McCann, D.J. and T.-P. Su. Distribution of (+)[^{3}H]SKF-10,047-labelled σ binding sites in rat brain subcellular fractions. Trans. Am Soc. Neurochem., Phoenix, Arizona, March 4-9, 1990.

McCann, D.J. and Su, T.-P.: Evidence that haloperidol inhibits the binding of (+)[^{3}H]SKF-10,047 to σ sites through a competitive interaction. FASEB J., 1990.

Wu, X.-Z. and Su, T.-P.: Guinea-pig vas deferens contains σ binding sites but not phencyclidine binding sites. Soc. Neurosci. Abstr. 15: 543, 1989.

Gund, T. M., Shuklar, K. and Su, T.-P.: Molecular modeling of σ receptor agonists and differentiation between σ and PCP receptor ligands. Abstr. International Chemical Cong., 1989.

Parish, D. W., Dodge, A. L., Petesch, M. M., Musallam, H. A., Koplovitz, I., Stewart, J. R. and Su, T.-P.: 1-[(1-arylcycloalkyl)methyl]amines: Synthesis, prophylactic efficacy against GD (*in vivo*), and PCP receptor affinity (*in vitro*). Bioscience Review. U.S. Army Medical Institute of Chemical Defense (USAMRICD), 1989.

Su, T.-P., Chien, S. F. and Oeltgen, P. R.: Hibernation induction trigger (HIT) extends preservation time in an autoperfusion multiorgan preparation. Abstr. Am. Soc. Clin. Pharmacol. Ther., 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00200-04 NPL (Previous DA 00201 & DA 00209 included)

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)

Cerebral Effects of Abused Drugs: Brain Imaging in Humans and Laboratory Animals

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI: London, E.D. (Chief, Neuropharmacology Lab, NPL, ARC)

Others: From NPL, ARC: Cascella, N.G. (Visiting Fellow), Sano, M. (Visiting Associate), Weissman, A.D. (Staff Fellow), Kimes, A.S. (Visiting Scientist), Phillips, R.L. (Visiting Scientist), Stapleton, J.S. (Staff Fellow)

From cooperating units, **ARC:** Jaffe, J.H. (Director), Rippetoe, L.R. (Head Nurse, Research Support Branch), Herning, R. (Visiting Scientist, Psychology of Vulnerability and Cognitive Studies Laboratory, VCS), Snyder, F.R. (Research Psychologist, VCS).

The Johns Hopkins Medical Institutions: Wong, D.F., Links, J., Dannals, R.F., La France, N.D., Grayson, R., Wagner, HN, Jr., Toung, J.K.T.

School of Pharmacy, University of Maryland: Marquis, K.L., Moreton, J.E.

LAB/BRANCH

Neuropharmacology Laboratory/ Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
3.8	2.65	1.15

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

This project uses brain imaging procedures to identify brain areas with functional alterations produced by abused drugs, and to relate drug effects on brain metabolism with effects on mood and electrical activities.

Human positron emission tomographic (PET) studies indicated that chronic heroin can produce persistent deficits in the regional cerebral metabolic rate for glucose (rCMRglc), measured by the PET [^{18}F]fluorodeoxyglucose (FDG) method, in the telencephalon. The deficits were apparently related to opioid effects on mood. Acute morphine or cocaine treatments, at euphorigenic doses, in placebo-controlled crossover studies, decreased rCMRglc in telencephalic areas, especially the cerebral cortex, and in whole brain. The findings support the view that a mechanism by which drugs of abuse produce euphoria involves a reduction of cortical rCMRglc. Future studies will test this hypothesis by using other abused drugs (e.g., nicotine). Other investigations will focus on imaging the effects of conditioned cues, which produce cocaine craving, and relating the time-course of the effects of cocaine on mood to those on regional cerebral blood flow.

Drug effects [nicotine, cocaine, amphetamine, 3,4-methylenedioxymethamphetamine (MDMA), phencyclidine (PCP)] on rCMRglc were studied by the deoxyglucose method in rats. Nicotine increased rCMRglc in a pattern matching distributions of [^{3}H]nicotine binding sites, defining the sites as receptors coupled to function. Chronic nicotine produced tolerance, seen as a reduced response in some areas. Cocaine increased rCMRglc in motor areas and reduced rCMRglc in the lateral habenula. Lewis rats were more sensitive to the effects than Fischer-344 rats, suggesting that genetic differences may influence susceptibility to cocaine abuse. MDMA produced rCMRglc effects which resembled those of cocaine, and self-administered PCP had effects which differed from those of acute PCP. While chronic drug exposure accounted for some differences, discrepancies between results in self-administering vs. non-self-administering rats with similar drug histories implicated some brain areas in drug self-administration training or behavior.

PUBLICATIONS
Z01 DA 00200-04 NPL

Cerebral Effects of Abused Drugs: Brain Imaging in Humans and Laboratory Animals

London, E.D., R.J. Connolly, M. Szikszay, J.K. Wamsley and Dam M.: Effects of nicotine on local cerebral glucose utilization in the rat. J. Neurosci. 8: 3920-3928, 1988.

London, E.D.: Effects of abused drugs on cerebral glucose metabolism. J. Neuropsychiat. Clin. Neurosci. 1, Suppl. 1: S30-S36, 1989.

Weissman, A.D., Marquis, K.L., Moreton, J.E., and London, E.D.: Effects of self-administered phencyclidine on regional brain uptake of 2-deoxy-D-[1-^{14}C]glucose. Neuropharmacology. 28: 575-583, 1989.

London, E.D., Margolin, R.A., Wong, D.F., Links, J.M., LaFrance, N.D., Cascella, N.G., Broussolle, E.P.M., Wagner, H.N., Jr., Snyder, F.R., and Jasinski, D.R.: Cerebral glucose utilization in human heroin addicts: Case reports from a positron emission tomographic study. Res. Comm. Substances of Abuse 10: 141-144, 1989.

Wilkerson, G. and London, E.D.: Effects of methylenedioxymeth-amphetamine 'Ecstasy' on local cerebral glucose utilization in the rat. Neuropharmacology 28: 1124-1138, 1989.

London, E.D., Casella, N.G., Wong, D.F., Phillips, R.L., Dannals, R.F., Links, J.M., Herning, R., Grayson, R., Jaffe, J.H., and Wagner, H.N.: Cocaine-induced reduction of glucose utilization in human brain: a study using positron emission tomography and [fluorine ^{18}]fluorodeoxyglucose. Arch. Gen. Psychiat., in press.

London, E.D., Broussolle, E.P.M., Links, J.M., Wong, D.F., Cascella, N.G., Dannals, R.F., Sano, M., Herning, R., Snyder, F.R., Rippetoe, L.R., Toung, T.J.R., Jaffe, J.H., and Wagner, H.N., Jr.: Morphine-induced metabolic changes in human brain - studies with positron emission tomography and 2-[^{18}F]fluoro-2-deoxy-D-glucose. Arch. Gen. Psychiat., in press.

London, E.D.: Effects of nicotine on cerebral metabolism. In: Ciba Foundation Symposium Biology of Nicotine Dependence, J. Marsh, ed., in press.

London, E.D., Wilkerson, G.W., Ori, C. and Kimes, A.S.: Central action of psychomotor stimulants on glucose utilization in extrapyramidal motor areas of the rat brain. Brain Res., in press.

London, E.D., Fanelli, R.J., Kimes, A.S., and Moses, R.L.: Effects of chronic nicotine on cerebral glucose utilization in the rat. Brain Res., in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00202-06 NPL

PERIOD COVERED: October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Physiological Effects of Opioids, and the Opioid Abstinence Syndrome

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI:	London, E.D.	Chief, Neuropharmacology Lab	NPL, ARC, NIDA
Others:	Kimes, A.S	Visiting Scientist	NPL, ARC, NIDA
	Bell, J.A.	Pharmacologist	NPL, ARC, NIDA
	Vaupel, D.B.	Pharmacologist	NPL, ARC, NIDA
	Della Puppa, A.	Visiting Fellow	NPL, ARC, NIDA
	De Souza, E.B.	Chief, Neurobiology Laboratory	NBL, ARC, NIDA

COOPERATING UNITS (if any)
Neurobiology Laboratory (NBL), Neuroscience Branch, ARC

LAB/BRANCH
Neuropharmacology Laboratory/Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.05	1.05	

CHECK APPROPRIATE BOX(ES)

☒ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

This project is aimed at delineating physiological effects of opioids and the central nervous system sites that mediate acute opioid agonist and antagonist effects, and the opioid abstinence syndrome.

The time course of verapamil interactions with morphine on physiological parameters and verapamil action on morphine pharmacokinetics have been studied in Fischer-344 rats. The interactions of verapamil with morphine on respiration, blood pH, blood pressure and heart rate were complex and unrelated to the influence of verapamil on morphine pharmacokinetics.

An interaction study of verapamil with the subjective and physiological effects of opioids in human opioid abusers was initiated. Preliminary data suggested that morphine-induced respiratory depression was partially antagonized by verapamil. Additional studies will focus on effects of the interaction on mood.

The deoxyglucose method was used to measure the regional metabolic rate for glucose (rCMRglc) in the rat. In animals tolerant to the analgesic effects of chronic morphine, metabolic tolerance in the brain was also evident. However, rCMRglc in many brain areas was stimulated during opioid abstinence. The hypermetabolism was reversed by small doses of clonidine, an α_2-adrenergic agonist.

Electrophysiological studies provided evidence that corticotropin releasing factor activates neurons in superficial dorsal horn presynaptic to motoneurons, directly depolarizes motoneurons, and could play a role in opioid abstinence. The dihydropyridine-type calcium channel antagonist, nifedipine, reduced capsaicin-induced depolarization without affecting the direct depolarizing effect of substance P on motoneurons, suggesting that calcium channel antagonists block substance P release and may reduce effects of opioid withdrawal in the neonatal rat spinal cord.

PUBLICATIONS
Z01 DA 00202-06 NPL

Physiological Effects of Opioids, and the Opioid Abstinence Syndrome

Kimes, A.S., and London, E.D.: Glucose utilization in the rat brain during chronic morphine treatment and naloxone-precipitated morphine withdrawal. J. Pharmacol. Exp. Ther. 248: 538-545, 1989.

Fanelli, R.J., Walovitch, R.C., Jasinski, D.R., and London, E.D.: Naloxone fails to alter local cerebral glucose utilization in the rat. Pharmacol. Biochem. Behav. 31: 481-485, 1989.

Bell, J.A., and De Souza, E.B.: Functional corticotropin-releasing factor (CRF) receptors in the neonatal rat spinal cord. Peptides 9: 1317, 1989.

Della-Puppa, A., Ford-Rice, F.Y., Snyder, F.R., Cone, E., and London, E.D.: Time course of verapamil interaction with morphine effects on physiological parameters in rats. J. Pharm. Pharmacol. 41:617-623, 1989.

Bell, J.A.: Naloxone-induced facilitation of C-fiber reflexes is reduced by chronic morphine. Eur. J. Pharmacol. 168: 101-105, 1989.

Kimes, A.S., Bell, J.A., and London, E.D.: Clonidine antagonizes increased glucose metabolism during naloxone-precipitated morphine withdrawal. Neuroscience, in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00003-05 NPL

PERIOD COVERED: October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Kappa and *Sigma* Properties of Antinociceptive Drugs in the Dog

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI:	D. B. Vaupel	Pharmacologist	NPL, ARC, NIDA
Others:	E. Cone	Chief	CDM, ARC, NIDA
	B. Nickel	Research Associate	Degussa Pharmaceuticals
	H. Shannon	Pharmacologist	E. Lilly, Inc.

COOPERATING UNITS (if any)
Degussa Pharmaceuticals, Frankfurt, West Germany
Laboratory of Chemistry & Drug Metabolism, Clinical Biology Branch, ARC
E. Lilly, Inc.

LAB/BRANCH: Neuropharmacology Laboratory/Neuroscience Branch

SECTION:

INSTITUTE AND LOCATION: ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.6	.6	0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The pharmacologic activity of *d,l*-ketocyclazocine is associated with the *l*-enantiomer, the *d*-form being inactive. We observed that this contrast is not due to pharmacokinetic differences. The pharmacologic profile of the κ-selective agonist U50,488H revealed that this drug has some actions that differ from *l*- and *d,l*-ketocyclazocine. To show that the actions of *l*-ketocyclazocine represent κ and not μ effects, selective antagonism studies with naltrexone were conducted. Low doses of naltrexone (0.01 mg/kg) antagonized morphine whereas high doses (1 mg/kg) were needed to antagonize *d,l*-ketocyclazocine, thus demonstrating that the agonist actions of *d,l*-ketocyclazocine are of the κ type in the chronic spinal dog. Publication of these findings will represent the termination of this project.

Flupirtine is an analgesic with an unknown mechanism of action. To assess the role of opioid mechanisms in flupirtine-induced antinociception, flupirtine was compared to the opioid, pentazocine, using single dose and naltrexone antagonism studies in the chronic spinal dog. It was concluded that flupirtine-induced antinociception is not opiate receptor-mediated and occurs primarily at supraspinal sites. Its antinociceptive potency was estimated to be 1/12 that of pentazocine in the dog. This work has been completed and the results published.

The acute interactions of pentazocine and tripelennamine, in ratios that have been abused by humans, were evaluated in the chronic spinal dog. No consistent pattern among the interactions emerged. Depending on the parameter, the interactions showed that effects of tripelennamine summated algebraically with or antagonized the effects of pentazocine. Naltrexone antagonized pentazocine but not tripelennamine, and tripelennamine failed to antagonize the σ-like activity of SKF-10047. Although such interactions may contribute to the abuse liability of tripelennamine and pentazocine, the tripelennamine component is not opioid-like. Furthermore, tripelennamine does not antagonize σ activity, which has been suggested as a mechanism for canine dysphoria. All experiments have been completed, and the findings published.

PUBLICATIONS
Z01 DA 00003-05 NPL

Kappa and *Sigma* Properties of Antinociceptive Drugs in the Dog

Vaupel, D.B., Nickel, B., and Becketts, K.: Flupirtine antinociception in the dog is primarily mediated by nonopioid supraspinal mechanisms. Eur. J. Pharmacol. 162: 447-456, 1989.

Vaupel, D.B.: Tripelennamine interactions with the psychotomimetic sigma agonist N-allylnormetazocine. Pharmacol. Biochem. Behav. 33: 717-720, 1989.

Vaupel, D.B.: Interactions between pentazocine and tripelennamine on autonomic and nociceptive measures in the dog. Pharmacol. Biochem. Behav. 33:245-251, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00206-05 NPL

PERIOD COVERED: October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Sigma and Phencyclidine Receptors

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI: Su, T.-P. Pharmacologist NPL, ARC, NIDA

Others: From NPL, ARC: London, E.D. (Chief, Pharmacologist), Majewska, M.D. (Sr. Staff Fellow), Spivak, C.E. (Pharmacologist), Vaupel, D.B. (Pharmacologist), Bell, J.A. (Pharmacologist), Wu, X.-Z. (Visiting Associate), Della Puppa, A. (Visiting Fellow), McCann, D. (Staff Fellow)

COOPERATING UNITS (if any)

LAB/BRANCH: Neuropharmacology Laboratory/Neuroscience Branch

SECTION

INSTITUTE AND LOCATION: ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
5.25	4.4	.85

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☒ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

This project examines molecular, electrophysiological, and *in vivo* interactions of ligands for *sigma* (σ) and phencyclidine (PCP) receptors.

Structure-activity relationship studies, using molecular modeling techniques, indicated that σ drugs have a hydrophobic moiety, an amine group with a lone pair of electrons, and an intermediate chain. Progesterone fit well into this model.

Potencies of σ and PCP ligands to produce inward currents in NCB-20 cells supported results of σ ligand binding in brain, but some drug stereoselectivities were inconsistent with binding. In NCB-20 cells, [^{3}H]*d*-N-allylnormetazocine ([^{3}H]*d*-NANM) labelled two sites. One seemed to be linked to tonic K^+ channels; the other was the σ receptor characterized before.

Sigma receptors were studied by subcellular fractionation, ontogenetically, and in assays of human brain, including postmortem tissue from schizophrenics. *Sigma* binding occurred in nonsynaptic organelles. While PCP receptors increased postnatally, σ sites were unchanged, rendering σ receptors unlike classical neurotransmitter receptors. *Sigma* receptors were lost in temporal cortex of schizophrenic brain, suggesting alteration of the σ system in psychosis.

In vivo studies, with the deoxyglucose technique in rats, indicated that σ sites are linked to brain function. BMY 14802 and BW 234U, antipsychotic candidate drugs which bind to σ sites, altered regional cerebral metabolic rate(s) for glucose (rCMRglc), an index of brain function. Changes in rCMRglc seemed driven by direct receptor interactions, as they followed the distribution of σ receptors.

Activity of the NMDA receptor complex, a site of PCP action, has been linked to pathology due to seizures or ischemia. Our studies focus on regulation of this complex. Regulation is effected by tissue redox phenomena, as reducing agents (e.g., ascorbic acid, glutathione) produced inactivation.

Future work includes studies on *in vivo* labelling of σ receptors, progesterone interactions with σ receptors, and factors that affect PCP receptor interactions.

PUBLICATIONS
Z01 DA 00206-05 NPL

***Sigma* and Phencyclidine Receptors**

Weissman, A.D., Su, T.-P., Hedreen, J.C. and London, E.D.: Sigma receptors in postmortem human brains. J. Pharmacol. Exp. Ther. 247: 29-33, 1988.

Majewska, M.D., Parameswaran, S., Vu, T., and London, E.D.: Divergent ontogeny of sigma and phencyclidine binding sites in the rat brain. Dev. Brain Res. 47: 13-18, 1989.

Weissman, A.D., Marquis, K.L., Moreton, J.E., and London, E.D.: Effects of self-administered phencyclidine on regional brain uptake of 2-deoxy-D-[1-^{14}C]glucose. Neuropharmacology. 28: 575-583, 1989.

Della Puppa, A. and London, E.D.: Cerebral metabolic effects of sigma ligands in the rat. Brain Res., in press.

Vu, T.H., Weissman, A.D., and London, E.D.: Pharmacological characteristics and distributions of sigma and phencyclidine binding sites in the animal kingdom. J. Neurochem., in press.

London, E.D.: Studies of σ receptors and metabolic responses to σ ligands in the brain. In: *Sigma*, PCP and NMDA Receptor Systems, E.B. DeSouza, E.D. London, and D. Clouet, eds. NIDA Research Monographs, in press.

Su, T.-P., London, E.D., and Jaffe, J.H.: Steroid binding at σ "opioid" receptor Response to a technical comment, Science, 246:1637-1638, 1989.

Su, T.-P. and Wu, X.-A.: Guinea-pig vas deferens contains σ but not phencyclidine receptors. Neurosci. Lett., in press.

Su, T.-P.: Pharmacological characterization of σ receptors. In: Sigma, PCP and NMDA Receptor Systems, E. B. De Souza, E. D. London, and D. Clouet, eds. NIDA Research Monographs, in press.

Su, T.-P., Shuklar, K. and Gund, T.: Steroid binding at σ receptors: CNS and immunological implications. In: Steroids and Neuronal Activity (Ciba Found. Symposium 153), D. J. Chadwick, ed., Wiley, Chicester, UK in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00207-05 NPL (includes previous Z01 DA 00217)

PERIOD COVERED: October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Nicotinic Receptors: Imaging, Regulation, and Structure Activity Relations

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI: E.D. London Chief, NPL, ARC

Others from NPL, ARC: Spivak , C.E. (Pharmacologist), Kimes, A.S. (Visiting Scientist), Majewska, M.D. (Senior Staff Fellow), Takayama, H. (Visiting Associate)

Others from cooperating units outside ARC: Waters, J.A. (Chemist, NIH), Gund, T.M. (Chemist, New Jersey Inst. Tech.), Magleby, K. (Biophysicist, Univ. Miami), Aronstam, R. (Biochemist, Med. Coll. Georgia)

COOPERATING UNITS (if any)
NIH, New Jersey Institute of Technology, University of Miami, Medical College of Georgia

LAB/BRANCH
Neuropharmacology Laboratory/Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.60	2.00	.6

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The objectives of this project are as follows: 1) to elucidate the molecular mechanisms of drug recognition at the nicotinic receptor (nAChr), 2) to determine how endogenous substances and drugs regulate the function of nAChr, and 3) to image nAChr in the brain.

In the course of this study, over 15 new agonists for nAChr have been synthesized, modeled, and assayed for potency in interosseal muscle from frog (*Rana pipiens*). Further insight into agonist actions was obtained by electrophysiological experiments, especially the patch clamp technique, which permitted recording of electrical currents through single ion channels.

Using receptor binding techniques, we determined the patterns of interaction of competitive and allosteric ligands binding to various domains of the receptor complex. Noncompetitive nAChr blockers, such as mecamylamine and chlorpromazine, bind inside of the cationic channel operated by this receptor, and their binding is a biochemical marker of the receptor activity. We discovered that the binding of radiolabeled mecamylamine and chlorpromazine is markedly increased by some purinergic nucleotides, suggesting that they may be important modulators of nAChr.

[^{3}H]*l*-nicotine was injected i.v. in mice which were killed at various times thereafter. Brains were dissected for measurement of radioactivity. Nonspecific binding was determined in mice pretreated with unlabelled *l*-nicotine. There was a rapid entry of [^{3}H]nicotine into the brain (maximum at 5 min) and specific binding was heterogeneously distributed. For example, levels were highest in medial and posterior cortex and thalamus/hypothalamus, intermediate in frontal cortex, cerebellum and caudate-putamen, and lowest in hippocampus and olfactory bulb. Nicotinic agonists significantly inhibited binding while several nicotinic antagonists were inactive. These results suggest that specific binding of [^{3}H]nicotine can be measured *in vivo* with radiolabelled nicotine.

PUBLICATIONS
Z01 DA 00207-05 NPL

Nicotinic Receptors: Imaging, Regulation, and Structure Activity Relations

McManus, O.B., Weiss, D.S., Spivak, C.E., Blatz, A.L. and Magleby, K.L.: Fractal models are inadequate for the kinetics of four different ion channels. Biophys. J. 54: 859-870, 1988.

Spivak, C.E., Yadav, J.S., Shang, W.C., Hermsmeier, M., and Gund, T.M.: Carbamyl analogues of potent nicotinic agonists: Pharmacology and computer-assisted molecular modeling study. J. Med. Chem. 32: 305-309, 1989.

Spivak, C.E., Waters, J.A. and Aronstam, R.S.: Binding of semirigid agonists to nicotinic and muscarinic receptors. Mol. Pharmacol. 36: 177-184, 1989.

McManus, O.B., Spivak, C.E., Blatz, A.L., Weiss, D.S., and Magleby, K.L.: Fractal models, Markov models, and channel kinetics. Biophys. J. 55: 383-385, 1989.

Broussolle, E.P., Wong, D.F., Fanelli, R.J., and London, E.D.: In vivo specific binding of [^{3}H]l-nicotine in the mouse brain. Life Sci. 44: 1123-1132, 1989.

London, E.D., Ball, M.J, and Waller, S.B.: Nicotinic binding sites in cerebral cortex and hippocampus in Alzheimer's dementia. Neurochem. Res. 14: 745-750, 1989.

Takayama, H., Majewska, M.D. and London, E.D.: Interactions of noncompetitive inhibitors with nicotinic receptors in the rat brain. J. Pharmacol. Exp. Ther., in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

Z01 DA 00208-05 NPL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Regulators of the $GABA_A$ Receptor Complex: Anxiolytics and Endogenous Steroids

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	London, E.D.	Chief, Neuropharmacology Lab	NPL, ARC, NIDA
Others:	Majewska, M.D	Senior Staff Fellow	NPL, ARC, NIDA
	Vaupel, D.B.	Pharmacologist	NPL, ARC, NIDA
	Spivak, C.E.	Pharmacologist	NPL, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Neuropharmacology Laboratory/ Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.6	.6	0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This project aims to assess the functional roles of various binding sites and endogenous substances which regulate the $GABA_A$ receptor. One question that was addressed was the relevance of type I vs. type II benzodiazepine (BZ) receptors. The deoxyglucose method was used to assay effects of diazepam (a nonselective BZ receptor agonist) and CL 218,872 (which has preferential activity at type I BZ receptors) on the regional cerebral metabolic rate for glucose (rCMRglc) in rats. Diazepam (2.5 mg/kg) decreased rCMRglc in 15 of 61 brain regions, but increased rCMRglc in the superior colliculus. A higher dose produced greater and more widespread decreases. Effects occurred preferentially in areas rich in type I BZ receptors compared to those with high densities of type II receptors. CL 218,872 produced similar results. The findings provide information about neuroanatomical sites that may be important to behavioral effects of BZ, and support a functional distinction between type I and type II BZ receptors.

Endogenous steroids are powerful modulators of $GABA_A$ receptors. Some behave as allosteric agonists, and others, such as pregnenolone sulfate, are antagonists. During pregnancy and the postpartum period, ligand binding to $GABA_A$ receptors in the brain and uterus was altered, suggesting steroid regulation. Such phenomena may influence the physiological changes accompanying pregnancy and the puerperium, and may regulate uterine activity. Our findings explain the altered sensitivity to anxiolytics and hypnotics during pregnancy and the puerperium.

It was known that pregnenolone sulfate behaves as an allosteric antagonist of the $GABA_A$ receptor complex *in vitro*. We observed further that this steroid also shows $GABA_A$ antagonistic actions *in vivo*, as it reduced barbiturate sleep time, suggesting that it can contribute to alterations of neuronal excitability and CNS arousal. We examined binding of [^{3}H]pregnenolone sulfate in the rat brain, noting that this steroid seems to interact at the interface of the receptor with associated phospholipids. Ongoing biochemical and electrophysiological studies focus on interactions of other steroids with $GABA_A$ receptors.

PUBLICATIONS
Z01 DA 00208-05 NPL

Regulators of the $GABA_A$ Receptor Complex: Anxiolytics and Endogenous Steroids

Majewska, M.D.: Interaction of ethanol with the $GABA_A$ receptor in the rat brain: Possible involvement of endogenous steroids. Alcohol 5: 269-273, 1988.

Majewska, M.D., Mienville, T.M., and Vicini, S.: Neurosteroid pregnenolone sulfate antagonizes electrophysiological responses to GABA in neurons. Neurosci. Lett. 90: 279-284, 1988.

Majewska, M.D., Ford-Rice, F., and Falkay, G.: Pregnancy-induced alterations of GABA-A receptor sensitivity in maternal brain: an antecedent of post-partum blues? Brain Res. 482: 397-401, 1989.

Majewska, M.D., Bluet-Payot, M.T., Robel, P.: Pregnenolone-sulfate: antagonists barbiturate-induced sleep. Pharmacol. Biochem. Behav. 33: 701-703, 1989.

Majewska, M.D., Falkay, G., and Baulieu, E.E.: Modulation of uterine $GABA_A$ receptors during gestational age and by tetrahydroprogesterone. Eur. J. Pharmacol. in press.

Majewska, M.D. and Vaupel, B.: Steroid control of uterine motility via $GABA_A$ receptor: a model mechanism. XXXI Intern. Congress of Physiology, Helsinki, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER
Z01 DA 00210-04 NPL
(includes previous
Z01 DA 00218)

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
HIV Infection and Drug Abuse Alter Expression/Function of Neuroimmune Receptors

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: London, E.D.	Chief, Neuropharmacology Lab	NPL, ARC, NIDA
Others: Kimes, A.S.	Visiting Scientist	NPL, ARC, NIDA
Smith, W.J.	Research Chemist, Biochemical Pharmacology Branch, USAMRICD, Aberdeen Proving Ground, MD	
Tabakoff, B.	Director, Intramural Research	NIAAA
Szabo, J.	Visiting Fellow	NIAAA

COOPERATING UNITS *(if any)*

USAMRICD, Aberdeen Proving Ground, MD
Intramural Research Program, NIAAA, Bethesda, MD

LAB/BRANCH
Neuropharmacology Laboratory/Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.4	.4	

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The effects of human immunodeficiency virus (HIV) infection in the central nervous system are characterized by severe dementia and deficits in motor functions. Cerebral metabolic effects of GP-120 (a HIV viral envelope glycoprotein), which binds to brain membranes and T-lymphocytes, were studied in rats. Intracerebroventricular injection of GP-120 reduced the regional cerebral metabolic rate for glucose in the lateral habenula and the suprachiasmatic nucleus, as well as producing an overall decrease in glucose metabolism. As reduced glucose metabolism is observed using positron emission tomography in humans presenting with HIV-associated dementia, the findings suggest that GP-120 can alter neuronal function and contribute to HIV-related dementia.

As intravenous drug abusers show an abnormally high incidence of HIV infection, animal studies were performed to determine if chronic exposure to abused substances alters immune function. Mice receiving chronic opioid treatment had fewer circulating T-lymphocytes (helpers and suppressor/cytotoxic) than concurrent controls. The effect was dose-dependent, was not blocked by treatment with the opioid antagonist naltrexone, and could be observed within 24 h of initiation of treatment. Oxymorphone caused a similar effect. Morphine treatment did not affect mitogen-stimulated lymphocyte proliferation. Scatchard analysis and inhibition studies of σ receptors in mouse splenocytes demonstrated binding characteristics that were similar but not identical to those of σ receptors in other species. Preliminary results suggested that chronic morphine treatment reduced the number of σ binding sites on mouse splenocytes. This work suggests that morphine compromises immunocompetency and that the use of opioids by intravenous drug abusers may increase the incidence of infection subsequent to exposure to bacterial and viral agents.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00212-05 NPL

PERIOD COVERED: October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Multiple Opioid Receptor Subtypes

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)
PI: Su, T.-P. Pharmacologist NPL, ARC, NIDA
Others: From NPL, ARC: London, E.D. (Chief, Pharmacologist), Vaupel, D.B. (Pharmacologist), Wu, X.-Z. (Visiting Associate)
From Cooperating Units
ARC: Goldberg, S.R. (Chief, Preclinical Branch), Schindler, C.W. (Staff Fellow)
University of Kentucky: Oeltgen, P.R. (Associate Professor), Chien, S.F. (Research Assistant Professor

COOPERATING UNITS (If any)
Preclinical Branch, ARC, NIDA
University of Kentucky

LAB/BRANCH
Neuropharmacology Laboratory/ Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.5	.5	0

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The development of supersensitivity to weekly injections of naltrexone in rats may involve a conditioning process. We observed changes in *kappa* and *delta* opioid receptors, and not *mu* receptors, suggesting that conditioning processes may include biochemical alterations of *kappa* and *delta* opioid receptors.

An endogenous substance (hibernation induction trigger, HIT), isolated from hibernating woodchucks, extended preservation time in an autoperfusion organ preparation. HIT-treated organs exhibited a mean survival time of 44 h in contrast to 14 h in the control group. HIT may act through opioid receptor because hibernation induced by HIT is reversible by naloxone. The mechanism may involve slowing of the metabolism and/or facilitation of microcirculation.

One possible mechanism of action of anesthetics is by alteration of neurotransmitter receptor function. Effects of N_2O and halothane on ligand binding to *mu* and *kappa* receptors were examined. N_2O increased Kd's for *mu* and *kappa* receptors, and decreased Bmax of *kappa* binding. Halothane increased Kd for *mu* receptors but decreased Kd for *kappa* receptors with a concomitant decrease in Bmax. Thus, volatile anesthetics interact with *mu* and *kappa* opioid receptors.

The effects of a *kappa* peptide BW942C on urine output were examined in humans, rats and squirrel monkeys. BW942C bound to *mu*, *kappa* and *delta* receptors, and was diuretic at low doses and antidiuretic at higher doses. The antidiuretic effect was antagonized by low doses of naltrexone; but the less efficacious diuretic effect of the *kappa* drug, U50488, was antagonized by high doses of naltrexone. The results suggested that BW942C is a partial *kappa* agonist and a *mu* agonist. This work is completed and the manuscript accepted for publication.

PUBLICATIONS
Z01 DA 00212-05 NPL

Multiple Opioid Receptor Subtypes

Ori, C., Ford-Rice, F.Y., and London, E.D.: Effects of nitrous oxide and halothane on *mu* and *kappa* opioid receptors in guinea-pig brain. Anesthesiology, 70: 541-544, 1989.

Oeltgen, P.R., Nuchols, P.A., Nilekani, S.P., Spurrier, W.A., and Su, T.-P.: Further studies on opioids and hibernation: *delta* opioid receptor ligand selectively induced hibernation in summer-active ground squirrels. Life Sci. 43: 1565-1574, 1988.

Oeltgen, P. R., Nilekani, S. P., Nuchols, P. A., Spurrier, W. A., Su, T.-P, Chien, S., Proffitt, G. E. and Mahony, C.: Identification of opioid receptor ligand(s) involved in summer-induced and natural winter hibernation. In: Living in the Cold, A. Malan and B. Canguihem, eds., pp. 97-104, Lolloque INSERM/John Libbey Eurotext Ltd., France, 1989.

Vaupel, D.B., Cone, E.J., Johnson, R.E., and Su T.-P.: *Kappa* opioid partial agonist activity of the enkephalin-like pentapeptide BW942C based on urination and *in vitro* studies in humans and animals. J. Pharmacol. Exp. Ther. in press.

Su, T.-P., Chien, S. F. and Oeltgen, P. R.: Hibernation induction trigger (HIT) extends preservation time in an autoperfusion multiorgan preparation. Clin. Pharmacol. Ther., in press.

Schindler, C.W., Wu, X.-Z., Su, T.-P., Goldberg, S.R. and Katz, J.L.: Enhanced sensitivity to the behavioral effects of naltrexone in rats: A conditioning phenomenon associated with opioid receptor changes. J. Pharmacol. Exp. Ther., in press.

3. Neurobiology Laboratory - Errol B. De Souza, Ph.D., Chief

Overview

The Laboratory of Neurobiology conducts research on the neurobiological underpinnings of drug abuse and addiction. At present, the Laboratory has three major areas of research which include 1) the study of the neuroendocrine aspects of addiction, with a focus on stress, hypothalamic peptides and drugs of abuse, 2) the study of the pharmacological and neurotoxic effects of drugs of abuse, and 3) the study of the interactions of the brain-neuroendocrine-immune axis and its related peptides, hormones, lymphokines and monokines. The Laboratory utilizes a multifaceted approach which includes biochemical, cellular, pharmacological, neuroendocrine and neuroanatomical techniques to investigate the problems outlined above.

Stress is a key factor which plays a major role in both the initiation and maintenance of drug abuse. Furthermore, stress produces profound and sustained neurochemical changes in the body and interacts in a complex manner with psychotropic and addictive drugs. A major effort of the Laboratory has been directed at understanding the basic mechanisms regulating stress responses. Corticotropin-releasing factor (CRF) is a critical hormone involved with stress responses. In addition to its role in regulating stress responses via the endocrine system, recent evidence suggests that CRF may act as a neurotransmitter in brain and function as an integrator of the overall stress response in the body. An important ongoing project is to establish CRF as a bona fide neurotransmitter in the CNS. Biochemical, cellular, pharmacological and neuroanatomical studies have been utilized for studying the characteristics and distributions of CRF and its receptors, the second messenger systems through which CRF produces its many effects, and establishing molecular neurobiological techniques to identify specific intracellular messenger RNA for CRF. We have identified high affinity binding sites for CRF in brain which are distributed throughout the CNS. In addition, we have demonstrated that the second messenger system through which CRF produces its effects in brain involves stimulation of adenylate cyclase activity. In biochemical studies, we have identified the ligand binding subunits of CRF receptors in brain and anterior pituitary of a number of species by chemical affinity cross-linking techniques. We have demonstrated using a combination of immunocytochemical, molecular biological and receptor autoradiographic techniques, that CRF is a major transmitter in the olivocerebellar pathway in a variety of species including humans. The production of neuroanatomical maps for CRF, mRNA for CRF and CRF receptors has set the basis for subsequent studies to examine the effects of various drugs of abuse that modulate CRF neurotransmission and stress responses.

Direct interactions between drugs of abuse and CRF have recently been observed by others in studies demonstrating that administration of

CRF, like stress, produces sensitization to the behavioral responses to amphetamine. Furthermore, the CRF antagonist severely attenuates the stress-induced sensitization in response to amphetamine. Studies carried out in the Neurobiology Laboratory in collaboration with Dr. Nick Goeders of Louisiana State University have demonstrated that chronic administration of cocaine in rats activates CRF in brain and down-regulates CRF receptors primarily in the mesolimbic/mesocortical dopaminergic system. CRF has been implicated in the etiology of a variety of human neuropsychiatric disorders including depression and anxiety and it is conceivable that the anxiety and depression observed during cocaine withdrawal may result, in part, through the actions of the drug on CRF release. Taken together, these data identify a novel transmitter in brain that may be involved in the initiation and maintenance of cocaine and amphetamine abuse. Furthermore, the data suggest that a CRF antagonist may be useful in attenuating both self-administration of the drug as well as in attenuating some of the withdrawal symptoms following chronic administration of cocaine. Neurochemical, neuroanatomical and pharmacological studies are currently underway to address some of these hypotheses.

To further investigate the role of CRF in brain, we have examined changes in various CRF markers in neurodegenerative disorders such as Alzheimer's disease, Parkinsons's disease, Huntington's disease and progressive supranuclear palsy. In our initial studies we found that in Alzheimer's disease, the concentrations of CRF-like immunoreactivity were reduced and that there were reciprocal increases in CRF receptor binding in affected cerebrocortical areas. Decreases in CRF-like immunoreactivity similar to those described for Alzheimer's disease were also seen in patients who died of Parkinson's disease and progressive supranuclear palsy. In contrast, patients who died of Huntington's disease did not show decrements in CRF-like immunoreactivity in the cerebral cortex but showed significant decreases in CRF content in the caudate/putamen. More recently, we have demonstrated abnormalities in CRF neurons in patients who died of Alzheimer's disease in that CRF-like immunoreactivity was localized to senile plaques. These results strongly support a neurotransmitter role for CRF in brain and suggest a possible role for CRF in the pathophysiology of various neurodegenerative disorders. In addition, they also suggest a role for CRF in brain in processes involving cognition and short-term memory.

The psychotomimetic agent 3,4-methylenedioxymethamphetamine (MDMA) has recently been the focus for a great deal of attention as it represents one of the most popular members of the class of abused substances known as designer drugs. A major research focus of the Laboratory has been to study the neurochemical mechanisms through which MDMA, 3,4-methylenedioxyamphetamine (MDA), and related amphetamine derivatives produce their psychotomimetic and neurotoxic effects. With regard to the psychotomimetic effects, we have demonstrated that MDMA and MDA have highest affinity for 5-HT_2

serotonin receptors in brain; these receptors have been shown to be the site of action through which a variety of amphetamine derivatives produce their hallucinogenic effects. In studies carried out in rats and monkeys, we have found that chronic administration of MDA and MDMA resulted in widespread and long-term destruction of serotonin nerve terminals in brain. Furthermore, our investigations revealed that MDMA is more potent, on a dose basis, in destroying serotonin terminals in primates than in rodents. Although regeneratin of serotonin neurons occurs in brain, the recovery occurs over a protracted period of time in rat; a 25% reduction is observed even at six months after treatment with MDMA. Again, rhesus monkeys appeared to be more susceptable to the effects of MDMA with decreases in the CSF concentrations of the serotonin metabolite 5-hydroxyindoleacetic acid (5-HIAA) and decreases (greater than 70%) in the brain concentrations of a variety of serotonergic markers evident even at four months after a short-term treatment. The neurotoxic effects of MDA and MDMA in rats could be prevented by pretreatment with a serotonin uptake blocker. In autoradiographic and immunocytochemical studies, we demonstrated that the effects of these compounds on destruction of serotonin neurons in brain were not diffuse but were rather limited to certain brain areas. Specifically, MDA and MDMA appeared to destroy serotonin terminals in ascending pathways while serotonin neurons in descending pathways, axons of passage and serotonin cell bodies appeared to be unaffected. Overall, these studies examining the effects of MDA and MDMA in rodents and primates demonstrate neurotoxic effects of these compounds in destroying serotonin neurons in brain and suggest their potential neurotoxic hazard in humans.

A variety of amphetamine derivatives are currently used to treat some psychiatric disorders. Some of these drugs include fenfluramine which is used for the treatment of obesity and is also used for the treatment of infantile autism. Another widely prescribed stimulant is methylphenidate (Ritalin) which is used for the treatment of attention deficit disorders in children and adolescents. Methlyphenidate abuse has also been reported in adults. Methylphenidate did not produce any long-term neurotoxic effects in rodents. In contrast, we observed that short-term fenfluramine treatment caused dose-dependent decreases in a variety of serotonergic markers (serotonin, 5-HIAA and serotonin uptake sites) in a variety of brain regions; no major effects of the drugs were noted on catecholamine markers. Immunocytochemical studies confirmed the neurochemical data demonstrating neurotoxic effects of fenfluramine resulting in profound reduction in fine-caliber serotonin-immunoreactive fibers and terminals with no major effects on cell bodies.

Dopamine and other brain monoamines have been implicated in mediating the reinforcing properties and behavioral effects of cocaine. While cocaine administration alters monoamine transmission in brain, there is an ongoing controversy regarding the neurotoxic effects of cocaine on brain monoamine neurons. Neuroanatomical and neurochemical

studies were carried out to assess the potential neurotoxic effects of high-dose repeated cocaine administration on brain monoamine neurons. The markers assessed for neurotoxicity included long-term changes in the content of monoamines and their metabolites in brain. Neurochemical studies did not demonstrate any marked or consistent changes in the concentrations of dopamine, norepinephrine, serotonin and their metabolites in a variety of brain regions examined at time points up to 48 days following chronic cocaine treatment. In addition, we did not observe any long-term alterations in the density of serotonin-like immunoreactive or tyrosine hydroxylase-like immunoreactive axons and terminals in a variety of brain regions. Furthermore, there was no evidence for morphology characteristic of degenerating axons and terminals in chronically cocaine treated rats. Overall, these data suggest that chronic cocaine treatment does not produce neurotoxic effects on brain monoamine neurons.

