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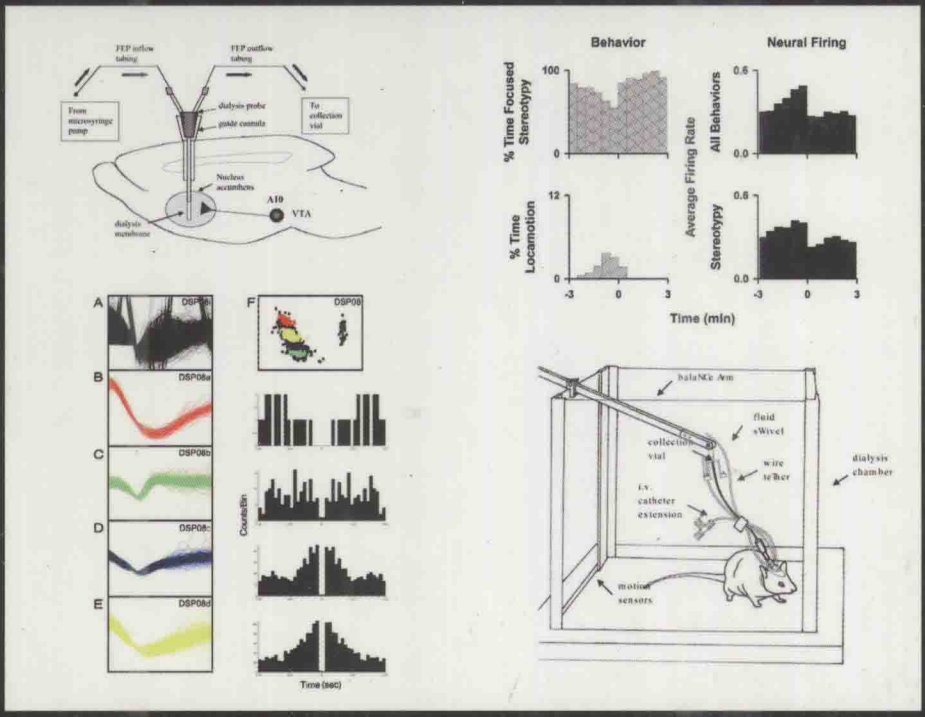
METHODS IN DRUG ABUSE RESEARCH

CELLULAR AND CIRCUIT LEVEL ANALYSES



Edited by Barry D. Waterhouse

METHODS & NEW FRONTIERS IN NEUROSCIENCE

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Our goal in creating the Methods & New Frontiers in Neuroscience series is to present the insights of experts on emerging experimental techniques and theoretical concepts that are, or will be, at the vanguard of neuroscience. Books in the series cover topics ranging from methods to investigate apoptosis, to modern techniques for neural ensemble recordings in behaving animals. The series also covers new and exciting multidisciplinary areas of brain research, such as computational neuroscience and neuroengineering, and describes breakthroughs in classical fields like behavioral neuroscience. We want these books to be the books every neuroscientist will use in order to get acquainted with new methodologies in brain research. These books can be given to graduate students and postdoctoral fellows when they are looking for guidance to start a new line of research.

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Sidney A. Simon, Ph.D.
Miguel A.L. Nicolelis, M.D., Ph.D.
Series Editors

Dedication

To Kathy for her unwavering love, companionship, and support through all my scientific endeavors.

About the Editor

Barry D. Waterhouse is a professor in the Department of Neurobiology and Anatomy and an associate dean of biomedical graduate studies at Drexel University College of Medicine (formerly MCP-Hahnemann University School of Medicine). After receiving his B.S. degree in biology in 1971 from Muhlenberg College, Dr. Waterhouse completed his Ph.D. in pharmacology at Temple University in 1977. From 1977 through 1987 he worked at Southwestern Medical School, University of Texas at Dallas, rising from postdoctoral fellow, to instructor, and then finally to assistant professor.

In 1987 he was recruited to the Department of Physiology and Biophysics as an associate professor at Hahnemann University School of Medicine, where in 1988 he developed and was subsequently appointed director of the university's graduate program in neuroscience, a post he held until 1994. In 1992 he was promoted to professor of physiology and biophysics, and in 1994, when Hahnemann University merged with Medical College of Pennsylvania (MCP), Dr. Waterhouse was invited to join the Department of Neurobiology and Anatomy in the newly formed university. He continued as director of the neuroscience graduate program at MCP-Hahnemann until 2001 and also served as vice-chair of the Department of Neurobiology and Anatomy from 1999 to the present. He was elected to the American College of Neuropsychopharmacology in 1996 and to the College on Problems of Drug Dependence in 1995. Throughout his research career Dr. Waterhouse has focused on the neurobiology of central monoaminergic systems and psychostimulant drug actions.

