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TOLERANCE TO, AND DEPENDENCE ON, SOME NON-OPIATE PSYCHOTROPIC DRUGS[1, 2]

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[1] We are indebted to the Documentation Section of the Addiction Research Foundation, under the direction of Mr. E. Polacsek, for assistance in collecting the reference material for this review. Systematic search of the literature covered publications up to August 31, 1970; occasional papers appearing after that time have been included if considered particularly relevant.

[2] A partial treatment of some aspects of this subject is contained in *Basic Aspects of Alcoholism*, edited by Y. Israel and J. Mardones, Wiley-Interscience Publishers, New York, 1971, in press.

I. INTRODUCTION

It has been known for centuries that regular drinkers generally become able to tolerate larger amounts of ethanol on repeated exposure. With the advent of barbiturates, other hypnotics and minor tranquilizers, the same tendency has been observed among regular users of these drugs. In addition, almost from their respective times of introduction into clinical medicine, these drugs were also found to resemble alcohol in being able to give rise to clinical pictures of varying degrees of severity, characterized by manifestations of hyperexcitability of the nervous system, on reduction or termination of a period of sustained intoxication. Although these hyperexcitable states have been known for centuries in relation to heavy use of alcohol, controversy continued until less than 20 years ago as to whether they were signs of chronic intoxication or of drug withdrawal. In this instance, common experience proved to be superior to many older clinical judgments, because alcoholics have long known that the signs can be abolished by ingestion of more alcohol and are manifestations of physical dependence upon it.

The factors influencing the development and loss of tolerance and dependence, in relation to ethanol as well as to other neurotropic drugs, have been investigated sporadically for over a century. However, significant progress could not be made until reliable methods had been developed for the production and measurement of tolerance and dependence in experimental subjects, so that the influence of various factors could be studied quantitatively. Some methods have been available for about 30 years, but the most reliable objective ones have been developed only during the past decade.

Behavioral scientists, on the other hand, have generally explored the phenomena of tolerance and dependence in behavioral terms, without reference to physiological or biochemical processes. A small but increasing body of knowledge indicates important functional relations between the effects of behavioral and pharmacological manipulations. The nature of these relations has only recently begun to be clarified.

Finally, experimental analysis of the fundamental biological mechanisms of tolerance and of dependence, and their relation to each other, must be considered rudimentary. A number of physiological and biochemical processes in brain and other tissues have been shown to change in a way which coincides roughly, in time and magnitude, with the changes in tolerance and dependence. They have been aptly designated "biochemical correlates" of tolerance (321). However, the exact relationship of these various processes to the integrated function of the

nervous system is far from clear. Therefore it is not yet possible to know which of the changes are causally related to the development of tolerance and dependence, which are consequences, and which are concomitant results of the same basic mechanisms.

The purpose of this review will be to summarize certain aspects of the present knowledge concerning each of these topics in turn, and to indicate important questions which have not yet been answered.

II. TOLERANCE

A. Definition

The term "tolerance," as used in relation to ethanol or any other drug, has two different connotations. The first is that of spontaneous or *initial tolerance*, as expressed by the amount of a drug which the subject must receive on first exposure to it, to produce a designated degree of effect. Though sometimes incorrectly called "innate tolerance," it reflects a variety of both congenital and environmental factors which contribute to the broad range of differences in tolerance among individuals, sexes, species, age groups, dietary and other treatment groups. The second connotation is that of an *acquired change in tolerance* within the same individual as a result of repeated exposure to the drug, so that an increased amount of drug is required to produce the same specified degree of effect, or less effect is produced by the same dose of drug. This definition of tolerance is valid only for a specified individual drug action, and not necessarily for the composite picture of all actions of a given drug on the whole organism. Examples of differential tolerance to various effects of the same drug will be given later in this review.

There is no evidence that an individual's capacity for increase in tolerance is proportional to his initial tolerance, nor is there any evidence to the contrary. Because of the lack of factual information, initial tolerance will not be examined in detail in this review, and *tolerance* will be used synonymously with *acquired increase in tolerance.*

The definition of tolerance given above carries no implications with respect to possible mechanisms. These can be divided into two classes. The first, designated *dispositional tolerance*, includes those changes in drug absorption, distribution, excretion and metabolism, which might lead to a reduction in the intensity and duration of contact between a given drug and the tissue on which it exerts its characteristic action ("target tissue"). The second includes those changes in the properties and functions of the target tissue which render it less sensitive to the same degree of exposure to the drug. This will be referred to as *functional tolerance*, in preference to the older term "tissue tolerance," because it makes no assumptions about the site of the underlying changes. This point will be considered in greater detail in section IV.