Schizophrenia is a mental disorder characterized by psychotic symptoms including delusions and hallucinations and disturbances of thinking and mood. Various neurochemical findings including alterations in dopaminergic neurotransmission and increased densities of D_2 dopamine receptors in the caudate/putamen have been associated with schizophrenia. In addition, evidence that drugs that interact with phencyclidine (PCP) and sigma binding sites can produce effects that resemble some of the psychotic symptoms of schizophrenia have implicated PCP and sigma binding sites in the clinical manifestations of this disorder. The potential role of sigma binding sites in schizophrenia is further supported by the high affinity of many established antipsychotic drugs, including haloperidol, for these sites. To further explore the role of sigma and PCP binding sites in schizophrenia, we assayed these sites in human postmortem brains. We observed statistically significant reductions in the density of sigma, but not PCP binding sites, in schizophrenics as compared to age- and postmortem interval-matched normal and suicide controls. Reductions in the density of sigma binding sites in schizophrenia were most prominent and consistent in the temporal cerebral cortex. The present data provide the first evidence for alterations in sigma binding sites in schizophrenia, and suggest that select sigma ligands may be useful in the treatment of the disorder.

Intravenous drug abusers are at higher risk for viral infections such as AIDS. While the disease is propagated through the use of contaminated needles, the potent immunosuppresant effects of a variety of substances of abuse, including opioids, may explain, in part, the increased progression of the disease in drug addicts. Furthermore, a variety of CNS functions appear to be affected in AIDS patients. The presence of neurotransmitters and receptors which are common to the immune, endocrine and central nervous systems suggest that the three systems may interact in a coordinated fashion. Since phencyclidine (PCP) and sigma opioids as well as chronic stress have been demonstrated to cause immunosuppression and to alter a variety of endocrine and CNS functions, we have examined the potential role of sigma drugs and CRF in modulating immune function. In addition,

we have examined the role of interleukin-1 (IL-1), a cytokine which is a key mediator of the immune response to AIDS infection, stress, and antigenic challenge. Sigma, CRF and IL-1 receptors were identified in immune, endocrine and CNS tissues. In the immune system, CRF receptors were found in mouse spleen, primarily in splenic macrophages. Also, sigma receptors were identified in human peripheral blood leukocytes and in rat spleen. IL-1 receptors were localized in mouse spleen primarily in lymphocytes. While PCP receptors were present in CNS, they were absent in immune and endocrine tissues. Since PCP acts on both PCP and sigma receptors, the results of our studies suggest that PCP may exert its immunomodulatory influence via sigma receptors and that endogenous sigma ligands, if in fact they exist, may play a role in modulating immune function. Furthermore, the data suggest the importance of stress, and the key players in stress response such as CRF and IL-1 in modulating immune function. Finally, we have identified IL-1 receptors in brain which may explain in part the behavioral and CNS actions that are seen in AIDS patients in which IL-1 concentrations are dramatically altered.

Summary of Ongoing Research

A. Corticotropin-Releasing Factor (CRF) as a Stress Neurotransmitter in the Central Nervous System

CRF is a critical hormone involved in stress responses and recent evidence suggests that CRF is a neurotransmitter in brain. Current and future studies are aimed at characterizing CRF binding sites at a molecular level and in examining the effects of a variety of treatments on these receptors. Specifically, we are utilizing a variety of chemical cross-linking techniques to study the molecular composition of CRF receptors, and studies have been initiated to purify the CRF receptor in an attempt to sequence the protein and clone the receptor. In addition, we are examining the modulation of CRF and its receptors both in vivo and in vitro. These studies include examining the effects of acute and chronic treatment with cocaine and a variety of amphetamine related derivatives on CRF markers in CNS and anterior pituitary. Furthermore, studies are being initiated to examine the effects of stress, CRF and CRF antagonists in altering self-administration and sensitization effects of stimulant drugs such as amphetamines and cocaine as well as in altering some of the withdrawal symptoms following chronic administration of these drugs.

B. The Role of Transmitters and Their Receptors in Human Neuropsychiatric Disorders and Neurodegenerative Diseases

Changes in specific neurotransmitters and their receptors play a key role in the pathophysiology of various neuropsychiatric disorders and neurodegenerative diseases. The ongoing studies

are aimed at examining the molecular and biochemical characteristics of CRF and its receptors in postmortem tissue obtained from individuals who died from Alzheimer's disease, depression, schizophrenia and age-matched controls. Specifically, we plan on examining the "functional nature" of CRF receptors in Alzheimer's tissue by examining the alterations in second messenger activity. In addition, parallel studies will be carried out in rodents to examine the effects of aging on various CRF markers in rat brain. Given the specific changes in sigma receptors in cerebral cortex in schizophrenia, we plan on following up this finding by carry on autoradiographic studies to localize the changes at a light microscopic level. In addition, we are also examining changes in sigma and PCP receptors in people that have died from PCP overdose.

C. Pharmacological and Neurotoxic Effects of MDA and MDMA

The designer drugs MDA and MDMA have potent, long-lasting, neurotoxic effects in brain. In addition, MDA, MDMA and related amphetamine derivatives produce a variety of behavioral effects through actions in the CNS. The goals of the project are to further assess the neurotoxic actions of these drugs and to determine the receptors in brain through which these drugs may produce the neurotoxic actions and behavioral effects. Studies are currently being carried out to examine the mechanisms responsible for the neurotoxic effects of the drugs. Neuroanatomical studies are being carried out to determine the serotonergic pathways that are affected and the long-term consequences of serotonin depletion in the affected pathways. In addition, studies are being carried out to identify MDA and MDMA binding sites in brain and to develop methods for the detection of these compounds in the periphery and in the CNS. We are also using a serotonin neuroblastoma cell line to examine ultrastructural changes following incubation with various doses of the amphetamines.

D. Neurochemical, Neuroendocrine and Neurotoxic Effects of Selected Drugs

Several drugs that are currently used to treat a variety of psychiatric disorders produce their effects through actions on monoaminergic systems in brain. Studies are currently underway to examine the effects of chronic administration of several antidepressant and appetite suppressant drugs that are currently in clincal use or are being reviewed for approval by the FDA on neurotoxicity to monoamine neurons in brain. The focus of these studies will be on fenfluramine, a clinically prescribed appetite suppressant. Changes in monoamine transmission will be correlated with changes in a variety of neuroendocrine parameters with a focus on the measurement of anterior pituitary hormones.

E. Interactions Between Brain-Endocrine-Immune Axis

The presence of neurotransmitters and their receptors in brain, endocrine and immune systems suggest that the three systems may interact in a coordinated fashion. In previous years, we have identified CRF and sigma receptors in the immune system. More recently, we have identified sigma receptors in very high concentrations in endocrine tissues with kinetic and pharmacological characteristics similar to those found in brain. In addition, we have identified interleukin-1 (IL-1) receptors in the brain-pituitary-immune axis. Studies which are currently underway will focus on identifying the cell type(s) in the endocrine and immune systems containing the various neurotransmitter receptors and on examining the role of the receptors in modulating hormone secretion and immune function. The effects of in vivo manipulations, such as application of stress and acute and chronic administration of drugs of abuse, will be examined on changes in CRF, sigma, PCP and IL-1 receptors in brain, endocrine organs, and immune tissue.

Publications

Wolfe, S.A. Jr., Kulsakdinun, C., Battaglia, G., Jaffe, J.H. and De Souza, E.B. Initial identification and characterization of sigma receptors in human peripheral blood leukocytes. J. Pharmacol. Exp. Ther. 247:1114-1119, 1988.

De Souza, E.B. Localization and modulation of brain and pituitary receptors involved in stress responses. Psychopharmacology Bulletin 24:360-364, 1988.

De Souza, E.B. Corticotropin-releasing factor receptors in brain and pituitary: implications for the stress response. In: Neuropeptides and Stress (Y. Tache, J.E. Morley and M.R. Brown, eds.), Springer-Verlag, New York, 1988, pp. 1-17.

De Souza, E.B. CRH defects in Alzheimer's and other neurologic disorders. Hospital Practice 23:59-71,1988.

Hsu, F.-L., Yeh, S.Y. and Munavalli, S. O-demethylation of pyrilamine. J. Pharmc. Sci. 77:727-728, 1988.

De Souza, E.B. and Battaglia, G. Corticotropin-releasing hormone receptors in brain. In: Mechanisms of Physical and Emotional Stress (G. Chroussos, D. Lynn Loriaux and P.W. Gold eds.), Plenum Press, New York 1988, pp 123-136.

Bell, J.A. and De Souza, E.B. Functional corticotropin-releasing factor (CRF) receptors in the neonatal rat spinal cord. Peptides 9:1317-1322, 1989.

Cummings, S., Young III, W.S., Bishop, G., De Souza, E.B. and King, J.S. Distribution of corticotropin releasing factor in the cerebellum and precerebellar nuclei of the opossum: a study utilizing immunohistochemistry, in situ hybridization, histochemistry and receptor binding. J. Comp. Neurol. 280:501-521, 1989.

Grigoriadis, D.E. and De Souza, E.B. Corticotropin-releasing factor receptors in intermediate lobe of the pituitary: biochemical and autoradiographic localization. Peptides 10:179-188, 1989.

Webster, E.L., Battaglia, G. and De Souza, E.B. Functional corticotropin-releasing factor (CRF) receptors in mouse spleen: evidence from adenylate cyclase studies. Peptides 10:395-401, 1989.

Wolfe, S.A., Jr., Culp, S.G. and De Souza, E.B. Sigma receptors in endocrine organs: identification, characterization, and autoradiographic localization in rat pituitary, adrenal, testis and ovary. Endocrinology 124:1160-1172, 1989.

Grigoriadis, D.E., Pearsall, D. and De Souza, E.B. Effects of chronic antidepressant and benzodiazepine treatment on corticotropin-releasing factor receptors in rat brain and pituitary. Neuropsychopharmacology 2:53-60, 1989.

Sharkey, J., Appel, N.M. and De Souza, E.B. Alterations in local cerebral glucose utilization following central administration of corticotropin-releasing factor (CRF) in rats. Synapse 4:80-87, 1989.

Insel, T.R., Battaglia, G., Johannessen, J.N., Marra, S. and De Souza, E.B. 3,4-Methylenedioxymethamphetamine (MDMA; "Ecstasy") selectively destroys brain serotonin terminals in Rhesus monkeys. J. Pharmacol. Exp. Ther. 249:713-720, 1989.

Appel, N.M., Contrera, J.F. and De Souza, E.B. Fenfluramine selectively and differentially decreases the density of serotonergic nerve terminals in rat brain: evidence from immunocytochemical studies. J. Pharmacol. Exp. Ther. 249:928-943, 1989.

Zaczek, R., Battaglia, G., Contrera, J.F., Culp, S. and De Souza, E.B. Methylphenidate and pemoline do not cause depletion of rat brain monoamine markers similar to that observed with methamphetamine. Toxicol. Appl. Pharmacol. 100:227-233, 1989.

Grigoriadis, D.E., Struble, R.G., Price, D.L. and De Souza, E.B. Normal pattern of labeling of cerebral cortical corticotropin-releasing factor (CRF) receptors in Alzheimer's disease: evidence from chemical cross-linking studies. Neuropharmacology 28:761-764, 1989.

Grigoriadis, D.E. and De Souza, E.B. Heterogeneity between brain and pituitary corticotropin-releasing factor (CRF) receptors is due to differential glycosylation. Endocrinology 125:1877-1888, 1989.

Han, Z., Kuyatt, B.L., Kochman, K.A., De Souza, E.B. and Roth, G.S. Effect of aging on concentrations of D_2-receptor-containing neurons in the rat striatum. Brain Research 498:299-307, 1989.

De Souza, E.B. and Van Loon, G.R. Rate sensitive glucocorticoid feedback inhibition of adrenocorticotropin and β-endorphin/β-lipotropin secretion in rats. Endocrinology 125:2927-2934, 1989.

Grigoriadis, D.E., Zaczek, R., Pearsall, D. and De Souza. Solubilization of high-affinity corticotropin-releasing factor (CRF) receptors from rat brain: Characterization of an active digitonin-solubilized receptor complex. Endocrinology 125:3068-3077, 1989.

Insel, T.R., Battaglia, G. and De Souza, E.B. Brain Corticotropin Releasing Factor and development. In: Molecular Biology of Stress (O. Zinder and S. Bresnitz, eds.), Alan R. Liss, New York, pp. 19-30, 1989.

Titeler, M., Leonhardt, S., Appel, N.M., De Souza, E.B. and Glennon, R.A. [^{125}I]DOI and [^{3}H]DOB: Novel agonist radioligand probes for $5HT_2$ and $5HT_{1c}$ receptors. Du Pont Biotech Update 4(6):20-24, 1989.

De Souza, E.B. and Battaglia, G. Effects of MDMA and MDA on brain serotonin neurons: evidence from neurochemical and autoradiographic studies. In: Pharmacology and Toxicology of amphetamine and related designer drugs (K. Asghar and E.B. De Souza, eds.), NIDA Research Monograph 94:196-222,1989.

Zaczek, R., Hurt, S., Culp, S. and De Souza, E.B. Characterization of brain interactions with methylenedioxyamphetamine and methylenedioxymethamphetamine. In: Pharmacology and Toxicology of Amphetamine and Related Designer Drugs (K. Asghar and E.B. De Souza, eds.), NIDA Research Monograh 94:223-239, 1989.

De Souza, E.B. The "Ecstacy" debate. Medical Aspects of Human Sexuality 23:40, 48,1989.

Battaglia, G. and De Souza, E.B. Pharmacologic profile of amphetamine derivatives at various brain recognition sites: Selective effects on serotonergic systems. In: Pharmacology and Toxicology of Amphetamine and Related Designer Drugs (K. Asghar and E.B. De Souza, eds.), NIDA Research Monographs 94:240-258, 1989.

Articles in Press

Wolfe, S.A. Jr. and De Souza, E.B. Sigma receptors in the brain-endocrine-immune axis. In: Sigma, PCP and NMDA receptor systems (E.B. De Souza, E.D. London and D.H. Clouet, eds.), NIDA Research Monographs (in press).

De Souza, E.B., Bissette, G., Whitehouse, P.J., Price, D.L., Vale, W.W. and Nemeroff, C.B. Role of corticotropin-releasing factor (CRF) in neurodegenerative diseases In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

Zaczek, R., Culp, S.G. and De Souza, E.B. Intrasynaptosomal sequestration of [^{3}H]-amphetamine and [^{3}H]-methylenedioxy-amphetamine: Characterization suggests the presence of a factor responsible for maintaining sequestration. J. Neurochem. (in press).

Appel, N.M., Mitchell, Wm. M., Contrera, J.F. and De Souza, E.B. Effects of high-dose fenfluramine treatment on monoamine uptake sites in rat brain: Assessment using quantitative autoradiography. Synapse (in press).

Zaczek, R., Battaglia, G., Contrera, J.F. and De Souza, E.B. Effects of repeated fenfluramine administration on indices of monoamine function in rat brain: Pharmacokinetics, dose response, regional specificity and time course data. J. Pharmacol. Exp. Ther. (in press).

De Souza, E.B. Autoradiographic localization of monoamine and corticotropin-releasing factor (CRF) receptors in the pituitary: effects of glucocorticoids and peripheral amines. In: Catecholamines and Other Neurotransmitters in Stress, (G.R. Van Loon, ed.), Gordon and Breach Science Publishers, New York, (in press).

De Souza, E.B. and Insel, T.R. Corticotropin-releasing factor (CRF) receptors in the rat central nervous system: autoradiographic localization studies. In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

De Souza, E.B. and Grigoriadis, D.E. Corticotropin-releasing factor (CRF) receptors in brain: characterization and regulation. In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

Battaglia, G., Webster, E.L. and De Souza, E.B. Characterization of second messengers coupled to corticotropin-releasing factor (CRF) receptors in brain. In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

Battaglia, G., Zaczek, R. and De Souza, E.B. MDMA effects in brain: Pharmacologic profile and evidence of neurotoxicity from neurochemical and autoradiographic studies. In: MDMA: "Ecstasy" and/or Human Neurotoxin? (S.J. Peroutka, ed.), Kluwer Academic Publishers, Boston, (in press)

De Souza, E.B. and Appel, N.M. Distribution of brain and pituitary receptors involved in mediating stress responses. In: Neurobiology and Neuroendocrinology of Stress (M.R. Brown, C. Rivier and G.F. Koob, eds.), Marcel Dekker, Inc., New York (in press).

Webster, E.L., Grigoriadis, D.E. and De Souza, E.B. Corticotropin-releasing factor receptors in the brain-pituitary-immune axis. In: Stress, Neuropeptides, and Systemic Disease (J.A. McCubbin, P.G. Kaufmann and C.B. Nemeroff, eds.), (in press).

De Souza, E.B. Neurotransmitter Receptor Imaging Techniques. In: Neuropeptides in Psychiatry (Progress in Psychiatry Series) (C.B. Nemeroff, ed.), (in press).

De Souza, E.B. Neuroendocrine effects of benzodiazepines. J. Psychiatric Research (in press).

Appel, N.M., Zaczek, R., Mitchell, W.M. and De Souza, E.B. Immunohistochemical and autoradiographic investigations of high-dose fenfluramine treatment on monoamine neurons in rat brain. In: Proceedings of Int. Symp. on Serotonin: From Cell Biology to Pharmacology and Therapeutics, Kluwer Academic Publishers, Boston (in press).

Titeler, M., Appel, N.M., De Souza, E.B. and Glennon, R.A. Receptor pharmacology of MDMA and related hallucinogens. Annals New York Acad. Sci. (The Neuropharmacology of Serotonin) (in press).

De Souza, E.B., Battaglia, G. and Insel, T.R. Neurotoxic effects of MDMA on brain serotonin neurons: Evidence from neurochemical and radioligand binding studies. Annals New York Acad. Sci. (The Neuropharmacology of Serotonin) (in press).

De Souza, E.B., Webster, E.L., Grigoriadis, D.E. and Tracey, D.E. Corticotropin-releasing factor (CRF) and Interleukin-1 (IL-1) receptors in the brain-pituitary-immune axis. Psychopharmacology Bulletin (in press).

Grigoriadis, D.E. and De Souza, E.B. Receptor binding techniques. In: Comprehensive textbook of neuroendocrinology (C.B. Nemeroff, ed.), The Telford Press, Caldwell, N.J. (in press).

Kuhar, M.J. and De Souza, E.B. Receptor autoradiography as an aid in explaining drug action. In: Imaging the functional neuroanatomy of drug action (E.D. London, ed.), The Telford Press, Caldwell, N.J. (in press).

De Souza, E.B., Grigoriadis, D.E. and Webster, E.L. Role of brain, pituitary and spleen corticotropin-releasing factor (CRF) receptors in the stress response. In: The stress of life, revisited (Methods and Achievements in Experimental Pathology) (G. Jasmin and M. Cantin, eds.), S. Karger, Switzerland (in press).

Abstracts Published

Yeh, S.Y. N-Depyridination and N-dedimethylaminoethylation of tripelennamine and pyrilamine in the rat: New metabolic pathways. Pharmacologist, 31:130, 1989.

Weissman, A.D., Casanova, M.F., Kleinman, J.B., London, E.D., and De Souza, E.B. Regional differences in PCP and σ binding in the brains of schizophrenics. Soc. for Neurosci. 14:104, 1988.

Yousif, M., Battaglia, G., De Souza, E.B., Glennon, R.A. Sigma receptors: a putative site of action of MDMA (3,4-methylenedioxymethamphetamine) and related analogs. Soc. for Neurosci. 14:524, 1988.

Contrera, J.F., Battaglia, G., Zaczek, R., and De Souza, E.B. Fenfluramine neurotoxicity: selective degeneration and recovery of brain serotonin neurons. Soc. for Neurosci. 14:556, 1988.

Appel, N.M. and De Souza, E.B. Fenfluramine selectively destroys serotonin terminals in brain: immunocytochemical evidence. Soc. for Neurosci. 14:556, 1988.

Zaczek, R., Battaglia, G., Contrera, J.F., and De Souza, E.B. Ritalin and pemoline do not cause monoamine terminal degeneration. Soc. for Neurosci. 14:557, 1988.

Culp, S., Zaczek, R., and De Souza, E.B. Incorporation of ^{3}H-methylenedioxyamphetamine (MDA) into rat brain synaptosomes. Soc. for Neurosci. 14:557, 1988.

Battaglia, G., Sharkey, J., Kuhar, M.J., and De Souza, E.B. Neuroanatomical specificity of MDA- and MDMA-induced degeneration of serotonin neurons in rat brain. Soc. for Neurosci. 14: 557, 1988.

Johannessen, J.N., Insel, T.R., Battaglia, G., Kuhar, M.J., and De Souza, E.B. MDMA selectively destroys brain serotonin terminals in Rhesus monkeys. Soc. for Neurosci. 14:557, 1988.

Grigoriadis, D.E. and De Souza, E.B. Deglycosylation of rat brain and pituitary corticotropin-releasing factor (CRF) receptors. Soc. for Neurosci. 14:667, 1988.

De Souza, E.B. and Grigoriadis, D.E. Characterization of corticotropin-releasing factor (CRF) receptors in the intermediate lobe of the pituitary. Soc. for Neurosci. 14:667, 1988.

Pearsall, D., Grigoriadis, D.E., and De Souza, E.B. Effects of chronic antidepressant and benzodiazepine treatment on corticotropin-releasing factor (CRF) receptors in rat brain. Soc. for Neurosci. 14:667, 1988.

Webster, E.L., Battaglia, G., and De Souza, E.B. Corticotropin-releasing factor (CRF) stimulated adenylate cyclase activity in mouse splenic macrophages. Soc. for Neurosci. 14:667, 1988.

Sharkey, J., Appel, N.M. and De Souza, E.B. Functional alterations in local cerebral glucose utilization following central administration of corticotropin-releasing factor (CRF) in rats. Soc. for Neurosci. 14:667, 1988.

Kant, G.J., Anderson, S.M., and De Souza, E.B. Effects of chronic stress on brain and pituitary corticotropin-releasing factor (CRF) receptors. Soc. for Neurosci. 14:668, 1988.

Tracey, D.E. and De Souza, E.B. Identification of interleukin-1 receptors in mouse pituitary cell membranes and AtT-20 pituitary tumor cells. Soc. for Neurosci. 14:1052, 1988.

Wolfe, S.A., Culp, S.G., and De Souza, E.B. Sigma and phencyclidine (PCP) receptors in rat endocrine organs. Soc. for Neurosci. 14:1176, 1988.

Tracey, D.E., Webster, E.L., Grigoriadis, D.E. and De Souza, E.B. Corticotropin-releasing factor (CRF) and interleukin-1 (IL-1) receptors in the brain-pituitary-immune axis. 27th Annual Meeting of Am. College of Neuropsychopharmacology, San Juan, Puerto Rico, p. 3, 1988.

Insel, T.R., Johannessen, J.N., Battaglia, G. and De Souza, E.B. MDMA selectively destroys brain serotonin terminals in rhesus monkeys. 27th Annual Meeting of Am. College of Neuropsychopharmacology, San Juan, Puerto Rico, p. 207, 1988.

De Souza, E.B. Neuroendocrine effects of benzodiazepines. McLean Hospital Symposium on Benzodiazepines: Therapeutic, Biologic and Psychosocial Issues, Boston, MA, September 30, October 1, 1988.

Wolfe, S.A. Jr., Culp, S. and De Souza, E.B. Sigma receptors in the brain-endocrine-immune axis. Fed. Proc. Abstract, 1989.

Kapcala, L.P. and De Souza, E.B. Glucocorticoid-induced decrease of corticotropin-releasing factor receptors in brain cell cultures, Winter Neuropeptide Conference, Breckenridge, Colorado, 1989.

Appel, N.M. and De Souza, E.B. Effects of fenfluramine on brain monoamine neurons: evidence from immunohistochemical and autoradiographic studies. Int. Symp. on Serotonin from Cell Biology to Pharmacology and Therapeutics. Florence, Italy, March 29 - April 1, 1989.

Kapcala, L.P. and De Souza, E.B. Glucocorticoid-induced decrease of corticotropin-releasing factor receptor concentrations in brain cell cultures. Am. Fed. Clin. Res., 1989.

Battaglia, G., Sharkey, J., Kuhar, M.J., and De Souza, E.B. Neuroanatomical specificity of MDA- and MDMA-induced degeneration of serotonin neurons in rat brain. Greater Chicago ASPET, 1989.

Titeler, M., Appel, N.M., De Souza, E.B. and Glennon, R.A. Receptor pharmacology of MDMA and related hallucinogens. N.Y. Acad. of Sci. Meeting on The Neuropharmacology of Serotonin. July 10-13, Abstract #45, 1989.

De Souza, E.B., Battaglia, G. and Insel, T.R. Neurotoxic effects of MDMA on brain serotonin neurons: Evidence from neurochemical and radioligand binding studies. N.Y. Acad. Sci. Meeting on The Neuropharmacology of Serotonin. July 10-13, Abstract #48, 1989.

Webster, E.L., Tracey, D.E., Wolfe, S.A. Jr., Jutila, M.A., and De Souza E.B. Corticotropin-releasing factor (CRF) receptors are present on mouse splenic macrophages. Soc. for Neurosci. 15:8, 1989.

Grigoriadis, D.E., Pearsall, D., and De Souza, E.B. Characterization of high affinity digitonin-solubilized corticotropin-releasing factor (CRF) receptors in rat cerebral cortex. Soc. for Neurosci. 15:186, 1989.

Wolfe, S.A. Jr. and De Souza, E.B. Sigma receptors in lymphocytes: Quantification on T and B cells and distribution in lymphoid tissues. Soc. for Neurosci. 15:381, 1989.

Johnson, J.E. Jr., De Souza, E.B., and Weissman, A.D. Fine structural effects of amphetamine and its analogs on cultured neuroblastoma--glioma cells (NG108-15). Soc. for Neurosci. 15:418, 1989.

Appel, N.M., Mitchell, Wm. M., Contrera, J.F., and De Souza, E.B. Effects of high-dose fenfluramine treatment on structural integrity of rat brain 5HT neurons: Assessment using quantitative autoradiography of ^{3}H-paroxetine-labeled 5HT uptake sites. Soc. for Neurosci. 15:419, 1989.

Weissman, A.D., Casanova, M.F., and De Souza, E.B. Phencyclidine (PCP) exposure in human addicts does not alter regional sigma binding. Soc. for Neurosci. 15:636, 1989.

De Souza, E.B., Zaczek, R., Owens, M., Culp, S., Appel, N.M., and Nemeroff, C.B. Effects of fenfluramine treatment on corticotropin-releasing factor (CRF) activity in rat brain. Soc. for Neurosci. 15:800, 1989.

Zaczek, R., Culp, S., and De Souza, E.B. Intrasynaptosomal sequestration of ^{3}H-methylenedioxyamphetamine (MDA) and ^{3}H-amphetamine (AMPH). Soc. for Neurosci. 15:814, 1989.

Grzanna, R., Zaczek, R., Fritschy, J.M., Culp, S., and De Souza, E.B. DSP-4 has different affinities for the norepinephrine (NE) carrier in cerebral cortex and hypothalamus. Soc. for Neurosci. 15:1009, 1989.

Culp, S., Grzanna, R., De Souza, E.B., Fritschy, J.M., and Zaczek, R. Cortical and hypothalamic norepinephrine (NE) transport processes differ in their kinetic and pharmacologic properties. Soc. for Neurosci. 15:1009, 1989.

Yeh, S.Y. and De Souza, E.B. Lack of neurochemical evidence for neurotoxic effects of repeated cocaine administration in rats on brain monoamine neurons. Soc. for Neurosci. 15:1098, 1989.

De Souza, E.B., Grigoriadis, D.E. and Webster, E.L. Corticotropin-releasing hormone receptors in the brain-pituitary-immune axis: Pharmacology, biochemistry and localization studies. In: Corticotropin-Releasing Hormone (CRH): Relevance to Human Physiology and Pathophysiology, Bethesda, MD Sept. 8, 9, 1989.

De Souza, E.B., Appel, N.M., Mitchell, Wm. M., Garlick, R.K., Glennon, R.A. and Titeler, M. Autoradiographic characterization of [^{125}I]DOI: A novel phenylisopropylamine derivative which labels both $5HT_2$ and $5HT_{1c}$ receptors. 1989 American College of Neuropsychopharmacology, Maui, Hawaii, p. 128, 1989.

Goeders, N.E. and De Souza, E.B. Effects of cocaine on corticotropin- releasing factor receptors in the rat brain. 1989 American College of Neuropsychopharmacology, Maui, Hawaii, p. 163, 1989.

Grigoriadis, D.E. and De Souza, E.B. Biochemical isolation, characterization and partial purification of corticotropin-releasing factor (CRF) receptors from rat brain. 1989 American College of Neuropsychopharmacology, Maui, Hawaii, p. 140, 1989.

Abstracts in Press

Appel, N.M., Zaczek, R., Owens, M., Culp, S., Nemeroff, C.B. and De Souza, E.B. Relationships between brain serotonin (5HT) and corticotropin-releasing hormone (CRH) in the anti-obesity effects of fenfluramine. 1989 Winter Neuropeptide Conference, Breckenridge, Colorado, 1989.

Zaczek, R., Culp, S., McCann, D. and De Souza, E.B. Sequestration of ^{3}H-amphetamine into rat brain synaptosomes. American Society of Neurochemistry Meeting, 1990.

Appel, N.M., Mitchell, Wm.M., Garlick, R.K., Glennon, R.A., Titeler, M., and De Souza, E.B. Autoradiographic characterization of ^{125}I-labeled 2,5-dimethoxy-4-iodophenylisopropylamine (DOI): A phenylisopropylamine derivative labeling both $5HT_2$ and $5HT_{1c}$ receptors. FASEB, 1990.

Wolfe, S.A. Jr., Aguayo, L.G. and De Souza, E.B. Sigma receptors in rat pineal gland: Electrophysiology and autoradiographic localization. FASEB, 1990.

Yeh, S.Y. and Haertzen, C.A. Sensitization and desensitization induced by chronic cocaine administration on locomotor activity in rats. FASEB 1990.

Yeh, S.Y. Metabolic profile of tripelennamine in man and identification of nortripelennamine carbamic and acid as its major metabolite: A new metabolic pathway. XIth Int. Cong. of Pharmacol., 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00300-02 NBL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Metabolism of Tripelennamine and Pyrilamine

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: S.Y. Yeh Pharmacologist NBL, ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH

Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.5	0.5	0.0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Only tentative evidence for N-depyridination and N-dedimethylaminoethylation of tripelennamine and pyrilamine was previously obtained because of the lack of suitable standards. In the present work, we used recently obtained known standards to provide conclusive evidence of additional new metabolic pathways of tripelennamine and pyrilamine in rats, namely N-depyridination and N-dedimethylaminoethylation. Urine of rats administered tripelennamine or pyrilamine was extracted with organic solvent. The extracts with or without derivatization with Tri-Sil-Z were examined by GC/MS. N-(2-dimethylaminoethyl)benzylamine and 2-benzylaminopyridine were identified as two new urinary metabolites of tripelennamine in the rat. 2-(4-methoxybenzylamino)-pyridine and N-(dimethylaminoethyl)-4-hydroxybenzylamine were identified as new urinary metabolites of pyrilamine. Thus, in addition to N- and O-demethylation, hydroxylation and glucuronidation, N-debenzylation, N-depyridination and N-dedimethylaminoethylation were shown to be novel pathways for metabolism of tripelennamine and pyrilamine. N-Debenzylation and N-dedimethylaminoethylation of tripelennamine and pyrilamine occurs via alpha-carbon oxidation following the known mechanism of N-demethylation of tertiary amines. N-depyridination may occur through epoxide and dihydrodiol intermediate and molecular rearrangement. These findings may have general significance for the metabolism of other tertiary amines with aromatic moieties.

GPO 914-918

PUBLICATIONS
ZO1 DA 00300-02 NBL

Metabolism of Tripelennamine and Pyrilamine

Hsu, F.-L., Yeh, S.Y. and Munavalli, S. O-demethylation of pyrilamine. J. Pharmc. Sci., 77:727-728, 1988.

Yeh, S.Y. N-Depyridination and N-dedimethylaminoethylation of tripelennamine and pyrilamine in the rat: New metabolic pathways. Pharmacologist, 31:130, 1989.

Yeh, S.Y. and Hsu, F.-L. Quantitative metabolic profile of tripelennamine and pyrilamine in the rat. Drug Metabolism and Disposition, 16:499-502, 1988.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00302-02 NBL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Neurotoxic Effects of MDA and MDMA (Ecstasy)

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI:	E.B. De Souza	Chief	NBL, ARC, NIDA
Others:	R. Zaczek	Staff Fellow	NBL, ARC, NIDA
	N.M. Appel	Staff Fellow	NBL, ARC, NIDA
	A. Weissman	Staff Fellow	NBL, ARC, NIDA
	S.Y. Yeh	Scientist	NBL, ARC, NIDA
	T. Insel	Scientist	LCS, NIMH

COOPERATING UNITS (if any)

Laboratory of Clinical Science, NIMH

LAB/BRANCH
Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3.00	2.2	0.8

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The goal of the project is 1) to study the neurochemical mechanisms through which 3,4-methylenedioxyamphetamine (MDA), 3,4-methylenedioxymethamphetamine (MDMA) and related amphetamine derivatives produce their neurotoxic effects in CNS and 2) to examine the pharmacological profile of amphetamines and related derivatives at various brain recognition sites.

1. Sub-chronic administration of MDA and MDMA produce selective decreases in both 5-HT and 5-HIAA with no major changes in catecholamines in discrete areas of rat and rhesus monkey brain, drastic reductions in 5-HT uptake sites and massive destruction of 5-HT preterminals. The autoradiographic and immunocytochemical data demonstrate that the neurotoxic effects of these compounds on destruction of serotonin terminals is not diffuse but rather is limited to certain brain areas. In other studies, we examined the potential neurotoxic effects of amphetamine, methamphetamine, MDA, MDMA, MDE and fenfluramine in the serotonin-containing neuroblastoma cell line NG108-15. All of the compounds listed above produced decreases in the number of viable neuroblastoma cells at 10^{-3}M, but had little effect on viability or cell morphology at lower doses. However, electron microscopy revealed cell degeneration and ultrastructural changes at pharmacologically relevant drug levels (10^{-6}M). Together, these *in vitro* and *in vivo* studies suggest neurotoxic effects of amphetamine class of drugs which goes beyond simple loss of cell monoamine content.

2. In other studies, we examined the incorporation of ^{3}H-MDA and ^{3}H-amphetamine into rat brain synaptosomes and identified two saturable sites with high capacities. Overall, the data indicate that incorporation of ^{3}H-MDA and ^{3}H-amphetamine represent a sequestration phenomenon. The intrasynaptosomal [illegible] of ^{3}H-MDA and ^{3}H-amphetamine may be important in the [illegible] mechanism of [illegible] release [illegible] by the amphetamines.

PUBLICATIONS
ZO1 DA 00302-02 NBL

Neurotoxic Effects of MDA and MDMA (Ecstasy)

O'Hearn, E., Battaglia, G., De Souza, E.B. Kuhar, M.J., and Molliver, M.E.: Methylenedioxyamphetamine (MDA) and methylenedioxymethamphetamine (MDMA) cause ablations of serotonin axon terminals in forebrain: immunocytochemical evidence. J. Neuroscience 8:2788-2803, 1988.

Insel, T.R., Battaglia, G., Johannessen, J.N., Marra, S. and De Souza, E.B. 3,4-Methylenedioxymethamphetamine (MDMA; "Ecstasy") selectively destroys brain serotonin terminals in Rhesus monkeys. J. Pharmacol. Exp. Ther. 249:713-720, 1989.

Zaczek, R. and De Souza, E.B.: Identification and characterization of MDA and MDMA incorporation in brain. In: K. Asghar and E.B. De Souza (Eds.): Pharmacology and Toxicology of amphetamine and related designer drugs. NIDA Research Monographs 94:223-239, 1989.

Battaglia, G. and De Souza, E.B.: Pharmacologic profile of amphetamine derivatives at various brain recognition sites: Selective effects on serotonergic systems. In: K. Asghar and E.B. De Souza (Eds.): Pharmacology and Toxicology of amphetamine and related designer drugs. NIDA Research Monographs 94:240-258, 1989

Titeler, M., Leonhardt, S., Appel, N.M., De Souza, E.B. and Glennon, R.A. [^{125}I]DOI and [^{3}H]DOB: Novel agonist radioligand probes for $5HT_2$ and $5HT_{1c}$ receptors. Du Pont Biotech Update 4(6):20-24, 1989.

De Souza, E.B. The "Ecstacy" debate. Medical Aspects of Human Sexuality 23:40, 48,1989.

Zaczek, R., Culp, S.G. and De Souza, E.B. Intrasynaptosomal sequestration of [^{3}H]-amphetamine and [^{3}H]-methylenedioxyamphetamine: Characterization suggests the presence of a factor responsible for maintaining sequestration. J. Neurochem. (in press).

De Souza, E.B. and Battaglia, G.: Effects of MDMA and MDA on brain serotonin neurons: evidence from neurochemical and autoradiographic studies. In: K. Asghar and E.B. De Souza (Eds.): Pharmacology and Toxicology of amphetamine and related designer drugs. NIDA Research Monographs, (in press).

Battaglia, G., Zaczek, R. and De Souza, E.B.: MDMA effects in brain: Pharmacologic profile and evidence of neurotoxicity from neurochemical and autoradiographic studies. In: S.J. Peroutka (Ed.): MDMA: "Ecstasy" and/or human neurotoxin? Kluwer Academic Publishers, Boston, (in press).

Neurotoxic Effects of MDA and MDMA (Ecstasy)

Titeler, M., Appel, N.M., De Souza, E.B. and Glennon, R.A. Receptor pharmacology of MDMA and related hallucinogens. Annals New York Acad. Sci. (The Neuropharmacology of Serotonin) (in press).

De Souza, E.B., Battaglia, G. and Insel, T.R. Neurotoxic effects of MDMA on brain serotonin neurons: Evidence from neurochemical and radioligand binding studies. Annals New York Acad. Sci. (The Neuropharmacology of Serotonin) (in press).

Yousif, M., Battaglia, G., De Souza, E.B., Glennon, R.A. Sigma receptors: a putative site of action of MDMA (3,4-methylenedioxymethamphetamine) and related analogs. Soc. for Neurosci. 14:524, 1988.

Culp, S., Zaczek, R., and De Souza, E.B. Incorporation of ^{3}H-methylenedioxyamphetamine (MDA) into rat brain synaptosomes. Soc. for Neurosci. 14:557, 1988.

Battaglia, G., Sharkey, J., Kuhar, M.J., and De Souza, E.B. Neuroanatomical specificity of MDA- and MDMA-induced degeneration of serotonin neurons in rat brain. Soc. for Neurosci. 14: 557, 1988.

Johannessen, J.N., Insel, T.R., Battaglia, G., Kuhar, M.J., and De Souza, E.B. MDMA selectively destroys brain serotonin terminals in Rhesus monkeys. Soc. for Neurosci. 14:557, 1988.

Insel, T.R., Johannessen, J.N., Battaglia, G. and De Souza, E.B. MDMA selectively destroys brain serotonin terminals in rhesus monkeys. 27th Annual Meeting of Am. College of Neuropsychopharmacology, San Juan, Puerto Rico, p. 207, 1988.

Battaglia, G., Sharkey, J., Kuhar, M.J., and De Souza, E.B. Neuroanatomical specificity of MDA- and MDMA-induced degeneration of serotonin neurons in rat brain. Greater Chicago ASPET, 1989.

Titeler, M., Appel, N.M., De Souza, E.B. and Glennon, R.A. Receptor pharmacology of MDMA and related hallucinogens. N.Y. Acad. of Sci. Meeting on The Neuropharmacology of Serotonin. July 10-13, Abstract #45, 1989.

De Souza, E.B., Battaglia, G. and Insel, T.R. Neurotoxic effects of MDMA on brain serotonin neurons: Evidence from neurochemical and radioligand binding studies. N.Y. Acad. Sci. Meeting on The Neuropharmacology of Serotonin. July 10-13, Abstract #48, 1989.

Neurotoxic Effects of MDA and MDMA (Ecstasy)

Johnson, J.E. Jr., De Souza, E.B., and Weissman, A.D. Fine structural effects of amphetamine and its analogs on cultured neuroblastoma--glioma cells (NG108-15). Soc. for Neurosci. 15:418, 1989.

Zaczek, R., Culp, S., and De Souza, E.B. Intrasynaptosomal sequestration of ^{3}H-methylenedioxyamphetamine (MDA) and ^{3}H-amphetamine (AMPH). Soc. for Neurosci. 15:814, 1989.

De Souza, E.B., Appel, N.M., Mitchell, Wm. M., Garlick, R.K., Glennon, R.A. and Titeler, M. Autoradiographic characterization of [^{125}I]DOI: A novel phenylisopropylamine derivative which labels both $5HT_2$ and $5HT_{1c}$ receptors. 1989 American College of Neuropsychopharmacology, Maui, Hawaii, p. 128, 1989.

Zaczek, R., Culp, S., McCann, D. and De Souza, E.B. Sequestration of ^{3}H-amphetamine into rat brain synaptosomes. American Society of Neurochemistry Meeting, 1990.

Appel, N.M., Mitchell, Wm.M., Garlick, R.K., Glennon, R.A., Titeler, M., and De Souza, E.B. Autoradiographic characterization of ^{125}I-labeled 2,5-dimethoxy-4-iodophenylisopropylamine (DOI): A phenylisopropylamine derivative labeling both $5HT_2$ and $5HT_{1c}$ receptors. FASEB, 1990.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

*Z01 DA 00303-02 NBL
Z01 DA 00310-01 NBL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

CRF in Addictive, Neuropsychiatric and Neurodegenerative Disorders

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: E. De Souza

Others:	D.E. Grigoriadis	Postdoctoral Fellow	NBL, ARC, NIDA
	D. Price	Professor	JHU
	N. Goeders	Associate Professor	LSU

COOPERATING UNITS *(If any)*

Neuropathology Laboratory, JHU
Department of Pharmacology, Louisiana State University Medical Center

LAB/BRANCH

Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3.0	2.0	1.0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☒ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Recent clinical and preclinical data suggest that corticotropin-releasing factor (CRF) plays a major role in several neuropsychiatric disorders including major depression and panic/anxiety disorders and in neurodegenerative diseases including Alzheimer's disease. Furthermore, recent preclinical data demonstrating that the administration of CRF, like stress, produces sensitization to the behavioral responses to amphetamine demonstrate direct interactions between drugs of abuse and CRF. We have carried out preclinical studies examining the effects of chronic treatment with antidepressants, benzodiazepines or cocaine on modulation of CRF receptors in discrete areas of rat brain and in anterior pituitary. In addition, we have examined changes in CRF and its receptors in postmortem human tissue obtained from controls, and patients who died of Alzheimer's disease, Parkinson's disease, progressive supranuclear palsy and Huntington's disease. Overall, the data support the hypothesis that antidepressants and benzodiazepines may produce some of their "therapeutic" effects by altering CRF secretion. Furthermore, the data suggest that cocaine activates CRF in brain primarily in the mesolimbic/mesocortical dopaminergic system. With regard to neurodegenerative disorders, changes in CRF and its receptors were noted in brain areas that are affected in the specific disorders.