Contributors

Michael H. Baumann
Medications Discovery Research Branch
National Institute on Drug Abuse-Intramural Research Program
National Institutes of Health
Baltimore, Maryland

Craig W. Berridge
Department of Psychiatry
University of Wisconsin
Madison, Wisconsin

Jason J. Burmeister
Center for Sensor Technology
University of Kentucky Chandler Medical Center
Lexington, Kentucky

David M. Devilbiss
Department of Neurobiology and Anatomy
MCP-Hahnemann University
Philadelphia, Pennsylvania

Steven I. Dworkin
Department of Psychology
University of North Carolina at Wilmington
Wilmington, North Carolina

Greg A. Gerhardt
Department of Anatomy and Neurobiology
University of Kentucky
Lexington, Kentucky

Alexander F. Hoffman
Cellular Neurobiology Branch
National Institute on Drug Abuse-Intramural Research Program
National Institutes of Health
Baltimore, Maryland

Patricia H. Janak
Ernest Gallo Clinic and Research Center
Department of Neurology
The University of California at San Francisco
San Francisco, California

Laura L. Peoples
Department of Psychology, Neuroscience Graduate Group
University of Pennsylvania
Philadelphia, Pennsylvania

John J. Rutter
Department of Biological Sciences
Truman State University
Kirksville, Missouri

Michael F. Salvatore
Department of Anatomy and Neurobiology
University of Kentucky
Lexington, Kentucky

Dustin J. Stairs
Department of Psychology
University of North Carolina
at Wilmington
Wilmington, North Carolina

Barry D. Waterhouse
Department of Neurobiology
and Anatomy
MCP-Hahnemann University
Philadelphia, Pennsylvania

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1 Overview

Barry D. Waterhouse and Laura L. Peoples

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1.1 INTRODUCTION

Drug addiction is a progressive disorder characterized by a transition from controlled to uncontrolled and compulsive drug seeking that continues despite knowledge of adverse consequences (Hoffman and Goldfrank, 1990; Leshner, 1997; McGinnis and Foege, 1999). It is also a chronic relapse disorder. Periods of successful drug abstinence for many individuals end with relapse to compulsive drug use. Drug addiction is a devastating disorder that has severe health costs to both the individual and the public (McLellan et al., 2001). Although environmental variables can influence an individual's risk for developing addiction, human and animal research show that addiction is fundamentally a disorder of the brain. Application of neuroscience approaches to the study of addiction is thus an integral part of efforts to understand and ultimately to treat the disorder. Two issues that are central to understanding the problem of drug abuse and addiction are 1) identification of drug actions that contribute to an initially positive drug experience and 2) elucidation of the neural mechanisms underlying the progression of addiction and the development of drug craving. Acute self-administration of addictive compounds produces a multitude of

transient effects on the brain, only some of which contribute to a positive drug experience and the desire to repeat that experience in a social setting. However, chronic use of these substances leads to long-lasting changes in nervous system function that are thought to contribute to the development and maintenance of compulsive and uncontrollable drug seeking.

1.1.1 Theories of Drug Addiction

Investigation of the acute and chronic effects of addictive drugs has led to the development of a number of formal theories of addiction. Two that are particularly influential today are the incentive motivation theory (DiChiara, 1998; Everitt et al., 1999; Robbins and Everitt, 1999; Robinson and Berridge, 1993; Stewart et al., 1984; Stewart, 1992) and the hedonic dysregulation theory (Koob and LeMoal, 1997). Evidence consistent with each theory can be found in the animal and human literature on addiction; however, important predictions of each have yet to be tested fully. Although the theories differ in a number of aspects, they are not necessarily mutually exclusive and actually share at least two common assumptions. First, it is proposed that repeated exposure to drugs produces long-lasting changes in the brain that contribute to the development of compulsive and uncontrollable drug taking. Second, these changes are proposed to occur in multiple regions and at several levels of organization, from the level of molecular regulation of protein synthesis and cellular function to the level of local neural networks and circuits involving interactions between multiple brain structures. These theories, and an additional hypothesis that is gaining increasing influence, are reviewed and critically evaluated relative to existing data in a number of publications (Robinson and Berridge, 1993; Jentsch and Taylor 1999; Wise, 1999; Koob and LeMoal, 2001). They will thus be only briefly summarized here because they are relevant to understanding the utility of the research methods described in this volume.