B. Measurement of Tolerance

1. VARIETIES OF TECHNIQUE. Since tolerance involves a change in the relation between the dose of a drug and the effect which it produces, any method suitable for measuring the acute effect should, in principle, be suitable for measuring the

acquisition of tolerance. In practice this is not always true, because tolerance does not necessarily develop at an equal rate to all the actions of a given drug. However, the assortment of useful methods is very large indeed.

Physiological measures have ranged from such gross indices as change in minimum lethal dose or LD50 (154, 163, 234, 300) to the very fine and sensitive changes in critical flicker frequency (157, 393). Spontaneous frequencies in the electroencephalogram (EEG) and response to photic stimulation (23, 93, 164, 395, 400) as well as the drug threshold and duration of electrical silence in the EEG (358, 383) have been used. Tolerance to various general depressants and anticonvulsants has been measured by the change in their effects on the thresholds for seizures induced by electroshock or pentylenetetrazole (3, 38, 39, 64, 115, 172, 362).

Perhaps the most commonly used indices for tolerance to depressant drugs are the time of induction and the duration of loss of righting reflex in animals (10, 163, 223, 234, 320), or duration of sleep in man (18). With somewhat lower doses which permit evaluation of drug effects on neuromuscular function, tolerance has been studied by means of various tests involving the ability of an animal to climb a pole (6), to remain on a tilted surface (7, 199) or a rotating narrow rod (73, 178), or to walk along a narrow elevated board (278, 310) without falling off. In man, the proprioceptive component of these tasks has been used separately, in the form of a quantified Romberg test of static ataxia (126).

For other classes of psychotropic drug, characterized by different physiological actions, other measures have been more appropriate in the study of tolerance. These have included thresholds for elicitation of tendon reflexes (315, 316), regulation of body temperature (316, 328, 368), food intake (369), pupillary diameter (315, 316), blood pressure and pulse rate (315, 316) and baroceptor reflexes (56, 219). There is no *a priori* reason why positional nystagmus, spinal reflexes of various types, or any other function on which a dose-dependent effect is demonstrable, could not be used for the same purpose.

Other behavioral tests of many different types have been used to measure the acute effects of drugs in man and in experimental animals. These may be classed roughly as tests of motor performance, sensory acuity, perception and discrimination, cognitive function, affect and mood. It is clearly beyond the scope of this review to consider these methods in detail; this has been done by various other reviewers (24, 117, 354, 382). One refinement which has been successfully employed recently is to study tolerance to one drug by observing the change in its effect on a behavior pattern induced by another drug, *e.g.*, tolerance to the effect of butyrophenone on amphetamine-induced stereotypy in mice (328). It is sufficient to point out that studies of tolerance have successfully employed many of these diverse tests both in man and in experimental animals. Specific examples are cited throughout the review.

2. ATTRIBUTES OF GOOD METHODS. In general the most useful methods are those yielding measures which: 1) are sensitive to small dose increments over the effective dose range of the drug; 2) are reproducible, and stable in the sense that they are subject to little uncontrolled variation either within or between observation periods; 3) permit identification and analysis of the components of the response

affected by the drug; and 4) are continuously variable rather than quantal or discrete.

Quantal or discrete responses are obtained in those tests in which only a single endpoint is possible. The rota-rod test of motor performance in rats or mice (73), for example, involves placing the animal on a thick rod which rotates about its longitudinal axis at constant speed. The test runs for a set time, and the animal is scored as either staying on the rod for the full time, or falling off. To establish a dose-response curve, therefore, it is necessary to have a sufficiently large number of animals at each dose to permit a statistically valid estimate of the proportion which fall off. A simpler version of the test consists of placing a rat at the middle of a fixed rod 1 m long and 1 cm thick, and observing whether it can successfully reach either end (278). The chimney-climbing test (22) and tests of lethality are other examples of quantal approach.