*These studies have been combined.

PUBLICATIONS
ZO1 DA 00303-02 NBL
ZO1 DA 00310-01 NBL

CRF in Addictive, Neuropsychiatric and Neurodegenerative Disorders

De Souza, E.B.: CRH defects in Alzheimer's and other neurologic diseases. Hospital Practice 23:59-71, 1988.

Grigoriadis, D.E., Struble, R.G., Price, D.L. and De Souza, E.B. Normal pattern of labeling of cerebral cortical corticotropin-releasing factor (CRF) receptors in Alzheimer's disease: evidence from chemical cross-linking studies. Neuropharmacology 28:761-764, 1989.

De Souza, E.B., Bissette, G., Whitehouse, P.J., Price, D.L., Vale, W.W., and Nemeroff, C.B.: Role of corticotropin-releasing factor (CRF) in neurodegenerative diseases. In: E.B. De Souza and C.B. Nemeroff (Eds.): Corticotropin-releasing factor: Basic and Clinical Studies of a Neuropeptide CRC Press, Boca, Raton, FL, (in press).

Grigoriadis, D.E., Pearsall, D. and De Souza, E.B. Effects of chronic antidepressant and benzodiazepine treatment on corticotropin-releasing factor receptors in rat brain and pituitary. Neuropsychopharmacology 2:53-60, 1989.

Pearsall, D., Grigoriadis, D.E., and De Souza, E.B. Effects of chronic antidepressant and benzodiazepine treatment on corticotropin-releasing factor (CRF) receptors in rat brain. Soc. for Neurosci. 14:667, 1988.

Goeders, N.E. and De Souza, E.B. Effects of cocaine on corticotropin-releasing factor receptors in the rat brain. 1989 American College of Neuropsychopharmacology, Maui, Hawaii, p. 163, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

*Z01 DA 00301-02 NBL
Z01 DA 00304-02 NBL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

CRF as a Stress Neurotransmitter in the Brain-Endocrine-Immune Axis

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.B. De Souza	Chief	NBL, ARC, NIDA
Others:	D.E. Grigoriadis	Postdoctoral Fellow	NBL, ARC, NIDA
	E.L. Webster	Staff Fellow	NBL, ARC, NIDA

COOPERATING UNITS *(If any)*

LAB/BRANCH

Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
3.5	2.8	0.7

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Corticotropin-releasing factor (CRF) is a critical hormone involved with stress responses. In addition to its role in regulating stress responses via the endocrine system, recent evidence suggest that CRF may act in brain to integrate the endocrine, autonomic, behavioral and immune responses of the body to stress. In addition, direct immunomodulatory actions of CRF have been demonstrated in the periphery. To provide additional evidence for CRF as a neurotransmitter/neuromodulator in brain and in immune tissues, we have carried out a series of studies to identify receptor binding sites for CRF in the CNS and in spleen. Biochemical, cellular, pharmacological and neuroanatomical studies have been utilized for studying the characteristics and distributions of CRF and its receptors, the second messenger systems through which CRF produces its many effects, and establishing molecular neurobiological techniques to identify specific intracellular messenger RNA for CRF. We have identified high affinity binding sites for CRF in brain and in spleen. The anatomical distribution of these sites in brain corresponds with the immunocytochemical distribution of CRF-containing terminals and the pharmacological sites of action of CRF in brain. In spleen, CRF binding sites appeared to be localized primarily to macrophages. In addition, we have demonstrated that CRF stimulates adenylate cyclase activity both in brain and in spleen. In biochemical studies, we have identified the ligand binding subunits of CRF receptors in brain and anterior pituitary of a number of species by chemical affinity cross-linking techniques. Studies are currently underway to purify the CRF receptor in brain.

*These studies have been combined.

PUBLICATIONS
Z01 DA 00301-02 NBL
Z01 DA 00304-02 NBL

CRF as a Stress Neurotransmitter in the Brain-Endocrine-Immune Axis

Insel, T.R., Battaglia, G., Fairbanks, D.W., and De Souza, E.B.: The development of brain receptors for corticotropin-releasing factor and their functional association with adenylate cyclase. J. Neurosci. 8:4151-4158, 1988.

Grigoriadis, D.E. and De Souza, E.B. Deglycosylation of rat brain and pituitary corticotropin-releasing factor (CRF) receptors. Soc. for Neurosci. 14:667, 1988.

De Souza, E.B. and Grigoriadis, D.E. Characterization of corticotropin-releasing factor (CRF) receptors in the intermediate lobe of the pituitary. Soc. for Neurosci. 14:667, 1988.

Pearsall, D., Grigoriadis, D.E., and De Souza, E.B. Effects of chronic antidepressant and benzodiazepine treatment on corticotropin-releasing factor (CRF) receptors in rat brain. Soc. for Neurosci. 14:667, 1988.

Sharkey, J., Appel, N.M. and De Souza, E.B. Functional alterations in local cerebral glucose utilization following central administration of corticotropin-releasing factor (CRF) in rats. Soc. for Neurosci. 14:667, 1988.

Kant, G.J., Anderson, S.M., and De Souza, E.B. Effects of chronic stress on brain and pituitary corticotropin-releasing factor (CRF) receptors. Soc. for Neurosci. 14:668, 1988.

Bell, J.A. and De Souza, E.B.: Functional corticotropin-releasing factor (CRF) receptors in the neonatal rat spinal cord: evidence from autoradiographic and electrophysiological studies. Peptides 9:1317-1322, 1989.

Cummings, S., Young III, W.S., Bishop, G., De Souza, E.B. and King, J.S.: Distribution of corticotropin releasing factor in the cerebellum and precerebellar nuclei of the opossum: a study utilizing immunohistochemistry, in situ hybridization histochemistry and receptor binding. J. Comp. Neurol. 280:501-521,1989.

Grigoriadis, D.E. and De Souza, E.B.: Corticotropin-releasing factor (CRF) receptors in intermediate lobe of the pituitary: biochemical characterization and autoradiographic localization. Peptides 10:179-188, 1989.

Sharkey, J., Appel, N.M. and De Souza, E.B. Alterations in local cerebral glucose utilization following central administration of corticotropin-releasing factor (CRF) in rats. Synapse 4:80-87, 1989.

CRF as a Stress Neurotransmitter in the Brain-Endocrine-Immune Axis

Grigoriadis, D.E. and De Souza, E.B. Heterogeneity between brain and pituitary corticotropin-releasing factor (CRF) receptors is due to differential glycosylation. Endocrinology 125:1877-1888, 1989.

Grigoriadis, D.E., Zaczek, R., Pearsall, D. and De Souza. Solubilization of high-affinity corticotropin-releasing factor (CRF) receptors from rat brain: Characterization of an active digitonin-solubilized receptor complex. Endocrinology 125:3068-3077, 1989.

Insel, T.R., Battaglia, G. and De Souza, E.B. Battaglia, G. and De Souza, E.B.: Brain Corticotropin Releasing Factor and Development. In: O. Zinder and S. Bresnitz (Eds.): Molecular Biology of Stress, Alan R. Liss, New York, NY, pp. 19-30, 1989.

Kapcala, L.P. and De Souza, E.B. Glucocorticoid-induced decrease of corticotropin-releasing factor receptors in brain cell cultures, Winter Neuropeptide Conference, Breckenridge, Colorado, 1989.

Kapcala, L.P. and De Souza, E.B. Glucocorticoid-induced decrease of corticotropin-releasing factor receptor concentrations in brain cell cultures. Am. Fed. Clin. Res., 1989.

Grigoriadis, D.E., Pearsall, D., and De Souza, E.B. Characterization of high affinity digitonin-solubilized corticotropin-releasing factor (CRF) receptors in rat cerebral cortex. Soc. for Neurosci. 15:186, 1989.

De Souza, E.B., Grigoriadis, D.E. and Webster, E.L. Corticotropin-releasing hormone receptors in the brain-pituitary-immune axis: Pharmacology, biochemistry and localization studies. In: Corticotropin-Releasing Hormone (CRH): Relevance to Human Physiology and Pathophysiology, Bethesda, MD Sept. 8, 9, 1989.

Grigoriadis, D.E. and De Souza, E.B. Biochemical isolation, characterization and partial purification of corticotropin-releasing factor (CRF) receptors from rat brain. 1989 American College of Neuropsychopharmacology, Maui, Hawaii, p. 140, 1989.

De Souza, E.B. and Insel, T.R. Corticotropin-releasing factor (CRF) receptors in the rat central nervous system: autoradiographic localization studies. In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

CRF as a Stress Neurotransmitter in the Brain-Endocrine-Immune Axis

De Souza, E.B. and Grigoriadis, D.E. Corticotropin-releasing factor (CRF) receptors in brain: characterization and regulation. In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

Battaglia, G., Webster, E.L. and De Souza, E.B. Characterization of second messengers coupled to corticotropin-releasing factor (CRF) receptors in brain. In: Corticotropin-Releasing Factor: Basic and Clinical Studies of a Neuropeptide, (E.B. De Souza and C.B. Nemeroff, eds.), CRC Press, Boca Raton, FL., (in press).

De Souza, E.B., Grigoriadis, D.E. and Webster, E.L. Role of brain, pituitary and spleen corticotropin-releasing factor (CRF) receptors in the stress response. In: The stress of life, revisited (Methods and Achievements in Experimental Pathology, (G. Jasmin and M. Cantin, eds.), S. Karger, Switzerland (in press).

Webster, E.L., Battaglia, G. and De Souza, E.B. Functional corticotropin-releasing factor (CRF) receptors in mouse spleen: evidence from adenylate cyclase studies. Peptides 10:395-401, 1989.

Webster, E.L., Grigoriadis, D.E. and De Souza, E.B. Corticotropin-releasing factor receptors in the brain-pituitary-immune axis. In: Stress, Neuropeptides, and Systemic Disease (J.A. McCubbin, P.G. Kaufmann and C.B. Nemeroff, eds.) (in press).

De Souza, E.B., Webster, E.L., Grigoriadis, D.E. and Tracey, D.E. Corticotropin-releasing factor (CRF) and Interleukin-1 (IL-1) receptors in the brain-pituitary-immune axis. Psychopharmacology Bulletin (in press).

De Souza, E.B., Grigoriadis, D.E. and Webster, E.L. Role of brain, pituitary and spleen corticotropin-releasing factor (CRF) receptors in the stress response. In: The stress of life, revisited (Methods and Achievements in Experimental Pathology) (G. Jasmin and M. Cantin, eds.), S. Karger, Switzerland (in press).

Webster, E.L., Battaglia, G., and De Souza, E.B. Corticotropin-releasing factor (CRF) stimulated adenylate cyclase activity in mouse splenic macrophages. Soc. for Neurosci. 14:667, 1988.

Webster, E.L., Tracey, D.E., Wolfe, S.A. Jr., Jutila, M.A., and De Souza, E.B. Corticotropin-releasing factor (CRF) receptors are present on mouse splenic macrophages. Soc. for Neurosci. 15:8, 1989.

CRF as a Stress Neurotransmitter in the Brain-Endocrine-Immune Axis

De Souza, E.B., Grigoriadis, D.E. and Webster, E.L. Corticotropin-releasing hormone receptors in the brain-pituitary-immune axis: Pharmacology, biochemistry and localization studies. In: Corticotropin-Releasing Hormone (CRH): Relevance to Human Physiology and Pathophysiology, Bethesda, MD Sept. 8, 9, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00305-02 NBL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Neurotoxicity of Selected Drugs to Monoamine Neurons in Brain

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.B. De Souza	Chief	NBL, ARC, NIDA
Others:	R. Zaczek	Staff Fellow	NBL, ARC, NIDA
	N.M. Appel	Staff Fellow	NBL, ARC, NIDA
	J.C. Contrera	Pharmacologist	FDA

COOPERATING UNITS *(if any)*

Food and Drug Administration, Rockville, MD

LAB/BRANCH
Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.0	1.5	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Several drugs that are currently used to treat a variety of psychiatric disorders produce their effects through actions on brain monoaminergic systems in brain. While these drugs may produce their beneficial effects through alterations in neurotransmission, the neurotoxic actions of the drugs on monoaminergic neurons following repeated use remain poorly assessed. Thus, the goal of the project is to assess the effects of chronic administration of several antidepressant and appetite suppressant drugs on neurotoxicity to monoamine neurons in the brain. These drugs include fenfluramine, Ritalin^{R} (methylphenidate), pemoline (Cylert^{R}), methamphetamine, Wellbutrim^{R} (buproprion), citalopram and paroxetine. We observed that short-term fenfluramine treatment caused dose-dependent reductions in a variety of serotonergic markers (5-HT, 5-HIAA and 5-HT uptake sites) in a variety of brain regions; no major effects of the drug were noted on catecholamine markers. Immunocytochemical studies confirmed the neurochemical data and demonstrated neurotoxic effects of fenfluramine resulting in a profound reduction in fine-caliber 5-HT-immunoreactive fibers and terminals with no major effect on cell bodies. Furthermore, we confirmed the neurotoxic effects of short-term methamphetamine administration in rats to cause significant depletion of dopamine, noradrenaline and serotonin markers. On the other hand, psychostimulants such as pemoline and ritalin which are used clinically to treat attention deficit disorders did not elicit any long-term decreases in the monoamine markers characteristic of neurotoxic effects of the drug.

PUBLICATIONS
Z01 DA 00305-02 NBL

Neurotoxicity of Selected Drugs to Monoamine Neurons in Brain

Appel, N.M., Contrera, J.F. and De Souza, E.B. Fenfluramine selectively and differentially decreases the density of serotonergic nerve terminals in rat brain: evidence from immunocytochemical studies. J. Pharmacol. Exp. Ther. 249:928-943, 1989.

Zaczek, R., Battaglia, G., Contrera, J.F., Culp, S. and De Souza, E.B. Methylphenidate and pemoline do not cause depletion of rat brain monoamine markers similar to that observed with methamphetamine. Toxicol. Appl. Pharmacol. 100:227-233, 1989.

Appel, N.M., Mitchell, Wm. M., Contrera, J.F. and De Souza, E.B. Effects of high-dose fenfluramine treatment on monoamine uptake sites in rat brain: Assessment using quantitative autoradiography. Synapse (in press).

Zaczek, R., Battaglia, G., Contrera, J.F. and De Souza, E.B. Effects of repeated fenfluramine administration on indices of monoamine function in rat brain: Pharmacokinetics, dose response, regional specificity and time course data. J. Pharmacol. Exp. Ther. (in press).

Appel, N.M., Zaczek, R., Mitchell, W.M. and De Souza, E.B. Immunohistochemical and autoradiographic investigations of high-dose fenfluramine treatment on monoamine neurons in rat brain. In: Proceedings of Int. Symp. on Serotonin: From Cell Biology to Pharmacology and Therapeutics, Kluwer Academic Publishers, Boston (in press).

Contrera, J.F., Battaglia, G., Zaczek, R., and De Souza, E.B. Fenfluramine neurotoxicity: selective degeneration and recovery of brain serotonin neurons. Soc. for Neurosci. 14:556, 1988.

Appel, N.M. and De Souza, E.B. Fenfluramine selectively destroys serotonin terminals in brain: immunocytochemical evidence. Soc. for Neurosci. 14:556, 1988.

Zaczek, R., Battaglia, G., Contrera, J.F., and De Souza, E.B. Ritalin and pemoline do not cause monoamine terminal degeneration. Soc. for Neurosci. 14:557, 1988.

Appel, N.M. and De Souza, E.B. Effects of fenfluramine on brain monoamine neurons: evidence from immunohistochemical and autoradiographic studies. Int. Symp. on Serotonin from Cell Biology to Pharmacology and Therapeutics. Florence, Italy, March 29 - April 1, 1989.

Neurotoxicity of Selected Drugs to Monoamine Neurons in Brain

Appel, N.M., Mitchell, Wm. M., Contrera, J.F., and De Souza, E.B. Effects of high-dose fenfluramine treatment on structural integrity of rat brain 5HT neurons: Assessment using quantitative autoradiography of ^{3}H-paroxetine-labeled 5HT uptake sites. Soc. for Neurosci. 15:419, 1989.

De Souza, E.B., Zaczek, R., Owens, M., Culp, S., Appel, N.M., and Nemeroff, C.B. Effects of fenfluramine treatment on corticotropin-releasing factor (CRF) activity in rat brain. Soc. for Neurosci. 15:800, 1989.

Grzanna, R., Zaczek, R., Fritschy, J.M., Culp, S., and De Souza, E.B. DSP-4 has different affinities for the norepinephrine (NE) carrier in cerebral cortex and hypothalamus. Soc. for Neurosci. 15:1009, 1989.

Culp, S., Grzanna, R., De Souza, E.B., Fritschy, J.M., and Zaczek, R. Cortical and hypothalamic norepinephrine (NE) transport processes differ in their kinetic and pharmacologic properties. Soc. for Neurosci. 15:1009, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00306-02 NBL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Neurotransmitter Receptors in the Pituitary Gland

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: De Souza, E.B. Chief NBL, ARC, NIDA

COOPERATING UNITS *(if any)*

None

LAB/BRANCH
Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.8	0.4	0.4

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Serotonin and catecholamines have been shown to play a major role in regulating pituitary hormone secretion both through effects in brain and direct actions on the pituitary. The goals of the project were to identify, characterize and localize, using in vitro autoradiography, the relative distribution of serotonin-2, dopamine-2, beta-2 adrenergic and alpha-1 adrenergic receptors in the rat pituitary gland. In order to define the role of adrenomedullary catecholamines in regulating pituitary function, we examined the effects of adrenalectomy on beta-2 adrenergic receptors in the rat pituitary gland. The identification of the various receptor described above provides further evidence of the importance of these neurotransmitters in regulating pituitary function and demonstrates conditions in which these receptors can be modulated.

PUBLICATIONS
ZO1 DA 00306-02 MPL

Neurotransmitter Receptors in the Pituitary Gland

De Souza, E.B.: Localization and modulation of brain and pituitary receptors involved in stress responses. Psychopharmacology Bulletin 24:360-364, 1988.

De Souza, E.B. Autoradiographic localization of monoamine and corticotropin-releasing factor (CRF) receptors in the pituitary: effects of glucocorticoids and peripheral amines. In: Catecholamines and Other Neurotransmitters in Stress, (G.R. Van Loon, ed.), Gordon and Breach Science Publishers, New York, (in press).

De Souza, E.B. and Appel, N.M. Distribution of brain and pituitary receptors involved in mediating stress responses. In: Neurobiology and Neuroendocrinology of Stress (M.R. Brown, C. Rivier and G.F. Koob, eds.), Marcel Dekker, Inc., New York (in press).

De Souza, E.B. Neuroendocrine effects of benzodiazepines. J. Psychiatric Research (in press).

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00307-02 NBL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effects of Cocaine on Monoamines and their Metabolites in Rat Brain

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	S.Y. Yeh	Pharmacologist	NBL, ARC, NIDA
Others:	E.B. De Souza	Chief	NBL, ARC, NIDA

COOPERATING UNITS *(If any)*

LAB/BRANCH
Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.5	0.5	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Dopamine and other brain monoamines have been implicated in mediating the reinforcing properties and behavioral effects of cocaine. While the effects of cocaine administration on the concentration of brain monoamines have been examined, the reports in the literature are conflicting. The purpose of this series of studies is to examine, in detail, the effects of cocaine administration on changes in various monoamine markers in brain and on several behavioral parameters. Rats were injected with cocaine (20 mg/kg, s.c. or i.p. for 8 days) and sacrificed at 1, 8, 15 or 48 days after the last injection. The concentrations of norepinephrine (NE), dopamine (DA), serotonin (5-HT) and their metabolites, assayed by HPLC-EC, in frontal cortex, hippocampus, striatum, hypothalamus, midbrain, pons-medula and spinal cord were not significantly different from those in the saline-injected controls at any of the time points examined. These data suggest that the repeated cocaine administration in rats does not produce any long-term depletion in brain catecholamine and 5-HT content characteristic of neurotoxic actions of the drug.

PUBLICATIONS
Z01 DA 00307-02 NBL

Effects of Cocaine on Monoamines and their Metabolites in Rat Brain

Yeh, S. Y. and De Souza, E.B.: Lack of neurochemical evidence for neurotoxic effects of repeated cocaine administration in rats on brain monoamine neurons. Soc. for Neurosci. 15:803, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER

*Z01 DA 00301-02 NBL
Z01 DA 00308-02 NBL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Role of Sigma Receptors in Endocrine Organs and Immune Tissue

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI: E.B. De Souza Chief NBL, ARC, NIDA

Others: S.E. Wolfe Jr. Staff Fellow NBL, ARC, NIDA

COOPERATING UNITS *(if any)*

None

LAB/BRANCH

Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER:
1.8	1.0	0.8

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☒ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

In addition to their psychotomimmetic effects in the CNS, PCP and N-allylnormetazocine have been reported to produce immunosuppressant effects and to alter neuroendocrine function. Since PCP and N-allylnormetazocine bind with high affinity to both PCP and sigma receptors, the identity of the receptor(s) mediating the endocrine and immune effects of these compounds is unknown. Furthermore, it is unclear whether the effects of these compounds are mediated in brain or through direct actions at peripheral target organs. The aim of this study is to identify, characterize and localize sigma and PCP receptors in a variety of rat endocrine organs including the pituitary gland, adrenal, testis and ovary and in immune tissues including human peripheral blood leukocytes (HPBL) and spleen. Studies demonstrate the absence of high-affinity PCP receptors in any of the peripheral tissues described above and the presence of high concentration of sigma receptors with kinetic and pharmacological characteristics similar to those previously reported in brain. In autoradiographic studies, sigma receptors were discretely localized to the anterior lobe of the pituitary, adrenal cortex, seminiferous tubules of testis, maturing follicles of ovary and primarily to lymphocytes in spleen. The presence of high concentrations of sigma receptors in endocrine and immune tissues suggest a physiological role of endogenous sigma ligands in the integration of the CNS-endocrine-immune axis. Also, the endocrine markers and human peripheral HPBL leukocytes may represent use for peripheral markers in humans for assessing the role of these receptors in brain.

*These studies have been combined.

PUBLICATIONS
ZO1 DA 00301-02 NBL
ZO1 DA 00308-02 NBL

Role of Sigma Receptors in Endocrine Organs and Immune Tissue

Wolfe, S.A., Jr., Culp, S.G. and De Souza, E.B. Sigma receptors in endocrine organs: identification, characterization, and autoradiographic localization in rat pituitary, adrenal, testis and ovary. Endocrinology 124:1160-1172, 1989.

Wolfe, S.A. Jr. and De Souza, E.B. Sigma receptors in the brain-endocrine-immune axis. In: Sigma, PCP and NMDA receptor systems (E.B. De Souza, E.D. London and D.H. Clouet, eds.), NIDA Research Monographs (in press).

Wolfe, S.A., Culp, S.G., and De Souza, E.B. Sigma and phencyclidine (PCP) receptors in rat endocrine organs. Soc. for Neurosci. 14:1176, 1988.

Wolfe, S.A. Jr., Kulsakdinun, C., Battaglia, G., Jaffe, J.H., and De Souza, E.B.: Initial identification and characterization of sigma receptors in human peripheral blood leukocytes. J. Pharmacol. Exp. Ther. 247:1114-1119, 1988.

Wolfe, S.A. Jr. and De Souza, E.B. Sigma receptors in the brain-endocrine-immune axis. In: Sigma, PCP and NMDA receptor systems (E.B. De Souza, E.D. London and D.H. Clouet, eds.) NIDA Research Monographs (in press).

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00309-02 NBL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Interleukin-1 in the Brain-Endocrine-Immune Axis

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.B. De Souza	Chief	NBL, ARC, NIDA
Others:	T. Takao	Visiting Fellow	NBL, ARC, NIDA
	E. Webster	Staff Fellow	NBL, ARC, NIDA
	D.E. Tracey	Visiting Scientist	Upjohn

COOPERATING UNITS *(if any)*
The Upjohn Co., Kalamazoo, MI

LAB/BRANCH
Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.0	1.5	0.5

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The cytokine interleukin-1 (IL-1) is one of the key mediators of the immune response to stress, infection or antigenic challenge. In addition, IL-1 has a variety of effects in brain including its ability to cause fever and to induce slow-wave sleep. More recently, IL-1 has been reported to stimulate the hypothalamic-pituitary-adrenocortical axis. Whether IL-1 induces pituitary-adrenocortical secretion by direct stimulation of cells in the pituitary or indirectly through hypothalamic stimulation of corticotropin-releasing factor is controversial. The purpose of this study is to examine the role of IL-1 and its receptors in modulating the brain-endocrine-immune responses to stress. The initial studies will involve the identification, characterization and localization of IL-1 receptors in CNS, pituitary and immune tissues including spleen and immune cell lines. In preliminary studies, we have identified IL-1 receptors in rat and mouse pituitary, testis and in brain with characteristics similar to previously identified IL-1 receptors in EL-4 immune cells. In localization studies, IL-1 receptors in the pituitary appear to be localized primarily in the anterior lobe. Current studies are aimed at studying the interactions between IL-1 and other factors including corticotropin-releasing factor and monoamines in regulating adrenocorticotropic hormone secretion from pituitary cells.

PUBLICATIONS
Z01 DA 00309-02 NBL

Interleukin-1 in the Brain-Endocrine-Immune Axis

Tracey, D.E. and De Souza, E.B. Identification of interleukin-1 receptors in mouse pituitary cell membranes and AtT-20 pituitary tumor cells. Soc. for Neurosci. 14:1052, 1988.

Tracey, D.E., Webster, E.L., Grigoriadis, D.E. and De Souza, E.B. Corticotropin-releasing factor (CRF) and interleukin-1 (IL-1) receptors in the brain-pituitary-immune axis. 27th Annual Meeting of Am. College of Neuropsychopharmacology, San Juan, Puerto Rico, p. 3, 1988.

De Souza, E.B., Webster, E.L., Grigoriadis, D.E. and Tracey, D.E. Corticotropin-releasing factor (CRF) and Interleukin-1 (IL-1) receptors in the brain-pituitary-immune axis. Psychopharmacology Bulletin (in press).

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00311-01 NBL

PERIOD COVERED

October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*

Sigma and PCP Receptors in Neuropsychiatric Disorders

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	E.B. De Souza	Chief	NBL, ARC, NIDA
Others:	A.D. Weissman	Staff Fellow	NBL, ARC, NIDA
	E.D. London	Chief, Neuropharmacology Laboratory	NPL, ARC, NIDA
	M. Casanova	Scientist	NIMH
	J. Kleinman	Deputy Chief, Clin. Brain Dis. Br.	NIMH

COOPERATING UNITS *(If any)*

Neuropharmacology Laboratory, ARC, NIDA
Clinical Brain Disorders Branch, St. Elizabeth's Hospital, NIMH

LAB/BRANCH

Laboratory of Neurobiology, Neuroscience Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.5	1.3	0.2

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☒ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The psychotomimetic effects of certain cycloalkyls and benzomorphans that interact with PCP and/or sigma receptors has led to the hypothesis that these sites may be important in the etiology of schizophrenia. To investigate this hypothesis, PCP and sigma receptors were measured in homogenates of seven regions of postmortem brains of patients diagnosed as belonging to several subclasses of schizophrenia and in age-matched controls. Specific sigma binding was significantly decreased when compared to controls in the parietal cortex, temporal cortex and dentate nucleus. In contrast, PCP receptors were not consistently altered in any of the brain regions examined. The data demonstrating selective and differential regional decreases in sigma but not PCP receptors in schizophrenia suggest the involvement of sigma receptors in this disorder. In other studies, we examined whether chronic exposure to PCP in human addicts would alter sigma and PCP binding in postmortem human brains. Age and postmortem interval matched individuals who had committed suicide, but had no history or postmortem evidence of abusing drugs, served as controls. No differences were observed in either the affinity or the density of sigma or PCP binding sites in occipital, frontal and anterior cingulate cortex as well as in hippocampus and cerebellum in PCP addicts when compared to controls. These results suggest that brain sigma and PCP receptors remain unchanged in humans despite repeated challenges with a potent psychotomimetic drug.

PUBLICATIONS
Z01 DA 00311-01 NBL

Sigma and PCP Receptors in Neuropsychiatric Disorders

Weissman, A.D., Casanova, M.F., Kleinman, J.B., London, E.D., and De Souza, E.B. Regional differences in PCP and σ binding in the brains of schizophrenics. Soc. for Neurosci. 14:104, 1988.

Weissman, A.D., Casanova, M.F., and De Souza, E.B. Phencyclidine (PCP) exposure in human addicts does not alter regional sigma binding. Soc. for Neurosci. 15:636, 1989.

4. Molecular Neurobiology Laboratory - George R. Uhl, M.D., Ph.D., Chief

Overview

Drugs impact the nervous system through interactions with the products of specific groups of genes, some of which have been cloned and characterized and many of which are unknown. During this year, the Laboratory of Molecular Neurobiology has been formed within the Neuroscience Branch in order to strengthen and enhance the application of molecular biologic techniques and principles to studies of the structure and regulation of genes involved in the actions of abused drugs. Major areas of concentration include: function-and drug-induced gene regulation within specific neuronal groups in the brain, and studies of drug and neurotransmitter receptor genes. Insights derived from these studies are applied in human clinical studies.

A principal working hypothesis motivating current experiments is that the detailed mechanisms of regulation of specific neurotransmission-related genes in particular neuronal pathways can reflect, or even possibly store, information about previous stimuli or drugs to which the nervous system has been exposed. A major thrust of the laboratory's effort thus studies specific patterns of regulation of genes related to neurotransmitters impacted by abused drugs within specific neural populations in the brain. Genes for neurotransmitters that are modulated by abused substances such as preproenkephalin, genes for transcription factors that could be involved in this modulation, such as the Jun family, and genes expressed in key neurons of central brain pathways for reinforcement reward have been studied during the past year. Knowledge of some of these regulated mechanisms, in turn, guides the formulation of clinical studies in attempts to identify whether such mechanisms can be discerned and possibly manipulated in man.

A second major hypothesis is that better understanding of the detailed molecular structures of the receptor molecules for abused drugs, and the ability to manipulate these structures, will come through the cloning of cDNAs encoding these molecules. Current efforts recognize the difficulties of directly purifying these membrane protein molecules. Accordingly, major efforts in the laboratory focus on expression-cloning strategies for obtaining these very rare cDNAs.

Collaborative studies include workers at Harvard Medical School, Johns Hopkins School of Medicine, Albert Einstein College of Medicine, Mount Sinai School of Medicine, Osaka and Hiroshima Universities' Schools of Dentistry, and co-workers within the Neuroscience, Clinical Pharmacology, and Etiology Branches of the Addiction Research Center. As the laboratory reaches full strength during the coming year, increased collaborations with workers in the Preclinical Branch in studies of transgenic animals, and with workers

in the Etiology Branch concerning possible linkage mapping of genes selectively involved in drug abuse behaviors in human populations are being formulated.

The laboratory is thus developing and employing strategies for identifying receptor genes required for the initial actions of abused drugs, and focusing on the neuronal regulation of the expression of genes intimately involved with abused drugs. Such strategies should enhance appreciation of mechanisms of drug action and provide new potential avenues for drug abuse therapies.

Summary of Ongoing Research

A. Receptor cDNA Expression Cloning Using Ligand Autoradiographic Screening.

Identifying genes encoding the cell surface receptors for abused drugs is an important step in the molecular biology of drug abuse. Since these rare membrance proteins are difficult to purify through conventional means, the laboratory is developing expression-cloning approaches for identification of these genes. They have achieved a 100-1000-fold purification of a cDNA encoding a neurotensin binding site using this approach, for example. Current efforts include optimizing DNA recovery after the ligand autoradiographic screening procedure, increasing the rapidity and reliability of the procedure, and screening libraries for expression of several neurotransmitter and drug receptors.

B. Receptor cDNA Expression Cloning Using Xenopus Oocyte Expression

A second approach to cloning drug receptors utilizes the powerful Xenopus oocyte expression system. Current efforts focus on subfractionation of libraries for "sib selection" of drug and neurotransmitter receptor cDNAs. For example, the laboratory has achieved a 100-1000-fold purification of a dopamine transporter/cocaine receptor using this approach. This procedure, and the ligand autoradiographic screening, should allow direct cloning of receptor cDNAS without requiring purification of receptor protein

C. Neuronal Expression of Genes for Neurotransmitters Related to Drug Abuse I: Regulation of Opioid Peptide Genes

Understanding the ways in which individual neurons regulate their expression of the neurotransmitter genes that are related to drug abuse can help to elucidate neurochemical mechanisms for drug action, including mechanisms of tolerance and dependence. The laboratory has defined changes in the neuronal expression of several neuropeptide genes in both animal and human tissues that may relate to drug action. The morphine-induced changes in

preproenkephalin expression that these workers have found in striatal neurons exhibit a time course consistent with a role for this agonist induced down regulation in opiate tolerance/dependence. However, these changes differ from brain region to brain region, underscoring the importance of understanding the specific mechanisms of gene regulation in individual neuronal populations. Examination of these mechanisms has identified a specific transcription factor pathway that is well positioned to play a key role in the rapid proenkephalin upregulation noted after sensory stimuli. Elucidation of drug-induced changes in opioid peptide gene expression in neurons in specific parts of the nervous system is likely to reveal specific "adaptive" biochemical mechanisms that could well play roles in tolerance/dependence and provide specific sites for targeted therapeutics for drug abuse.

D. Neuronal Expression of Genes for Neurotransmitters Related to Drug Abuse II: Genes Implicated in Central Brain Pathways of Reinforcement/Reward

Specific neuronal pathways in the brain are selectively and importantly implicated in the reinforcing and rewarding properties of a number of abused drugs, especially cocaine. However, detailed elucidation of the way in which these circuits function, and the way in which they are influenced by abused drugs is lacking.

The laboratory has begun to approach these problems by examining the expression of genes encoding peptide neurotransmitters, cholecystokinin and neurotensin, in the ventral tegmental area, which contains the cells that form a key part of these important circuits. These studies show a substantial diversify of gene expression among the different subnuclei of the VTA and clearly point toward the possibility that different neuronal subpopulations in this area could display substantially different activities. Such studies are an important precursor to examination of drug effects on the expression of these genes in these important neurons.

Publications

Uhl G. An approach to in situ hybridication using oligonucleotide cNDA probes. In: Van Leeuwen FW, Buijs RM, Pool CW and Pach O (eds.), Molecular Neuroanatomy. Elsevier Science Publishers B.V., Amsterdam, 1988: 25-41.

Reppert, S.M. and Uhl, G.R. The Vasopression Gene is Expressed Prior to regulation in the supraoptic nuclei of fetal rats. Brain Res. 456: 392-396, 1988.

Uhl G.R., Navia B., and Douglass J. Differential expression of preproenkephalin and preprodynorphin mRNAs striatal neurons: High levels of preproenkephalin expression depend on cerebral cortical afferents J. Neurosci. 12: 4755-4764, 1988.

Nishimori T., Moskowitz M.A., Uhl G.R. Opioid peptide gene expression in rat trigeminal nucleus caudalis neurons: Normal distribution and effects of trigeminal deafferentation. J. Comp. Neurol. 274 (1): 142-150, 1988.

Uhl G.R., Ryan J., Schwartz J. Morphine alters preproenkephalin gene expression. Brain Res. 459: 391-397, 1988.

Voigt M.M., Uhl G.R. Preprocholecystokinin mRNA in rat brain: regional expression includes thalamus. Molecular Brain Research 4: 247-253, 1988.

Uhl G.R. Neuropeptide systems in Parkinson's disease and tardive dyskinesia. In: C. Nemeroff (ed.) Neuropeptides in Neuropsychiatric Disorders. Baltimore: Johns Hopkins University Press, 1988: 157-177.

Uhl, G.R. In Situ Hybridization: Quantitation Using Radiolabeled Hybridization Probes. In: P. M. Conn (ed.), Neuroendocrine Peptide Methodology, New York, Academic Press, 1989 pp 135-146.

Linnik M.D., Sakas D.E., Uhl G.R., Moskowitz M.A. Subarachnoid Blood and
Headache: Altered Trigeminal Tachykinin Gene Expression. Ann. Neurol. 25: 179-184, 1989.

Rivkees S.A., Chaar M.R., Hanley D.F., Maxwell M., Reppert S.M., Uhl G.R.
Localization and regulation of vasopressin mRNA in human neurons. Synapse 3: 246-254,1989.

Uhl G. In situ Hybridization: Quantitation using radiolabeled hybridization probes. In: P. M. Conn (ed.), Methods in Enzymology, New York, Academic Press, 1989, pp. 741-752.

Nishimori, T., Buzzi, M.G., Moskowitz, M.A. and Uhl, G.R. Preproenkephalin mRNA Expression in Nucleus Caudalis Neurons is Enhanced By Trigeminal Stimulation. Mol. Br. Res. 6: 203-210, 1989.

Rattray, M., Lautar S.L. and Uhl G.R. A new method of screening receptor cDNAs: Influence of plasmid competition on receptor expression. Biochem. Soc. Trans. 17: 1068-1069, 1989.

Articles in Press

Schwartz, J.P., Simantov, R., Ryan, J.P. and Uhl, G.R.: Control of Striatal Enkephalin Gene Expression. Proceed. VII Int. Congress of Endocrinology, (In press).

Rattray, M., Lautar, S.L. and Uhl, G.R. Ligand autoradiographic receptor expression screening I: assessment of transiently-expressed receptors by radioligand binding to replicas of transfected COS cell colonies. Mol. Br. Res. (In press).

Uhl, G.R., O'Hara, B., Shimada, S., Zacek, R., DiGiorgianni, J. and Nishimori, T. Dopamine Transporter: Expression in Xenopus oocytes. Mol. Br. Res. (In press).

Uhl G. Neurotensin receptors. In: A. Bjorklund, T. Hokfelt, M. Kuhar (eds.). Handbook of Chemical Neuroanatomy IV. New York, Elsevier, (In press).

Price D.L., Whitehouse P.J., Struble R.G., Hedreen J.C., Uhl G.R. Transmitter systems in selected types of dementia in N. Kopp and J. Pearson (eds). Human Neurotransmission in Health and Disease. New York: Oxford University Press, (In press).

Uhl G.R. Principles of assessing neurotransmitter receptors in disease. In: J. Frost and H. Wagner, eds. Quantitative Imaging of Neuroreceptors. New York: Raven Press (In press).

Uhl G.R. mRNA localization with the microscope: in situ hybridization using radiolabeled probes. In: Yamamura and Kuhar (eds.) Methods in Receptor Analysis, (In press).

Uhl G.R. Parkinson's disease: Neurotransmitter and neurotoxin receptors and their genes. European Neurology (In press).

Uhl G.R., Jayaraman A., Nishimori T., Shoichi S., Rattray M. and O'Hara B. Applications of neuropharmacologic techniques in the molecular neurobiology of schizophrenia. In: SC Schultz and CA Tamminga (eds.) Schizophrenia (In press).

Uhl, G.R. and Nishimori, T. Neuropeptide Gene Expression and Neural Actiivty: Assessing a working hypothesis in nucleus caudalis and dorsal horn neurons expressing preproenkephalin and preprodynorphin. Mol. Cell Neurobiol. (In press).

Jayaraman, A., Nishimori, T., Dobnar, P. and Uhl, G. Cholecystokinin and Neurotensin mRNAs are differentially expressed in subnuclei of the ventral tegmental area. J. Comp. Neurol. (In press).

Nishimori, T., Buzzi, M.G., Moskowitz, M.A. and Uhl, G.R. Differential effects of small- and large-caliber inputs on nucleus caudalis preproenkephalin expression. J. Neuroscience (In press).

Abstracts

Uhl, G.R.: Bind and clone: Ligand-autoradiographic receptor expression screening. Society for Neuroscience Abstract 14(1): 318, 1988.

Rattray, M., Lautar, S.L., and Uhl, G.R. Ligand autoradiographic receptor screening: validation using beta-adrenergic receptor cDNA. Society for Neuroscience Abstracts 15(2): 1270, 1989.

Rattray, M., Lautar, S.L. and Uhl, G.R. A new method of screening receptor cDNAs. The Biochemist (Abstract Supplement) 11(2) #80, P.57, 1989.

Linnik, M.D., Sakas, D.E., Uhl, G.R. and Moskowitz, M.A.: Subarachnoid blood alters preprotachykinin gene expression in rabbit trigeminal ganglia. FASEB J 2 (5): A1382, 1988.

Nishimori T., Moskowitz M., Borsook D., Maciewicz R. and Uhl G. Trigeminal and PAC modulation of opioid peptide gene expression in nucleus caudalis. Society for Neuroscience Abstracts 14(2): 857, 1988.

Borsook D., Nishimori T., Maciewicz R. and Uhl, G. Analgesic PAG stimulation modulates neuronal opioid peptide gene expression in raphe magnus and trigeminal nucleus caudalis. Neurology 39 (Suppl 1): 359, 1989.

Nishimori, T., Buzzi, G., Moskowitz, M. and Uhl, G.R. N. caudalis prepro-ENK mRNA: biphasic enhancement by afferents. Society for Neuroscience Abstracts 15 (1) 343,1989.