1.1.1.1 Incentive Motivation Theory of Addiction

Incentive motivation theories of drug addiction propose that the disorder reflects a pathological responsivity of individuals to the influences of drug-associated conditioned stimuli on behavior. The theories further assert that the abnormal responsivity to drug stimuli is caused by acute and long-lasting actions of addictive drugs on brain. Acute drug actions are proposed to amplify mechanisms that contribute to stimulus–reward learning and lead to abnormally powerful conditioning of stimuli associated with drug administration. It is also proposed that long-lasting neuroplasticity induced by drugs facilitates this drug-induced amplification of learning (for review see DiChiara, 1998; Everitt et al., 1999; Robbins and Everitt, 1999; Robinson and Berridge, 1993; Stewart et al., 1984; Stewart, 1992). These proposals are based, in part, on evidence that stimuli associated with drugs undergo conditioning and can facilitate drug seeking in both animals and humans (Arroyo et al., 1998; Davis and Smith, 1987; deWit and Stewart, 1981; Ehrman et al., 1992; Goldberg et al., 1976; Ranaldi and Roberts, 1996; Stewart et al., 1984; Tiffany, 1990). Additionally, animal studies show that acute actions of addictive drugs amplify the gain of behavioral

responses to conditioned stimuli and may facilitate conditioning (Harmer and Phillips, 1998; O'Brien et al., 1998; Panililio et al., 1998; Robbins et al., 1989; Taylor and Robbins, 1986; Taylor and Horger, 1999; Weiss et al., 2000). Furthermore, repeated administration of addictive drugs sensitizes animals to various effects of those drugs including acute reinforcing effects and gain-amplifying effects on responsivity to conditioned stimuli (Lorrain and Vezina, 2000; Taylor and Horger, 1999; Wyvell and Berridge, 2001).

1.1.1.2 Hedonic Dysregulation of Reward and Allostasis

The hedonic dysregulation theory (Koob and LeMoal, 1997 and 2001) proposes that addiction is a self-regulatory condition. The theory is based on principles of homeostatic self-regulation and allostasis (Sterling and Eyer, 1988) and has roots in the opponent process theory of motivation described by Solomon and Corbit (1974). Homeostasis corresponds to the mechanisms that maintain stability within physiological systems and hold all parameters of an organism's internal milieu within adaptive limits. Certain parameters are held at a constant set point by local negative feedback responses to deviations from the set point. Other parameters are allowed to vary within a wide range so as to maintain balanced function within particular physiological systems. In contrast, the principle of allostasis involves the stabilization or balancing of function by a resetting of the set point in response to a chronic demand on homeostatic mechanisms that are insufficient to fully compensate for the deviations in the original set point. These allostatic changes can compensate for the demand, but they can also lead to an abnormal state when the demand is removed and are proposed to contribute directly to the development of drug addiction.

More specifically, acute actions of addictive drugs are proposed to overactivate reward pathways in the brain. This overactivation leads to a homeostatic response that involves down-regulation of neurochemical systems involved in mediating the drug-induced overactivation. However, the homeostatic changes in reward neurotransmitters are hypothesized to be insufficient to maintain balanced function within the reward system. This insufficiency, in conjunction with a chronic demand on these homeostatic mechanisms, leads to the onset of an allostatic process. This process involves changes that tend toward reestablishing the balanced reward function by changing the reward set point that is normally guarded by homeostatic mechanisms. The increase in reward set point is proposed to be mediated, in part, by recruitment of nonreward systems, specifically brain stress systems that are normally involved in negative emotional states. The recruitment and changes in the brain stress systems tend to counteract the drug-induced overactivation of the reward system but engender a negative mood or state in the absence of drug. This negative mood state is thought to set up a negative reinforcement mechanism. That is, individuals begin to seek drug in order to avoid the negative mood state. It is proposed that these allostatic responses grow in magnitude with repeated drug use. Thus, addiction is proposed to be a feed-forward cycle in which increases in drug intake are followed by increases in the magnitude of the allostatic response and resetting of the reward set point, which leads to further increases in drug intake and the reinitiation of the cycle.

This theory is based primarily on two observations. First, acute withdrawal from addictive drugs is commonly associated with negative affective states including dysphoria, depression, irritability, and anxiety. Second, after cessation of intravenous self-administration, animals exhibit an increased threshold for intracranial self-stimulation (ICSS) reward (i.e., a higher level of stimulation is required for the stimulation to be reinforcing). Additional evidence consistent with the hypothesis is described by Koob and LeMoal (2001).