In contrast, a recent modification of the rota-rod test (178) consists of applying a constant acceleration to the motor which rotates the rod, so that each animal will eventually reach a point at which it falls off. That point can be measured on a continuous scale of time or speed of rotation. An analogous transformation can be made in the inclined plane test. As a test of muscle paralysis produced by meprobamate (199), it was used in quantal fashion by recording whether each animal was able to remain for 10 sec on a board set at a fixed tilt of 40° from horizontal. It was employed to give a continuously variable response (7) by tilting the board at constant angular velocity and measuring the angle at which the rat began to slide off. This type of measurement permits more precision, flexibility and economy of tests in establishing a dose-response relationship, and most recent workers have preferred such techniques.

Much valuable information, with respect to acute effects of ethanol as well as to tolerance, was obtained by Newman and his colleagues (283–287) by the use of what may be called a quasi-quantitative technique, which provided results in the form of apparently continuously variable behavioral scores. The observers became skilled at discriminating different degrees of motor impairment in dogs, ranging from slight unsteadiness of gait to complete inability to stand. They ranked these in order of increasing severity, assigned numerical ratings from 1 to 10, and used the numerals as scores to be plotted against alcohol dose or alcohol concentration in the blood. This procedure assumes that each interval in the rating scale represents an equal increment of impairment of the motor system, but this was never shown to be true.

Among the more widely used techniques which do employ genuinely continuous measurement scales are sleeping time (9, 74, 272, 360, 383), the inclined plane test (7, 185), tests of spontaneous (25, 128, 301) and forced locomotor activity (127), shuttlebox avoidance (33, 160, 256, 352) and operant bar-pressing on single (6, 36, 311, 312, 333) and multiple (331, 387) schedules of reinforcement.

3. BASIS OF SELECTION. With such a wide variety of techniques available, the selection of methods for a particular study of drug tolerance at times appears somewhat arbitrary. However, the range of appropriate choices may be narrowed by use of a few general principles.

The first is appropriateness of the dose range within which a particular test is

applicable. Measurement of LD50, for example, involves doses which are much above those used to develop tolerance to some drugs. Tolerance to opiates is of such magnitude that an escalating scale of dosage, as used to develop tolerance in behavioral effects, does indeed go far beyond the normal LD50 (163) so that increase in LD50 is also an appropriate index of tolerance. With ethanol, barbiturates and many other types of drug, however, the maximal degree of functional tolerance which can be attained is rather small (see below). Administration of single, near-lethal doses of certain barbiturates may cause irreversible neurological damage (216), and repeated daily administration of comparable doses of ethanol causes rapid deterioration in the general state of the animals (205). Chronic experiments therefore involve smaller doses. This may be one reason why very little increase in the minimum lethal dose of ethanol (234) or the LD50 of barbiturates (138) has been observed. An alternative explanation is suggested in section IV C.

A second factor to be considered is that some of the possible measurement procedures, such as those involving unit recording at various levels in the nervous system (79, 327), involve surgical intervention which makes it impossible to study the same animal more than once. Tolerance would then have to be assessed by comparing the measured effects of a drug in different groups of animals, before and after chronic administration. This increases greatly both the difficulty of the experiment and the variability of results.

Specificity of the phenomenon is another important consideration. Change of food or water intake during chronic administration and withdrawal of barbiturates (86, 161) or ethanol (340) is a non-specific indication of the subject's general state of well-being. There may be depression of food intake during the initial stage of drug action before tolerance has developed, and also during drug withdrawal. In contrast, food intake is relatively specifically diminished by amphetamine and related compounds (212), and hyperorexia occurs during withdrawal (214, 348). Therefore food intake would be a rather insensitive index for studying tolerance to depressants, but is quite appropriate for the amphetamines (26, 145, 212, 369).

A final consideration in the choice of methods is the purpose for which the study is intended. Relatively crude but simple methods, such as change in LD50, or rough scoring of ataxia, may be quite adequate for qualitative answers as to whether or not tolerance occurs. In contrast, a kinetic study of the rate of development of tolerance, especially during treatment with moderate doses, requires a technique which is objectively quantifiable and able to discriminate between the effects of small dose increments.

C. Identification of type

As noted in section II A, the measurement of tolerance may give little or no clue concerning its cause. To differentiate dispositional tolerance from functional tolerance, it is necessary to relate the effect to the concentration of the drug in the blood or brain, rather than to the administered dose. Earlier chemical methods of drug analysis required samples of a size which could be conveniently and

repeatedly obtained only from large animals. In smaller animals cardiac puncture or decapitation was needed, and for some drugs it was necessary to pool blood from several animals (156). This would obviously be a serious difficulty in any experiment in which the subjects required extensive training on a behavioral task to be used for the study of drug effects. It is not surprising, therefore, that relatively few of the older studies of drug tolerance included measurement of potent drugs in body fluids or tissues.