Fan S.G., Kushner L., Lerma J., O'Hara B.F., Uhl G.R., Bennett M.V.L. and Zukin R.S. Identification of size classes of mRNA encoding kainate and neurotensin receptors. Society for Neuroscience Abstracts 15 (1) 672, 1989.

Jayaraman A., Nishimori T., Dobnar P. and Uhl G.R. Differential expression of CCK and neurotensin (NT) mRNAs in the subnuclei of rat VTA. Society for Neuroscience Abstracts 15 (1) 902, 1989.

O'Hara B.F., DiGiorgianni J.M., Shimada S., Kushner L., Spivak C.E., Lerma, J., Zukin R.S., Bennett M.V.L. and Uhl G.R. Expression of the neurotensin receptor in Xenopus oocytes with RNA transcribed from lambda and pCDM8 libraries. Society for Neuroscience Abstracts 15 (1) 672, 1989.

Shimada S., O'Hara B.F., Nishimori T., DiGiorgianni J.M. and Uhl G.R. Dopamine transporter mRNA and cDNA: Xenopus oocyte expression. Society for Neuroscience Abstracts 15 (1) 106, 1989.

Abstracts in Press

Jayaraman A. and Uhl G.R. Cholecystokinin (CCK) mRNA: Prominent expression in the rostral nigral neurons. Proceedings First International Congress of Movement Disorders Abstract 1990 (In press).

Uhl G.R., Shimada S., O'Hara B., DiGiorgianni J., and Nishimori T. Dopamine transporter mRNA and cDNA: Strategy for expression cloning a selective neurotoxin concentrator. Proceedings First International Congress of Movement Disorders Abstract 1990 (In press).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00114-02-MNL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Receptor cDNA Expression Cloning Using Ligand Autoradiographic Screening

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

Uhl, G.R., Chief Laboratory of Molecular Neurobiology, ARC.
Others: Rattray, M.A.N., Foreign Fellow, MPL, ARC; Lautar, S., Research Lab Manager; MPL, ARC; Lin, Glen, Research Technician (guest worker) Dept. of Neuroscience, Johns Hopkins University School of Medicine, Baltimore, MD 21205.

COOPERATING UNITS *(if any)*

LAB/BRANCH
Laboratory of Molecular Neurobiology, Neuroscience Branch

SECTION
Gene Neuroscience Unit

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2	1	1

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Understanding the receptor molecules that recognize abused drugs and the neurotransmitters impacted by drugs is an important step in determining the molecular mechanisms underlying drug abuse. Little is known about many of these molecules, because they have proven very difficult to purify through conventional approaches. The laboratory thus continues to work to establish a method for directly cloning these molecules based on their ligand binding properties.

Over the past year, these workers have made substantial progress toward this end. Previously, a cloned beta adrenergic receptor cDNA was used to document and optimize expression of the beta adrenergic receptor binding site in COS cells. These studies have been completed during this year. Current efforts focus on improved DNA recovery techniques. With electroporation and other modifications; the plasmid present in very small number of cells can frequently be recovered. Progress in adapting the method for use with tritiated radioligands recognizing the cocaine and PCP receptors, and showing correlations between positives obtained from this method and the Xenopus oocyte injection paradigm has also been made. Goals for the current year include: improving the efficiency of recovery of expressed cDNAs, further screening and subfractionating the libraries based on this approach, and adaptation of the polymerase chain reaction to aid in recovery of the very small numbers of plasmids present. This approach is technically difficult, and its success depends on pushing forward the state of the art in several areas. Nevertheless, it continues to provide promise for allowing direct cloning of genes key to the action of several classes of abused substances.

PUBLICATIONS
Z01 DA00114-02-MNL

Receptor cDNA Expression Cloning Using Ligand Autoradiographic Screening

Rattray, M., Lautar S.L. and Uhl G.R. A new method of screening receptor cDNAs: Influence of plasmid competition on receptor expression. Biochem. Soc. Trans. 17: 1068-1069, 1989.

Uhl, G.R.: Bind and clone: Ligand-autoradiographic receptor expression screening. Society for Neuroscience Abstract 14(1): 318, 1988.

Rattray, M., Lautar, S.L., and Uhl, G.R. Ligand autoradiographic receptor screening: validation using beta-adrenergic receptor cDNA. Society for Neuroscience Abstracts 15(2): 1270, 1989.

Rattray, M., Lautar, S.L. and Uhl, G.R. A new method of screening receptor cDNAs. The Biochemist (Abstract Supplement) 11(2) #80, P.57, 1989.

Rattray, M., Lautar, S.L. and Uhl, G.R. Ligand autoradiographic receptor expression screening I: assessment of transiently-expressed receptors by radioligand binding to replicas of transfected COS cell colonies. Mol. Br. Res. (in press).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER
Z01 DA00115-02-MNL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Receptor cDNA Expression Cloning Using Xenopus Oocyte Expression

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator) (Name, title, laboratory, and institute affiliation)*

Uhl, G.R., Chief Laboratory of Molecular Neurobiology, ARC
Others: Shimada, S., Visiting Scientist, LMN, ARC , O'Hara, B., Staff Fellow, LMN, ARC and DiGiorgianni, J., Technician (guest worker), Johns Hopkins University, Dept. of Neuroscience, Johns Hopkins School of Medicine; Spivak C., Pharmacologist , NPL, ARC, Drs. Suzanne Zukin and Michael Bennett, Albert Einstein College of Medicine, Dr. Emmanuel Landau, Mt. Sinai School of Medicine, New York.

COOPERATING UNITS *(If any)*

Neuropharmacology Laboratory, ARC.

LAB/BRANCH
Laboratory of Molecular Neurobiology, Neuroscience Branch

SECTION
Gene Neuroscience Unit

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.5	1	0.5

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Expression cloning is an attractive approach to identifying the genes and cDNAs encoding receptors for abused substances and for the neurotransmitters implicated in drug abuse (see above). The ability of the Xenopus oocyte to appropriately translate, post-translationally modify, and appropriately insert several receptors into its membrane has led us to establish this system as a screening tool for these cDNAs and receptors.

Progress in this project has included: 1) Production of cDNA libraries in vectors containing active promoters for RNA polymerases whose transcription yields capped mRNAs of appropriate size, 2) Obtaining electrophysiologic signals for neurotensin, cholecystokinin, and kainic acid from oocytes injected with these synthetic mRNA transcripts, 3) Assessment of the cocaine receptor/dopamine transporter by injection of mRNA, and subsequently by injection of synthetic RNAs transcribed from a cDNA library. The electrically induced responses have appropriate temporal and electrical characteristics. The dopamine transport induced has the appropriate pharmacologic and physiologic characteristics to represent the physiologic dopamine transporter/cocaine receptor molecule. We can thus assess physiologic activities conferred to the oocyte by the translation products of specific cDNAs or mRNAs. These approaches should enhance our abilities to clone these DNAs through "sib selection" techniques.

PUBLICATIONS
Z01 DA00115-02-MNL

Receptor cDNA Expression Cloning Using Xenopus Oocyte Expression

Uhl, G.R., O'Hara, B., Shimada, S., Zacek, R., DiGiorgianni, J. and Nishimori, T. Dopamine Transporter: Expression in Xenopus oocytes. Mol. Br. Res. (In press).

O'Hara B.F., DiGiorgianni J.M., Shimada S., Kushner L., Spivak C.E., Lerma, J., Zukin R.S., Bennett M.V.L. and Uhl G.R. Expression of the neurotensin receptor in Xenopus oocytes with RNA transcribed from lambda and pCDM8 libraries. Society for Neuroscience Abstracts 15 (1) 672, 1989.

Shimada S., O'Hara B.F., Nishimori T., DiGiorgianni J.M. and Uhl G.R. Dopamine transporter mRNA and cDNA: Xenopus oocyte expression. Society for Neuroscience Abstracts 15 (1) 106, 1989.

Fan S.G., Kushner L., Lerma J., O'Hara B.F., Uhl G.R., Bennett M.V.L. and Zukin R.S. Identification of size classes of mRNA encoding kainate and neurotensin receptors. Society for Neuroscience Abstracts 15 (1) 672, 1989.

Uhl G.R. Parkinson's disease: Neurotransmitter and neurotoxin receptors and their genes. European Neurology (In press).

Uhl G.R., Shimada S., O'Hara B., DiGiorgianni J., and Nishimori T. Dopamine transporter mRNA and cDNA: Strategy for expression cloning a selective neurotoxin concentrator. Proceedings First International Congress of Movement Disorders Abstract 1990 (In press).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER
Z01-DA00116-02-MNL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Genes Related to Drug Abuse I: Regulation of Opioid Peptide Genes.

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*
Uhl, G.R. Others: Appleby, D., LMN, Research Technician, ARC/NIDA, DiGiorgianni, J., Technician (guest worker) ; Moskowitz, M., Associate Professor of Neurology & Neurosurgery*.

*Departments of Neurology, Neurosurgery, Massachusetts General Hospital, Harvard Medical School and Department ofNeuroscience, Johns Hopkins University School of Medicine .

COOPERATING UNITS *(If any)*

LAB/BRANCH
Molecular Neurobiology Laboratory, Neuroscience Branch

SECTION
Gene Neuroscience Unit

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.0	1	1

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Drugs alter the expression of many important genes in the brain. These changes in gene expression are likely to contribute to long-term drug effects, such as tolerance and dependence. Continuing studies of the genes encoding endogenous morphine - like peptides suggest that drug-induced gene regulation depends on the circuits connecting to the neuron and the duration of drug exposure.

During the past year, the laboratory has monitored cellular levels of neuropeptide mRNAs and of the transcription factor genes that may alter expression of these mRNAs to assess gene regulation related to functional activity in these neurons. Previously, reduced striatal proenkephalin mRNA following chronic systemic administration of morphine was found. In initial studies completed during this year, the same regimen has led to upregulation of preproenkephalin expression in pain-modulating neurons of the nucleus caudalis of the spinal tract of the trigeminal, while acute local administration of the drug through an intrathecal catheter down regulated the same gene. The transcription factors likely to be involved in this regulation are now being identified. Stimulation of primary afferents to the nucleus caudalis results in a remarkable, time dependent, biphasic enhancement of proenkephalin expression. Furthermore, a specific member of the Jun transcription factor family, Jun B, shows increased expression that correlates with, and could help to cause, the early up regulation of proenkephalin expression. These results provide one of the first examples, in the brain, of a correlation between transsynaptic regulation of a transcription factor and of a possible target gene. Understanding such mechanisms is increasingly important for attempts to therapeutically manipulate these drug influences on gene expression. Preliminary results from a human study using opiate antagonists to change the expression of these opioid peptide genes, for example, provide evidence for the efficacy of such targeted therapies.

PUBLICATIONS
Z01-DA00116-02-MNL

Genes Related to Drug Abuse I: Regulation of Opioid Peptide Genes

Uhl G. An approach to in situ hybridication using oligonucleotide cNDA probes. In: Van Leeuwen FW, Buijs RM, Pool CW and Pach O (eds.), Molecular Neuroanatomy. Elsevier Science Publishers B.V., Amsterdam, 1988: 25-41.

Reppert, S.M. and Uhl, G.R. The Vasopression Gene is Expressed Prior to regulation in the supraoptic nuclei of fetal rats. Brain Res. 456: 392-396, 1988.

Uhl G.R., Navia B., and Douglass J. Differential expression of preproenkephalin and preprodynorphin mRNAs striatal neurons: High levels of preproenkephalin expression depend on cerebral cortical afferents J. Neurosci. 12: 4755-4764, 1988.

Nishimori T., Moskowitz M.A., Uhl G.R. Opioid peptide gene expression in rat
trigeminal nucleus caudalis neurons: Normal distribution and effects of trigeminal deafferentation. J. Comp. Neurol. 274 (1): 142-150, 1988.

Uhl G.R., Ryan J., Schwartz J. Morphine alters preproenkephalin gene expression. Brain Res. 459: 391-397, 1988.

Voigt M.M., Uhl G.R. Preprocholecystokinin mRNA in rat brain: regional expression includes thalamus. Molecular Brain Research 4: 247-253, 1988.

Uhl G.R. Neuropeptide systems in Parkinson's disease and tardive dyskinesia. In: C. Nemeroff (ed.) Neuropeptides in Neuropsychiatric Disorders. Baltimore: Johns Hopkins University Press, 1988: 157-77.

Uhl, G.R. In Situ Hybridization: Quantitation Using Radiolabeled Hybridization Probes. In: P.M. Conn (ed.) Neuroendocrine Peptide Methodology New York, Academic Press, 1989 pp 135-146.

Linnik M.D., Sakas D.E., Uhl G.R., Moskowitz M.A. Subarachnoid Blood and
Headache: Altered Trigeminal Tachykinin Gene Expression. Ann. Neurol. 25: 179-184, 1989.

Rivkees S.A., Chaar M.R., Hanley D.F., Maxwell M., Reppert S.M., Uhl G.R.
Localization and regulation of vasopressin mRNA in human neurons. Synapse 3: 246-254,1989.

Uhl G. In situ Hybridization: Quantitation using radiolabeled hybridization probes. In: P.M. Conn (ed.) Methods in Enzymology, New York, Academic Press, 1989, pp. 741-752.

Genes Related to Drug Abuse I: Regulation of Opioid Peptide Genes

Nishimori, T., Buzzi, M.G., Moskowitz, M.A. and Uhl, G.R. Preproenkephalin mRNA Expression in Nucleus Caudalis Neurons is Enhanced By Trigeminal Stimulation. Mol. Br. Res. 6: 203-210, 1989.

Linnik, M.D., Sakas, D.E., Uhl, G.R. and Moskowitz, M.A.: Subarachnoid blood alters preprotachykinin gene expression in rabbit trigeminal ganglia. FASEB J 2 (5): A1382, 1988.

Nishimori T., Moskowitz M., Borsook D., Maciewicz R. and Uhl G. Trigeminal and PAC modulation of opioid peptide gene expression in nucleus caudalis. Society for Neuroscience Abstracts 14(2): 857, 1988.

Borsook D., Nishimori T., Maciewicz R. and Uhl, G. Analgesic PAG stimulation modulates neuronal opioid peptide gene expression in raphe magnus and trigeminal nucleus caudalis. Neurology 39 (Suppl 1): 359, 1989.

Nishimori, T., Buzzi, G., Moskowitz, M. and Uhl, G.R. N. caudalis prepro-ENK mRNA: biphasic enhancement by afferents. Society for Neuroscience Abstracts 15 (1) 343,1989.

Schwartz, J.P., Simantov, R., Ryan, J.P. and Uhl, G.R.: Control of Striatal
Enkephalin Gene Expression. Proceed. VII Int. Congress of Endocrinology, (In press).

Uhl G. Neurotensin receptors. In: A. Bjorklund, T. Hokfelt, M. Kuhar (eds.). Handbook of Chemical Neuroanatomy IV. New York, Elsevier, (In press).

Price D.L., Whitehouse P.J., Struble R.G., Hedreen J.C., Uhl G.R. Transmitter systems in selected types of dementia in N. Kopp and J. Pearson (eds). Human Neurotransmission in Health and Disease. New York: Oxford University Press, (In press).

Uhl G.R. Principles of assessing neurotransmitter receptors in disease. In: J. Frost and H. Wagner, eds. Quantitative Imaging of Neuroreceptors. New York: Raven Press (In press).

Uhl G.R. mRNA localization with the microscope: in situ hybridization using radiolabeled probes. In: Yamamura and Kuhar (eds.) Methods in Receptor Analysis, (In press).

Uhl G.R., Jayaraman A., Nishimori T., Shoichi S., Rattray M. and O'Hara B. Applications of neuropharmacologic techniques in the molecular neurobiology of schizophrenia. In: SC Schultz and CA Tamminga (eds.) Schizophrenia (In press).

Genes Related to Drug Abuse I: Regulation of Opioid Peptide Genes

Uhl, G.R. and Nishimori, T. Neuropeptide Gene Expression and Neural Actiivty: Assessing a working hypothesis in nucleus caudalis and dorsal horn neurons expressing preproenkephalin and preprodynorphin. Mol. Cell Neurobiol. (In press).

Nishimori, T., Buzzi, M.G., Moskowitz, M.A. and Uhl, G.R. Differential effects of small- and large-caliber inputs on nucleus caudalis preproenkephalin
expression. J. Neuroscience (In press).

Reppert, S.R. and Uhl, G.: Vasopressin mRNA in the supraoptic and suprachiasmatic nuclei: appearance and circadian regulation during development. The Society for Pediatric Research, (Submitted).

Rivkees, S.A., Chaar, M., Reppert, M. and Uhl, G.R: Regulated vasopressin gene expression in human hypothalamus. Society for Pediatric Research, (Submitted).

Linnik, M.D., Sakas, D.E., Uhl, G.R. and Moskowitz, M.A.: Preprotachykinin Gene Expression in Sensory Neurons: Regulatory Role of Blood Components. American Society for Pharmaceutical Experimental Therapeutics, (Submitted).

Jayaraman A. and Uhl G.R. Cholecystokinin (CCK) mRNA: Prominent expression in the rostral nigral neurons. Proceedings First International Congress of Movement Disorders Abstract 1990 (Submitted).

Uhl G.R., Shimada S., O'Hara B., DiGiorgianni J., and Nishimori T. Dopamine transporter mRNA and cDNA: Strategy for expression cloning a selective neurotoxin concentrator. Proceedings First International Congress of Movement Disorders Abstract 1990 (Submitted).

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER
Z01-DA00117-01-MNL

PERIOD COVERED
October 1, 1988 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Genes Related to Drug Abuse II: Central Brain Pathways of Reinforcement/Reward.

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

Uhl, G.R. Others: Appleby, D., Research Technician, LMN, ARC NIDA, Rao, J., Dept. of Neurology, Louisiana State University, New Orleans, LA.

COOPERATING UNITS *(If any)*

LAB/BRANCH
Molecular Neurobiology Laboratory

SECTION
Gene Neuroscience Unit

INSTITUTE AND LOCATION
ARC, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER:

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Neurons in the ventral tegmental area are major constituents of central brain pathways of drug reinforcement. Despite this potential central role in the actions of many abused drugs, however, the detailed expression of important neurotransmission genes in these neurons has not been elucidated. The expression of the genes encoding two of the dopamine-cotransmitter peptides, cholecystokinin and neurotensin, have thus been mapped to neurons in this area during this year. In these studies, a surprising region-to-region variation in the expression of neurotensin and CCK genes in these neuronal subdivisions has been found. Such detailed studies are necessary before defining activities of cocaine and other abused drugs on this expression. These approaches thus allow detailed examination of the ways in which drugs and other physiologic processes influence gene regulatory mechanisms that could serve as a store for some of the information about prior drug use that may accompany tolerance and dependence.

The potential function of these neurons in human drug-induced reinforcement/reward is being studied as well. Parkinson's disease patients, whose brains lose VTA neurons, are assessed after administration of methylphenidate. Blunting of drug effects on mood in these patients would support a central role for these neurons in reinforcement/reward in man.

PUBLICATIONS
Z01 DA00117-01-MNL

Genes Related to Drug Abuse II: Central Brain Pathways of Reinforcement/Reward

Voigt M.M., Uhl G.R. Preprocholecystokinin mRNA in rat brain: regional
expression includes thalamus. Molecular Brain Research 4: 247-253, 1988.

Uhl G.R. Neuropeptide systems in Parkinson's disease and tardive dyskinesia. In: C. Nemeroff (ed.) Neuropeptides in Neuropsychiatric Disorders. Baltimore: Johns Hopkins University Press, 1988: 157-177.

Uhl, G.R. In Situ Hybridization: Quantitation Using Radiolabeled Hybridization Probes. In: P. M. Conn (ed.) Neuroendocrine Peptide Methodology, New York, Academic Press, 1989 pp 135-146.

Rivkees S.A., Chaar M.R., Hanley D.F., Maxwell M., Reppert S.M., Uhl G.R.
Localization and regulation of vasopressin mRNA in human neurons. Synapse 3: 246-254,1989.

Uhl G. In situ Hybridization: Quantitation using radiolabeled hybridization probes. In: P. M. Conn (ed.) Methods in Enzymology, New York, Academic Press, 1989, pp. 741-752.

Jayaraman A., Nishimori T., Dobnar P. and Uhl G.R. Differential expression of CCK and neurotensin (NT) mRNAs in the subnuclei of rat VTA. Society for Neuroscience Abstracts 15 (1) 902, 1989.

Uhl G.R. mRNA localization with the microscope: in situ hybridization using radiolabeled probes. In: Yamamura and Kuhar (eds.) Methods in Receptor Analysis, (In press).

Uhl G.R. Parkinson's disease: Neurotransmitter and neurotoxin receptors and their genes. European Neurology (In press).

Uhl G.R., Jayaraman A., Nishimori T., Shoichi S., Rattray M. and O'Hara B. Applications of neuropharmacologic techniques in the molecular neurobiology of schizophrenia. In: SC Schultz and CA Tamminga (eds.) Schizophrenia (In press).

Jayaraman, A., Nishimori, T., Dobnar, P. and Uhl, G. Cholecystokinin and Neurotensin mRNAs are differentially expressed in subnuclei of the ventral tegmental area. J. Comp. Neurol. (In press).

Jayaraman A. and Uhl G.R. Cholecystokinin (CCK) mRNA: Prominent expression in the rostral nigral neurons. Proceedings First International Congress of Movement Disorders Abstract 1990 (In press).

Preclinical Pharmacology Branch

Steven R. Goldberg, Ph.D., Chief

Introduction

The Preclinical Pharmacology Branch conducts research in experimental animals on the behavioral modes of action of drugs of abuse both in producing reinforcing, punishing and discriminative stimuli and in altering established behavior controlled by non-drug events such as food or electric shock. It also studies the role of genetics in determining the effects of drugs of abuse. Studies cover a wide range of topics, including the pharmacology of opioid, psychomotor stimulant and benzodiazepine dependence, alterations in the acquisition and retention of classically conditioned behavioral and physiological responses by drugs of abuse, consequences of repeated drug administration, and environmental and genetic determinants of drug-seeking and drug-taking behavior. Drugs are also evaluated to characterize physiological and toxic actions which accompany acute and prolonged administration, delineate the mechanisms responsible for these effects and establish methods for preventing or reversing the effects. Research is carried out in both nonhuman primates (rhesus and squirrel monkeys) and non-primates. New drugs are evaluated for abuse potential by comparison of reinforcing, aversive and discriminative stimulus effects, and by comparison of effects of prototypic drugs of abuse on neurophysiologic systems. Emphasis is placed on the use of pharmacological, environmental and genetic interventions to alter the effects of drugs of abuse. These aims are intended to further the overall goals of the Addiction Research Center by providing a background of information to be used in developing rational clinical procedures for the prevention and treatment of drug abuse.

In 1989, the Preclinical Pharmacology Branch consisted of one laboratory, the Behavioral Pharmacology and Genetics Laboratory. At the end of 1989, a reorganization of the Branch into two laboratories was initiated with a division of personnel from the above laboratory allowing the formation of a Psychobiology Laboratory. The two laboratories will be subdivided into functional and collaborative units with emphasis on behavioral pharmacology, drug self-administration, physiological psychology, pharmacogenetics, behavioral and biochemical genetics, medications development, and neuropsychopharmacology and toxicity. Since this reorganization has only recently been initiated, the present report covers the activities of the Behavioral Pharmacology and Genetics Laboratory for 1989. In addition, in October, 1989, Dr. Larry Sharpe was reassigned to the Preclinical Pharmacology Branch from another branch and laboratory (the Psychology and Biology of Vulnerability Laboratory). Although the projects conducted by Dr. Sharpe were not a part of the Preclinical Pharmacology Branch program in 1989, the final project

reports are included here; these projects have now been terminated and promising areas of study are being incorporated into new or ongoing projects within the Branch.

Behavioral Pharmacology and Genetics Laboratory - Steven R. Goldberg, Ph.D., Chief

Overview

The Behavioral Pharmacology and Genetics Laboratory is responsible for research in experimental animals on the reinforcing effects of drugs of abuse, the influence of such drugs on learned operant behavior, and the discriminative stimulus effects of these drugs. The roles of drugs of abuse from different pharmacological classes, including psychomotor stimulants, opioids and benzodiazepines are investigated with respect to how the opportunity for occasional drug self-administration leads to long sequences of integrated behavior culminating in self-administration of the drug and how administration of these drugs alters ongoing behavior controlled by non-drug events. The positive reinforcing as well as the punishing properties of these drugs are studied to develop an understanding and technology of how drug-seeking becomes strong and persistent and how it might be weakened by pharmacologic and behavioral means. These objectives are pursued using a variety of experimental procedures, including (1) assessing the reinforcing effects of these drugs using intravenous self-administration procedures, (2) examining their effects as noxious stimuli using schedules of punishment of ongoing behavior by i.v. drug injections, (3) quantifying the behavioral effects using fixed-interval and fixed-ratio schedules of food presentation or electric shock delivery or postponement as baselines, and (4) determining their effects as discrminative stimuli using two-lever choice situations.

Collaborative studies are pursued with various laboratories. For example, collaborative studies of neurochemical correlates of the behavioral actions of psychomotor stimulants are pursued with the Neuroscience Branch utilizing studies of receptor binding. Comparative studies of repeated sequences of drug-seeking behavior controlled by administration of various doses of nicotine, cocaine, morphine or placebo under simple fixed-ratio or complex second-order schedules in humans and in non-human primates are pursued jointly with the Biology of Dependence Laboratory.

The long term goals of the Behavioral Pharmacology and Genetics Laboratory continue to focus on environmental conditions which determine whether drugs have positive reinforcing or aversive effects, on the use of complex second-order schedules of drug injection in humans and non-human primates to investigate the control of drug-seeking behavior by associated environmental stimuli, and on determination of the pharmacological mechanisms of behavioral effects of drugs of abuse.

The Behavioral Pharmacology and Genetics Laboratory also conducts behavioral, pharmacological and biochemical studies using animal models to investigate the contribution of genetic factors to drug abuse, the central mechanisms of drugs of abuse, and the commonality between various drug-related behaviors. The overall goals are: 1) to determine the extent to which genetic factors contribute to drug effects and substance abuse; 2) to identify gene products, such as receptors and synthesizing enzymes, which are involved in the reinforcing and toxic effects of drugs; and 3) to identify gene loci related to various drug effects.

These goals are being met through progress along several specific research pathways. A comprehensive genetically and operantly defined database of drug self-administration data is being produced, utilizing both mice and rat strains as experimental subjects. In future experiments, Preclinical Pharmacology investigators and others can incorporate this information into studies examining the biochemical and environmental mediators of drug self-administration in these same strains. Similarly, work is progressing to produce a similarly defined database of schedule controlled behavior and other acute drug effect data within the same genetically defined strains of rats and mice. These results will allow comparisons to be made between the effects of drugs on operant responding and other acute responses to drugs.

Through the use of genetic correlations and other genetic methods, such as Mendelian analysis, qualitative and quantitative estimates of genetic contributions to drug effects are determined. In this way, the genetic and biological relationships between drug self-administration and other drug-related phenotypes, or the degree of common genetic control of these various drug effects can be assessed. In some cases, the mode of transmission of drug related phenotypes can also be elucidated.

A recent expansion of neuropharmacological efforts within the laboratory allows systematic receptor binding and autoradiographic studies designed to explore the relationship between various neuronal systems with several drug effects, particularly drug-seeking behavior, but also including other effects such as stimulation, seizures and lethality.

Summary of Ongoing Research

A. **Control of Behavior by Drug Injections: Goldberg, S.R., Katz, J.L., Schindler, C.W., Spear, D. and Prada, J.A.**

Drugs of abuse can control large amounts of behavior by acting as either reinforcing stimuli to maintain behavior that leads to their administration, or by functioning as discriminative stimuli that are associated with conditions under which behavior is consistently reinforced by other relevant stimuli, such as

presentation of food or avoidance of electric shock. In many situations, drugs of abuse probably function through multiple mechanisms to persistently sustain long sequences of drug seeking behavior that may be very resistant to extinction. Schedule-controlled performances provide a meaningful way to analyze these long sequences of drug-seeking behavior in the same way as operant behavior maintained by other events such as food or electric shock.

Pharmacological modifications of the reinforcing effects of cocaine. One series of experiments involved attempts at pharmacological modification of cocaine self-administration. In cardiovascular studies in squirrel monkeys, (see project summary), we found that treatment with calcium channel blockers such as nimodipine will reverse or prevent cardiovascular changes produced by cocaine. Presession treatments with nimodipine that were sufficient to reverse cardiovascular effects of cocaine were without effect on cocaine-maintained behavior or on food-maintained behavior that was suppressed by cocaine administration.

In another series of experiments conducted in collaboration with Dr. L. Porrino of the Laboratory of Cerebral Metabolism, NIMH, Bethesda, MD, and Dr. M.J. Kuhar of the Neuroscience Branch, the effects of administration of serotonergic drugs on rates of responding by rats self-administering cocaine or amphetamine under a fixed-ratio schedule of drug injection in daily 4 hour sessions were compared. We compared the effects of fluoxetine, an inhibitor of serotonin reuptake, and cinanserin, a $5HT_2$ receptor antagonist, on cocaine and amphetamine self- administration in the same rats. Fluoxetine pretreatment (2.5, 5 and 10 mg/kg) significantly decreased rates of responding for amphetamine, but had no effect on responding maintained by cocaine at any of the doses tested. Cinanserin pretreatment (3, 10 and 17.5 mg/kg) decreased rates of amphetamine self-administration only at the highest dose tested, but had no effect on cocaine self-administration. These data suggest a differential sensitivity of cocaine and amphetamine self-administration to pharmacological manipulation of central serotonin systems. They are consistent with biochemical data which demonstrate a negative correlation between the reinforcing potency of amphetamine-like drugs, but not cocaine-like drugs, and their potency at serotonin binding sites. In future investigations we need to examine not only the effects of other serotonergic manipulations, but also of serotonergic drugs against a full range of psychomotor stimulant doses.

Structure-activity relationships for the reinforcing effects of cocaine and its analogs. In a series of experiments conducted in collaboration with Dr. M.J. Kuhar of the Neuroscience Branch, we examined the potency and the efficacy with which cocaine and its optical and geometric isomers as well as its n-desmethyl

metabolite, norcocaine, were able to act as reinforcers under a fixed-ratio schedule of i.v. drug injection in squirrel monkeys. Under this procedure both l-cocaine and norcocaine produced a dose-related increase in responding. Low levels of behavior, which were below those maintained by optimal doses of cocaine and above those maintained by drug vehicle, were generally maintained by d-pseudococaine. In contrast, neither l-pseudococaine nor d-cocaine maintained behavior leading to their injection at any dose tested. For comparative purposes the disruptive effects of these analogs on similar responding maintained by a fixed-ratio schedule of food presentation was also examined. The following order of potency was observed: l-cocaine > norcocaine > d-pseudococaine = l-pseudococaine > d-cocaine. This work provides further support for the concept that the reinforcing effects of cocaine are due to actions of cocaine at dopamine reuptake sites.

Modification of the reinforcing effects of nicotine by caffeine. Caffeine is widely used in conjunction with tobacco, and epidemiologic studies have shown that amount of coffee drinking is related to amount of tobacco consumption. The results of clinical studies on the effects of caffeine on smoking, however, have often been contradictory. We conducted a series of studies in squirrel monkeys to determine whether caffeine treatment could alter the effectiveness of nicotine as a reinforcer of drug-seeking behavior. A group of three squirrel monkeys was studied under a fixed-ratio schedule in which 30 lever-press responses were required to produce an i.v. injection of nicotine and each injection was followed by a 4-min timeout. To evaluate whether the effects of caffeine pretreatment were specific to behavior maintained by nicotine, a second group of three monkeys was studied under an identical schedule of food presentation. Comparable rates and patterns of responding were maintained by nicotine injections and by food presentations. Caffeine had little effect on rates of responding maintained by food until a high dose of 100 mg/kg was reached that decreased responding. In contrast, pretreatment with 3 to 10 mg/kg of caffeine markedly increased overall rates of responding maintained by nicotine injection. Thus, caffeine selectively facilitated nicotine self-administration. In other studies, the timeout value of the schedule and the dose of nicotine were reduced to values that resulted in only very low rates of nicotine-maintained responding, rates similar to those maintained by saline injections. When caffeine was given, there was over a 600% increase in rates of nicotine-maintained responding but no effect on rates of saline-maintained responding. Thus, caffeine appears to have a specific effect on nicotine-maintained responding rather than a general effect in increasing low rates of responding.

These studies indicate that concurrent use of caffeine can have significant effects on responding maintained by nicotine and, further, that these effects can be most prominent when the behavior controlled by nicotine is poorly maintained.

Additionally, caffeine tolerance studies (see below) indicate that a history of caffeine use may itself be sufficient to alter behavioral effects of nicotine. Further studies are needed to explore the behavioral mechanisms of caffeine's facilitating effects on nicotine self-administration and to determine whether these effects of caffeine are specific to nicotine or are a more general effect of caffeine on drug self-administration behavior. Since the use of caffeine is so ubiquitous, further studies of its interactions with drug self-administration behavior seem warranted in both the animal laboratory and in controlled clinical situations. In these studies one might expect to find the most prominent effects of caffeine under situations where behavior is poorly maintained by drugs.

B. Suppression of Behavior by Drug Injections: Katz, J.L., Goldberg, S.R. and Prada, J.A.

Many psychoactive drugs, including cocaine, nicotine and nalorphine, can function effectively as positive reinforcers or as punishers within the same dose range. Which effect becomes manifest depends on the context of environmental conditions and the history of the subject. Systematic evaluation of the environmental conditions which determine the type and direction of behavioral effects with a variety of drugs may have practical implications for the control of drug abuse by humans.

Recent studies within this project have concentrated on the punishing effects of benzodiazepine receptor ligands. Several benzodiazepine inverse agonists produce effects resembling anxiety and it has been suggested that anxiety production may be a primary mechanism by which these compounds exert their behavioral effects.

Since stress and anxiety may play a role in the initiation and maintenance of drug abuse, we have initiated extensive studies of the behavioral effects of ethyl-β-carboline-3-carboxylate (β-CCE), a prototype benzodiazepine inverse agonist. These studies have shown that response produced injections of β-CCE function as punishing stimuli indicating an aversive effect of the drug. Additionally, injections of β-CCE increase the aversiveness of otherwise ineffective stimuli (proconflict effect). Each of these effects is antagonized by the benzodiazepine receptor antagonist, flumazenil.

Analysis of the antagonism data is continuing in order to assess the affinity of the antagonist for the receptor through which β-CCE is exerting its effects. These results will yield information on whether these two actions of β-CCE are mediated by different receptor mechanisms. Importantly, comparisons of these effects with data on the antagonism of benzodiazepine agonist actions will give information on whether the inverse agonist and agonist actions are mediated by common receptor mechanisms.

Studies also have concentrated on the punishing effects of corticotropin releasing factor (CRF). CRF had dose-dependent proconflict effects after i.c.v. administration in rodents. Additionally, a rapid profound tolerance developed to these effects, which is currently being studied further.

This project is terminated with the continuing studies of benzodiazepine inverse agonists and CRF incorporated within the project on benzodiazepines (Z01 DA00007-05 BPL).

C. **Effects of Drugs on Schedule-Controlled Behavior of Experimental Animals: Goldberg, S.R., Prada, J.A., Katz, J.L. and Schindler, C.W.**

General information on the behavioral pharmacology of a drug in a pertinent species is necessary to evaluate quantitatively how the drug functions as a reinforcer or a punisher as well as to establish a profile of behavioral effects. Multiple schedules of food presentation with both fixed-interval and fixed-ratio components have been most frequently used in this type of study since they generate a wide range of rates and patterns of responding within a session and provide stable, long-term baselines for chronic studies in individual animals. Second-order schedules of food presentation are useful for comparative studies of behavior maintained by food presentation or i.v. drug injection and may provide a particularly sensitive baseline for analyzing the rate-increasing effects of psychomotor stimulants such as cocaine. The present project involves the assessment of both the acute and chronic effects of a variety of drugs, under multiple or second-order schedules of food presentation in squirrel monkeys and rats. The drugs studied include psychomotor stimulants, such as cocaine, various cocaine analogs, nicotine, various nicotine metabolites and analogs and caffeine. Since the behavioral effects of certain drugs depend on the type of event that maintains behavior, the effects of drugs are also studied on comparable performances maintained under fixed-interval schedules by either delivery of electric shock or by termination of a stimulus associated with electric shock. Finally, the discriminative stimulus effects of selected drugs, such as nicotine and cocaine, are explored with a focus on the actions of their metabolites and analogs. These procedures provide stable, long-term sensitive baselines for quantitative assessment of both stimulant and depressant drug effects. The long-term nature of these baselines also makes them ideal for studying tolerance and cross-tolerance development to chronic treatment with various drugs, including cocaine, caffeine, adenosine analogs, and benzodiazepines for studying the mechanims of action through pharmacological interaction with specific agonists and antagonists, and for studying the effects on behavior of various combinations of psychoactive drugs.

In one series of experiments, squirrel monkeys trained to discriminate i.v. injections of l-nicotine from saline were tested with different doses of l-nicotine, d-nicotine, l-nornicotine, and l-cotinine. The order of potency of the drugs was l-nicotine > l-nornicotine = d-nicotine >> l-cotinine. The potency of l-cotinine could be accounted for entirely by the l-nicotine impurity in this preparation. Previous studies of the psychomotor stimulant effects of these compounds in squirrel monkeys have found increases in response rates under fixed-interval schedules with l-nicotine and l-cotinine, which were approximately equipotent, but not with d-nicotine. The present results suggest that these effects of l-cotinine were not mediated by nicotinic mechanisms and that nicotinic agonist action alone (e.g., d-nicotine) is not sufficient activity for a psychomotor stimulant effect.

In future studies, we plan to train animals to discriminate injections of cotinine from saline and then either test for generalization to nicotine and other psychoactive compounds or attempt to block the discrimination with specific antagonists, such as mecamylamine. Additionally, further experiments are being conducted to replicate the findings of low dose stimulant effects of cotinine in squirrel monkeys and their possible reversal by nicotinic antagonists, such as mecamylamine, or by non-nicotinic antagonists.

In another series of experiments we determined the effects of caffeine pretreatment on the behavioral effects of nicotine in squirrel monkeys. In one study the effects of caffeine and nicotine on responding by squirrel monkeys were studied under a multiple schedule of food presentation. Given alone caffeine and nicotine showed qualitatively similar effects on behavior. At intermediate doses, each drug produced about a 50% increase in fixed-interval responding; higher doses decreased both fixed-interval and fixed-ratio responding. When nicotine was given in combination with caffeine, there were very large increases in fixed-interval responding, increases that were almost as large as those produced by maximally effective doses of amphetamine or cocaine. In contrast, the rate-decreasing effects on responding during fixed-ratio components produced by combinations of caffeine and nicotine were no different than those produced by nicotine alone.

In another study, the effects of nicotine-caffeine combinations were tested while responding under a fixed-interval schedule was controlled by delivery of electric shock. When nicotine was given in combination with caffeine, there were very large increases in fixed-interval responding. In fact, the combinations of nicotine and caffeine were as effective as d-amphetamine in increasing rates of responding. Thus, the pronounced effects of nicotine-caffeine combinations appear to be relatively independent of the event that maintains responding. Some squirrel monkeys were

placed on a chronic caffeine regimen for one month. Complete tolerance developed to caffeine's rate-increasing effects on behavior but maximal increases in responding produced by nicotine were as large as those produced by optimal dose combinations of nicotine and caffeine in nontolerant monkeys. These results suggest the potentiation of the psychomotor stimulant effects of nicotine by caffeine are not necessarily related to the psychomotor stimulant effects of caffeine and moreover that nicotine has psychomotor stimulant actions that are affected by secondary but not direct effects of caffeine.

D. **Comparative Studies of Drug Self-Administration in Squirrel Monkeys and Humans: Goldberg, S.R., Henningfield, J.E., Katz, J.L., Schindler, C.W. and Heishman, S.**

Self-administration studies permit an assessment of the relative contributions of environmental and pharmacologic factors to the self-administration of drugs, as well as to changes in response to drug due to tolerance and sensitization. Parallel comparative studies in monkeys and humans in which subjects are given the opportunity to self-administer comparable doses of drugs under similar behavioral schedules and experimental conditions provide a means to assess the generality of biological variables influencing drug self-administration. These studies allow an opportunity to directly compare the roles of environmental variables and conditioning in human drug-taking behavior and in animal models of drug taking. We have been conducting a series of experiments which have shown that drug self-administration behavior is maintained in human subjects in a manner similar to which it is maintained in non-human primates. Additionally, drug self-administration behavior in humans and monkeys appears to be a function of similar variables.

Second-order schedules with fixed-ratio components have been used to compare responding maintained by drug administration in humans to that maintained in monkeys. Under this complex schedule, responding by the subject under a fixed-ratio schedule (FR) results only in brief presentations of a stimulus (light/tone; S) which have been previously associated with injection of drug; according to a second schedule the first fixed ratio completed after a fixed interval of time has passed or a fixed number of FR components have been completed results in both the presentation of the brief stimulus and actual injection of drug. The use of second-order schedules permits repeated sequences of behavior controlled by drug-associated stimuli with relatively little disruption by the direct effects of drug administration. Since drugs can be injected only at the end of each session, it is also possible to avoid the use of catheters by utilizing intramuscular or other routes of administration. These studies provide an opportunity to evaluate the role of conditioned stimuli in the initiation, maintenance and resistance to extinction of drug-seeking behavior in human and non-human subjects.

Additionally, all human self-reported subjective effects are evaluated for dose-dependent drug effects.

We have recently completed a series of studies on morphine self-administration by squirrel monkeys and by human volunteers under second-order schedules of drug administration. The studies in monkeys demonstrated that environmental stimuli associated with either morphine injection or food presentation can prevent the extinction of drug-seeking behavior (see opioid project description for more details). We found that under second-order schedules with drug injected or food presented only at the end of each daily session, animals will continue to respond during extinction as they did when either morphine or food had been used to reinforce responding. However, responding does cease if the brief stimuli which had been paired with reinforcement are no longer presented during the early portions of the second-order schedule. After responding ceases, reinstating these brief-stimuli does not lead to an increase in responding and subsequent periods of extinction do lead to decreases in responding. Similar effects were found in previous studies of humans self-administering i.v. cocaine under similar second-order schedules. We are currently testing the effects of presession injections of the opioid antagonist naltrexone on morphine-maintained behavior under this schedule. Since all the morphine is given at the end of the session, any effect of naltrexone can be attributed to an effect on morphine-maintained responding rather than a direct action on the effects of morphine itself.