1.1.1.2.1 Hypoactivity in Cortical Inhibitory Mechanisms

Neuroimaging studies in humans show that individuals addicted to drugs such as cocaine show evidence of reduced activity and lower cell density in cortical regions such as the anterior cingulate and orbitofrontal cortex (Volkow, 1991; Childress et al., 1999; Franklin et al., 2002). These and other cortical structures are involved in willed or executive control of behavioral selection. Executive control of behavior involves dynamic emotional and cognitive analyses of past and expected events and the influence of these analyses on decisions about future actions. This online willed control comes into play when automatic or habitual behaviors controlled by subcortical brain structures are not sufficient to guide adaptive behavior. Moreover, it can be important in the initiation of actions, persistence of adaptive actions in the absence of reward, and inhibition of impulses to engage in alternative but less beneficial behaviors. The anterior cingulate cortex (ACC) and orbitofrontal cortex contribute to the generation of emotion and to executive control of the influence of these emotions on behavior. Abnormalities in these brain regions are associated with a range of disorders involving disturbances in emotion and action. In light of these data, it is hypothesized that the cortical abnormalities observed in addicted individual contribute to addiction (Volkow, 1991; Childress et al., 1999; Rogers et al., 1999; Volkow and Fowler, 2000; London et al., 2000).

Consistent with this hypothesis, addicted individuals exhibit symptoms associated with insults to the ACC and orbitofrontal cortices including anhedonia and an inability to make adaptive decisions regarding future actions (e.g., Grant et al., 1996; Rogers et al., 1999). In fact, a hallmark of drug addiction is compulsive and uncontrollable drug use, despite knowledge of adverse consequences. It is thus possible that addicted individuals, like others who suffer from hypoactivity in these brain regions, are unable either to experience normal affective responses to future events or to exert executive control over the adaptive influence of those emotions on selection of beneficial actions. Perhaps most important in the case of addiction, the deficiencies in emotional regulation may limit the ability of addicted individuals to inhibit responses to rewards, including drug, in order to avoid harm (e.g., incarceration or death). There is some evidence that these neural abnormalities are induced by drug exposure, but it is also possible that some or all of the brain abnormalities are present in the individual prior to drug exposure and perhaps enhance vulnerability to addiction. Finally, it has been proposed that addiction may reflect a combination of weakened cortical inhibitory mechanisms and the overactive (sensitized) responses of subcortical mediated responses to drug-associated stimuli (for review see Jentsch and Taylor, 1999).

1.1.1.2.2 *Experimental Approaches for Studying Drugs of Abuse*

Research into the problem of drug abuse and drug addiction represents an interdisciplinary enterprise that to date includes social scientists, behavorists, biochemists, pharmacologists, physiologists, anatomists, neuroimaging specialists, and cell and molecular biologists. Within these disciplines many different techniques and experimental preparations have been utilized to address fundamental biological questions regarding the actions of acute and chronic drug administration. Initial studies using behavioral approaches characterized drug-related behaviors and defined assays for evaluating the reinforcing properties of addictive compounds. Biochemical and pharmacological investigations determined the cellular targets and time course of drug actions within brain tissue. Early anatomical and more recent imaging studies have identified specific pathways and regions of the brain that are activated directly or subsequent to acute or chronic drug administration or during drug withdrawal or craving. Experiments employing molecular biological techniques have identified gene products in brain tissue that represent genomic responses to chronic drug administration. In many cases, the appearance of these products has been linked to specific drug-related behaviors. Additional studies have provided evidence of genetic predisposition to compulsive behaviors and susceptibility to drug addiction. The reader is referred to numerous excellent reviews that document progress made with each of the abovementioned approaches (Altman et al., 1996; Amara and Sonders, 1998; Bozarth, 1987; Carroll and Mattox, 1997; Cloninger and Dinwiddie, 1993; Gatley and Volkow, 1998; George, 1997; Hyman et al., 2001; Johanson and Fischman, 1989; Koob et al., 1998; Lukas, 1997; Lyons et al., 1997; Markou et al., 1993; Nestler, 1992; Pickens et al., 1978; Self and Nestler, 1998; Vrana and Vrana, 1997; White and Kalivas, 1998; Wise and Bozarth, 1987;).

Despite the advances that have resulted from the use of these varied experimental applications, they do not address the question of how addictive drugs initiate and consolidate system-wide changes in neuronal functions that underlie drug-related behavior. However, combinations of electrophysiological, neurochemical, and behavioral techniques are uniquely capable of providing information that can fill this considerable gap in our understanding.

1.2 RATIONALE FOR CELLULAR AND CIRCUIT LEVELS OF ANALYSIS OF DRUGS OF ABUSE

Implicit in any description of drug addiction is the fact that circuits within the brain must be reorganized so as to generate compulsive behaviors for acquisition and self-administration of rewarding compounds. Such changes are clearly long-lasting and not readily reversible, as evidenced by the chronic relapsing character of drug addiction. The physical and psychological symptoms that emerge during withdrawal from chronic drug use and the sensitization that is evident with many addictive compounds further underscore the likelihood that drug addiction is a behavioral manifestation of fundamental changes in cellular biological processes. Many studies have in fact begun to identify neuron-specific gene products that are synthesized in response to chronic