More modern approaches to drug analysis include gas chromatographic (139, 222), immunochemical (194), fluorescent (123, 220), enzymatic (41, 349), and other methods (386). Many of these are so sensitive and accurate that they can be performed repeatedly on a single rat during one period of drug action, or during the development of tolerance to ethanol (225) or pentobarbital (251). However, the difficulty in measuring such drugs as LSD and tetrahydrocannabinol in body fluids and tissues still impedes the study of tolerance to these agents.

D. Dispositional tolerance

1. ABSORPTION, DISTRIBUTION AND EXCRETION. (*a*) *Changes during chronic treatment.* The literature on ethanol in relation to this topic has been reviewed recently (184, 388, 389). It is now quite clear that ethanol can be absorbed through any mucosal or serosal surface by simple diffusion, and is rapidly distributed in the same manner throughout all body water, including the cerebrospinal fluid. Absorption is more rapid in the small intestine than in the stomach, so that delay in gastric emptying tends to slow the absorption. After chronic administration the rate of ethanol absorption from the gastrointestinal tract is normal or even increased, perhaps because adaptation to it results in less delay in gastric emptying (104, 105, 226, 283, 370). There is little doubt that ethanol tolerance does not result from reduced absorption. This appears to be equally true of isopropanol (228), and methanol (221).

The suggestion (248) that tolerance might arise through reduced entry of ethanol into the brain has been disproven by direct measurements, which have shown equal concentrations of ethanol in the brains of normal and tolerant animals after administration of equal doses (226, 233). The concentration of ethanol in the cerebrospinal fluid after a test dose rose slightly more rapidly in alcoholic patients than in non-alcoholics (104, 105), reflecting the more rapid rise in blood level, and confirming the absence of a diffusion barrier to alcohol even after chronic ingestion.

Similarly, tolerance cannot be explained by increased elimination of ethanol in the urine, breath and sweat. Elimination by these routes is also a process of simple physical diffusion (184, 388), and is therefore a function of the blood ethanol level. Excretion rates in tolerant subjects have been found to be either normal or reduced (287, 302). Reduction could be explained by an increased rate of metabolism of ethanol, with a corresponding reduction of the total area under the alcoholemia-time curve; this possibility is considered below.

The possibility of reduced absorption as a factor in the development of tolerance to barbiturates and other drugs does not appear to have been considered

seriously. Perhaps this indicates a greater degree of sophistication in the study of newer drugs, as a result of the experience gained in the earlier investigation of ethanol. The only available evidence is indirect. Butler *et al.* (31) found that in people with clear signs of tolerance to phenobarbital the blood levels conformed to the predicted values based on the assumption of complete absorption and first-order elimination. Since other evidence (see below) shows that elimination of unmetabolizable barbiturates does not change during chronic administration, by inference the absorption must also be normal.

The levels of ^{14}C-activity in brain, plasma and urine at various times after injection of ^{14}C-barbital were found to be unaltered in rats which had become tolerant as a result of daily injection of increasing doses of barbital over a period of 13 to 36 days (74). This is the only reasonably complete study reported. However, the results reported by two other groups raise a question about its applicability to all barbiturates. Timar *et al.* (367) also administered ^{14}C-barbital to control rats and to others treated with barbital or phenobarbital for 8 or 15 days. Although their results are not presented with adequate detail about numbers and variability, they suggest a lower brain:blood ratio of ^{14}C activity in the chronically treated animals than in the controls, 24 hr after the injection.

A similar relationship is suggested by the mean concentrations of pentobarbital in plasma and brain, 20 min after injection of a 50 mg/kg dose (29). The brain:serum ratio appeared to be reduced by about one-third in animals pretreated with phenobarbital for 4 days (29). Admittedly this suggestion is based on the ratio of group mean values for blood and brain, rather than the mean of ratios in individual animals. However, when the variance is small, as in the study cited (29), the ratio of the means approaches the mean ratio fairly closely. Therefore the possibility of altered distribution cannot be dismissed out of hand.