We have also completed a study in humans using second-order schedules of i.m. morphine injection. The studies were a collaboration between Drs. R.J. Lamb, K.L. Preston and J.E. Henningfield of the Clinical Pharmacology Branch and Drs. S.R. Goldberg, C.W. Schindler and J.L. Katz of the Preclinical Pharmacology Branch. The reinforcing and subjective effects of various doses of morphine were determined in human volunteers with a history of i.v. heroin abuse responding under a FR 30 (FR100:S) schedule of intramuscular injection. Under this schedule once each weekday subjects could work to obtain an intramuscular injection of morphine or placebo. Each drug dose was available for one week. Under these conditions placebo did not maintain responding across days. The lowest dose of morphine tested (3.75 mg) maintained responding in four of five subjects. The higher doses of morphine tested (7.5, 15 and 30 mg) maintained responding in all five subjects. Subjective effects measures, including measures of drug liking, euphoria (MBG scale of the ARCI), drug detection and identification, were obtained in these same subjects. Using these measures, subjects did not report subjective effects different from placebo for the lowest dose of morphine, while the highest dose of morphine produced clear drug related effects including reports of drug liking, increased MBG scores, and dope identifications. These results indicate that

there can be a substantial separation of the reinforcing effects of opioids and the subjective effects of opioids as these are traditionally measured. This separation of the reinforcing and subjective effects of morphine has profound implications for theories of opioid abuse, particularly those based on the notions that the reinforcing effects of opioids are causally related to the euphoric effects of opioids or the ability of opioids to relieve opioid withdrawal symptoms. This separation of the subjective and reinforcing effects of opioids, also, has implications for the laboratory assessment of the abuse liability of drugs from this class.

Much of the research on the subjective and behavioral effects of abused drugs in humans has focused on high doses that clearly produce feelings of euphoria and function as powerful reinforcers. However, the data from previous intravenous nicotine self-administration studies and intramuscular morphine self-administration studies described in this report, have shown that drug self-administration can persist at high rates, involving high levels of work, for doses that produce mild or even unreliable subjective effects.

E. Behavioral Effects of Opioid Agonists, Opioid Mixed Agonist-Antagonists and other...: Schindler, C.W., Goldberg, S.R. and Katz, J.L.

While it has been known for some time that chronic opioid antagonist treatment will produce behavioral supersensitivity in squirrel monkeys, the effect of opioid antagonist treatment in rats has been less clear. We have been investigating this phenomenon in rats using a cumulative dosing procedure where a single dose-effect function (1.0-100.0 mg/kg) can be determined in a single session. Initially, only a dose of 100.0 mg/kg naltrexone will affect fixed-ratio responding for food. However, even with naltrexone given only once per week during the determination of the dose-effect function, over a period of 8 weeks, a dose as low as 10 mg/kg will produce a complete cessation in responding. This effect is long lasting and appears to be the result of learning as substituting saline for the higher doses of naltrexone leads to recovery of responding to the lower naltrexone doses. It has been reported previously that chlordiazepoxide will partially antagonize the supersensitivity seen in squirrel monkeys, however this does not appear to be the case in rats. Morphine pretreatments do produce some antagonism of the sensitivity however. In collaboration with Dr. Su in the Neuroscience Branch we have shown that the naltexone dosing regimen can also affect kappa and delta opioid receptors, but not mu opioid receptors. Finally, we have recently demonstrated the pharmacological specificity of the supersensitivity in that only the opioid antagonist naltrexone shows cross-sensitivity to naltrexone.

F. **Abuse Liability and Behavioral Effects of Benzodiazepines: Katz, J.L., Witkin, J.M., Spear, D. and Prada, J.A.**

Benzodiazepines are among the most widely prescribed drugs. The widespread use of these compounds leads to concerns regarding their possible abuse. The present studies are designed to provide a characterization of the possible conditions that promote benzodiazepine abuse as well as information relevant to the mechanisms of benzodiazepine action and dependence. Specific areas that have been addressed in these studies are: 1) receptor specificity of various actions of benzodiazepines including ataxic, anxiolytic, anticonvulsant and dependence producing effects; and 2) anxiogenic actions of benzodiazepine-receptor antagonists and inverse agonists.

There are several characteristic behavioral effects of benzodiazepines, among them antianxiety, anticonvulsant, muscle relaxant and sedative effects. Recently, several potential therapeutic agents have been developed that have differing efficacies towards some of these actions, suggesting that the therapeutic actions of these drugs may be separable from unwanted side effects. Indeed, there have been suggestions that these different effects of benzodiazepines are due to actions at different receptor subtypes. The present series of studies is designed to assess the effects of different benzodiazepine agonists with regard to these actions, and the antagonism of these effects. A pA_2 analysis of the antagonism data will be conducted in order to assess if the actions of the various agonists are due to effects at different receptor subtypes. One side effect of the therapeutic actions of benzodiazepines is physiological dependence. The present studies are also directed at an assessment of whether the dependence produced by benzodiazepine agonists is related to actions at one of the putative benzodiazepine receptor subtypes.

The benzodiazepine-receptor inverse agonists have many physiological and biochemical effects that are opposite of those of benzodiazepine agonists. Further, the effects of these drugs are antagonized by flumazenil. One the basis of the anxiolytic effects of benzodiazepine agonists, several investigators have suggested that the benzodiazepine inverse agonists, when examined in vivo, would have "anxiogenic" effects. The results of studies designed to assess these anxiogenic effects have been mixed, and, with little justification, have been generally interpreted as consistent with that suggestion. The present studies are designed to functionally define some of the effects that have been interpreted as "anxiogenic" and to analyze those effects within that framework.

1. Receptor specificity of actions of benzodiazepines.

A series of studies has been initiated to characterize

benzodiazepine (Bz) receptor subtype-specific effects of Bz agonists. The anticonvulsant and ataxic behavioral actions of Bz agonists have been antagonized by the relatively pure Bz antagonist, flumazenil. Effects of chlordiazepoxide, zolpidem, CGS 9896, quazepam, and alprazolam have been studied in combination with several doses of flumazenil. For each of the effects described below, a pA_2 analysis of the effects of flumazenil was conducted in order to determine if the effects of the various agonists are mediated by different receptor mechanisms.

Ataxic effects. The apparatus for the inverted screen test for motor ataxia consists of four 15 x 15 cm pieces of wire mesh mounted 15 cm apart on a rod, 35 cm above the table top. For each trial, one mouse is placed on each screen and the rod is rotated 180° over 10 sec. Mice which fail to climb to the top of the screen within 60 sec are scored as ataxic. Each mouse is injected with a drug and twenty minutes later given a test trial. The mice are then injected with antagonists and retested 10 min later.

Anticonvulsant effects. Each mouse is injected with a dose of an agonist followed 20 min later by a dose of antagonist or vehicle. After an additional 10 min, the mice are injected with 80 mg/kg of pentylenetetrazole. Mice which do not develop clonic convulsions within 5 min of receiving pentylenetetrazole are scored as being protected by the agonist. Results of these studies are currently being analyzed in detail. Preliminary indications are that there are no appreciable differences in the pA_2 values of zolpidem and chlordiazepoxide for ataxia or anticonvulsant effects.

2. Anxiogenic actions of benzodiazepine inverse agonists and antagonists.

Studies have been initiated to investigate the discriminative effects of benzodiazepine receptor ligands. In these studies, rats or pigeons were trained to discriminate the effects of the inverse agonists β-CCE or the relatively pure antagonist flumazenil, respectively. A study using a conditioned taste-aversion procedure to develop drug discriminative control in rats was unsuccessful using β-CCE as the training drug.

Discriminative control with flumazenil has been developed. Discriminative control was established with a relatively low dose of the drug (100 mg/kg) using a more conventional procedure with pigeons. Several benzodiazepine agonists and antagonists agonists do not share discriminative effects with flumazenil. However, certain antagonists with structures closely related to those of flumazenil do substitute as

discriminative stimuli. Studies of the pharmacological specificity of the discriminative effect are continuing.

G. Cardiovascular Changes Induced by Cocaine: Schindler, C.W., Goldberg, S.R. and Tella, S.R.

Over the past few years, there has been an increase in the number of deaths due to cocaine abuse. Many of these deaths have been related to the effects of cocaine on cardiovascular function. The effects of cocaine on cardiovascular function are being investigated in squirrel monkeys and rats with chronic indwelling catheters in the iliac artery and vein. Heart rate and blood pressure can be measured from the arterial catheter and cocaine or other drugs can be delivered through the venous catheter. In addition, EKGs can be obtained simultaneously with these other measures.

The results for squirrel monkeys indicate that cocaine (0.3-3.0 mg/kg) produces an immediate increase in mean blood pressure of 10-30 mm and a delayed increase in heart rate of 30-80 bpm. At the higher doses of cocaine, the increase in heart rate is occasionally preceded by a decrease in heart rate. A slow infusion of cocaine as opposed to the bolus infusion eliminates that initial reduction in heart rate indicating it may be due to a direct effect of cocaine on the heart. Phentolamine will antagonize the blood pressure increasing effect of cocaine and propranolol will antagonize the heart rate increasing effects of cocaine indicating that these effects are mediated through adrenergic systems. While anesthesia will attenuate the effects of cocaine, hexamethonium pretreatment does not alter the cocaine response on either heart rate or blood pressure, indicating that these effects are mediated via peripheral adrenergic systems. Haloperidol does partially antagonize the effects of cocaine on heart rate, indicating that dopaminergic systems may also be involved in the cardiovascular response. A variety of calcium channel antagonists (nimodipine, verapamil and diltiazem) will also antagonize the cardiovascular response of cocaine in both squirrel monkeys and rats, however, these drugs do not alter the lethal response to cocaine in rats. Further, the calcium channel antagonists also do not alter the behavioral response to cocaine in squirrel monkeys, including cocaine self-administration behavior. As such, the utility of the calcium channel antagonists in treatment may be restricted to cocaine's cardiovascular effects. We have also recently shown that the squirrel monkey can be used as a model for the cardiovascular response to smoked free-base cocaine (i.e., crack). We have shown that smoked cocaine does lead to an increase in both heart rate and blood pressure.

H. **Behavioral Pharmacology and Toxicology of Psychomotor Stimulants: Witkin, J.M., Goldberg, S.R., Katz, J.L., Schindler, C.W. and Shores, E.I.**

Involvement of Dopamine in Cocaine Self-administration, Cocaine Discrimination, and Effects on Schedule-Controlled Behavior. Studies on the relevance of dopamine receptor subtypes in the pharmacology of cocaine have been coordinated along several dimensions including behavioral and cardiovascular pharmacology (see Cardiovascular project summary), studies of tolerance/cross-tolerance (see below), and toxicology (see below). Both D_1 and D_2 antagonists (haloperidol and SCH 23390) antagonized the rate-increasing effects of cocaine in squirrel monkeys. However, the antagonism was not always complete with either compound, even at doses that decreased responding when given alone. In addition, neither drug altered the disrupted temporal patterning of responding or the rate-suppressant effects of cocaine. While these results point to possible differences in the mechanisms whereby cocaine stimulates or depresses ongoing behavior, they are not consistent with an exclusive dopaminergic action of cocaine.

Correlational studies in collaboration with Dr. M.J. Kuhar of our Neurosciences Branch, have indicated a positive association between the affinity of cocaine, cocaine analogs, and related compounds for dopamine reuptake sites and their reinforcing potencies (See also Control of Behavior by Drug Injections project summary).

The role of dopamine receptor subtypes in the discriminative control of behavior by cocaine also appears uncertain. In rats trained to discriminate 10 mg/kg cocaine from saline, cocaine and amphetamines produce dose-dependent increases in cocaine-lever responses; however, neither mixed nor selective D_1 or D_2 agonists reliably produced cocaine-lever responses. However, SCH 23390 appears to function as a better antagonist than haloperidol of the discriminative effects of cocaine.

Involvement of Muscarinic Receptors in the Pharmacological Actions of Cocaine. Recent findings suggest that pharmacological actions of cocaine may be reciprocally regulated by tonic inhibitory influences of acetylcholine. The muscarinic antagonist, benactyzine, markedly potentiates the increases in fixed-interval responding of rats produced by cocaine. The rate-decreasing effects of cocaine are also attenuated by benactyzine. Studies with the muscarinic agonist, oxotremorine, and other muscarinic compounds are ongoing. Lethal effects of cocaine are also modified by muscarinic agents (see below).

Dopaminergic and Muscarinic Involvement in the Lethal and Neurotoxic Effects of Cocaine. Dopamine D_1 receptors were implicated in the lethal effects of cocaine in rats. SCH 23390

shifted the cocaine lethality function to the right, increasing the LD 50 in rats by 20 mg/kg. At 100 mg/kg, the lethality of cocaine was reduced from over 90% to approximately 50%. Although haloperidol was inactive against cocaine-induced lethality, haloperidol protected against lethal effects of amphetamines. The inactive enantiomer, SCH 23380, was also devoid of protective effects. SCH 23390 was not active against the lethal effects of lidocaine, indicating that non-specific actions of SCH 23390 against local anesthetic actions of cocaine were not involved in its protective effects.

Muscarinic compounds also modify the lethal effects of cocaine in rats possibly by modifying sympathetic/parasympathetic balances. Physostigmine (0.1 and 0.? mg/kg) reduces the lethal effects of cocaine. Oxotremorine does not produce protection. However, both compounds markedly potentiate cocaine-induced lethality at doses devoid of convulsant or lethal effects of their own. Additional muscarinic agonists and cholinesterase inhibitors will be investigated to clear up the discrepancy between the results with physostigmine and oxotremorine. These studies are in collaboration with Dr. M.J. Kuhar, Neuroscience Branch.

In contrast to recent reports, we have been unable to detect evidence of neurotoxicity as reflected in changes in dopamine levels in striatum or frontal cortex of rats after a 7 day regimen of cocaine (10 or 20 mg/kg, twice/day). These studies are in collaboration with Dr. G.A. Ricaurte, Johns Hopkins University School of Medicine.

Consequences of Repeated Administration. Tolerance and cross-tolerance studies in rats have indicated that tolerance to rate-decreasing effects of cocaine may involve metabolic factors and that cocaine tolerance may not involve general psychomotor stimulant actions of this compound. Whereas only a small shift in the cocaine dose-effect function was observed after repeated daily cocaine administration, there were large differences in the recovery from disruptive behavioral effects, with cocaine-treated subjects showing faster recovery. At the lower doses, there were also differences in the initial effects of cocaine in chronically-treated rats relative to controls. Tolerance to cocaine conferred cross-tolerance to apomorphine but not to d-amphetamine. There was also no evidence of exclusive involvement of either D_1 or D_2 receptors in the expression of cocaine tolerance. Rats tolerant to cocaine did not show cross tolerance to compounds with selectivity for D_1 or D_2 receptors. Additional work involves 1) assessment of cross tolerance to compounds that block dopamine reuptake, 2) evaluation of tolerance/ cross- tolerance relationships under conditions in which cocaine can increase behavioral output, and 3) pharmacokinetic alterations in chronically-treated subjects.

Route of Administration. We have begun investigations of the relation between route of cocaine administration and its discriminative stimulus effects. Squirrel monkeys trained to discriminate iv cocaine infusions from saline show dose-related increases in cocaine- appropriate responses when exposed to vaporized cocaine or intramuscular injections. Complete time- and dose-effect information is being collected for correlation with blood-level analysis. These experiments are part of a collaborative effort with Dr. L.G. Sharpe and Dr. J.H. Jaffe.

Medications Development: We have recently begun a focused research effort devoted to providing preclinical support to the NIDA Medication Development Program. Specifically, our research agenda involves the systematic evaluation of behavioral and toxic effects of the treatment compounds proposed by NIDA. The ability of these compounds to modify both behavioral and toxic effects of cocaine is the major focus of our research efforts. Other compounds will also be studied as dictated by our data and developments in the pharmaceutical industry and by changes in NIDA policy.

Several test systems are used to evaluate specific aspects of the behavioral or toxic effects of cocaine. Behavioral and toxicity models are employed to provide prediction of alterations in subjective effects (eg. euphoria, depressant actions), psychomotor stimulation, reinforcing properties (eg. drug taking or self-administration), dependence, convulsant actions, and lethality. Tests are generally administered in a sequential fashion so as to limit tests with minimal throughput to compounds that show promise in other drug screens.

Thus far we have conducted some tests with mazindol, carbamazepine, sulpiride, SCH 23390, and buprenorphine. Both SCH 23390 and buprenorphine were effective in blocking lethal effects of cocaine. Additional studies with these and other compounds are in progress.

Conclusions: These findings have led us to conclude that (1) The rate-enhancing effects of cocaine appear to be pharmacologically distinct from its behavioral depressant actions; (2) The involvement of dopamine or of dopamine receptor subtypes in the behavioral effects of cocaine are neither robust nor straightforward; the interaction of D_1 and D_2 sites may be relevant; (3) Muscarinic receptors may be involved in both behavioral and lethal effects of cocaine; investigation of receptor selective compounds may yield new insights into mechanisms of action; (4) D_1 receptors appear to be more relevant than D_2 receptors to the lethal effects of cocaine; (5) Unlike amphetamines, cocaine does not appear to have neurotoxic activity; (6) Studies with different routes of administration are feasible and may help elucidate pharmacokinetic determinants in the behavioral effects of cocaine that may be relevant to its abuse;

(7) Several compounds may be valuable in prohylactic treatment of behavioral and physiological toxicity of cocaine.

I. **Behavioral and Neurotoxic Effects of Substituted Amphetamines: Katz, J.L., Witkin, J.M., Shores, E.I., Ricaurte, G.A. and Castagnoli, Jr., N.**

The present studies are designed to examine behavioral effects of substituted amphetamines that may contribute to drug abuse and how those effects relate to neurotoxic actions of the drugs. The specific objectives of this project are to study: (1) Reinforcing effects of i.v. drug injections scheduled as consequences of behavior. Various substituted amphetamines, as well as other reference drugs of abuse, will be studied for possible reinforcing effects under fixed-ratio schedules of drug injection. (2) Effects on operant behavior will be assessed in standard procedures in which subjects are trained to respond under schedules of reinforcement during daily experimental sessions. Once performances are stable, the effects of the drugs will be assessed by routine presession injections. (3) Neurotoxic effects of these compounds will be assessed, through standard techniques. The long-term objectives of this research include the assessment of abuse liability of various substituted amphetamines, to better define the duration of the neurotoxic effects of (±)-3, 4-methylenedioxymethamphetamine (MDMA) in the primate, and to develop functional correlates of neurotoxicity by assessing the effects of drugs active on serotonergic systems before and after a regime of MDMA.

Reinforcing Effects of Drugs. Studies of the reinforcing effects of isomers of MDMA and MDA are in preliminary stages. Responding was well maintained by either isomer of MDMA, with maximal rates of responding at a dose of 0.3 ug/kg/inj. Results have also been obtained with (±)-N,N-dimethylamphetamine (NNDMA), a designer drug for which there have been recent incidents of abuse. This drug maintains maximal rates of behavior at doses of 3.0 to 10.0 µg/kg/inj indicating a relative potency of one tenth that of its close analog methamphetamine.

Effects on Operant Behavior. Recent studies have compared the effects of methamphetamine and NNDMA. Methamphetamine was ten times more potent than NNDMA in decreasing rates of schedule-controlled responding in rats. Similar potency differences between effects on operant behavior were obtained in squirrel monkeys. Methamphetamine displayed greater efficacy than NNDMA in producing psychomotor stimulant effects. The indications of relative potency were surprising since methamphetamine was approximately only three times more potent than NNDMA in producing lethality.

Effects of several drugs have been assessed in squirrel monkeys, before and during recovery from treatment with MDMA, which a dose

that has toxic effects on serotonergic neurons. A dose of 5.0 mg/kg (s.c.) of (±)-MDMA was administered twice daily to monkeys for 4 consecutive days. Effects of drugs active on serotonergic systems were assessed at several times points after MDMA administration. These studies have indicated no important changes in effects of drugs active on serotonergic systems following a regimen of MDMA that produces a significant serotonergic neurotoxicity.

Neurotoxicity. Both methamphetamine and N,N-dimethyl amphetamine produced degeneration of dopaminergic nerve terminals in mouse striatum without cell body loss in the substantia nigra. The N-methylated analog was approximately one-eighth as potent as the parent compound. In rats, 25 mg/kg of N,N-dimethylamphetamine twice daily for 4 days failed to produce a long-term depletion of serotonin in somatosensory cortex, in marked contrast to the effects of the same dose of methamphetamine. A higher dose of N,N-dimethylamphetamine was lethal.

J. **Genetic Factors in Response to Chronic Drug Treatment: Marley, R.J., Goldberg, S.R. and Goodman, N.L.**

In spite of the widespread recognition that there are strong individual differences in liability for drug abuse, relatively few studies designed to elucidate genetic factors associated with chronic drug use have been conducted. We have begun a series of studies designed to evaluate pharmacogenetic differences in response to the chronic administration of drugs of abuse among genetically defined strains of mice and rats. The use of a pharmacogenetic approach not only facilitates the understanding of individual differences in response to chronic drug administration, it also provides a tool for understanding the biochemical mechanisms underlying these responses. The drugs to be evaluated in these studies will include both CNS stimulants (cocaine and amphetamine) and CNS depressants (benzodiazepines and barbiturates). Chronic-stimulant treatment will be examined with special reference to understanding the mechanisms underlying genetic differences in the development of increased seizure susceptibility (pharmacological kindling) and behavioral sensitization. Chronic treatment with the benzodiazepines and barbiturates will similarly facilitate understanding the mechanisms underlying differences in the development of tolerance and physical dependence. Prevention or reversal of the effects of long-term drug treatment by agents suggested to have therapeutic value (e.g., carbamazepine, flumazenil) will also be evaluated in different genotypes subjected to chronic drug treatment. NMDA and GABA receptors and voltage-dependent sodium channels will be evaluated using ion flux and receptor binding techniques. These measures will be conducted on various genotypes and subjected to genetic correlational analyses. The use of genetic correlations has proven quite valuable for identifying possible mechanisms underlying particular behavioral or physiological responses.

These correlations also provide a means of ascertaining whether a common mechanism is associated with a particular response to different classes of drugs.

While these studies are still in the early stages, the following progress can be reported:

1. There are large genetic differences in susceptibility to seizures induced by acute cocaine administration. To allow a more accurate determination of the best dose of cocaine to use in our kindling paradigm, four inbred mouse strains were screened for acute sensitivity to cocaine's convulsive properties. These initial studies revealed that C57 mice were highly susceptible and SJL mice highly resistant to cocaine-induced seizures. BALB and DBA mice showed an intermediate degree of seizure susceptibility.

2. Genetic differences in response to chronic treatment of these same mouse strains with doses of cocaine below the seizure threshold have also been observed. The repeated administration of cocaine resulted in the rapid sensitization to cocaine- induced seizures (kindling) in two of the inbred strains (C57 and SJL). In contrast, kindling developed at a much slower rate in the DBA and BALB strains. Sensitivity to cocaine kindling does not appear to correlate directly with susceptibility to seizures induced by the acute administration of cocaine.

3. Repeated administration of cocaine to animals after they have kindled to cocaine revealed another, previously unreported, phenomenon. In the two strains in which kindling developed at a rapid rate, tolerance to cocaine-kindled seizures developed upon further exposure to the drug. In contrast, in the two strains in which kindling proceeded at a slower rate, no tolerance was observed following repeated administration of cocaine.

4. It has been suggested that the convulsant properties of cocaine can be attributed to its local anesthetic properties. To further evaluate this hypothesis, we have evaluated the seizure-inducing and epileptogenic properties of the pure local anesthetic, lidocaine, using the same protocols described above. The strain segregation patterns, both for acute seizure susceptibility and for the time course of the development of kindling, were not the same for the two drugs. This would suggest that the convulsant and epileptogenic properties of cocaine can not be explained entirely by its local anesthetic actions.

K. **Neural Substrates of Behavior Maintained by Intravenous Psychomotor Stimulants: Sharpe, L.G., Goodman, N.L., Jaffe, J.H., and Porrino, L.L.**

Intravenous self-administration of drugs is a paradigm that has been used frequently to assess the reinforcing properties of drugs in several animal species. The purpose is to determine the neurochemical basis of this paradigm to several psychomotor stimulants. Our main emphasis centers around a drug development program in which FDA-approved drugs will be used to investigate how they modify self-administration of cocaine and other abused drugs. Quite often, approved drugs that are targeted to treat one illness are efficacious in other illnesses. For example, the antiepileptic carbamazepine is also used to treat trigeminal neuralgia, mania and other neurological disorders. The antihypertensive clonidine has been used in the clinical management of opiate and nicotine withdrawal. Recent data from clinical trials suggest that carbamazepine may be effective in treating patients dependent on cocaine. Even though carbamazepine seems to influence the release and uptake of dopamine, the exact mechanisms remain unknown. However, by emphasizing the use of FDA-approved drugs we may accelerate the process by which drug intervention can be used to help treat people dependent on drugs. By using such drugs in our model, we can compare it with clinical data to better understand the drug's mode of action and to develop strategies for testing drugs that alter drug seeking behavior.

In our paradigm, rats are trained to lever press for cocaine (1 mg/kg/inf) via chronic indwelling catheters. Their behavior is shaped to an FR 10 schedule of reinforcement with a time-out period of 15 sec. Daily test sessions last 2 hr. Saline is substituted for cocaine periodically to evaluate the reinforcing properties of cocaine. This dose of cocaine (1 mg/kg) is used first because it occupies the center of the dose-response curve such that any pretreatment drug which may increase or decrease performance would be detected.

Clinical trials have been conducted by other investigators to test the antidepressant properties of amfonelic acid (AFA), a non-amphetamine class of psychomotor stimulant. There are no reports that it is abused by humans. However, there are reports that AFA stimulates locomotor behavior, generalizes to cocaine and amphetamine in a drug discrimination paradigm, and lowers the threshold for rewarding brain stimulation in rats. Together, these results suggest that AFA, like other psychostimulants, can have effects on central reinforcement systems. We found that AFA was self-administered at a dose 9.4 times lower than that of cocaine. The data indicate that amfonelic acid can act as a reinforcer in rats, and further suggest that it may have abuse potential in humans.

Calcium channel antagonists are another class of FDA-approved antihypertensive agents that have had several clinical uses. Based on several years of research, two classes of calcium channels have been identified in several invertebrate and vertebrate cells, type I and type II. One factor that distinguishes the two is that the dihydropyridines (nifedipine, nitredipine, nimodipine) interact with the type II but not the type I calcium channels. A few animal studies suggest that the dihydropyridine antagonists reduce dopaminergic transmission. Clinical evidence from ARC suggest that the "rush" produced by cocaine may be altered by nifedipine. Preliminary evidence from ARC's Preclinical Branch show that calcium channel blockers attenuate the cardiovascular effects of cocaine in monkeys. Preliminary evidence from our laboratory showed that if rats were pretreated with the dihydropyridine antagonists nifedipine and nimodipine, they increased the number of cocaine reinforcements during a 2-hr session. The effective doses ranged from 0.25 to 2 mg/kg, ip, but the apparent potentiation was not dose dependent. Otherwise, the effect resembles that of pretreatment with dopamine D_1 and D_2 antagonists at low doses. One hypothesis is that dopamine antagonists increase responding for cocaine and amphetamine, presumably because more cocaine is required to compensate for partial receptor blockade by the antagonist. Nifedipine and nimodipine may reduce calcium-dependent release of dopamine caused by cocaine. The two non-dihydropyridine antagonists, Verapamil and diltiazem (10 to 60 mg/kg), either depressed or had no effect on self-injections of cocaine, suggesting that type II calcium channels are primarily involved in the reinforcing effects of cocaine.

Our overall strategy is to compare the effects of drug pretreatment in human with that of animals self-injecting cocaine as an effort to strenghten the use of animal models for discovering drugs that have a strong potential in the treatment of drug dependence. For example, if a non-cocaine like drug is found to reduce craving for cocaine, then its effects in our paradigm may reveal a strategy for developing drugs that reduce craving, not only for cocaine, but for several classes of abused substances. Our plan is to use several classes of drugs that act not only at selective receptor types, but also those that influence primarily the second messengers (ion channels, cAMP, cGMP, G protein, Kinase C, etc.). Drugs that interact with the excitatory amino acid receptors (N-methyl-D-aspartic, kainic and quisqualic acids) will also be explored as potential agents for intervention with PCP abusers, who are also receiving treatment at ARC outpatient programs.

This project is terminated with extensions of significant findings to be incorporated within other projects within the branch.

L. **Neurochemical Mechanisms Controlling the Morphine Abstinence Syndrome: Sharpe, L.G. and Jaffe, J.H.**

The neurokinins (substance P, neurokinin A and B, Physalaemin, etc.) may play an important role in the opiate abstinence syndrome because morphine inhibits and naloxone increases their release in the morphine-dependent rat. The purpose of this study is to investigate this possibility by administering, to morphine-dependent rats (before naloxone), drugs that would be expected to either increase or decrease the efficacy of endogenous neurokinins. In a completed study, we found that captopril (0.3 mg/kg i.p.), a drug that increases peripheral levels of substance P, was found to enhance the secretory signs of abstinence in the morphine-dependent rat. Moreover, pretreatment with capsaicin (125 mg/kg) prevented these enhanced withdrawal signs caused by captopril. This animal model may contribute to the development of drugs that could aid in the clinical management of opiate detoxification.

A study has been completed in which we investigated the effects of ibogaine on the morphine abstinence syndrome. Ibogaine is an alkaloid of Tabernanthe iboga H. Bn. and reportedly interacts with several receptor systems. A patent application for the use of ibogaine to interrupt abuse syndromes in humans has been filed. We found that ibogaine, in non-tremorogenic doses (5 and 10 mg/kg) had no effect on naloxone-precipitated withdrawal in morphine-dependent rats.

This project is terminated with extensions of significant findings to be incorporated within other projects within the branch.

M. **Self-administration of Drugs in Aerosol Form in Rats: L.G. Sharpe, Weinhold, L.L., Jaffe, J.H. and Jaffe, A.B.**

Most drugs that are reinforcing to humans are administered by the pulmonary or intranasal routes (e.g., opiates, hallucinogens, cocaine, PCP, nicotine, cannabis). Indeed, it has become the route of choice for many because of its immediate reinforcing effects and because intravenous drug use is often associated with those at risk for AIDS. The use of smoked cocaine has increased about 4 fold between 1985 to 1988 in 21 major US cities (DAWN data). The purpose of this project is to develop an animal model for the self-administration of non-volatile inhaled drugs. Our goal is to understand the importance of this route and to use this model in ways that are advantageous over the more traditional paradigm of intravenous drug self-administration.

An ultrasonic nebulizer is used to create a drug vapor. Rats are trained to lever press on an FR 5 schedule of reinforcement for a drug vapor (5 sec duration) which remained in the chamber for 15 sec (mean particle sizes = 1.5 um). An exhaust fan was then activated for 60 sec to vent the vapor. Lever presses during this

80 sec time-out period were counted but did not produce more vapor episodes. Training sessions lasted 13 to 15 hr overnight with no shaping procedures used. Rats began on an FR 1 schedule which was gradually increased on subsequent sessions until an FR 5 schedule was obtained in which a minimum of 1 vapor episode per hr had to occur (at least 75 lever presses overnight). The rats were then changed to daily test sessions lasting 2 hr.

After several failed attempts using psychostimulant drugs, our first successful attempt at training rats to self-administer a drug vapor was with sufentanil, a highly potent and selective mu opioid agonist that is self-administered intravenously by monkeys. Surprisingly, rats learn rather quickly to lever-press for sufentanil vapor going from an FR 1 to an FR 5 in 3 to 9 overnight sessions. Rats given access to water vapor in overnight training sessions perform significantly more poorly. Responding maintained by sufentanil vapor during the daily test sessions was dose dependent in that a linear regression analysis revealed a significant inverse relation ($r = -0.65$) between the concentrations of sufentanil (25 to 75 ug/ml) and the number of sufentanil-vapor presentations (24 to 7 per 2-hr session). Naloxone (1mg/kg, ip) given immediately before the session reduced responding within 5 to 7 sessions to levels observed for water vapor. On the first day of naloxone pretreatment, 7 out of 8 animals showed no change in lever-pressing compared with sufentanil alone. Substituting water vapor for all concentrations of sufentanil significantly reduced responding within 5 to 20 2-hr sessions. The pattern of responding showed that the behavior was controlled by the reinforcing properties of sufentanil and not by nonconditioned or stimulative properties. For example, post-reinforcement pauses were prevalent and its length dose dependent. From 83 to 93 per cent of the lever presses occurred within a 30-sec period before sufentanil-vapor reinforcements.

One of the major reasons for developing this method is that self administration of drugs can be investigated with animals living continuously in larger spaces (habitat) either as individuals or in groups of two or more. Information from these studies would reveal if self administration of drugs is related to circadian rhythm, becomes self regulated or toxic over time, and is influenced by social interactions. In a completed study, we investigated whether rats caged as isolates since weaning will self-administer sufentanil vapor in a manner different from rats reared as social pairs. Previous reports have shown that rats reared in isolation self inject amphetamine more frequently than rats caged together. We also determined if patterns of drug taking behavior were related to dominance behavior as measured by video taping social pairings before and after the drug session. Social behavior was video taped for 10 min after paring animals that are familiar or unfamiliar with each other because most of the social interactions related to dominance or "play" aggression appears during this initial period. We scored the frequency and

duration of pinning behavior as an index of dominance behavior. The primary aim is to investigate if this "aggression profile" relates to individual differences in sufentanil vapor self administration. We found that individually caged rats acquired the self-administration of sufentanil vapor significantly faster than did rats that were housed as pairs.

Our plans are to use this method to test a variety of drugs that are abused by humans through inhalation (i.e., cocaine, PCP, nicotine, THC). Some methodological problems still exist and are being worked out. Techniques to obtain smaller particle sizes, to get drugs into solution, to make rats less sensitive to the higher concentrations of drug vapor, and to optimize the schedule contingencies are currently under investigation. With some modifications, we believe this method offers advantages over the more conventional intravenous route. Aside from the social experiments in progress, we will characterize the 24-hr response pattern for vaporized sufentanil (or other drugs) in rats living alone (or in pairs) for 2 to 4 weeks in a large habitat. Once a rat learns to respond in a stable manner for 2 vaporized drugs we also plan to introduce a choice paradigm to see if it prefers one drug over the other or if instead it becomes a multiple drug user.

This project is terminated with extensions of significant findings to be incorporated within other projects within the branch.

N. Pharmacogenetics: Acute Responses to Drug Administration: George, F.R., Goldberg, S.R., Ritz, M.C. and Elmer, G.I.

We have completed strain analyses of acute changes in locomotor activation by cocaine in five rat stocks and twelve mouse stocks. In addition to the expected differences in potency and efficacy there are several other novel and significant findings of this work. First, a rat strain (NBR) has been identified which shows an extreme activation response to cocaine, with scores approaching one order of magnitude greater than any other rats tested. Second, a mouse line, the LS/Ibg, has been found to show no cocaine-induced increases in activity, across a wide range of doses. Third, we have identified a mouse strain, the C57BL/6J, which shows a severe and unique seizure response to cocaine. Interestingly, a closely related strain, the C57BL/6ByJ, does not show this response. The strain rank order data, and data from F1, F2 and backcrosses suggests that this unique seizure response in the C57BL/6J mice is due to a single gene.

We examined genetic differences in response to cocaine by studying the effects of a range of cocaine doses, 1-75 mg/kg, on locomotor activity in LS/Ibg (LS) and SS/Ibg (SS) mice. At the lowest doses, activity was depressed in both lines, but to a greater extent in LS mice. As the dose of cocaine was increased, activity returned to baseline, and at the highest doses, increases in locomotor activity were found, but only in SS mice. In LS mice,

cocaine was ineffective in increasing locomotor activity at any of the doses tested. Thus, novel findings were obtained in that LS mice failed to show any locomotor stimulant effects of cocaine even across a broad range of doses. These mice did not show a stimulant response to cocaine at any of the time points tested. Thus, this lack of stimulant effect was not due to a brief but significant stimulant effect being masked by a prolonged testing period. Since LS mice show a low-dose depressant response to cocaine but not a stimulant response, it appears that cocaine-induced locomotor activation and depression are not dose-related aspects of a single behavioral and biological continuum, but are instead two distinct responses to cocaine mediated by different neuronal mechanisms or pathways. Whether this difference in response to cocaine is mediated by some of the genes responsible for the differences in sensitivity to alcohols between the LS and SS mice remains to be determined.

We examined the effects of relatively low doses of cocaine, in the range of 0.1-10 mg/kg, on locomotor activity in C57BL/6J and DBA/2J mice. A biphasic dose response curve was seen for both strains. At the lowest doses, activity was depressed. As the dose of cocaine increased, activity returned to baseline, and at the highest doses, increases in locomotor activity were found. DBA/2J mice were depressed at a lower dose of cocaine than were C57BL/6J mice, however, C57BL/6J mice showed locomotor depression over a more broad range of doses. Activity was maximally depressed at 0.1 mg/kg for DBA/2J mice, and maximally depressed at 0.3 mg/kg for C57BL/6J mice. Thus, low doses of cocaine are shown to produce significant decreases in locomotor activity in two strains of mice. It is postulated that these low doses of cocaine which depress locomotor activity do so via inhibition of serotonin uptake, resulting in potentiation of serotoninergic activity.

Cocaine and amphetamine produce several similar behavioral effects, most notably locomotor stimulation. Biochemically, evidence suggests specific involvement of dopaminergic systems, although not necessarily identical sites, in mediating cocaine and amphetamine induced locomotor stimulation. This study examined the effects of cocaine or amphetamine on locomotor activity in rats from the ACI, F344, LEW and NBR inbred strains. Dose-dependent increases in locomotor activity were found for both drugs in all strains. However, large and efficacy differences were found. Further, significant strain by drug interactions were found, in that the strain rank order for stimulant response to the two drugs was not identical. Since striatal dopaminergic neurons influence locomotor activity, we also assessed ligand affinity and receptor density of dopamine transporters and dopaminergic D_1 and D_2 receptors in striatal tissue from these same strains of rats. No differences in these receptor binding parameters were found. These findings support the conclusion that these two drugs produce their locomotor stimulant effects through different sites of action, and that genetic differences in response to these drugs at

the behavioral level do not appear to be mediated significantly by differences in structure or number of striatal dopaminergic sites. The further use of genetic methods, however, may aid in determining the specific sites of action of these widely used stimulant drugs.

At high doses, cocaine produces convulsions and death. While evidence suggests specific involvement of dopaminergic systems in mediating cocaine-induced locomotor stimulation, cocaine is known to bind, with varying degrees of affinity, with several sites, including muscarinic and sigma. Thus, cocaine may be producing its toxic effects through similar or distinct pathways from those which mediate the stimulant effects of this drug. One approach to answering this question is the use of pharmacogentic designs which compare relative responses to different drug effects across several defined genotypes. Genetic differences in the locomotor stimulant response to cocaine have previously been reported for four inbred strains of rats. This study examined the lethal effects of cocaine using the same four rat strains, namely the ACI, F344, LEW and NBR. Significant genetic differences were found in cocaine-induced lethality, with two-fold difference seen between the most sensitive (NBR) and least sensitive (LEW) strains. However, lethality was not correlated with activation. Since striatal dopaminergic neurons influence locomotor activity, we also assessed ligand affinity and receptor density of dopamine transporters and dopaminergic D_1 and D_2 receptors in striatal tissue obtained from these same strains of rats. No differences in these receptor binding parameters were found. These findings indicate that large genetic differences exist in rats in terms of their lethal response to cocaine, and support the conclusion that cocaine produces its locomotor stimulant and lethal effects through different sites of action.

We have studied the effects of morphine on locomotor activity in seven mouse genotypes, and have found large differences in the behavioral activating and behavioral depressant effects of morphine. In addition, we have identified genotypes which show no behavioral activating response to morphine, which should provide a valuable tool in elucidating the mechanisms of this behavior.

Cocaine administered i.v. or i.p. produces dose-related changes in spontaneous locomotor behavior. However, the extent to which p.o. administered cocaine affected locomotor activity and how the potency and efficacy of p.o. administration compared to i.p. effects was not known. We thus determined the effects of cocaine administered either i.p. or p.o. on ambulatory behavior. The i.p. injections of cocaine resulted in dose-related increases in locomotor activity with significant increases beginning at 10 mg/kg and activity peaking at 30 mg/kg. Some mice had seizures at 56 mg/kg. The p.o. injections resulted in a significant decrease in activity at 3 and 10 mg/kg, modest increases in activity at 10 and 30 mg/kg, and a significant increase at 75 mg/kg. There were

no seizures in the p.o. group. While both routes of administration produced increases in activity, there was a significant shift to the right in the dose response curve for p.o. cocaine vs i.p. cocaine.

Sensitization to the effects of cocaine on locomotor activity occurs with repeated dosing. Since self-administration studies involve repeated self-exposures to orally delivered cocaine, we determined the effects of chronic p.o. exposure to cocaine on locomotor activity. We also determined the direction of sensitization to repeated p.o. administration cocaine, since it had not been determined whether repeated administration of low doses of cocaine would enhance, reverse or not effect the initial decreases in locomotor activity observed in the above experiments. We found differences in both initial response and direction of sensitizing effect as a function of p.o. cocaine dose. At the lowest dose, 3 mg/kg, a significant decrease in locomotor activity was found on the first day of testing. However, across days there were no significant changes in activity, indicating that no sensitization had occurred. Six mg/kg cocaine p.o. produced an initial decrease in locomotor activity. A significant sensitization to this effect was seen with chronic dosing. This sensitization effect declined over time. At 10 mg/kg the locomotor activity scores on Day 1 were not different from control, but across days a significant decrease in activity was found, suggesting that a sensitizing effect had occurred. At 30 mg/kg an initial increase in activity was seen, and this effect was significantly enhanced across days.

Ethanol-induced increases in the production of cyclooxygenase (CO) metabolites are proposed to be important in mediating the effects of ethanol. The purpose of the following studies were to determine the potency correlation between <u>in vitro</u> anti-enzyme activity, <u>in vivo</u> therapeutic potency and the ability of prostaglandin synthetase inhibitors (PGSI) to antagonize the effect of ethanol on schedule- controlled behavior (SCB) and ethanol-induced loss of the righting reflex (LORR). Ethanol (1.5 g/kg) decreased fixed ratio 20 responding for water to 16% of their saline baseline levels. Eight PGSIs of diverse chemical class and structure antagonized the depressant effects of ethanol in a dose dependent manner. In addition, the same PGSIs decreased the duration of LORR in a dose dependent manner when given 15 min prior to 3.5 g/kg. Importantly, the potency of these PGSI's to antagonize the effects of ethanol were significantly correlated with their <u>in vitro</u> IC_{50} ability to inhibit CO activity ($r = 0.87$ (SCB) and $r = 0.74$ (LORR)) and <u>in vivo</u> ED_{50} anti-inflammatory values ($r = 0.96$ (SCB) and $r = 0.82$ (LORR)). Significant correlations with <u>in vitro</u> and <u>in vivo</u> measures of CO inhibition suggest a common underlying mechanism of the behavioral effects of ethanol and the ability of CO to form prostaglandins.

Inhibition of CO activity decreases the production of prostanoids and increases AA substrate conversion to lipoxygenase (LO) metabolites. The purpose of these experiments were to systematically explore the contribution of individual eicosanoid metabolites in mediating the effects of ethanol. The dual LO/CO inhibitors (BW755C, phenidone), specific LO inhibitors (NDGA, ETYA) and the leukotriene receptor antagonists (oxatomide, LY17883) did not consistently alter the duration of LORR when given 15 min prior to 3.5 g/kg ethanol. These data suggest that LO metabolites do not mediate the acute narcotic effects of ethanol and that the ability of PGSIs to antagonize the effects of ethanol is not related to the shunting of AA substrate into the LO cascade. The receptor antagonists L640-035 (TXA, PGE, PGF) and SQ29548 (TXA) as well as the agonists 16-,diMe PGE2 (PGE), U44069 (TXA), carbacylin (PGI) and BW245C (PGD) were investigated in order to characterize the specific role of CO metabolites. The TXA receptor agonists and antagonists as well as the PGI agonist were ineffective. Importantly, the PGE antagonists significantly decreased duration of LORR and the PGE and PGD receptor agonists increased LORR four and two-fold, respectively. These data support a strong role for the specific eicosanoids PGE and PGD in mediating the acute hypnotic effect of ethanol.

We have utilized biochemical genetic methods to determine whether dopaminergic receptors and transporters might influence observed genetic differences in locomotor response to cocaine. Since striatal dopaminergic neurons influence locomotor activity, we hypothesized that dopaminergic neuronal systems might mediate this genetic difference in cocaine-induced locomotion observed. Thus, we assessed ligand affinity and receptor density of dopamine transporters and dopaminergic D_1 and D_2 receptors in striatal tissue obtained from the LS and SS mice, two selected lines which we have shown differ greatly in stimulant response to cocaine. No differences in these receptor binding parameters were found.

In another study, we examined the effects of cocaine or amphetamine on locomotor activity in rats from the ACI, F344, LEW and NBR inbred strains. Dose-dependent increases in locomotor activity were found for both drugs in all strains. However, large potency and efficacy differences were found. Further, significant strain by drug interactions were found, in that the strain rank order for stimulant response to the two drugs was not identical. These findings support the conclusion that these two drugs produce their locomotor stimulant effects though different sites of action. Since striatal dopaminergic neurons influence locomotor activity, we also assessed ligand affinity and receptor density of dopamine transporters and dopaminergic D_1 and D_2 receptors in striatal tissue from these same strains of rats. No differences in these receptor binding parameters were found. Taken together, these studies suggest that genetic differences in response to these drugs at the behavioral level do not appear to be mediated significantly by differences in structure or number of striatal

dopaminergic sites. The further use of genetic methods, however, may aid in determining the specific sites of action of these widely used stimulant drugs.

Cocaine is a powerfully addicting drug of abuse. However, its use has increasingly been associated with serious toxic effects. In particular, the incidence and reporting of cocaine-induced seizures associated with ingestion of large doses of this drug has increased significantly over the past several years. The goal of this study was to identify which cocaine binding sites in the brain mediate the seizurogenic effects of this drug.

Cocaine is known to interact with multiple brain receptor sites, including sigma, cholinergic muscarinic M_1 and M_2 receptors, and dopamine, norepinephrine and serotonin transporters. Incorporating a multifaceted approach using pharmacological correlations followed by multiple regression analyses, we assessed the potency of cocaine and various chemically and pharmacologically related compounds in producing seizures. We then determined the potencies of these same compounds at the six receptor sites with which cocaine is known to directly interact, including dopamine, norepinephrine and serotonin transporter sites, and M_1 and M_2 cholinergic and sigma opiate receptor sites. Utilizing multiple regression methods, we were then able to determine the relative degree of influence of each of these sites on seizures produced by these compounds.

Our analyses of these data indicates that four binding sites are significantly influencing the potency of the test drugs to produce seizures. These sites are serotonin transporters, sigma opiate receptors, and muscarinic M_1 and M_2 cholinergic receptors. The serotonin transporter is the primary site significantly associated with the potency of cocaine related drugs to produce seizures, accounting for 80% of the variance in the potency of cocaine related drugs to produce seizures. The results also indicate that affinity at sigma opiate, muscarinic M_1 and M_2 cholinergic receptors also influences potency to produce seizures. Specifically, drug binding at any of these sites is significantly inversely related to seizurogenesis, accounting for an additional 12-15% of the variance in seizure potency. Taken together, the binding of cocaine and related rugs at serotonin transporters, sigma opiate receptors, and muscarinic M_1 and M_2 cholinergic receptors accounts for over 99% of the variance in potency to produce seizures.

Preliminary pharmacological studies have confirmed the results of the biochemical experiments and suggest that cocaine-induced seizures may be antagonized by manipulations of the predicted neuronal systems. The results of subsequent pharmacological studies indicate that pretreatments with cinanserin, SKF 10047, or pirenzapine significantly decrease the percentage of seizures induced by 100 mg/kg cocaine, relative to saline-treated controls.

In summary, these experiments indicate the neuronal sites which mediate cocaine-induced seizures. Drug binding to the serotonin transporter is the primary factor mediating the potency of cocaine and related drugs in seizuregenesis. However, drug binding at sigma opiate receptors, and muscarinic M_1 and M_2 cholinergic receptors can attenuate this effect. These biochemical findings are substantiated by pharmacological results indicating that blockade of serotonergic activity, as well as blockade of muscarinic or sigma sites, can effectively antagonize the seizurgenic properties of cocaine.

O. **Pharmacogenetic Factors in Drug Reinforcing Behavior: George, F.R., Goldberg, S.R. and Elmer, G.I.**

The present studies are designed to examine genetic factors in substance abuse using animal models of drug seeking behavior. Most self-administration studies with non-alcohol drugs have used the intravenous route for administration of the drug. However, because of the large number of animals required for genetic studies, it was important to develop other less invasive and longer lasting models of self-administration. Drawing upon experience with models of ethanol self-administration, we developed procedures for oral delivery of cocaine, etonitazine (ETZ, a potent, orally effective opiate agonist) and amphetamine.

These studies have produced the following important findings:

1. There are large genetic differences in the reinforcing efficacy of ETZ. In particular, LEWIS rats show strong levels of responding for ETZ under various conditions, while F344 rats have not shown any consistent patterns of responding for ETZ regardless of the condition or training procedure used. This report will be the first to demonstrate in a systematic, conclusive manner that reinforcement from non-alcohol drugs is mediated significantly by genetic factors.

2. Cocaine serves as a reinforcer across several routes of administration and species. However, whether orally delivered cocaine serves as a positive reinforcer has not been established, and the relationship between reinforcing doses of cocaine and the effects of these doses on other behaviors has not been systematically examined. We determined the extent to which contingent access to orally delivered cocaine would maintain lever pressing behavior in C57BL/6J mice. The findings demonstrate that orally delivered cocaine can serve as a reinforcer of operant behavior. Post-prandial training strategies used with other species and substances were successful in inducing pharmacologically significant intakes of cocaine under a fixed ratio (FR) schedule of drug access. When post-prandial induction of cocaine intake was removed, responding for cocaine continued under non-induced

conditions. As FR size was increased, proportionate increases in responding occurred except at the highest FR value. Responding maintained by cocaine significantly exceeded responding maintained by vehicle, with the mice typically consuming 6-10 mg/kg cocaine per 30 min session. Comparisons between drug intake under these conditions and changes in locomotor activity following acute or chronic i.p. or p.o. cocaine indicate that amounts of cocaine consumed in the self-administration studies had significant effects on locomotor activity. The utilization of inbred strains and the procedures followed in the present studies should prove useful in determining the extent of both genetic and environmental influences on various behavioral effects of cocaine and their mechanisms of action.

3. The procedures developed above in (2) are now being used to examine orally delivered cocaine as a reinforcer across several rat strains. Importantly, we are using the same strains used in the locomotor activity studies, which will aid in developing a comprehensive pharmacogenetic database of use to researchers interested in the substrates of substance abuse. We are also using these same strains in a study examining reinforcement from cocaine using the i.v. route. This work will integrate well with existing i.v. data on cocaine self-administration, and will also allow us to compare results obtained using the i.v. route with results using the oral route. The results obtained to date from this study indicates that substantial genetic differences exist with regards to cocaine-reinforced behavior. Some strains will lever press to obtain significant amounts of cocaine at Fixed Ratio sizes up to and including 64. Other strains will extinguish responding for cocaine when the Fixed Ratio is increased above 2. There are also drug concentration effects which correlate well with the genetic differences in sensitivity to cocaine seen in the activity studies.

   Opiate addiction and drug abuse in general has recently been shown to be influenced by genetic factors. The goal of our opiate drug self-administration projects is to utilize recombinant inbred strains to investigate the role of opiate receptor subtypes and cosegregating opiate-related phenotypes in opiate self-administration. Opiate related phenotypes such as disruption of schedule-controlled responding, analgesia, hypothermia, activation and opiate-reinforced behavior currently being investigated in the parental strains C57BL/6 and BALB/C mice along with the CxBk subline. The CxBk mice are known to be deficient in mu opioid binding sites and to be significantly less sensitive than the parental strains to the analgesic and locomotor stimulatory effects of opiates. These data will allow for the generation of phenotypic strain distribution patterns for <u>in vitro</u> and <u>in vivo</u> correlates of opiate reinforced behavior.

Publications

Elmer, G.I. and George, F.R.: Indomethacin posttreatment antagonizes ethanol-induced sleep time. Annals of the New York Academy of Sciences 559: 441-443, 1989.

Elmer, G.I., Meisch, R.A., Goldberg, S.R. and George, F.R.: A fixed ratio analysis of oral ethanol reinforced behavior in inbred mouse strains. Psychopharmacology 96: 431-436, 1988.

Elsmore, T.F., Parkinson, J.K., Leu, J.R. and Witkin, J.M.: Atropine effects on delayed discrimination performance of rats. Pharmacology, Biochemistry and Behavior 32: 971-975, 1989.

Fukagawa, Y., Katz, J.K. and Suzuki, T.: Effects of a selective κ-opioid agonist, U-50,488H on morphine dependence in rats. European Journal of Pharmacology 170: 47-51, 1989.

Galbicka, G., Elsmore, T.F. and Witkin, J.M.: Tolerance to behavioral effects of physostigmine under interval schedules of schedules of positive or negative reinforcement. Psychopharmacology, 97: 448-455, 1989.

Genovese, R.F., Elsmore, T.F. and Witkin, J.M. Environmental influences on the development of tolerance to the effects of physostigmine on schedule-controlled behavior. Psychopharmacology 96: 462-467, 1988.

George, F.R.: The role of arachidonic acid metabolites in mediating ethanol self-administration and intoxication. Annals of the New York Academy of Sciences 559: 382-391, 1989.

George, F.R.: Cocaine produces low-dose locomotor depressant effects in mice. Psychopharmacology 99: 147-150, 1989.

George, F.R.: Prostaglandin synthetase inhibitors specifically modulate ethanol self-administration. Annals of the New York Academy of Sciences 559: 449-450, 1989.

George, F.R. and Goldberg, S.R.: Genetic factors in response to cocaine. In Clouet, D., Asgher, K. and Brown, R. (Eds.): Mechanism of Cocaine Abuse and Toxicology. NIDA Research Monograph Series, No. 88, 1988, pp. 239-249.

George, F.R. and Goldberg, S.R.: Genetic approaches to the analysis of addiction processes. Trends in the Pharmacological Sciences, 10: 78-83, 1989.

Goldberg, S.R., Risner, M.E., Stolerman, I.P. and Reaville, C.: Nicotine and some related compounds: Effects on schedule-controlled behavior and discriminative properties of rats. Psychopharmacology 97: 295-302, 1989.

Jaffe, A.B., Sharpe, L.G. and Jaffe, J.H.: Rats self-administer sufentanil in aerosol form. Psychopharmacology 99: 289-293, 1989.

Katz, J.L.: Drugs as reinforcers: Pharmacological and behavioral factors. In Liebman, J.M. and Cooper, S.J. (Eds.): The Neuropharmacological Basis of Reward. Oxford, U.K., Oxford University Press, 1989, pp. 164-213.

Katz, J.L.: Effects of drugs on stimulus control of behavior under schedules of reinforcement. In: Thompson, T., Dews, P.B. and Barrett, J.E. (Eds.): Advances in Behavioral Pharmacology, Vol. 7. Hillsdale, NJ, Lawrence Earlbaum Associates, 1989, pp. 13-38.

Katz, J.L.: Two types of bias in psychophysical detection and recognition procedures: Nonparmetric indices and effects of drugs. Psychopharmacology 97: 202-205, 1989.

Katz, J.L.: Interactions of clonidine and naloxone on schedule-controlled behavior in opioid-naive mice. Psychopharmacology 98: 445-447, 1989.

Meisch, R.A. and George, F.R.: Influence of genetic factors on drug reinforced behavior in animals. In Pickens, R. and Svikis, D. (Eds.): Biological Vulnerability to Drug Abuse. National Institute on Drug Abuse Monograph 89: 9-24, 1989.

Porrino, L.J., Goodman, N.L. and Sharpe, L.G.: Intravenous self-administration of the indirect dopaminergic agonist amfonelic acid by rats. Pharmacology, Biochemistry and Behavior 31: 623-626, 1989.

Porrino, L.J., Ritz, M.C., Goodman, N.L., Sharpe, L.G., Kuhar, M.J. and Goldberg, S.R.: Differential effects of the pharmacological manipulation of serotonin systems on cocaine and amphetamine self-administration in rats. Life Sciences 45: 1529-1535, 1989.

Ricaurte, G.A., DeLanney, L.E., Irwin, I., Witkin, J.M., Katz, J.L. and Langston, J.W.: Evaluation of the neurotoxic potential of N,N-dimethylamphetamine: An illicit analog of methamphetamine. Brain Research 490: 301-306, 1989.

Ritz, M.C., Boje, J., George, F.R. and Kuhar, M.J.: Cocaine binding sites related to drug self-administration. NIDA Research Monograph, Committee on Problems of Drug Dependence, 1989, pp. 239-246.

Ritz, M.C., George, F.R. and Meisch, R.A.: Ethanol self-administration in ALKO rats: I Effects of selection and concentration. Alcohol 6: 227-233, 1989.

Ritz, M.C., George, F.R. and Meisch, R.A.: Ethanol self-administration in ALKO rats: II Effects of selection and fixed ratio size. Alcohol 6: 235-239, 1989.

Ritz, M.C. and Kuhar, M.J.: Monoamine uptake inhibition mediates amphetamine self-administration: Comparison with cocaine. Journal of Pharmacology and Experimental Therapeutics 248: 1010-1017, 1989.

Ritz, M.C., Lamb, R.J., Goldberg, S.R. and Kuhar, M.J.: Cocaine self-administration appears to be mediated by dopamine uptake inhibition. Progress in Neuropsychopharmacology and Biological Psychiatry 12: 233-239, 1988.

Sharpe, L.G. and Jaffe, J.H.: Captopril and capsaicin modify opioid withdrawal in the morphine-dependent rat. Pharmacology, Biochemistry and Behavior 33: 899-902, 1989.

Solomon, R.E., Goodrich, J.E. and Katz, J.L.: Opioid receptor subtype-specific cross tolerance to the effects of morphine on schedule-controlled behavior in mice. Psychopharmacology 96: 218-222, 1989.

Suzuki, T., Fukagawa, Y., Yoshii, T., Yanaura, S. and Katz, J.L.: Modification of the effects of naloxone in morphine-dependent mice. Life Sciences 45: 1237-1246, 1989.

Suzuki, T. George, F.R. and Meisch, R.A.: Differential establishment and maintenance of oral ethanol reinforced behavior in Lewis and Fischer 344 inbred rat strains. Journal of Pharmacology and Experimental Therapeutics 245: 164-170, 1989.

Swedberg, M.D.B., Shannon, H.E. Nickel, B. and Goldberg, S.R.: Pharmacological mechanisms of action of flupertine: A novel centrally acting non-opioid analgesic evaluation by its discriminative effects in the rat. Journal of Pharmacology and Experimental Therapeutics 246: 1067-1074, 1988.

Takada, K., Suzuki, T., Hagen, T., Cook, J.M. and Katz, J.L.: Behavioral effects of benzodiazepine antagonists in chlordiazepoxide tolerant and non-tolerant rats. Life Sciences 44: 289-299, 1989.

Takada, K., Swedberg, M.D.B., Goldberg, S.R. and Katz, J.L.: Discriminative stimulus effects of intravenous l-nicotine and nicotine analogs or metabolites in squirrel monkeys. Psychopharmacology 99: 208-212, 1989.

Tortella, F.C., Witkin, J.M. and Musacchio, J.M.: Caramiphen, a non-opioid antitussive with potent anticonvulsant properties in rats. European Journal of Pharmacology 155: 69-75, 1988.

Weiss, S.J. and Schindler, C.W.: Integrating control generated by positive and negative reinforcement: Appetitive-aversive interactions. Animal Learning and Behavior 17, 433-446, 1989.

Weiss, S.J., Schindler, C.W., and Eason, R.: The integration of habits maintained by food and water reinforcement through stimulus compounding. Journal of the Experimental Analysis of Behavior 50: 237-247, 1988.

Witkin, J.M.: Central and peripheral muscarinic actions of physostigmine and oxotremorine on avoidance behavior of squirrel monkeys. Psychopharmacology 97: 376-382, 1989.

Witkin, J.M., Alvarado-Garcia, R., Perez, L.A. and Witkin, K.M.: Central oxotremorine antagonist properties of pirenzepine. Life Sciences 42: 2467-2473, 1988.

Witkin, J.M., Goldberg, S.R. and Katz, J.L.: Lethal effects of cocaine are reduced by the dopamine-1 receptor antagonist SCH 23390 but not by haloperidol. Life Sciences 44: 1285-1291, 1989.

Witkin, J.M., Goldberg, S.R., Katz, J.L. and Kuhar, M.J.: Modulation of the lethal effects of cocaine by cholinomimetics. Life Sciences 45: 2295-2301, 1989.

Witkin, J.M., Lee, M.A. and Walczak, D.D.: Anxiolytic properties of amygdaloid kindling unrelated to benzodiazepine receptors. Psychopharmacology 96: 296-301, 1988.

Witkin, J.M., Markowitz, R.A. and Barrett, J.E. Physostigmine-insensitive behavioral excitatory effects of atropine in squirrel monkeys. Pharmacology, Biochemistry and Behavior 32: 309-315, 1989.

Witkin, J.M. and Tortella, F.C.: Behavioral effects of non-opioid antitussive anticonvulsants. Drug Development Research 18: 57-65, 1989.

Woods, J.H., Winger, G. and Katz, J.L.: Abuse of benzodiazepines. Journal of the American Medical Association 261: 2956-2957, 1989.

Articles in Press

Barrett, J.E. and Witkin, J.M.: Buspirone in animal models of anxiety. In Tunnicliff, G., Eison, A.S. and Taylor, D.P. (Eds.): Buspirone: Mechanisms and Clinical Aspects. New York, NY, Academic Press, 1989, In press.

Elmer, G.I., Meisch, R.A., Goldberg, S.R. and George, F.R.: Ethanol self-administration in long sleep and short sleep mice: Evidence for genetic independence of neurosensitivity and reinforcement. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

George, F.R.: The use of genetic tools in the study of substance abuse. Alcoholism: Clinical and Experimental Research 12, 1989, In press.

George, F.R.: Genetic approaches to studying drug abuse: Correlates of drug self-administration. Alcohol, 1989, In press.

George, F.R.: Is there a common genetic basis for reinforcement from alcohol and other drugs? Advances in Alcoholism and Substance Abuse, 1989, In press.

George, F.R., Elmer, G.I., Meisch, R.A. and Goldberg, S.R.: Oral self-administration of cocaine in C57BL/6J mice and the relationship between intake and behavioral effects. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

George, F.R. and Meisch, R.A.: The use of behavior genetic methods in the study of behavioral pharmacology. In Thompson, T., Dews, P.B. and Barrett, J.E. (Eds.): Advances in Behavioral Pharmacology, Vol. 7. New York, NY, Academic Press, 1989, In press.

George, F.R. and Ritz, M.C.: Cocaine produces locomotor stimulation in SS/Ibg but not LS/Ibg mice. Psychopharmacology, 1989, In press.

George, F.R., Ritz, M.C. and Elmer, G.I.: The pharmacogenetics of drug effects and addiction. In Pratt, J. (Ed.): The Biological Basis of Drug Tolerance and Development. New York, NY, Academic Press, 1989, In press.

George, F.R., Ritz, M.C. and Meisch, R.A. The effects of ethanol on schedule-controlled responding in AA and ANA rats. Psychopharmacology, 1989, In press.

Goldberg, S.R., Schindler, C.W. and Lamb, R.J.: Second-order schedules and the analysis of human drug-seeking behavior. Drug Development Research, 1989, In press.

Katz, J.L., Dworkin, S., Dykstra, L.A., Carter, R.B. and Witkin, J.M.: Some behavioral effects of repeated d-amphetamine administration. Drug Development Research, 1989, In press.

Katz, J.L. and Goldberg, S.R.: Second-order schedules of drug injection: Implications for understanding reinforcing effects of abuse. In Mello, N.K. (Ed.): Advances in Substance Abuse. Vol 3. Greenwich CT, JAI Press, Inc, 1989, In press.

Schindler, C.W., Witkin, J.M., Tella, S.R. and Goldberg, S.R.: Effects of cocaine alone and in combination with haloperidol and SCH 23390 on cardiovascular function and schedule-controlled behavior in squirrel monkeys. Psychopharmacology, 1989, In press.

Schindler, C.W., Wu, X.-Z, Su, T.S., Goldberg, S.R. and Katz, J.L.: Enhanced sensitivity to the behavioral effects of naltrexone in rats. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

Suzuki, T., George, F.R. and Meisch, R.A.: Inbred rat strains differ in the establishment of etonitazine as reinforcer. Psychopharmacology, 1989, In press.

Swedberg, M.D.B., Henningfield, J.E. and Goldberg, S.R.: Evidence of nicotine dependency from animal studies: Self-administration, tolerance and withdrawal. In Russell, M.A.H., Stolerman, I.P. and Wannacott, S. (Eds.): Nicotine: Actions and Medical Implication. Oxford, U.K., Oxford University Press, 1989, In press.

Tella, S.R., Schindler, C.W. and Goldberg, S.R.: The role of central and autonomic neural mechanisms in the cardiovascular effects of cocaine in conscious squirrel monkeys. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

Tella, S.R., Schindler, C.W. and Goldberg, S.R.: Neural mechanims in the cardiovascular effects of cocaine. In Thadani, P.V. (Ed.): Cardiovascular Toxicity of Cocaine: Underlying Mechanisms. National Institute on Drug Abuse Research Monograph, 1989, In press.

Tortella, F.C., Witkin, J.M. and Musacchio, J.M.: Caramiphen, a non-opioid antitussive with potent anticonvulsant properties in rats. European Journal of Pharmacology, 1989, In press.

Thomas, D.A., Weiss, S.J. and Schindler, C.W.: The effects of chlordiazepoxide and Ro 15-1788 on preference for punished and unpunished response alternatives in rats. Psychopharmacology, 1989, In press.

Trouve, R., Nahas, G.G., Vinyard, C. and Goldberg, S.R.: Cocaine and cardiovascular changes in the squirrel monkey. Proc. Soc. for Exp. Biol. Med., 1989, In press.

Witkin, J.M.: Central and peripheral muscarinic actions of physostigmine and oxotremorine on avoidance behavior of squirrel monkeys. Psychopharmacology, 1989, In press.

Witkin, J.M.: Behavioral pharmacology of compounds affecting muscarinic cholinergic receptors. In: Thompson, T., Dews, P.B. and Barrett, J.E. (Eds.): Advances in Behavioral Pharmacology, Vol. 7. Hillsdale, NJ, Lawrence Erlbaum, 1989, In press.

Witkin, J.M. and Katz, J.L.: Analysis of behavioral effects of drugs. Drug Development Research, 1989, In press.

Witkin, J.M. and Perez, L.A.: Comparison of effects of buspirone and gepirone with benzodiazepines and antagonists of dopamine and serotonin receptors on punished behavior of rats. Behavioural Pharmacology, 1989, In press.

Witkin, J.M., Ricaurte, G.A. and Katz, J.L.: Behavioral effects and lethality of N-methylamphetamine and N, N-dimethylamphetamine in rats and squirrel monkeys. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

Abstracts Published

Aulisi, E.F., Wehby, R.G., Katz, J.L. and Valentino, R.J.: Selective proconflict effect of corticotropin-releasing factor (CRF). Society for Neuroscience Abstracts, 1989.

Elmer, G.I. and George, F.R.: Ethanol-induced narcosis antagonized by posttreatment with indomethacin. Problems of Drug Dependence, 1988, Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph 90: 385, 1988.

Elmer, G.I. and George, F.R.: Antagonism of ethanol induced narcosis: The combined effects of indomethacin and Ro 15-4513. Alcoholism: Clinical and Experimental Research 13: 313, 1989.

Elmer, G.I. and George, F.R.: Cyclooxygenase inhibitors antagonize ethanol's depressant effects: Potency correlations with scheduled controlled behavior. Society for Neuroscience Abstracts 15(1): 36, 1989.

George, F.R.: Mediation of ethanol self-administration by prostaglandin synthetase inhibitors. Problems of Drug Dependence, 1988, Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph 90: 345, 1988.

Goldberg, S.R., Kuhar, M.J., Katz, J.L. and Witkin, J.M.: Modulation of the lethal effects of cocaine by cholinomimetics. Society for Neuroscience Abstracts 15: 252, 1989.

Griffiths, J., Witkin, J.M. and Katz, J.L.: Behavioral effects of repeated cocaine injections. FASEB Journal 3: A295, 1989.

Jaffe, J.H., Witkin, J.M., Goldberg, S.R. and Katz, J.L.: Potential toxic interactions of cocaine and mazindol. The Lancet II 8654: 111, 1989.

Katz, J.L., Ricaurte, G.A. and Witkin, J.M.: Behavioral pharmacology and toxicology of N,N-dimethylamphetamine (NNDMA). FASEB Journal 3: A1035, 1989.

Katz, J.L. Sharpe, L.G., Jaffe, J.H. and Witkin, J.M.: Discriminative stimulus effects of inhaled cocaine in squirrel monkeys. Society for Neuroscience Abstracts 15, 253, 1989.

Rao, T.S., Schindler, C.W. and Goldberg, S.R.: Autonomic neural mechanisms in the cardiovascular effects of cocaine in conscious squirrel monkeys. FASEB Journal 3, A297, 1989.

Ricaurte, G.A., Molliver, M.E., Witkin, J.M., Molliver, D.D., Wilson, M.A. and Katz, J.L.: d-Fenfluramine produces long-term effects on central serotonin neurons in nonhuman primates. Society for Neuroscience Abstracts 15: 419, 1989.

Ricaurte, G.A., Witkin, J.M. and Katz, J.L.: Assessment of neurotoxic effects of cocaine. FASEB Journal 3: A298, 1989.

Ritz, M.C., Boje, J.W., Carroll, F.I., Lewin, A.H. and Kuhar, M.J.: [^{3}H] WIN 35,065-2: A ligand for cocaine receptors in striatum. Society for Neuroscience Abstracts 15, 1989.

Ritz, M.C. and George, F.R.: Ethanol self-administration is not related to behavioral or biochemical sensitivity to ethanol. Problems of Drug Dependence, 1988, Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph 90: 361, 1988.

Schindler, C.W., Witkin, J.M., Rao, T.S. and Goldberg, S.R.: Effects of cocaine alone and in combination with haloperidol and SCH 23390 on cardiovascular function and schedule-controlled behavior in squirrel monkeys. FASEB Journal 3: A296, 1989.

Sharpe, L.G., Weinhold, L.L. and Jaffe, J.H.: Capsaicin desensitization facilitates self-administration of amphetamine vapor in rats. CPDD, 1989.

Sharpe, L.G., Goodman, N.L. and Jaffe, J.H.: Calcium channel antagonist alter self-administration of cocaine in rats. Society for Neuroscience Abstracts 104.20, 1989.

Witkin, J.M.: Use of aversive stimuli in animal experimentation. In Guttman, H.N., Mench, J.A. and Simmonds, R.C. (Eds.): Science and Animals: Addressing Contemporary Issues, Scientists Center for Animal Welfare, Bethesda, MD, pp. 115-116, 1989.

Witkin, J.M., Goldberg, S.R., Jaffe, J.H. and Katz, J.L.: Dopamine-1 receptor specific involvement in the lethal effects of cocaine. Society for Neuroscience Abstracts 15: 253, 1989.

Witkin, J.M., Goldberg, S.R. and Katz, J.L.: Lethal effects of cocaine are reduced by the dopamine-1 receptor antagonist SCH 23390 but not by haloperidol. FASEB Journal 3: A298, 1989.

Abstracts In Press

Elmer, G.I. and George, F.R.: Indomethacin antagonizes the effects of ethanol: Effect of route of administration. Problems of Drug Dependence, 1989, Proceedings of the 51st Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph, 1989, In press.

George, F.R. and Meisch, R.A.: Orally delivered cocaine as a reinforcer in mice, rats and rhesus monkeys. Neuropsychopharmacology, 1989, In press.

George, F.R. and Ritz, M.C.: Cocaine-induced lethality: Mediation by dopamine uptake inhibition and direct action at muscarinic receptors. Federation Proceedings, 1989, In press.

Pickworth, W.B., Klein, S.A., George, F.R. and Henningfield, J.E.: Acetaminophen and ethanol interactions in humans. Journal of Clinical Pharmacology and Therapeutics, 1989, In press.

Ritz, M.C. and George, F.R.: Cocaine-induced seizures: Mediation by specific CNS receptors. Neuropsychopharmacology, 1989, In press.

Ritz, M.C. and George, F.R.: Cocaine-induced seizures are initiated by serotonin uptake inhibition, but attenuated by direct action at sigma and muscarinic receptors. Federation Proceedings, 1989, In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00001-05 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Control of Behavior by Drug Injection

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
Others:	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
	C.W. Schindler	Staff Fellow	BPGL, NIDA, ARC
	J.A. Prada	Research Psychologist	BPGL, NIDA, ARC
	D. Spear	Staff Fellow	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*
None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.30	1.00	0.30

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Schedule-controlled performances provide a meaningful way to analyze drug-seeking behavior in the same way as operant behavior maintained by other events such as food or electric shock. In the present project with squirrel monkeys and rhesus monkeys, the rates and patterns of responding maintained by various drugs, including cocaine, nicotine, methohexital, morphine and chlordiazepoxide are being compared using simple fixed-ratio and fixed-interval schedules and complex second-order schedules with brief stimulus presentation in which the role of brief stimuli in maintaining extended sequences can be assessed. The effects of presession treatments with a range of doses of pharmacologic agonists and antagonists, such as caffeine, specific D-1 and D-2 dopamine antagonists, serotonergic reuptake inhibitors, alpha-adrenergic antagonists, and calcium channel blockers, on responding maintained by i.v. psychomotor stimulant injection or food presentation under fixed-interval, fixed-ratio and second-order schedules will be studied. The interactions of naloxone or naltrexone with behavior maintained under extended second-order schedules of morphine self-administration or food presentation will be explored. These experiments with long second-order schedules in which drug is injected only at the end of the session will be extended to study the reinforcing effects of other drugs, including benzodiazepines and barbiturates. Studies of pharmacological and environmental means of weakening established behavior maintained by different drugs will be continued.

GPO 914-919

Z01 DA00001-05 BPL

Control of Behavior by Drug Injection

Publications

Elmer, G.I., Meisch, R.A., Goldberg, S.R. and George, F.R.: A fixed ratio analysis of oral ethanol reinforced behavior in inbred mouse strains. Psychopharmacology 96: 431-436, 1988.

George, F.R. and Goldberg, S.R.: Genetic factors in response to cocaine. In Clouet, D., Asgher, K. and Brown, R. (Eds.): Mechanism of Cocaine Abuse and Toxicology. NIDA Research Monograph Series, No. 88, 1988, pp. 239-249.

Goldberg, S.R., Risner, M.E., Stolerman, I.P. and Reaville, C.: Nicotine and some related compounds: Effects on schedule-controlled behavior and discriminative properties of rats. Psychopharmacology 97: 295-302, 1989.

Goldberg, S.R., Schindler, C.W. and Lamb, R.J.: Second-order schedules and the analysis of human drug-seeking behavior. Drug Development Research, 1989, In press.

Katz, J.L.: Drugs as reinforcers: Pharmacological and behavioral factors. In Liebmand, J.M. and Cooper, S.J. (Eds.): The Neuropharmacological Basis of Reward. Oxford, U.K., Oxford University Press, 1989, pp. 164-213.

Katz, J.L. and Goldberg, S.R.: Second-order schedules of drug injection: Implications for understanding reinforcing schedules of abused drugs. In Mello, N.K. (Ed.): Advances in Substance Abuse, Vol. 3. Greenwich, CT, JAI Press, Inc., 1989, In press.

Porrino, L.J., Ritz, M.C., Goodman, N.L., Sharpe, L.G., Kuhar, M.J. and Goldberg, S.R.: Differential effects of the pharmacological manipulation of serotonin systems on cocaine and amphetamine self-administration in rats. Life Sciences 45: 1529-1535, 1989.

Swedberg, M.D.B., Henningfield, J.E. and Goldberg, S.R.: Evidence of nicotine dependency from animal studies: Self-administration, tolerance and withdrawal. In Russell, M.A.H., Stolerman, I.P. and Wannacott, S. (Eds.): Nicotine: Actions and Medical Implication. Oxford, U.K., Oxford University Press, 1989, In press.

Takada, K., Swedberg, M.D.B., Goldberg, S.R. and Katz, J.L.: Discriminative stimulus effects of intravenous l-nicotine and nicotine analogs or metabolites in squirrel monkeys. Psychopharmacology 99: 208-212, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00002-05 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Suppression of Behavior by Drug Injections

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	J.A. Prada	Research Psychologist	BPGL, NIDA, ARC

COOPERATING UNITS *(If any)*

Hahnemann University (R.J. Valentino)

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.30	0.20	0.10

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Many psychoactive drugs, including cocaine, nicotine and nalorphine, can function effectively as positive reinforcers or as punishers within the same dose range depending on the context of environmental conditions. Systematic evaluation of the environmental conditions which determine the type and direction of behavioral effects with a variety of drugs may have practical implications for the control of licit or illicit drug use by humans.

Recent studies have examined the punishing effects of ethyl-β-carboline-3-carboxylate (β-CCE), a drug that produces anxiety-like effects in animals. Since stress and anxiety may play a role in drug abuse, we have examined this drug in detail. These studies have shown that flumazenil antagonizes the punishing effects of β-CCE, and that the antagonism is dose-related. Response independent administration of β-CCE increases the aversive effects of ineffective stimuli. These effects depend on the intensity of the ineffective stimulus, and are also antagonized by flumazenil. These studies will provide information on the regulation of anxiety by the benzodiazepine system and may provide insights into the role of that system in the initiation and maintenance of drug abuse.

Z01 DA00002-05 BPL

Suppression of Behavior by Drug Injection

Publications

Aulisi, E.F., Wehby, R.G., Katz, J.L. and Valentino, R.J.: Selective proconflict effect of corticotropin-releasing factor (CRF). Society for Neuroscience Abstracts, 1989.

Takada, K., Swedberg, M.D.B., Goldberg, S.R. and Katz, J.L.: Discriminative stimulus effects of intravenous l-nicotine and nicotine analogs or metabolites in squirrel monkeys. Psychopharmacology 99: 208-212, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00003-05 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Effects of Drugs on Schedule-Controlled Behavior of Experimental Animals

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
Others:	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
	J.A. Prada	Research Psychologist	BPGL, NIDA, ARC
	C.W. Schindler	Staff Fellow	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*
None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.45	1.15	0.30

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

General information on the behavioral pharmacology of a drug in a pertinent species is necessary to evaluate quantitatively how the drug functions as a reinforcer or a punisher as well as to establish a profile of behavioral effects. Multiple schedules of food presentation with both fixed-interval and fixed-ratio components have been most frequently used in this type of study since they generate a wide range of rates and patterns of responding within a session and provide stable, long-term baselines for chronic studies in individual animals. Second-order schedules of food presentation are useful for comparative studies of behavior maintained by food presentation or i.v. drug injection and may provide a particularly sensitive baseline for analyzing the rate-increasing effects of psychomotor stimulants such as cocaine. The present project involves the assessment of both the acute and chronic effects of a variety of drugs, under multiple or second-order schedules of food presentation in squirrel monkeys and rats. The drugs studied include psychomotor stimulants, such as cocaine, various cocaine analogs, nicotine, various nicotine metabolites and analogs and caffeine. Since the behavioral effects of certain drugs depend on the type of event that maintains behavior, the effects of drugs are also studied on comparable performances maintained under fixed-interval schedules by either delivery of electric shock or by termination of a stimulus associated with electric shock. Finally, the discriminative stimulus effects of selected drugs, such as nicotine and cocaine, are explored with a focus on the actions of their metabolites and analogs. These procedures provide stable, long-term sensitive baselines for quantitative assessment of both stimulant and depressant drug effects. The long-term nature of these baselines also makes them ideal for studying tolerance and cross-tolerance development to chronic treatment with various drugs, including cocaine, caffeine, adenosine analogs, and

GPO 814-818

benzodiazepines for studying the mechanims of action through pharmacological interaction with specific agonists and antagonists, and for studying the effects on behavior of various combinations of psychoactive drugs.

Publications

Goldberg, S.R., Risner, M.E., Stolerman, I.P. and Reaville, C.: Nicotine and some related compounds: Effects on schedule-controlled behavior and discriminative properties in rats. Psychopharmacology 97: 295-302, 1989.

Katz, J.L. and Goldberg, S.R.: Second-order schedules of drug injection: Implications for understanding reinforcing effects of abused drugs. In Mello, N.K. (Ed.): Advances in Substance Abuse, Vol. 3. Greenwich, CT, JAI Press, Inc., 1989, In press.

Swedberg, M.D.B., Henningfield, J.E. and Goldberg, S.R.: Evidence of nicotine dependency from animal studies: Self-administration, tolerance and withdrawal. In Russell, M.A.H., Stolerman, I.P. and Wannacott, S. (Eds.): Nicotine: Actions and Medical Implication. Oxford, U.K., Oxford University Press, 1989, In press.

Swedberg, M.D.B., Shannon, H.E., Nickel, B. and Goldberg, S.R.: Pharmacological mechanisms of action of flupirtine: A novel, centrally acting non-opioid analgesic evaluated by its discriminative effects in rats. Journal of Pharmacology and Experimental Therapeutics 246: 1067-1074, 1988.

Takada, K., Swedberg, M.D.B., Goldberg, S.R. and Katz, J.L.: Discriminative stimulus effects of intravenous l-nicotine and nicotine analogs or metabolites in squirrel monkeys. Psychopharmacology 99: 208-212, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00004-05 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Comparative Studies of Drug Self-Administration in Squirrel Monkeys and Humans

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
Others:	J.E. Henningfield	Chief	BDL, NIDA, ARC
	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
	C.W. Schindler	Staff Fellow	BPGL, NIDA, ARC
	S. Heishman	Staff Fellow	BDL, NIDA, ARC

COOPERATING UNITS *(if any)*
Biology of Dependence Laboratory, Clinical Pharmacology Branch

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.50	0.50	0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Self-administration studies permit an assessment of the relative contribution of environmental and pharmacologic factors to the self-administration of drugs, and to changes in response to drug due to tolerance and sensitization. Parallel comparative studies in squirrel monkeys and humans in which subjects are given the opportunity to self-administer comparable doses of cocaine, morphine and nicotine and other drugs under similar behavioral schedules and experimental conditions provide a means to assess the generality of biological variables influencing drug self-administration. These studies allow an opportunity to evaluate the role of environmental variables and the role of conditioning in human drug taking behavior and whether those roles differ from the roles of those variables in animal models of drug taking. We found that under second-order schedules with drug injected or food presented only at the end of each daily session, animals will continue to respond during extinction as they did when either morphine or food had been used to reinforce responding. However, responding does cease if the brief stimuli which had been paired with reinforcement are no longer presented during the early portions of the second-order schedule. We also completed a study in humans using second-order schedules of i.m. morphine injection. The reinforcing and subjective effects of various doses of morphine were determined in human volunteers with a history of i.v. heroin abuse. Results indicate that there can be a substantial separation of the reinforcing effects of opioids and the subjective effects of opioids as these are traditionally measured. This separation of the reinforcing and subjective effects of morphine has profound implications for theories of opioid abuse, particularly those based on the notions that the reinforcing effects of opiods are causally related to the euphoric effects of opioids or the ability of opioids to relieve opioid withdrawal symptoms. This project has been terminated with continuing comparative studies conducted as part of project Z01 DA00001-05 BPL.