One suggestion has been made (215) that increased tolerance to barbital in a single dog was due, at least in part, to an increase in the rate of urinary excretion of unchanged barbital. This claim was based on the finding that barbital excretion continued for some 10 days following temporary interruption of administration early in the experiment, but continued for only 3 days when administration was finally stopped after 7 months. However, the total barbital output during the 3 days was much too small to account for the drug administered even on the last day, and there appeared to be a marked oliguria during the week after withdrawal. Since no barbital metabolites were found, and the administered drug could not be accounted for, it is unwarranted to conclude that excretion was more rapid.

A similar disagreement is noted in the few studies of distribution of minor tranquilizers in tolerant animals. Rats given daily injections of meprobamate for periods varying from 1 to 20 days showed increasing degrees of tolerance with increasing length of treatment, but the brain:serum ratio (*i.e.*, the ratio of mean values for brain and serum) of meprobamate concentration remained virtually constant (198). In contrast the data of Hoogland *et al.* (155) on the distribution of chlordiazepoxide-2-^{14}C in the tissues of tolerant and non-tolerant rats again suggest that the brain:plasma ratio is consistently about one-third

lower in the tolerant animals throughout a 4-hr period after injection of the test dose.

We have found only one study (345) of tissue distribution of amphetamine, at 1 and 4 hr after injection of *dl*-^{3}H-amphetamine into tolerant and non-tolerant cats. The data are insufficient to provide a valid statistical estimate of variability, but there does not appear to be any difference between the two groups.

No distribution studies on phenothiazines, antidepressants, tetrahydrocannabinol (THC), LSD, mescaline, or other hallucinogenic substances appear to have been carried out so far in tolerant subjects. In view of the prolonged retention of chlorpromazine, THC, and some of the other substances in the body at the same time that tolerance occurs, it is perhaps surprising that such studies have not been done.

(*b*) *Possible significance of altered distribution.* As noted above, the evidence for a reduced brain:blood or brain:plasma ratio of drug distribution as a concomitant of tolerance to barbiturates and chlordiazepoxide is scant and at best suggestive. Nevertheless it should not be overlooked. It is conceivable that chronic administration of these drugs can induce the synthesis not only of the hepatic microsomal enzymes which metabolize them, but also of non-specific binding sites including plasma proteins. Similar suggestions were made many years ago by Ehrlich (329) and Santesson (323) and more recently in the form of postulated "silent receptors" (44).

A given plasma concentration of the drug might then correspond to a lower concentration of free diffusible drug in the tolerant than the non-tolerant subject. Although this seems hardly likely to be a major factor in the development of tolerance, it underlines the importance of measuring drug levels in the brain whenever possible in the study of tolerance.

2. METABOLISM. A number of excellent reviews on the metabolism of ethanol have appeared in the past few years (236, 240, 241, 325, 380, 389), to which the reader is referred for detailed information. Our concern here is only with the question of the role which changes in metabolism might play in the production of tolerance.

(*a*) *Rate of metabolism.* An increase in the rate of *ethanol* metabolism as a result of prolonged administration has been reported by many investigators, while many others have found no change or a decrease (for references, see 149, 380, 389). Controversy has continued, but recent work (196, 207, 237, 379) has tended to confirm the view that chronic ingestion of ethanol leads to an increase in the rate of its metabolism unless overt liver disease supervenes. It seems likely, therefore, that disagreement about an increase in rate of ethanol metabolism during chronic exposure results from differences in diet and schedule of alcohol administration, with consequent differences in hepatic biochemistry (205).

Since the alcohol dehydrogenase activity of the liver cytoplasm has long been considered the major determinant of the rate of ethanol metabolism, it is not surprising that the same disagreement exists about the effect of chronic ethanol treatment on alcohol dehydrogenase activity as on the rate of ethanol metabolism (149, 276, 381). The reasons for the disagreement may be the same as

noted above. An alternative explanation is suggested by the recent finding (379) that dinitrophenol increases the rate of ethanol metabolism in slices of normal rat liver, but does not enhance further the increased rate found in slices from rats made tolerant to ethanol. In the latter, the increase in rate of metabolism may result, at least in part, from an increase in the rate of $NADH_2$ oxidation by liver mitochondria as the final stage of the pathway initiated by alcohol dehydrogenase. However, our concern at this point is with extent, rather than with the mechanism of increase.