Z01 DA00004-05 BPL

Comparative Studies of Drug Self-Administration in Squirrel Monkeys and Humans

Publications

Goldberg, S.R., Schindler, C.W. and Lamb, R.J.: Second-order schedules and the analysis of human drug-seeking behavior. Drug Development Research, 1989, In press.

Katz, J.L.: Drugs as reinforcers: Pharmacological and behavioral factors. In: J.M Liebman and S.J. Cooper (Eds.). The Neuropharmacological Basis of Reward. Oxford, U.K., Oxford University Press, 1989, pp. 164-213.

Katz, J.L. and Goldberg, S.R.: Second-order schedules of drug injection: Implications for understanding reinforcing effects of abuse. In Mello, N.K. (Ed.): Advances in Substance Abuse. Vol 3. Greenwich, CT, JAI Press, Inc., 1989, In press.

Swedberg, M.D.B., Henningfield, J.E. and Goldberg , S.R.: Evidence of nicotine dependency from animal studies: Self-administration, tolerance and withdrawal. In Russell, M.A.H., Stolerman, I.P. and Wannacott, S. (Eds.): Nicotine: Actions and Medical Implication. Oxford, U.K., Oxford University Press, 1989, In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00006-05 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Behavioral Effects of Opioid Agonists, Opioid Mixed Agonist-Antagonists and other...

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	C.W. Schindler	Staff Fellow	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC

COOPERATING UNITS *(If any)*

None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.10	0.80	0.30

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This project is directed at determining the various effects of opioid agonists, antagonists, and mixed agonists-antagonists on behavior, including their ability to modify opioid self-administration behavior. A primary finding over the past year has been that naltrexone, an opioid antagonist, produces clear behavioral supersensitivity when its effects are determined on schedule-controlled beahvior in rats. Initially, only a dose of 100 mg/kg naltrexone suppressed food-maintained behavior in rats. After repeated treatment, however, a dose of 10 mg/kg completely suppressed behavior. Unlike previous demonstrations of naltrexone supersensitivity in rats, the supersensitivity was long-lasting, persisting for at least 10 weeks following the last naltrexone injection. Two opioid agonists, morphine and ethylketocyclazocine, were able to partially antagonize the enhanced sensitivity to naltrexone. However, the enhanced snesitivity appears to be related primarily to conditioning processes, as an extinction procedure reversed the sensitivity which developed to naltrexone. In collaboration with Dr. Su in the Neuroscience Branch, we have also observed changes in kappa and delta receptor populations in the brain following naltrexone treatment. Finally, we have recently shown that this sensitivity in pharmacologically specific. Complete generalization to naltrexone sensitivity occurs with naltrexone, but not with a wide range of other compounds. These results indicate that sensitivity to naltrexone can result from conditioning processes and may be an important factor in opioid abuse treatment with naltrexone.

GPO 914-918

Behavioral Effects of Opioid Agonists, Opioid Mixed Agonist-Antagonists and other...

Publications

Fukagawa, Y., Katz, J.K. and Suzuki, T.: Effects of a selective κ-opioid agonist, U-50,488H on morphine dependence in rats. European Journal of Pharmacology 170: 47-51, 1989.

Goldberg, S.R., Schindler, C.W. and Lamb, R.J.: Second-order schedules and the analysis of human drug-seeking behavior. Drug Development Research, 1989, In press.

Katz, J.L.: Drugs as reinforcers: Pharmacological and behavioral factors. In Liebman, J.M. and Cooper, S.J. (Eds.): The Neuropharmacological Basis of Reward. Oxford, U.K., Oxford University Press, 1989, pp. 164-213.

Katz, J.L.: Interactions of clonidine and naloxone on schedule-controlled behavior in opioid-naive mice. Psychopharmacology 98: 445-447, 1989.

Schindler, C.W. and Harvey, J.A.: The use of classical conditioning procedures in behavioral pharmacology. Drug Development Research, 1989, In press.

Schindler, C.W., White, M.F. and Goldberg, S.R.: Effects of morphine, ethylketocyclazocine, N-allylnormetazocine and naloxone on locomotor activity in the rabbit. Psychopharmacology, 1989, In press.

Schindler, C.W., Wu, X.-Z, Su, T.S., Goldberg, S.R. and Katz, J.L.: Enhanced sensitivity to the behavioral effects of naltrexone in rats. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

Solomon, R.E., Goodrich, J.E., and Katz, J.L.: Opioid receptor subtype-specific cross tolerance to the effects of morphine on schedule-controlled behavior in mice. Psychopharmacology 96: 218-222, 1989.

Suzuki, T., Fukagawa, Y., Yoshii, T., Yanaura, S. and Katz, J.L.: Modification of the effects of naloxone in morphine-dependent mice. Life Sciences 45: 1237-1246, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00007-05 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Abuse Liability and Behavioral Effects of Benzodiazepines

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
Others:	J.M. Witkin	Senior Staff Fellow	BPGL, NIDA, ARC
	D. Spear	Staff Fellow	BPGL, NIDA, ARC
	J.A. Prada	Research Psychologist	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*

Department of Chemistry, University of Milwaukee at Madison (J.M. Cook)

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.50	1.40	0.10

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Benzodiazepines are among the most widely prescribed drugs. The widespread use of these compounds leads to concerns regarding their possible abuse. The present studies are designed to provide a characterization of the possible conditions that promote benzodiazepine abuse as well as information relevant to the mechanisms of benzodiazepine action and dependence. The receptor specificity of effects of benzodiazepines was addressed by a pA_2 analysis of the various actions of benzodiazepines including ataxic, and anticonvulsant effects. These studies suggest no appreciable differences in the mechanisms by which chlordiazepoxide and zolpidem produce these effects. In addition, the behavioral significance of the benzodiazepine system will be assessed in studies of benzodiazepine receptor inverse agonists. The anxiogenic actions of these compounds are being assessed in studies of discrimination as well as studies of the aversive effects of anxiogenic agents. Studies of the discrminative effects of the benzodiazepine antagonist flumazenil suggest that it has unique actions that are related neither to benzodiazepine agonist, inverse agonist, or antagonist effects.

GPO 914-918

Z01 DA00007-05 BPL

Abuse Liability and Behavioral Effects of Benzodiazepines

Publications

Aulisi, E.F., Wehby, R.G., Katz, J.L. and Valentino, R.J.: Selective proconflict effect of corticotropin-releasing factor (CRF). Society for Neuroscience Abstracts, 1989.

Katz, J.L.: Two type of bias in psychophysical detection and recognition procedures: nonparametric indices and effects of drugs. Psychopharmacology 97: 202-205, 1989.

Katz, J.L.: Effects of drugs on stimulus control of behavior under schedules of reinforcement. In Thompson, T., Dews, P.B. and Barrett, J.E. (Eds.): Advances in Behavioral Pharmacology. Vol. 7, Hillsdale, NJ, Lawrence Earlbaum Associates, 1989, pp. 13-38.

Katz, J.L., Winger, G.D. and Woods, J.H.: Abuse liability of benzodiazepines. In Brandon, S. and Hindmarch, I. (Eds.): Benzodiazepines: Current Perspectives, New York, NY, John Wiley, 1990, In press.

Takada, K., Suzuki, T., Hagen, T., Cook, J.M. and Katz, J.L.: Behavioral effects of benzodiazepine antagonists in chlordiazepoxide tolerant and non-tolerant rats. Life Sciences 44: 289-299, 1989.

Takada, K., Swedberg, M.D.B., Goldberg, S.R. and Katz, J.L.: Discriminative stimulus effets of intravenous l-nicotine and nicotine analogs or metabolites in squirrel monkeys. Psychopharmacology 99: 208-212, 1989.

Tortella, F.C., Witkin, J.M. and Musacchio, J.M.: Caramiphen, a non-opioid antitussive with potent anticonvulsant properties in rats. European Journal of Pharmacology 155: 69-75, 1988.

Witkin, J.M., Alvarado-Garcia, R., Perez, L.A. and Witkin, K.M.: Central oxotremorine antagonist properties of pirenzepine. Life Sciences 42: 2467-2473, 1988.

Witkin, J.M. and Katz, J.L.: Analysis of behavioral effects of drugs. Drug Development Research, 1989, In press.

Witkin, J.M., Lee, M.A. and Walczak, D.D.: Anxiolytic properties of amygdaloid kindling unrelated to benzodiazepine receptors. Psychopharmacology 96: 296-301, 1988.

Witkin, J.M. and Perez, L.A.: Comparison of effects of buspirone and gepirone with benzodiazepines and antagonists of dopamine and serotonin receptors on punished behavior of rats. Behavioural Pharmacology, 1989, In press.

Woods, J.H., Winger, G. and Katz, J.L.: Abuse of benzodiazepines. Journal of the American Medical Association 261: 2956-2957, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00009-03 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Cardiovascular Changes Induced By Cocaine

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	C.W. Schindler	Staff Fellow	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	S.R. Tella	Foreign Fellow	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*

None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.25	1.75	1.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The effects of cocaine on a number of physiological parameters are being studied in squirrel monkeys and rats. The results for squirrel monkeys indicate that cocaine (0.3-3.0 mg/kg) produces an immediate increase in mean blood pressure of 10-30 mm and a delayed increase in heart rate of 30-80 bpm. Phentolamine will antagonize the blood pressure increasing effect of cocaine and propranolol will antagonize the heart rate increasing effects of cocaine indicating that these effects are mediated through adrenergic systems. While anesthesia will attenuate the effects of cocaine, hexamethonium pretreatment does not alter the cocaine response on either heart rate or blood pressure, indicating that these effects are mediated via peripheral adrenergic systems. Haloperidol does partially antagonize the effects of cocaine on heart rate, indicating that dopaminergic systems may also be involved in the cardiovascular response. A variety of calcium channel antagonists (nimodipine, verapamil and diltiazem) will also antagonize the cardiovascular response to cocaine in both squirrel monkeys and rats, however, these drugs do not alter the lethal response of cocaine in rats. Further, the calcium channel antagonists also do not alter the behavioral response to cocaine in squirrel monkeys, including cocaine self-administration behavior. As such, the utility of the calcium channel antagonists in treatment may be restricted to cocaine's cardiovascular effects. We have also recently shown that the squirrel monkey can be used as a model for the cardiovascular response to smoked free-base cocaine (i.e., crack). We have shown that smoked cocaine does lead to an increase in both heart rate and blood pressure.

GPO 914-918

Z01 DA00009-03 BPL

Cardiovascular Changes Induced by Cocaine

Publications

Rao, T.S., Schindler, C.W. and Goldberg, S.R.: Autonomic neural mechanisms in the cardiovascular effects of cocaine in conscious squirrel monkeys. FASEB Journal 3: A297, 1989.

Schindler, C.W., Tella, S.R. and Goldberg, S.R.: Cardiovascular effects of cocaine: Doapmine antagonist, calcium channel antagonist and individual effects. In Thadani, P.V. (Ed.): Cardiovascular Toxicity of Cocaine: Underlying Mechanisms. National Institute on Drug Abuse Research Monograph, 1989, In press.

Schindler, C.W., Witkin, J.M., Rao, T.S. and Goldberg, S.R.: Effects of cocaine alone and in combination with haloperidol and SCH 23390 on cardiovascular function and schedule-controlled behavior in squirrel monkeys. FASEB Journal 3: A296, 1989.

Schindler, C.W., Witkin, J.M., Tella, S.R. and Goldberg, S.R.: Effects of cocaine alone and in combination with haloperidol and SCH 23390 on cardiovascular function and schedule-controlled behavior in squirrel monkeys. Psychopharmacology, 1989, In press.

Tella, S.R., Schindler, C.W. and Goldberg, S.R.: The role of central and autonomic neural mechanisms in the cardiovascular effects of cocaine in conscious squirrel monkeys. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

Tella, S.R., Schindler, C.W. and Goldberg, S.R.: Neural mechanims in the cardiovascular effects of cocaine. In Thadani, P.V. (Ed.): Cardiovascular Toxicity of Cocaine: Underlying Mechanisms. National Institute on Drug Abuse Research Monograph, 1989, In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00010-02 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Behavioral Pharmacology and Toxicology of Psychomotor Stimulants

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	J.M. Witkin	Senior Staff Fellow	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
	C.W. Schindler	Staff Fellow	BPGL, NIDA, ARC
	E.I. Shores	Research Psychologist	BPGL, NIDA, ARC

COOPERATING UNITS *(If any)*
None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.60	1.10	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This project is comprised of studies directed at an understanding of cocaine's behavioral effects, the consequences of its repeated administration, adverse effects of cocaine including neurotoxicity and lethality, and modulation of these effects of cocaine by behavioral and pharmacological factors. Integrated within this work are experiments designed to delineate the neuropharmacological mechanisms of action of cocaine. The primary findings to date are: (1) Whereas both dopamine D_1 and D_2 receptor antagonists prevent rate-increasing effects of cocaine, neither antagonist alters the rate-decreasing effects or the disruption of the temporal pattern of responding produced by cocaine; (2) Neither D_1 nor D_2 antagonists alter discriminative stimulus effects of cocaine at doses below those producing marked behavioral effects of their own; (3) The self-administration of cocaine by squirrel monkeys is positively associated with its actions as an inhibitor of dopamine reuptake; (4) Muscarinic antagonists potentiate rate-increasing effects of cocaine and prevent to some extent their rate-decreasing effects; (5) Tolerance to behavioral effects of cocaine may involve an important metabolic component; (6) Tolerance to cocaine confers cross-tolerance to apomorphine but not to selective D_1 or D_2 agonists; (7) Protection against the lethal effects of cocaine is conferred by the D_1 antagonist SCH 23390 but not by the D_2 antagonist haloperidol; (8) The cholinomimetics, oxotremorine and physostigmine, potentiate lethality of cocaine, and at lower doses, physostigmine protects against cocaine-induced lethality; (9) Cocaine regimens previously reported to produce neurotoxicity do not in our hands alter striatal or cortical dopamine levels; (10) Inhaled cocaine produces discriminative stimulus effects qualitatively similar to i.v. cocaine in squirrel monkeys.

Z01 DA00010-02 BPL

Behavioral Pharmacology and Toxicology of Psychomotor Stimulants

Publications

Katz, J.L., Dworkin, S., Dykstra, L.A., Carter, R.B. and Witkin, J.M.: Some behavioral effects of repeated d-amphetamine administration. Drug Development Research, 1989, In press.

Porrino, L.J., Ritz, M.C., Goodman, N.L., Sharpe, L.G., Kuhar, M.J. and Goldberg, S. R. Differential effects of the pharmacological manipulation of serotonin systems on cocaine and amphetamine self-administration in rats. Life Sciences 45: 1529-1535, 1989.

Ritz, M.C., Lamb, R.J., Goldberg, S.R. and Kuhar, M.J.: Cocaine self-administration appears to be mediated by dopamine uptake inhibition. Progress in Neuropsychopharmacology and Biological Psychiatry 12: 233-239, 1988.

Witkin, J.M., Goldberg, S.R. and Katz, J.L.: Lethal effects of cocaine are reduced by the dopamine-1 receptor antagonist SCH 23390 but not by haloperidol. Life Sciences 44: 1285-1291, 1989.

Witkin, J.M., Goldberg, S.R., Katz, J.L. and Kuhar, M.J.: Modulation of the lethal effects of cocaine by cholinomimetics. Life Sciences 45: 2295-2301, 1989.

Witkin, J.M. and Katz, J.L.: Analysis of behavioral effects of drugs. Drug Development Research, 1989, In press.

Witkin, J.M., Markowitz, R.A. and Barrett, J.E.: Physostigmine-insensitive behavioral excitatory effects of atropine in squirrel monkeys. Pharmacology Biochemistry and Behavior 32: 309-315, 1989.

Witkin, J.M. and Perez, L.A.: Comparison of effects of buspirone and gepirone with benzodiazepines and antagonists of dopamine and serotonin receptors on punished behavior of rats. Behavioural Pharmacology, 1989, In press.

Witkin, J.M., Ricaurte, G.A. and Katz, J.L.: Behavioral effects and lethality of N-methylamphetamine and N, N-dimethylamphetamine in rats and squirrel monkeys. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

Abstracts

Goldberg, S.R., Kuhar, M.J., Katz, J.L. and Witkin, J.M.: Modulation of the lethal effects of cocaine by cholinomimetics. Society for Neuroscience Abstracts 15: 252, 1989.

Griffiths, J., Witkin, J.M. and Katz, J.L.: Behavioral effects of repeated cocaine injections. FASEB Journal 3: A295, 1989.

Jaffe, J.H., Witkin, J.M., Goldberg, S.R. and Katz, J.L.: Potential toxic interactions of cocaine and mazindol. The Lancet II 8654: 111, 1989.

Katz, J.L., Ricaurte, G.A. and Witkin, J.M.: Behavioral pharmacology and toxicology of N, N-dimethyl-amphetamine. FASEB Journal 3: A1035, 1989.

Abstracts (cont'd)

Katz, J.L., Sharpe, L., Jaffe, J.H., and Witkin, J.M.: Discriminative stimulus effects of inhaled cocaine in squirrel monkeys. Society for Neuroscience Abstracts 15: 253, 1989.

Ricaurte, G.A., Witkin, J.M. and Katz, J.L.: Assessment of neurotoxic effects of cocaine. FASEB Journal 3: A298, 1989.

Schindler, C.S., Witkin, J.M., Rao, T.S. and Goldberg, S.R.: Effects of cocaine alone and in combination with haloperidol and SCH 23390 on cardiovascular function and schedule-conrolled behavior in squirrel monkeys. FASEB Journal 3: A296, 1989.

Witkin, J.M., Goldberg, S.R., Jaffe, J.H. and Katz, J.L.: Dopamine-1 receptor specific involvement in the lethal effects of cocaine. Society for Neuroscience Abstracts 15: 253, 1989.

Witkin, J.M., Goldberg, S.R. and Katz, J.L.: Lethal effects of cocaine are reduced by the dopamine-1 receptor antagonist SCH 23390 but not by haloperidol. FASEB Journal 3: A298, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00011-02 BPL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Behavioral and Neurotoxic Effects of Substituted Amphetamines

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	J.L. Katz	Research Psychologist	BPGL, NIDA, ARC
Others:	J.M. Witkin	Senior Staff Fellow	BPGL, NIDA, ARC
	E.I. Shores	Research Psychologist	BPGL, NIDA, ARC

COOPERATING UNITS *(If any)*

Dept. of Neurology, Johns Hopkins Univ. Sch. of Medicine, (G.A. Ricaurte); Dept. of Chemistry, VA Polytechnic Institute & State University (N. Castagnoli)

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.90	0.40	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The present studies are designed to examine behavioral and neurotoxic effects of substituted amphetamines. Abuse potential of these compounds are studied by assessing the reinforcing effects of these drugs when delivered i.v. to subjects trained to self-administer cocaine. Various substituted amphetamines, as well as other reference drugs of abuse, were studied for their reinforcing effects under fixed-ratio schedules of drug injection. The psychomotor stimulant effects of these compounds were examined in squirrel monkeys and rats trained to respond under fixed-interval schedules of reinforcement.

Methamphetamine was about ten times more potent than its N-methylated analog, N N-dimethylamphetamine (NNDMA), in producing disruptions in operant behavior, reinforcing effects, discriminative effects and dopaminergic neurotoxicity. Methamphetamine was only three times more potent in producing lethality. Importantly, NNDMA was devoid of serotonergic neurotoxicity. These results suggest that alterations in N-substituents can dramatically change the actions of β-phenethylamines.

Studies of the effects of drugs active on serotonergic systems before and after long-term treatment with (±)-3, 4-methylenedioxymethamphetamine (MDMA) indicated no change in these effects, suggesting that the functional consequences of serotonergic neurotoxicity induced by MDMA are subtle.

Z01 DA00011-02 BPL

Behavioral and Neurotoxic Effects of Substituted Amphetamines

Publications

Katz, J.L.: Drugs as reinforcers: Pharmacological and behavioral factors. In Liebman, J.M. and Cooper, S.J. (Eds.): The Neuropharmacological Basis of Reward. Oxford, U.K., Oxford University Press, 1989, pp. 164-213.

Katz, J.L., Dworkin, S.I., Dykstra, L.A., Carter, R.B. and Witkin, J.M.: Some behavioral effects of repeated d-amphetamine administrations. Drug Development Research, 1989, In press.

Katz, J.L. and Goldberg, S.R.: Second-order schedules of drug injection: Implications for understanding reinforcing effects of abused drugs. In Mello, N.K. (Ed.): Advances in Substance Abuse, Vol. 3, Greenwich, CT, JAI Press Inc., 1989

Katz, J.L., Ricaurte, G.A. and Witkin, J.M.: Behavioral pharmacology and toxicology of N,N-dimethylamphetamine (NNDMA). FASEB Journal 3: A1035, 1989.

Ricaurte, G.A., DeLanney, L.E., Irwin, I., Witkin, J.M., Katz, J.L. and Langston, J.W.: Evaluation of the neurotoxic potential of N,N-dimethylamphetamine: An illicit analog of methamphetamine. Brain Research 490: 301-306, 1989.

Ricaurte, G.A., Molliver, M.E., Witkin, J.M., Molliver, D.C. and Wilson, M.A.: d-Fenfluramine produces long-term effects on central serotonin neurons in nonhuman primates. Abstracts of the 19th Annual Meeting of the Society for Neuroscience Abstracts 15: 419, 1989.

Witkin, J.M., Ricaurte, G.A. and Katz, J.L.: Behavioral effects and lethality of N-methylamphetamine and N,N-dimethylamphetamine in rats and squirrel monkeys. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00012-01 BPL

PERIOD COVERED
June 15, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Genetic Factors in Response to Chronic Drug Treatment

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	R.J. Marley	Staff Fellow	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	N.L. Goodman	Res. Pharmacologist	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*

None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.50	1.00	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The purpose of this project is to evaluate individual differences in response to chronic administration of drugs of abuse using pharmacogenetic techniques. Genetic differences in sensitization to the effects of cocaine are presently being evaluated by examining the development of increased susceptibility to the seizure-inducing properties of cocaine following repeated administration of the drug (pharmacological kindling). Differences among inbred mouse strains in the rate of cocaine kindling have been found and strains of mice that sensitize at different rates identified. Additionally, we have observed that tolerance to cocaine-kindled seizures develops in the rapidly sensitizing strains. These genetic differences in response to chronic cocaine treatment do not correlate with differences in acute sensitivity to the drug. Similar experiments using lidocaine have also been completed. The results suggest that cocaine's convulsant and epileptogenic properties can not be explained entirely by its local anesthetic actions. Further evaluation of these findings has been proposed and will include: (1) assessment of genetic differences in response to carbamazepine treatment alone and in conjunction with chronic cocaine treatment; (2) evaluation of genetic differences in response to pharmacological kindling with other CNS stimulants, notably amphetamine and benzodiazepine inverse agonists; and (3) neurochemical analyses of possible changes in NMDA, GABA and voltage-dependent sodium channel binding parameters and function following these chronic treatment paradigms.

Genetic Factors in Response to Chronic Drug Treatment

Publications

Marley, R.J., Freund, R.K. and Wehner, J.M.: Differential response to flurazepamin in long-sleep and short-sleep mice. Pharmacology, Biochemistry and Behavior 31: 453-458, 1988.

Marley, R.J. and Gallager, D.W.: Chronic diazepam alters GABA-stimulated chloride influx in cortex, but not cerebellum. Society for Neuroscience Abstracts 14: 812, 1988.

Marley, R.J. and Gallager, D.W.: Chronic diazepam treatment produces regionally specific changes in GABA-stimulated chloride influx. European Journal of Pharmacology 159: 217-223, 1989.

Marley, R.J., Heninger, C. and Gallager, D.W.: The long-term, continuous release of FG 7142 increases GABA-stimulated chloride uptake in cortical membrane preparations. Society for Neuroscience Abstracts 14, 1989.

Marley, R.J., Stichcomb, A. and Wehner, J.M.: Further characterization of the benzodiazepine receptor in long-sleep and short-sleep mice. Life Sciences 43: 1223-1232, 1988.

Martin, B.J., Marley, R.J., Miner, L.L. and Wehner, J.M.: Classical genetic analysis of GABA-related seizures. Pharmacology, Biochemistry and Behavior 29: 501-507, 1988.

Wehner, J.M., Martin, B.J., Marley, R.J. and Pounder, J.I.: Behavioral studies of GABAergic responses in LS and SS mice: Are ethanol sensitivity and responses to GABAergic agents regulated by common mechanisms? In Deitrick, R.A. (Ed.): Proceeding of the Conference on Initial Sensitivity to Ethanol. 1989, In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00110-02 BPVL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Neural Substrates of Behavior Maintained by Intravenous Psychomotor Stimulants

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	L.G. Sharpe	Research Psychologist	BPVL, NIDA, ARC
Others:	N.L. Goodman	Pharmacologist	BPVL, NIDA, ARC
	J.H. Jaffe	Acting Chief	BPVL, NIDA, ARC

COOPERATING UNITS *(if any)*
National Institutes of Health, Mental Lab of Cerebral Metabolism (L.L. Porrino)

LAB/BRANCH
Clinical Pharmacology Branch, Biology and Psychology of Vulnerability Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.80	1.30	0.50

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Intravenous self-administration (IVSA) is a paradigm that has been used frequently to assess the reinforcing properties of drugs in several animal species. The purpose of the study was to determine the neuroanatomical and neurochemical basis of IVSA to several psychomotor stimulants. we found that amfonelic acid, a non-amphetamine class of psychomotor stimulant, was self-administered at doses 9.4 times lower than that of cocaine. The data indicate that amfonelic acid can act as a reinforcer in rats, and further suggest that amfonelic acid may have abuse potential in humans.

In a completed study, we investigated whether calcium channel blockers would reduce the reinforcing effect of cocaine self-administration in rats. Four calcium channel blockers were administered before the test session to rats whose responding was maintained by cocaine on an FR 10 schedule of reinforcement. Pretreatment with several doses of the type II antagonists, nifedipine and nimodipine (0.1 to 4 mg/kg, i.p.), significantly increased the number of cocaine self-injections but this effect was not dose dependent. Of the type I antagonists, verapamil depressed responding at 20 mg/kg, whereas diltiazem (3 to 60 mg/kg, i.p.) was without effect. The type II antagonists, nifedipine and nimodipine, may reduce calcium-dependent release of dopamine caused by cocaine. This project is terminated with extension of significant findings to be incorporated within other projects within the branch.

ZO1 DA00110-02 BPVL

Neural Substrates of Behavior Maintained by Intravenous Psychomotor Stimulants

Publications

Porrino, L.J., Goodman, N.L. and Sharpe, L.G.: Intravenous self-administration of the indirect dopaminergic agonist amfonelic acid by rats. Pharmacology, Biochemistry and Behavior 33: 623-626, 1989.

Porrino, L.J., Ritz, M.C., Goodman, N.L., Sharpe, L.G., Kuhar, M.J. and Goldberg, S.R.: Differential effects of the pharmacological manipulation of serotonin systems on cocaine and amphetamine self-administration in rats. Life Sciences 45: 1529-1535, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00111-02 BPVL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Neurochemical Mechanisms Controlling the Morphine Abstinence Syndrome

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	L.G. Sharpe	Research Psychologist	BPVL, NIDA, ARC
Others:	J.H. Jaffe	Acting Chief	BPVL, NIDA, ARC

COOPERATING UNITS *(if any)*
None

LAB/BRANCH
Clinical Pharmacology Branch, Biology and Psychology of Vulnerability Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.30	0.20	0.10

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The neurokinins (substance P, neurokinin A and B, Physalaemin, etc.) may play an important role in the opiate abstinence syndrome because morphine inhibits and naloxone increases their release in the morphine-dependent rat. The purpose of this study is to investigate this possibility by administering, to morphine-dependent rats (before naloxone), drugs that would be expected to either increase or decrease the efficacy of endogenous neurokinins. In a completed study, we found that captopril (0.3 mg/kg i.p.), a drug that increases peripheral levels of substance P, was found to enhance the secretory signs of abstinence in the morphine-dependent rat. Moreover, preteatment with capsaicin (125 mg/kg) prevented these enhanced withdrawal signs caused by captopril. This animal model may contribute to the development of drugs that could aid in the clinical management of opiate detoxification.

A study has been completed in which we investigated the effects of ibogaine on the morphine abstinence syndrome. Ibogaine is an alkaloid of Tabernanthe iboga H. Bn. and reportedly interacts with several receptor systems. A patent application for the use of ibogaine to interrupt abuse syndromes in humans has been filed. We found that ibogaine, in non-tremorogenic doses (5 and 10 mg/kg) had no effect on naloxone-precipitated withdrawal in morphine-dependent rats. This project is terminated with extension of significant findings to be incorporated within other projects within the branch.

ZO1 DA00111-02 BPVL

Neurochemical Mechanisms Controlling the Morphine Abstinence Syndrome

Publications

Sharpe, L.G. and Jaffe, J.H.: Captopril and capsaicin modify opioid withdrawal in the morphine-dependent rat. Pharmacology, Biochemistry and Behavior 33: 899-902, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00113-02 BPVL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Self-administration of Drugs in Aerosol Form in Rats

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	L.G. Sharpe	Research Psychologist	BPVL, NIDA, ARC
Others:	L.L. Weinhold	Staff Fellow	BPVL, NIDA, ARC
	J.H. Jaffe	Acting Chief	BPVL, NIDA, ARC
	A.B. Jaffe	Summer Student	BPVL, NIDA, ARC

COOPERATING UNITS *(if any)*

None

LAB/BRANCH
Clinical Pharmacology Branch, Biology and Psychology of Vulnerability Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
1.80	1.30	0.50

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Most drugs that are reinforcing in humans are administered by the pulmonary or intranasal routes (e.g., opiates, hallucinogens, cocaine, PCP, nicotine, cannabis). Indeed it has become the route of choice for many because of its immediate reinforcing effects and because intravenous drug use is often associated with those at risk for AIDS. Our goal is to develop an animal model for the self-administration of inhaled drugs so that we can explore the importance and advantages of this paradigm of drug seeking behavior. An ultrasonic nebulizer is used to create a drug vapor. The rat is trained to either lever press or nose poke on an FR 5 schedule of reinforcement for 2 to 5 sec of drug vapor. In a completed study, we found that when given access to sufentanil vapor, rats would press a lever for this opioid in a dose-dependent manner (10 to 75 mg/ml). Naloxone antagonizes this behavior and substituting water vapor for all concentrations of sufentanil significantly reduces responding within 5 to 20 2-hr sessions.

We have completed a study investigating whether rats reared in isolation would self-administer sufentanil vapor in a manner different from rats reared together. We tested whether patterns of drug taking behavior were related to dominance behavior quantified from video tapes of social pairings before and after the drug sessions. We found that individually caged rats acquired the self-administration of sufentanil vapor significantly faster than did rats that were housed as pairs. This project is terminated with extension of significant findings to be incorporated within other projects within the branch.

Z01 DA00113-02 BPVL

Self-administration of Drugs in Aerosol Form in Rats

Publications

Jaffe, A.B., Sharpe, L.G. and Jaffe, J.H.: Rats self-administer sufentanil in aerosol form. Psychopharmacology 99: 289-293, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00001-04 BGL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Pharmacogenetics: Acute Responses to Drug Administration

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	F.R. George	Senior Staff Fellow	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	M.C. Ritz	Staff Fellow	BPGL, NIDA, ARC
	G.I. Elmer	Staff Fellow	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*
None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.50	1.75	0.75

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

With the exception of ethanol, genetic factors have not been widely examined with other abused substances, but existing reports do indicate large genetic differences in both acute sensitivity and predisposition to self-administer drugs, particularly narcotics. In the present project with rats and mice, acute behavioral effects of drug administration are being systemically explored. The drugs being studied include opiate agonists, stimulants, especially cocaine, benzodiazepines, barbiturates, ethanol and phencyclidine. Drug effects are being studied using a variety of behavioral tasks, including open field activity, rotational behavior, tail flick, sleep time, rate altering effects, seizures and lethality. Simple physiological measures such as body temperature, blood pressure, heart rate and respiration rate may also be assessed. For each drug tested, several doses are examined to obtain dose-response patterns across a wide range of drug effects. Several inbred strains of mice and/or rats are being included in all experiments to determine an estimate of the genetic variation for each behavioral or physiological measure. Where appropriate, these measures will correlated with each other to estimate the commonalty among the acute responses studied. In all of these studies, genotype is incorporated as a independent variable. As the initial strain studies are completed, further genetic designs such as Mendelian analysis, are conducted to obtain estimates of the number of loci involved in determining a particular trait, as well as its mode of transmission. Where appropriate, biochemical studies are performed to determine the neural sites of action of the above drugs. Recent findings include: (1) A seizure response to cocaine mediated by a single gene; (2) A genetically determined depressant effect of cocaine; (3) A mouse genotype insensitive to cocaine; (4) sites of action for cocaine-induced seizures; and (5) sites of action for cocaine-induced lethality.

Z01 DA00001-04 BGL

Pharmacogenetics: Acute Responses to Drug Administration

Publications

Elmer, G.I. and George, F.R.: Indomethacin posttreatment antagonizes ethanol-induced sleep time. Annals of the New York Academy of Sciences 559: 441-443, 1989.

George, F.R.: Cocaine produces low-dose locomotor depressant effects in mice. Psychopharmacology 99: 147-150, 1989.

George, F.R. and Clouet, D.: Behavioral and biochemical genetic issues in substance abuse. Advances in Alcoholism and Substance Abuse, 1989, In press.

George, F.R. and Goldberg, S.R.: Genetic factors in response to cocaine. In Clouet, D., Asghar, K. and Brown, R. (Eds.): Mechanisms of Cocaine Abuse and Toxicity. National Institute on Drug Abuse Monograph 88: 239-249, 1988.

George, F.R. and Ritz, M.C.: Cocaine produces locomotor stimulation in SS/Ibg but not LS/Ibg mice. Psychopharmacology, 1989, In press.

Abstracts

Elmer, G.I. and George, F.R.: Ethanol-induced narcosis antagonized by posttreatment with indomethacin. Problems of Drug Dependence, 1988, Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph 90: 385, 1988.

Elmer, G.I. and George, F.R.: Antagonism of ethanol induced narcosis: The combined effects of indomethacin and Ro 15-4513. Alcoholism: Clinical and Experimental Research 13: 313, 1989.

Elmer, G.I. and George, F.R.: Cyclooxygenase inhibitors antagonize ethanol's depressant effects: Potency correlations with scheduled controlled behavior. Society for Neuroscience Abstracts 15(1): 36, 1989.

Elmer, G.I. and George, F.R.: Indomethacin antagonizes the effects of ethanol: Effect of route of administration. Problems of Drug Dependence, 1989, Proceedings of the 51st Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph, 1989, In press.

Pickworth, W.B., Klein, S.A., George, F.R. and Henningfield, J.E.: Acetaminophen and ethanol interactions in humans. Journal of Clinical Pharmacology and Therapeutics, 1989, In press.

Ritz, M.C. and George, F.R.: Cocaine-induced seizures: Mediation by specific CNS receptors. Neuropsychopharmacology, 1989, In press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA00002-04 BGL

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Pharmacogenetic Factors in Drug Reinforced Behavior

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	F.R. George	Senior Staff Fellow	BPGL, NIDA, ARC
Others:	S.R. Goldberg	Chief	BPGL, NIDA, ARC
	G.I. Elmer	Staff Fellow	BPGL, NIDA, ARC

COOPERATING UNITS *(if any)*
None

LAB/BRANCH
Preclinical Pharmacology Branch, Behavioral Pharmacology and Genetics Laboratory

SECTION
None

INSTITUTE AND LOCATION
Addiction Research Center, National Institute on Drug Abuse, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.00	1.25	0.75

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☒ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

The objectives of the proposed research are to identify and study factors that control drug reinforced behavior using genetically divergent rat and mouse populations. The methodology and principles of operant conditioning and pharmacogenetic analysis will be used. The studies will be limited to conditions in which the drug is taken orally and functions as a positive reinforcer. The focus will be on the variables that control drug reinforced behavior, especially genetic variables, but also including pharmacological variables and environmental variables, e.g., drug concentration and fixed ratio size. Emphasis will be given to systematically studying variables and over a range of values and interactions among variables will be parametrically explored. The proposed studies are important because (1) drug intake will be examined under conditions in which it is taken orally and functions as a reinforcer; (2) they will explore genetic and environmental factors and their interactions which contribute to drug self-administration; and (3) the use of genetically defined animals will provide information concerning the degree to which genetic factors regulate drug-seeking behavior, and will contribute to a systematized body of knowledge that will aid in the analysis of the complex problems of drug abuse. Recent findings include (1) Defining the existence of genetic differences in reinforcement from cocaine; and (2) Defining genetic differences in reinforcement from opiates.

Z01 DA00002-04 BGL

Pharmacogenetic Factors in Drug Reinforced Behavior

Publications

Elmer, G.I., Meisch, R.A., Goldberg, S.R. and George, F.R.: A fixed ratio analysis of oral ethanol reinforced behavior in inbred mouse strains. Psychopharmacology 96: 431-436, 1988.

George, F.R.: The role of arachidonic acid metabolites in mediating ethanol self-administration and intoxication. Annals of the New York Academy of Sciences, 559: 382-391, 1989.

George, F.R.: Prostaglandin synthetase inhibitors specifically modulate ethanol self-administration. Annals of the New York Academy of Sciences, 559: 449-450, 1989.

George, F.R.: Genetic approaches to studying drug abuse: Correlates of drug self-administration. Alcohol, 1989, In press.

George, F.R.: Is there a common genetic basis for reinforcement from alcohol and other drugs? Advances in Alcoholism and Substance Abuse, 1989, In press.

George, F.R., Elmer, G.I., Meisch, R.A. and Goldberg, S.R.: Oral self-administration of cocaine in C57BL/6J mice and the relationship between intake and behavioral effects. Journal of Pharmacology and Experimental Therapeutics, 1989, In press.

George, F.R. and Goldberg, S.R.: Genetic factors in response to cocaine. In Clouet, D., Asghar, K. and Brown, R. (Eds.): Mechanisms of Cocaine Abuse and Toxicity. National Institute on Drug Abuse Monograph, 88: 239-249, 1988.

George, F.R. and Goldberg, S.R.: Genetic approaches to the analysis of addiction processes. Trends in the Pharmacological Sciences, 10: 78-83, 1989.

George, F.R., Ritz, M.C. and Elmer, G.I.: The pharmacogenetics of drug effects and addiction. In Pratt, J. (Ed.): The Biological Basis of Drug Tolerance and Development. New York, NJ, Academic Press, 1989, In press.

Meisch, R.A. and George, F.R.: Influence of genetic factors on drug reinforced behavior in animals. In Pickens, R. and Svikus, D. (Eds.): Biological Vulnerability to Drug Abuse. National Institute of Drug Abuse Monograph, 89: 9-24, 1989.

Meisch, R.A., George, F.R. and Lemaire, G.: Oral self-administration of cocaine by rhesus monkeys. Pharmacology, Biochemistry and Behavior, 1989, In press.

Ritz, M.C., George, F.R. and Meisch, R.A.: Ethanol self-administration in ALKO rats: I Effects of selection and concentration. Alcohol 6: 227-233, 1989.

Ritz, M.C., George, F.R. and Meisch, R.A.: Ethanol self-administration in ALKO rats: II Effects of selection and fixed ratio size. Alcohol 6: 235-239, 1989.

Pharmacogenetic Factors in Drug Reinforced Behavior

Publications (cont'd)

Ritz, M.C., George, F.R., Boja, J. and Kuhar, M.J.: Cocaine binding sites on dopamine transporters: Interaction with other variables mediating reinforcement. In: Problems of Drug Dependence, NIDA Research Monograph, 1990, In press.

Suzuki, T. George, F.R. and Meisch, R.A.: Differential establishment and maintenance of oral ethanol reinforced behavior in Lewis and Fischer 344 inbred rat strains. Journal of Pharmacology and Experimental Therapeutics 245: 164-170, 1988.

Abstracts

George, F.R.: Mediation of ethanol self-administration by prostaglandin synthetase inhibitors. Problems of Drug Dependence, 1988, Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph 90: 345, 1988.

George, F.R. and Meisch, R.A.: Orally delivered cocaine as a reinforcer in mice, rats and rhesus monkeys. Neuropsychopharmacology, 1989, In press.

Ritz, M.C. and George, F.R.: Ethanol self-administration is not related to behavioral or biochemical sensitivity to ethanol. Problems of Drug Dependence, 1988, Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, Inc. National Institute on Drug Abuse Monograph 90: 361, 1988.

Treatment and Early Intervention Branch

David A. Gorelick, M.D., Ph.D., Chief

Introduction

The Treatment and Early Intervention Branch conducts research on psychopharmacologic and psychosocial treatments for drug abuse, especially for cocaine and I.V. heroin use. Research is carried out on both the ARC residential unit and the outpatient clinic. In addition, the Branch collaborates with other treatment facilities in the Baltimore area. The Treatment and Early Intervention Branch includes the Pharmacotherapy Laboratory (David A. Gorelick, M.D., Ph.D., Acting Chief).

Overview

Branch research studies explore the safety and efficacy of interventions with a view toward the replication of effective strategies by the treatment community. Detailed diagnostic and biopsychosocial characterization of subjects is done to identify predictors of treatment response, develop optimum matching of patients to treatment method, and determine the extent to which biomedical and psychosocial consequences of drug abuse are affected by treatment. These objectives are being pursued using a variety of research techniques, including (1) use of single-blind and double-blind procedures with pharmacologic interventions, (2) use of experimental designs employing random assignment to control and/or comparison groups, (3) obtaining and quantifying observational data to clarify behaviors significant to the conduct of drug abuse treatment. When possible, long-term followup is obtained on subjects after active treatment has ended, in order to assess the persistence of treatment effects.

Studies are frequently conducted in collaboration with other laboratories of the ARC, including the Chemistry and Drug Metabolism and Neuroendocrinology Laboratories (Clinical Pharmacology Branch) and the Cognitive Studies and Human Performance Laboratory (Etiology Branch). In addition, several programs of the Maryland Substance Abuse Administration have indicated a willingness to collaborate with the Branch as appropriate.