The metabolism of a wide range of other psychoactive agents, including *barbiturates, other hypnotics and sedatives, major and minor tranquilizers and antihistaminics*, is known to be capable of rapid and substantial increase by induction of hepatic microsomal drug-metabolising systems. Excellent reviews of this subject have appeared within the last few years (30, 47, 307), and the reader is referred to these for details. It is sufficient for the purposes of the present review to recall only the main conclusions.

The duration of action of hexobarbital, pentobarbital, amobarbital and other intermediate- or short-acting barbiturates, glutethimide, meprobamate, chlordiazepoxide and various other drugs is determined mainly by the rate at which they are metabolized by the hepatic microsomal mixed-function oxidase system. On repeated administration, many of these drugs act as inducers of the microsomal system, and so increase the rate of their own metabolism. In addition, since this microsomal system has a low order of substrate specificity, its induction by one drug enhances the rate of metabolism of many other drugs, giving rise to metabolic cross-tolerance. Among the effective inducers of microsomal drug metabolism are substances which may form an unintentional and unrecognized part of the experimental treatment, such as terpenes contained in soft-wood shavings used for bedding the experimental animals (101, 375) or pesticide residues in food (146–148). The quantitative importance of such metabolic tolerance or cross-tolerance varies with drug, species, sex, age, and experimental conditions. It is important to note that the occurrence of this form of tolerance does not preclude the simultaneous development of functional tolerance (198, 338).

Some drugs undergo fairly complex sequences of metabolic conversion, effected by enzyme systems which respond differently to chronic administration of the drugs. For example, chlorpromazine gives rise to at least eight metabolites in the rat (82) but only one of these (chlorpromazine sulfoxide primary amine) is formed in increased amount in the tolerant animal. This is offset by a reduced excretion of the sulfoxide. The total urinary excretion of chlorpromazine and its metabolites is therefore unchanged and the fecal excretion is actually reduced in the tolerant animal. Thus, even though chlorpromazine is an effective inducer of hepatic microsomal metabolism of other drugs (200, 201, 391), and its metabolism is induced by phenobarbital pretreatment (317), it does not cause significant induction of its own.

Tolerance to LSD-25 is developed extremely rapidly, three or four consecutive daily doses being enough to abolish all response (40, 113, 398). There are no data available yet, to indicate whether increased rate of metabolism plays any role

in this process. The fact that LSD-tolerant subjects are cross-tolerant to mescaline (14, 275, 398), psilocybin (166), N,N-dimethyltryptamine (315), and *p*-bromomethamphetamine (209), but not to amphetamine (316) suggests that the tolerance is related to the mechanism of action rather than to drug metabolism.

Repeated daily administration of rather large doses of tetrahydrocannabinol has recently been shown to give rise to tolerance at different rates in rats, mice (21, 279) and pigeons (262, 263). Identification of the nature of this tolerance is not yet possible, for reasons to be discussed later. However, one suggestion of a metabolic component arises from the observation that the injection of SKF-525A into a dog which had developed tolerance resulted in restoration of the effects of Δ^1-THC (66). Since this was a single instance, so far unconfirmed, it cannot be considered more than a suggestion. Moreover, prevention of drug metabolism should at least partially overcome functional tolerance as well as metabolic tolerance, by raising the effective drug concentration.

Although there has been considerable research devoted to the metabolic pathways of amphetamine and related compounds in man and other species, no study appears to have been made of possible changes in their metabolism in relation to amphetamine tolerance. SKF-525A given 40 to 45 min before amphetamine has been found to decrease the urinary excretion of *p*-hydroxyamphetamine in the rat by about 90% (51), and of total metabolites by about 20% (19). Pretreatment with phenobarbital for 5 to 6 days, ending 24 hr before the amphetamine was given, was reported to cause a significant increase in the excretion of metabolites (19, 235) but two other groups (51, 137) found no increase after 3 to 4 days' pretreatment with either phenobarbitone or benzo[a]pyrene. This discrepancy has not yet been explained, and leaves uncertain the question of whether amphetamine might act as an inducer of its own metabolism, and thus contribute to the development of tolerance.

(*b*) *Significance of increased rate of metabolism.* In most of the studies cited above, in which metabolic tolerance has been demonstrated, its extent has been rather small. For example, the maximum increases in rate of metabolism of ethanol (197, 237, 379) and of meprobamate (199) are of the order of 60 to 100% of the control rate. Even the largest increases in rate of microsomal metabolism of barbiturates *in vitro*, which do not necessarily parallel exactly the rate of metabolism *in vivo*, are about 100 to 200% of the control rate in male rats (308, 355), and dogs (309).