Long-term goals of the Branch will continue to be the exploration of issues significant to the treatment process and the examination of interventions that have promise for improved treatment. Specific areas of interest will include the impact of AIDS on drug abuse treatment, concurrent psychiatric diagnosis in drug addicts, and the effort to provide effective treatment for cocaine abusers. Many of these efforts will be in cooperation with NIDA's Medication Development and AIDS programs.

Summary of Ongoing Research

A. Effects of pharmacologic agents on cocaine abuse treatment: Weddington, W.W., Brown, B.S., and Jaffe, J.H.

Systematic investigation comparing different pharmacologic regimens for cocaine dependence with each other under blind conditions are only beginning to be reported in the literature. This study compared desipramine hydrochloride and amantadine hydrochloride in regimens suggested as useful in earlier open-trial investigations. A third group received placebo drug. The treatment regimen for the three randomly assigned groups lasted 12 weeks and involved twice weekly counseling in addition to daily medication.

The measures in this study assessed cocaine craving, sleep satisfaction, mood, drug use, psychological symptoms, and depression on a weekly or more frequent basis. Reports of side effects, blood pressure and pulse were obtained routinely. In addition, bloods were drawn periodically to assess compliance with the therapeutic regimen.

This study provided an assessment of two of the major pharmacologic strategies that have been suggested for use in outpatient treatment of cocaine abusers. There were no significant group differences on any dependent variable, thus failing to support the usefulness of these medications in the treatment of cocaine abuse. In addition, there were no significant differences in treatment retention or mood state between patients who were HIV antibody positive vs HIV antibody negative. Followup is now being done to assess patients' functioning six and twelve months after the completion of treatment.

B. Use of pharmacologic agents for cocaine dependence in methadone maintenance clients: Kolar, A., Brown, B.S. and Jaffe, J.H. (070)

Cocaine use has long been associated with opiate dependence and the two drugs have been used together to achieve particular psychic states. Increasingly, there have been reports from methadone programs of significant levels of cocaine use by persons stabilized on methadone. The first phase of this study systematically surveyed 12 methadone maintenance clinics, and found 17.1% of patients had a recent urine sample positive for cocaine. Such cocaine use hinders recovery and promotes dropout of patients from treatment. Clarification of the role played by available pharmacologic agents in permitting patients to be retained in methadone treatment can be significant not only to improving drug abuse rehabilitation, but to the containment of AIDS and HIV infection as well.

The second phase of this study explored, under double-blind conditions, the efficacy of amantadine hydrochloride, desipramine hydrocholoride, and a placebo condition in the treatment of cocaine-dependent methadone clients. Subjects from an area treatment program were randomly assigned to each of the three treatment groups, and received both methadone and study medication daily. Counseling and all other clinic activities were available to all groups.

The following measures were assessed at least weekly: cocaine craving, sleep satisfaction, mood, psychological symptoms,

depression, and drug use. Reports of side effects and blood pressure and pulse were obtained routinely. In addition, medication blood levels were obtained weekly for both safety and determination of treatment compliance.

All groups showed significant decreases in cocaine use, cocaine craving, and depression, with no significant differences among groups, except that patients receiving desipramine were more likely to stay in treatment and be cocaine-free at the end of the 3-month study period.

C. **Behavioral and physiological effects associated with acute cessation of cocaine abuse: Weddington, W.W., Cone, E., Dax, E. Herning, R.I. and Levin, F. (068)**

Acute cocaine cessation is a period of high risk for relapse, and has been associated with significant psychological, but not physical, abnormalities. These observations have been largely restricted to uncontrolled outpatient populations. The current investigation is intended to clarify behavioral and physiological functioning associated with the cessation of cocaine use under controlled, i.e. inpatient, conditions.

Individuals meeting criteria for cocaine dependence are admitted for study lasting up to six weeks. Measures are made at prescribed intervals to assess cognitive performance, neuroendocrine functioning, cocaine and cocaine-metabolite excretion, psychological status, depressive ideation, drug craving, sleep satisfaction/dissatisfaction, and cardiovascular functioning. Subjects will be available for followup assessments every other month up to one year post-discharge.

Among 12 male cocaine addicts studied to date, mood-distress and cocaine craving declined gradually after admission. The addicts differed from 10 non-addicted control subjects in resting heart rate, but not in any other physiological or sleep variable. These results suggest that cocaine, unlike alcohol or opiates, does not produce a prominent physical withdrawal syndrome.

D. **Characteristics of waiting list clients and behaviors: Brown, B.S., Hickey, J.E., Jaffe, J.H. (067)**

In much of the country, drug abuse treatment programs report maintaining lengthy waiting lists that delay entrance into treatment. To date, no studies had been conducted to examine the behaviors of waiting list applicants with regard to social functioning, efforts to locate alternative treatment opportunities and/or intentions to remain available to the treatment program initially contacted. The attitudes and behaviors of persons on a waiting list can have significance for their later program compliance and functioning in the community. These issues are relevant to the threat of HIV infection posed by intravenous drug users.

Studies of the behaviors of individuals awaiting entry into treatment can help clarify individuals' continuing accessability to treatment programs and the cost to society of maintaining waiting lists. They can lay the groundwork for the development of strategies to permit clients to remain available to treatment programs. In addition, such studies can clarify the extent to which IV drug users are practicing risk reduction behaviors in relationship to HIV infection and have made efforts to modify risk taking behaviors.

This project drew a random sample of 29 applicants on the waiting list for an area residential cocaine abuse treatment program. The sample was stratified by gender and length of time on the waiting list. Data were collected on the demographics and psychological functioning of waiting list clients; their functioning in the community with regard to drug use, employment and antisocial activities; efforts to obtain drug abuse treatment; and their risk taking/risk reduction behaviors in relationship to HIV infection. In addition, the study explored issues of community pressures for and against entry into treatment.

This study found few differences between applicants awaiting treatment for 3 months or less and those waiting 4-6 months. Nearly half the subjects reported reduced drug use in association with applying for treatment, while slightly more than half expressed pessimism about long-term recovery and less interest in entering treatment.

E. Buprenorphine/Methadone Comparison - Maintenance and Detoxification: Johnson, R.E., Fudala, P.J., Lange, W.R. (090)

The partial agonist burprenorphine is a clinically useful parenteral and sublingual analgesic approximately 25 to 40 times more potent than morphine. It has a wide therapeutic index and low toxicity, even in an overdose situation. Buprenorphine offers several advantages in drug-abuse treatment. Its opiate agonist properties make it acceptable to an addict population and offer cross-tolerance (blockade) to the effects of other opiates. As a partial agonist, the maximal effects of buprenorphine on behavioral and physiologic measures are less than those expected from a pure agonist. Buprenorphine has also been shown to attenuate the self-administration of heroin in humans.

The use of intravenously administered illicit drugs has been positively correlated with the spread of HIV infection; therefore, effective drug abuse treatment strategies can have a favorable impact on this spread. However, many intravenous opiate abusers refuse to enter treatment due to the limited pharmacotherapies available. Thus, the development of additional therapeutic interventions is warranted.

The purpose of this study is to determine the utility of buprenorphine in maintaining opiate-dependent individuals in outpatient treatment as compared to the prototypic treatment drug

methadone. A rapid buprenorphine dose-induction procedure, previously shown to be effective in an inpatient investigation, will be evaluated in an outpatient population. The ability of burprenorphine and methadone to decrease illicit opiate and cocaine use will be assessed, along with their effectiveness in maintaining individuals in treatment; and the number of subject-reported withdrawal and opiate-related side effects of the two treatments, as well as pharmacokinetic data, will be obtained from blood and/or urine analyses. Although previous investigations have indicated that the abrupt termination of buprenorphine produced only mild to moderate opiate withdrawal symptomatology, the present study uses a gradual dose reduction schedule in an effort to further enhance the acceptability of the treatment.

This study, enrolling over 160 subjects, is the largest clinical trial to date assessing the effectiveness of buprenorphine for the treatment of opiate dependence. It is hoped to potentially lead to the approval of buprenorphine as a pharmacotherapy for opiate dependence.

F. Assessment of Methadone Maintenance Treatment: Ball, J.C. and Brown, B.S.

Methadone maintenance has continued as both a controversial and widely employed treatment for opiate dependence. The controversy stems from concerns about the efficacy of methadone maintenance treatment and the level and types of services provided in methadone programs. Clarification of the efficacy of methadone maintenance treatment in reducing IV drug use, and thereby in halting the spread of HIV infection, and increased understanding of the role played by differing treatment and program variables will be essential aids in the improvement of drug abuse treatment programming and services.

A comprehensive schema for evaluation of methadone maintenance treatment has been developed, assessing 89 variables in 4 areas: patient history and characteristics, program characteristics, treatment service provided, and outcome. This schema was used to study the efficacy of treatment and the variables associated with successful outcome.

The subject population was 633 methadone patients at six methadone maintenance programs in Baltimore, New York and Philadelphia. Face-to-face interviews were conducted with all subjects on two occasions. In addition, data was gathered on the program services offered patients and characteristics of the treatment environment. Analysis was made of the extent to which successful outcome, i.e., diminution of drug taking behavior, occurred and the patient and program variables associated with successful outcome.

Treatment was found to be effective in reducing IV drug use and needle sharing among most heroin addicts. Of 388 patients who remained in treatment for one year or more, 71 percent had ceased IV use. Conversely, 82 percent of patients who left treatment

relapsed rapidly to IV drug use. Marked differences in the effectiveness of various programs were observed: current IV use varied from less than 10 percent to over 57 percent of patients in particular treatment programs. This differential effectiveness was related both to length of patient's stay and to the quality of treatment provided.

G. **Efficacy of fluoxetine and desipramine in the treatment of cocaine and PCP dependence: Covi, L., Brown, B.S., Jaffe, J.H. (081)**

Efforts to contain cocaine dependence and craving have focused on use of pharmacologic adjuncts with a primary impact on the dopaminergic system. Data from the field of neurosciences also implicate the serotonergic system as disrupted by cocaine and phencyclidine (PCP). In order to understand the efficacy of serotonergic blockers for limiting craving and use of cocaine and PCP, fluoxetine was selected as an antidepressant with particular potential. Double-blind studies were begun to compare the efficacy of fluoxetine, desipramine and a placebo drug (low dose diphenhydramine), assessing variables such as drug use, craving, psychological functioning, sleep disturbance, and physical health. All subjects received weekly counseling sessions.

Because of the interest in the use of fluoxetine, the initial study focused on comparison of fluoxetine and placebo. Fifty-three cocaine-dependent subjects were randomly assigned to fluoxetine (37) or placebo (16) groups. Preliminary data analysis suggests no significant group differences, whether subjects are grouped by fluoxetine dose (20, 40, or 60 mg daily) or achieved plasma level.

The second phase of this study, which is still ongoing, involves separate comparisons between fluoxetine and placebo in PCP abusers and desipramine and placebo in cocaine abusers. In this phase, factors which motivate entrance into treatment are being assessed, in order to determine possible predictive factors for treatment compliance and outcome.

H. **Spread of Cocaine Use in Adult and Adolescent Populations: Hickey, J., Brown, B.S., Kolar, A.F. (064)**

Spread of cocaine use through both adolescent and adult communities has given rise to widespread concern and a considerable investment of resources in an effort to contain that spread. Nonetheless, little is known of the way in which cocaine use has spread in the community and the extent to which factors involved in the spread of cocaine may differ for adults and aolescents. In addition, in spite of considerable effort to develop prevention programming, little is known about various factors which may influence cocaine use in vulnerable populations, or about the sources of information about the consequences of cocaine use which may be available to those populations and may be seen by them as being credible.

This study examined characteristics of adult and adolescent cocaine abusers in terms of initiation into, involvement with,

consequences from, and sources of information about cocaine use. Ninety adults (26 to 53 years old) and 20 youths (21 and below) stratified by gender and ethnicity were drawn from Baltimore area residential and outpatient treatment programs. Structured, closed-ended interviews were administered to assess issues in cocaine use as described above, plus relevant background and demographic characteristics.

In both groups, friends were the main providers of cocaine for the initial experience (youth: 55.0%, adult: 62.2%). Both groups reported few friends who did not use cocaine at the time of their initiation. Among the adults, 37.8% reported using heroin prior to cocaine initiation, compared with 15% of the youths. The adolescents were more involved with marijuana (youth: 95.0%, adult: 73.3%) and PCP (youth: 45%, adult: 6.7%) prior to cocaine initiation. More of the adults injected cocaine at initiation, (youth: 10.0%, adult: 35.5%). No adults and 2 adolescents initiated cocaine use by smoking.

87.7% of the adults and 80.0% of youth had experienced at least one negative consequence of their cocaine use, other than addiction, prior to entry into treatment, the commonest being loss of reality testing. While the youth had entered treatment within a year of first cocaine use, adults entered treatment 7.9 years after first use, and reported an average of 6.6 years of cocaine use before experiencing the first negative consequence.

Both adolescents and adults rated books and magazines as the most accurate source of information about cocaine, with television a close second. In terms of amount of information about cocaine use, adolescents ranked television first and friends second while adults reversed the order, ranking friends first and television second.

I. **Impact of Differing Intensitites of Drug Abuse Counseling: Covi, L., Baker, C., Hess, J.M. (098)**

This study is designed to evaluate the influence of frequency of individual counseling on the effectiveness of drug abuse treatment. Subjects meeting DSM-III-R criteria for cocaine dependence will be randomonly assigned to one of three treatment conditions: a) twice weekly counseling and urine monitoring for 12 weeks, b) once weekly counseling and urine monitoring for 12 weeks, or c) placement on a waiting list for 12 weeks with random assignment to either twice weekly urine monitoring or no services. After 12 weeks all subjects on the waiting list will be offered random assignment to one of the two counseling groups. All counseling will be delivered according to a specified therapy manual integrating aspects of interpersonal, cognitive, and behavioral approaches to drug abuse counseling.

Treatment outcome will be assessed by twice weekly collection of data on urine toxicology, self-reported drug use, alcohol breath analysis, craving for cocaine, and depressive symptoms. Follow-up will be done at 3, 6, and 12 months after active treatment in order to determine any long-term effects.

Currently, the counseling manual is being refined in pilot testing with three subjects, prior to beginning the large scale randomly assigned study design.

Publications

Ball, J.C., Lange, W.R. and Brown, B.S.: The prevalence of IV drug use and needle sharing among heroin addicts during and after methadone maintenance treatment. Proceedings of the 50th Annual Scientific Meeting, Committee on Problems of Drug Dependence, in press.

Ball, J.C. and Corty, E.: Policy issues pertaining to the treatment of heroin addicts in the United States - With particular reference to methadone maintenance therapy. Proceedings of the Dutch-American Conference. The Effectiveness of Drug Abuse Treatment, in press.

Ball, J.C., Lange, W.R., Myers, C.P. and Friedman, S.R.: Reducing the risk of AIDS through methadone maintenance treatment. Journal of Health and Social Behavior, 29:214-226, 1988.

Ball, J.C. and Ross, A.: The similarity of crime rates among heroin addicts in New York City, Philadelphia and Baltimore. Journal of Drug Issues, R. Rachin (ed.), Fall 1990 (Guest Issue), in press.

Ball, J.C.: A schema for evaluating methadone maintenance programs. Proceedings of the 51st Annual Scientific Meeting of the Committee on Problems of Drug Dependence, 1989. L.S. Harris (ed.). NIDA Research Monograph, in press.

Ball, J.C.: A comprehensive evaluation of methadone maintenance programs in New York City, Philadelphia and Baltimore. Advances in Alcohol & Substance Abuse (Guest Issue), in press.

Brown, B.S., Rose, M., Weddington, W. and Jaffe, J.: Kids and cocaine - A treatment dilemma. Jounal of Substance Abuse Treatment, in press.

Brown, B.S., Hickey, J.E., Chung, A.,S., Craig, R.D. and Jaffe, J.H.: The Functioning of Individuals on a Drug abuse Treatment Waiting List. Am J Drug Alcohol Abuse, 15(3):261-274, 1989.

Fudala, P.J., Iwanmoto, E.T.: Conditioned aversion after delay place conditioning with amphetamine. Pharmacol Biochem Behav, in press.

Fudala, P.J., Jaffe, J.H., Dax E., Johnson, R.E.: Use of buprenorphine in the treatment of opiate addiction II: Physiologic and behavioral effects of daily and alternate-day administration and abrupt withdrawal. Clin Pharmcol Ther, in press.

Herning, R.I., Hickey, J.E., Pickworth, W.B. and Jaffe, J.H.: Auditory event-related potentials in adolescents at risk for drug abuse. Biological Psychiatry, in press.

Hickey, J.E., Brown, B.S., Chung, A.S., Kolar, A.F., and Michaelson, B.S.: Perceived Risk and Sources of Information Regarding Cocaine. International Journal of the Addictions, in press.

Jasinski, D.R., Fudala, P.J., Johnson, R.E.: Sublingual versus subcutaneous buprenorphine in opiate abusers. Clin Pharmacol Ther, 1989; 45:513-519.

Johnson, R.E., Cone, E.J., Henningfield, J.E., Fudala, P.J.: Use of buprenorphine in the treatment of opiate addiction I: Physiologic and behavioral effects during a rapid dose induction. Clin Pharmacol Ther, 1989; 46:335-43.

Kolar, A.F., Brown, B.S., Weddington, W.W., and Ball, J.C.: A treatment crisis: Cocaine use by clients in methadone maintenance programs. Journal of Substance Abuse Treatment, in press.

Kolar, A.F., Brown, B.S., Weddington, W.W., Haertzen, C.A., and Michaelson, M.A.: Treatment of cocaine dependence in methadone maintenance clients: A double blind pilot study comparing desipramine and amantadine. American Journal of Drug and Alcohol Abuse, in press.

Kolar, A.F.: Management of cocaine abuse in methadone maintenance programs. Maryland Medical Journal, 12:1067-1068, 1989.

Lange, W.R., Ball, J.C., Pfeiffer, M.B., Snyder, F.R. and Cone, E.J.: The Lexington addicts, 1971-1972; Demographic characteristics, drug use patterns, and selected infectious disease experience. International Journal of the Addictions, 24(7):609-626, 1989.

Levin, F., Levin, H.R., and Nagoshi, C.: Assessment of autonomic functioning using spectral analysis in a cigarette smoking population. Biological Psychiatry, in press.

Platt, J.J., Buhringer, G., Kaplan, C.D., Brown, B.S., and Taube, D.O.: The prospects and limitations of compulsory treatment for drug addiction: Results of a German-American workshop. Journal of Drug Issues, in press.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Hess, J.A., and Kolar, A.F.: DSM-III-R Psychiatric Diagnosis of Cocaine Addicts Seeking Treatment. Archives of General Psychiatry, in press.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Cone, E.D., Dax, E.M., Herning, R.I., and Michaelson, M.A.: Changes in mood, craving, and sleep during acute abstinence reported by male cocaine addicts: A controlled, residential study. Archives of General Psychiatry, in press.

Weddington, W.W., Haertzen, C.A., Hess, J.M., Brown, B.S.: Psychological reactions and retention by cocaine addicts during treatment according to HIV serostatus: A matched-control study. Am J Drug Alcohol Abuse, in press.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Hess, J.M., Mahaffey, J.R., Kolar, A.F., Jaffe, J.H.: Comparison of amantadine and desipramine combined with psychotherapy for treatment of cocaine dependence. Am J Drug Alcohol Abuse, in press.

Presentations and Abstracts

Ball, J.C.: The effectiveness of methadone maintenance treatment in the United States - An overview. What Works Conference, 1989.

Ball, J.C.: Bridging the troubled waters. The Sixth Annual Methadone Maintenance Regional Conference, 1989.

Ball, J.C.: Reducing the risk of AIDS through methadone maintenance treatment: The impact of successful programs. Annual Meeting of the American Medical Society on Alcoholism and Other Drug Dependencies (AMSAODD), Atlanta, April, 1989.

Ball, J.C.: The evaluation of program success in methadone maintenance treatment. 51st Annual Scientific Meeting of the Committee on Problems of Drug Dependence, Keystone, CO, June 1989.

Ball, J.C.: Cocaine and heroin use by methadone maintenance patients. 51st Annual Scientific Meeting of the Committee on Problems of Drug Dependence, Keystone, CO, June 1989.

Ball, J.C.: The evaluation and findings of methadone maintenance treatment in three cities. The Southeastern School Seminar on Innovative Approaches to Methadone Treatment, Athens, Georgia, 1989.

Ball, J.C.: Reducing the risk of AIDS though methadone maintenance treatment. Alcohol & Drug Problems Association of North America, Washington, D.C., 1989.

Ball, J.C.: Cocaine treatment in a methadone center/challenges. The Sixth Annual Northeast Regional Methadone Conference, Newport, Rhode Island, 1989.

Hickey, J.E., Kolar, A.F., Michaelson, B.S., Chung, A., Haynie, C., Brown, B.S.: Spread of Cocaine among adults and adolescents. American Psychiatric Association, 142nd Annual Meeting, San Francisco, 1989.

Hickey, J.E., Brown, B.S., Chung, A.S., Kolar, A.F., Michaelson, B.S.: Cocaine: Perceived risk and sources of information. American Psychiatric Association, 142nd Annual Meeting, San Francisco, 1989.

Kolar, A.F., Ball, J.C., and Lane, R.: Cocaine treatment in a methadone center/challenges. The Sixth Annual Methadone Maintenance Regional Conference, 1989.

Levin, F., Weddington, W.W., Haertzen, C.A., McDuff, D., and Cohen, A.: A substance abuse consultation/liaison service: Characteristics of patients and pedagogical potential. AMERSA Annual Meeting, Rockville, MD, 1989.

Weddington, W.W. and Brown, B.S.: HIV-antibody testing of patients seeking treatment for drug abuse. Institute on Hospital and Community Psychiatry, Philadelphia, 1989.

Weddington, W.W., Brown, B.S., Hess, J., Kolar, A., and Haertzen, C.: DSM-III-R Diagnoses of Outpatient Cocaine Addicts. American Psychiatric Association, 142nd Annual Meeting, San Francisco, 1989.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Hess, J., and Kolar, A.F.: Amantadine and desipramine for cocaine dependence. American Psychiatric Association, 142nd Annual Meeting, San Francisco, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00065-02 TEI

PERIOD COVERED: January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.) The Treatment of Cocaine Abuse: with Amantadine Hydrochloride, Tyrosine or Placebo and Desipramine

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI:	W.W. Weddington	Visiting Scientist	Clin. Trials
Others:	B.S. Brown	Chief	TEI, NIDA
	J.H. Jaffe	Director	ARC, NIDA

COOPERATING UNITS (if any)

LAB/BRANCH: TEIB

SECTION:

INSTITUTE AND LOCATION: Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL:	OTHER.
2.90	.25	2.65

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

We conducted a single-blind, random assignment, placebo-controlled, twelve-week comparison of desipramine hydrochloride and amantadine hydrochloride as adjunctive treatments to counseling for cocaine dependence. Subjects were 54 outpatients who met DSM-III-R criteria for current cocaine dependence. Subjects treated with fixed doses of 200 mg/day desipramine (N=17), 400 mg/day amantadine (N=16), or placebo (N=21) did not differ at intake in cocaine use, lifetime histories of psychopathology, admission scores on psychometric assessments, and sociodemographics. All treatment groups demonstrated dramatic and persistent decreases in cocaine use, craving for cocaine, and psychiatric symptoms compared to intake. There were no significant differences among treatment groups regarding retention in treatment, craving for cocaine and cocaine use (confirmed by urine toxicology). There was a trend for subjects treated with desipramine to maintain longer periods of cocaine abstinence. Mean plasma concentration of desipramine in subjects was less than that recommended for treatment of depression; consequently further study of desipramine using higher dosages is suggested. The mean plasma concentration of amantadine in our subjects matched the steady state plasma concentrations reported for prevention of influenza in healthy young adults, suggesting that amantadine was not a useful aid in the treatment of cocaine dependence.

Six-month and one-year followup is being conducted on all subjects who volunteered for the study to assess longer term effects of treatment participation. As of 9/30/89, 61% of subjects had completed 6-month followup and 67% one-year followup.

Z01 DA00065-02 TEI

The Treatment of Cocaine Abuse: with Amantadine Hydrochloride, Tyrosine or Placebo and Desipramine

Publications

Weddington, W.W., Brown, B.S., Haertzen, C.A., Hess, J.M., Mahaffey, J.R., Kolar, A.F., Jaffe, J.H.: Comparison of amantadine and desipramine combined with psychotherapy for treatment of cocaine dependence. Am J Drug Alcohol Abuse, in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00081-2 TEI

PERIOD COVERED

January 1, 1988 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.) Double-Blind Comparison of Desipramine, Fluoxetine and Bromocriptine for the Treatment of Cocaine and PCP Abuse

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PIs:	B.S. Brown	Chief	TEI, NIDA
	J.H. Jaffe	Director	ARC, NIDA
Acting PI:	L. Covi	Visiting Scientist	TEI, NIDA

COOPERATING UNITS (if any)

LAB/BRANCH

Treatment and Early Intervention Branch

SECTION

INSTITUTE AND LOCATION

Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL.	OTHER:
2.5	0.5	2.0

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither

☐ (a1) Minors

☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

The first phase of this study examined the efficacy of fluoxetine in reducing craving and promoting abstinence for abusers of cocaine. Double-blind comparison was made to an active placebo, diphenhydramine 12.5 mg. All subjects visited the outpatient clinic for 12 weeks three times weekly for urine toxicology testing and nursing examination for side effects and vital signs. Two weekly visits included a 50-minute individual counseling session (Rounsaville's adaptation of interpersonal psychotherapy).

Fifty-three subjects (ages 21-60) were randomly assigned to receive one of three fluoxetine doses - 20 mg (11), 40 mg (14), 60 mg (12) or placebo (16). Preliminary data analysis found no significant group differences whether subjects were grouped by fluoxetine dose or achieved plasma level (above or below 100 mg/ml). These results do not suggest a role for fluoxetine in the outpatient treatment of cocaine abuse.

The second phase of this study involves separate double-blind comparisons between fluoxetine - 20 mg daily and placebo in PCP abusers and between desipramine 300 mg daily and placebo in cocaine abusers. For the cocaine study, subjects visit the outpatient clinic twice weekly with only one individual counseling session weekly. Forty-five subjects have entered the PCP study and 46 the cocaine study.

Three-, 6-, and 12-month follow-up is being conducted on all subjects who volunteer for the study, to assess longer term effects of treatment participation. As of 9/30/89, 68% of subjects had completed 3-month follow-up and 78% 6-month follow-up.

PHS 6040 (Rev 1/84) GPO 914-911

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00068-03 TEI

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT (80 characters or less. Title must fit on one line between the borders.)
Physiological and Psychological Aspects of Cocaine Cessation

PRINCIPAL INVESTIGATOR (List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)

PI:	W. Weddington	Visiting Scientist	TEI, NIDA
Others:	E. Dax	Chief	NI, NIDA
	F.R. Levin	Fellow	TEI, NIDA
	E. Cone	Chief	CDM, NIDA
	R.I. Herning		CHP, NIDA

COOPERATING UNITS (if any)

Chemistry and Drug Metabolism Laboratory, Cognitive Studies and Human Performance Laboratory, Neuroendocrinology Laboratory

LAB/BRANCH
Treatment and Early Intervention Branch

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL	OTHER
.65	.25	.40

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK (Use standard unreduced type. Do not exceed the space provided.)

We examined changes over 28 days in mood states, craving for cocaine, and sleep during acute abstinence reported by 12 male, predominantly intravenous-using cocaine addicted subjects residing in a research facility. For comparison, we examined 10 non-addicted control subjects. There were no significant differences between cocaine addicts and controls regarding demographics and selected DSM-III-R diagnoses other than psychoactive substance use disorder and antisocial personality disorder. There were significantly higher scores of psychiatric symptoms reported by addicts one week prior to admission. Mood-distress and depression scores recorded at admission and during acute abstinence were significantly greater than those reported by controls. Addicts' mood-distress scores and craving for cocaine were greatest at admission and decreased gradually and steadily during the 28-day study. There were no significant differences between groups regarding reports of sleep other than difficulty falling asleep and clear-headedness on arising. Although there were significant differences in resting heart rate at admission and over time, there were no significant differences in weight gain or blood pressure.

Given the absence of a classical "withdrawal" pattern, "acute abstinence" may be the more appropriate classificiation of psychological and physical phenomena experienced by cocaine addicts who initiate abstinence in a controlled environment.

Z01 DA00068-03 TEI

Physiological and Psychological Aspects of Cocaine Cessation

Publications

Weddington, W.W., Brown, B.S., Haertzen, C.A., Hess, J.A., and Kolar, A.F.: DSM-III-R Psychiatric Diagnosis of Cocaine Addicts Seeking Treatment. Archives of General Psychiatry, in press.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Cone, E.D., Dax, E.M., Herning, R.I., and Michaelson, M.A.: Changes in mood, craving, and sleep during acute abstinence reported by male cocaine addicts: A controlled, residential study. Archives of General Psychiatry, in press.

Weddington, W.W., Haertzen, C.A., Hess, J.M., Brown, B.S.: Psychological reactions and retention by cocaine addicts during treatment according to HIV serostatus: A matched-control study. Am J Drug Alcohol Abuse, in press.

Weddington, W.W., Brown, B.S., Haertzen, C.A., Hess, J.M., Mahaffey, J.R., Kolar, A.F., Jaffe, J.H.: Comparison of amantadine and desipramine combined with psychotherapy for treatment of cocaine dependence. Am J Drug Alcohol Abuse, in press.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00070-02 TEI

PERIOD COVERED
January, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Cocaine Abuse Treatment for Clients Receiving Methadone Maintenance: Double-Blind Trial with Amantadine and Desipramine

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	A. Kolar	Staff Fellow	CT, NIDA
Others:	B.S. Brown	Chief	TEI, NIDA
	J.H. Jaffe	Director	ARC, NIDA

COOPERATING UNITS *(if any)*
Maryland Substance Abuse Administration (Man Alive Program)

LAB/BRANCH
Clinical Trials Laboratory

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
2.90	.40	2.50

CHECK APPROPRIATE BOX(ES)
☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

In the first phase of this study, 12 methadone maintenance programs with a total patient population of 2,634 were surveyed to examine the extent of cocaine abuse among their clients and the therapeutic methods employed to decrease it. The percentage of patients with at least one urine sample positive for cocaine during the previous month was 17.1% (450/2,634). The proportion of patients using cocaine per program during the prior month ranged from 5.9% to 33%. Programs' use of urine testing, contingency contracting, individual and group psychotherapy, inpatient hospitalization, and a mandatory discharge did not appear to substantially alter the extent of cocaine use.

The second phase of this study was a double-blind, placebo-controlled, randomly assigned, 12-week comparison of desipramine (N=8), amantadine (N=5), and placebo (N=9) for treatment of cocaine dependence in patients receiving methadone maintenance. Subjects on desipramine received a mean dose of 174 mg daily and achieved a mean desipramine plasma level of 213 ng/ml. Subjects treated with amantadine received a mean amantadine dose of 200 mg daily, and achieved a mean amantadine plasma level of 225 ng/ml.

Subjects in all groups showed significant reductions in self-reported and urinalysis detected cocaine use, craving for cocaine, and depression. Subjects receiving desipramine were significantly more likely to remain in treatment and be cocaine-free at the end of the 12-week study period. There were no other significant differences among groups in terms of abstinence from cocaine, self-reported cocaine use, craving for cocaine, or depressive symptoms.

Z01 DA00070-02 TEI

Cocaine Abuse Treatment for Clients Receiving Methadone Maintenance: Double-Blind Trial with Amantadine & Desipramine

Publications

Kolar, A.F., Brown, B.S., Weddington, W.W., and Ball, J.C.: A treatment crisis: Cocaine use by clients in methadone maintenance programs. Journal of Substance Abuse Treatment, in press.

Kolar, A.F., Brown, B.S., Weddington, W.W., Haertzen, C.A., and Michaelson, M.A.: Treatment of cocaine dependence in methadone maintenance clients: A double blind pilot study comparing desipramine and amantadine. American Journal of Drug and Alcohol Abuse, in press.

Kolar, A.F.: Management of cocaine abuse in methadone maintenance programs. Maryland Medical Journal, 12:1067-1068, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00064-02 TEI

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Spread of Cocaine Use in Adult and Adolescent Populations

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	J. Hickey	Social Worker	RS, NIDA
Others:	B.S. Brown	Chief	TEI, NIDA
	A.F. Kolar	Staff Fellow	TEI, NIDA

COOPERATING UNITS *(if any)*

Maryland Substance Abuse Administration (Epoch Counseling Centers, X-Cell Program)

LAB/BRANCH
Early Intervention Laboratory

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
.50	.25	.25

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This study examined characteristics of adult and adolescent cocaine abusers in terms of initiation into, involvement with, consequences from, and sources of information about cocaine use. Ninety adults (26 to 53 years old) and 20 youths (21 and below) stratified by gender and ethnicity were drawn from Baltimore area residential and outpatient treatment programs. Structured, closed-ended interviews were administered to assess issues in cocaine use as described above, plus relevant background and demographic characteristics.

In both groups, friends were the main providers of cocaine for the initial experience (youth: 55.0%, adult: 62.2%). Both groups reported few friends who did not use cocaine at the time of their initiation. Among the adults, 37.8% reported using heroin prior to cocaine initiation, compared with 15% of the youths. The adolescents were more involved with marijuana (youth: 95.0%, adult: 73.3%) and PCP (youth: 45%, adult: 6.7%) prior to cocaine initiation. More of the adults injected cocaine at initiation, (youth: 10.0%, adult: 35.5%). No adults and 2 adolescents initiated cocaine use by smoking.

87.7% of the adults and 80.0% of youth had experienced at least one negative consequence of their cocaine use, other than addiction, prior to entry into treatment, the commonest being loss of reality testing. While the youth had entered treatment within a year of first cocaine use, adults entered treatment 7.9 years after first use, and reported an average of 6.6 years of cocaine use before experiencing the first negative consequence.

Both adolescents and adults rated books and magazines as the most accurate source of information about cocaine, with television a close second. In terms of amount of information about cocaine use, adolescents ranked television first and friends second while adults reversed the order, ranking friends first and television second.

Z01 DA 00064-02 TEI

Spread of Cocaine Use in Adult and Adolescent Populations

Publications

Hickey, J.E., Brown, B.S., Chung, A.S., Kolar, A.F., and Michaelson, B.S.: Perceived Risk and Sources of Information Regarding Cocaine. International Journal of the Addictions, in press.

Hickey, J.E., Kolar, A.F., Michaelson, B.S., Chung, A., Haynie, C., Brown, B.S.: Spread of Cocaine among adults and adolescents. American Psychiatric Association, 142nd Annual Meeting, San Francisco, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00098-01 TEI

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Impact of Differing Intensities of Drug Abuse Counseling

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	L. Covi	Visiting Scientist	NIDA/ARC
Others:	C. Baker	MS	RS, NIDA
	J. M. Hess	MA	TEI, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH
Treatment and Early Intervention

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS:	PROFESSIONAL:	OTHER:
0.5	0.1	0.4

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

This study is designed to evaluate the influence of frequency of individual counseling on the effectiveness of drug abuse treatment. Subjects meeting DSM-III-R criteria for cocaine dependence will be randomly assigned to one of three treatment conditions: a) twice weekly counseling and urine monitoring for 12 weeks, b) once weekly counseling and urine monitoring for 12 weeks, or c) placement on a waiting list for 12 weeks with random assignment to either twice weekly urine monitoring or no services. After 12 weeks all subjects on the waiting list will be offered random assignment to one of the two counseling groups. All counseling will be delivered according to a specified therapy manual integrating aspects of interpersonal, cognitive, and behavioral approaches to drug abuse counseling.

Treatment outcome will be assessed by twice weekly collection of data on urine toxicology, self-reported drug use, alcohol breath analysis, craving for cocaine, and depressive symptoms. Follow-up will be done at 3, 6, and 12 months after active treatment in order to determine any long-term effects. Currently, the counseling manual is being refined in pilot testing with three subjects, prior to beginning the large scale randomly assigned study design.

PHS 6040 (Rev. 1/84) GPO 914-918

DEPARTMENT OF HEALTH AND HUMAN SERVICES · PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00067-03 TEI

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Characteristics of Waiting List Clients and Behaviors

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	J. Hickey	Social Worker	RS, NIDA
Others:	B.S. Brown	Chief	TEI, NIDA
	J. H. Jaffe	Director	ARC, NIDA

COOPERATING UNITS *(if any)*

Maryland Substance Abuse Administration (X-Cell Program)

LAB/BRANCH
Clinical Trials Laboratory

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS	PROFESSIONAL:	OTHER:
0.50	0.25	0.25

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Using structured interviews and the SCL-90R, study was made of the behaviors of 29 applicants to a residential treatment program for cocaine abuse who had been placed on that program's waiting list for periods ranging from 1 to 6 months. It was hypothesized that applicants awaiting treatment for 3 months or less (N=16) would be more likely to view themselves as treatment candidates and would show behaviors different from those waiting 4-6 months (N=13). Being on the waiting list a longer period was associated with greater evidence of criminal justice involvement, but with few other differences. Nearly half the total sample (48.3%) reported having significantly reduced drug use in association with their applying for treatment, but most applicants (58.6%) were pessimistic about their long-term capacity to remain free of drug-related difficulty. The pattern of SCL-90R scores for all subjects suggested significant psychiatric symptoms, including depression. Nonetheless, a majority of all applicants (51.7%) reported themselves as having become less interested in entering treatment. Nearly all applicants reported high levels of encouragement for their decision to enter treatment from persons with whom they were living and about half reported encouragement from friends. Of the 23 applicants who were IV drug users, 10 (41.7%) reported knowing someone who had contracted AIDS, 87.0% reported having changed behaviors (chiefly needle sharing) to reduce the risk of infection, and 69.6% reported having obtained HIV testing. The difficulty encountered in locating a random sample of applicants suggests the problem of maintaining a useful waiting list for treatment.

PHS 6040 (Rev. 1/84) GPO 914-911

Z01 DA00067-03 TEI

Characteristics of Waiting List Clients and Behaviors

Publications

Brown, B.S., Hickey, J.E., Chung, A.,S., Craig, R.D. and Jaffe, J.H.: The Functioning of Individuals on a Drug abuse Treatment Waiting List. Am J Drug Alcohol Abuse, 15(3):261-274, 1989.

DEPARTMENT OF HEALTH AND HUMAN SERVICES - PUBLIC HEALTH SERVICE

NOTICE OF INTRAMURAL RESEARCH PROJECT

PROJECT NUMBER: Z01 DA 00090-01 TEI

PERIOD COVERED
January 1, 1989 to December 31, 1989

TITLE OF PROJECT *(80 characters or less. Title must fit on one line between the borders.)*
Buprenorphine/Methadone Comparison - Maintenance and Detoxification

PRINCIPAL INVESTIGATOR *(List other professional personnel below the Principal Investigator.) (Name, title, laboratory, and institute affiliation)*

PI:	R.E. Johnson	Chief	RS, NIDA
Others:	P.J. Fudala	Deputy Chief	RS, NIDA
	W.R. Lange	Medical Director	ARC, NIDA

COOPERATING UNITS *(if any)*

LAB/BRANCH
Research Support/Treatment

SECTION

INSTITUTE AND LOCATION
Addiction Research Center, NIDA, Baltimore, MD 21224

TOTAL MAN-YEARS.	PROFESSIONAL.	OTHER:
11.2	2.8	8.4

CHECK APPROPRIATE BOX(ES)

☐ (a) Human subjects ☐ (b) Human tissues ☐ (c) Neither
☐ (a1) Minors
☐ (a2) Interviews

SUMMARY OF WORK *(Use standard unreduced type. Do not exceed the space provided.)*

Previous residential studies conducted at the ARC have indicated that heroin-dependent individuals may be rapidly inducted onto buprenorphine without producing clinically significant opiate-withdrawal symptoms. These studies indicated that subjects may be maintained on daily or alternate-day buprenorphine dosing schedules and that the abrupt withdrawal of buprenorphine produced a mild to moderate withdrawal syndrome. Results from dose-ranging studies that indicated an appropriate dose for use in maintenance treatment were applied to a non-residential study.

The purpose of this study was to determine the effectiveness of bruprenorphine in maintaining opiate-dependent individuals in non-residential treatment as compared to the prototypic treatment drug methadone. The primary outcome measure was urine samples positive for opiates. Secondary outcome measures included urine samples positive for cocaine, missed clinic visits, retention time in the study, and subject-reported withdrawal and opiate-related side effects. Blood and urine chemistries were obtained to assess the safety of each treatment.

In this study, 162 opiate-dependent individuals were enrolled in a 180-day treatment/detoxification protocol. Although the final subject has been admitted to the protocol, the study blind will not be broken until the last subject has completed the protocol. Subjects were randomly assigned to receive buprenorphine or methadone after stratification by age, sex, and the results of a naloxone challenge test. Doses are given under double-blind, double-dummy (both oral and sublingual dosage forms given) conditions. Subjects receive routine non-medical treatment services according to a relapse prevention counseling strategy. Urine samples are obtained 3 times weekly and assessed for illicit drug use. This study is the largest clinical trial to date assessing the effectiveness of buprenorphine for the treatment of opiate dependence.

Z01 DA00090-01 TEI

Buprenorphine/Methadone Comparison - Maintenance and Detoxification

Publications

Fudala, P.J., Jaffe, J.H., Dax E., Johnson, R.E.: Use of buprenorphine in the treatment of opiate addiction II: Physiologic and behavioral effects of daily and alternate-day administration and abrupt withdrawal. Clin Pharmcol Ther, in press.

Johnson, R.E., Cone, E.J., Henningfield, J.E., Fudala, P.J.: Use of buprenorphine in the treatment of opiate addiction I: Physiologic and behavioral effects during a rapid dose induction. Clin Pharmacol Ther, 1989; 46:335-43.

LIBRARY
NIH
Amazing Research.
Amazing Help.

http://nihlibrary.nih.gov

10 Center Drive
Bethesda, MD 20892-1150
301-496-1080

NIH LIBRARY
3 1496 01023 9179