The effect of such increases on the apparent drug effect *in vivo* depends on how the effect is measured. For example, assuming a 60 to 100% increase in the rate of ethanol metabolism, the concentration of ethanol in the blood after a given dose in tolerant subjects would be expected to return to zero in one-half to two-thirds the time required in normals. Other things being equal, the total duration of exposure of the nervous system, and of the period of intoxication, should be shortened accordingly. However, the typical blood alcohol curve in the fasting subject (184, 388) shows its maximum concentration at 30 to 60 min after oral ingestion of a small or moderate dose, and up to 120 min or later after a large dose. In the rat, metabolizing ethanol at a normal rate of about 275 mg/

kg/hr, a 100% increase would mean a rate of about 0.5 g/kg/hr, so that one-fourth of a 2 g/kg dose could be metabolized by the time the peak level would normally be reached. This should result in a slightly lower and earlier peak level than in the previously untreated animal, and could thus contribute to a slight reduction in observed behavioral effects. The larger the dose, the lower would be the proportional reduction in peak height.

After intraperitoneal injection of 2 g/kg or less, the peak blood alcohol level is reached within minutes, and the maximum behavioral impairment on the treadmill test appears at 10 to 15 min (225). Under these conditions, increased metabolism would make no appreciable difference to the maximum effect, since even a 100% increase would mean that not more than 0.1 g/kg could be metabolized in that time.

Similar considerations apply to the actions of barbiturates, meprobamate, chlordiazepoxide, and other drugs for which metabolic tolerance is demonstrable. This is especially true in experimental animals, to which the drugs are most commonly given by intraperitoneal or intravenous injection. Therefore metabolic tolerance cannot explain increases in the time taken for loss of righting reflex in tolerant animals, or the failure of some of them to lose the reflex at all in a dose which is effective in controls (56, 135, 138, 189), since onset of "sleep" normally occurs within 5 min of drug administration. The same consideration applies to other drug effects which are measured within minutes of drug administration (225, 358, 383). In contrast, shortening of the duration of "sleep" is quite consistent with increased drug metabolism (308, 355).

In summary, increased rate of drug metabolism could possibly account for a slight reduction in the maximum pharmacological effect measured after oral administration of small doses, but would have negligible influence on the early effects found after large doses or after parenteral administration. On the other hand, it would be expected to contribute significantly to a shortening of the duration of drug effect.

(*c*) *Changes in pathways of metabolism.* Possible changes in the pathways of ethanol metabolism in tolerant individuals are of interest for another reason. It has long been known that ethanol-tolerant subjects also show increased tolerance to barbiturates, volatile anesthetics and a variety of other drugs with more or less similar actions. Recently it has been proposed that this cross-tolerance may result from the ability of hepatic microsomal enzyme systems to metabolize both ethanol and the other drugs in question (237, 238, 291, 318), and to be induced by chronic treatment with either ethanol or these other drugs. In support of this view, it has been reported that not only pentobarbital and meprobamate (274), but also tolbutamide and various other drugs (197) are metabolized more rapidly after chronic intake of ethanol.

However, several other groups (170, 206, 313) have presented evidence suggesting that the metabolism of ethanol by liver microsomes is an artifactual result of tissue homogenization and does not occur *in vivo*. Other reports are consistent with this suggestion: inhibitors and inducers of drug metabolism were found to have no effect on the rate of ethanol disappearance *in vivo* (204, 208,

364), and chronic ethanol pretreatment did not affect pentobarbital metabolism *in vivo* or by liver slices (189). Destruction of the hepatic microsomal ethanol-oxidizing system by a single dose of carbon tetrachloride which left the alcohol dehydrogenase activity intact, had no effect on *in vivo* ethanol metabolism in the rat (207).

The apparent contradiction may be explained in part by the fact that one report of metabolic cross-tolerance (197) was based on a study of alcoholic patients, rather than subjects to whom alcohol was given experimentally. A substantial proportion of alcoholics are also heavy users of barbiturates and other drugs (65), some of which may be very effective inducers of hepatic microsomal systems involved in drug metabolism (47).

Another possible explanation may be that the microsomal mixed function oxidase (MMFO) system may not be the rate-limiting step in the metabolism of some of the drugs which have been tested. Tolbutamide, for example, is converted to hydroxytolbutamide, which is further oxidized by hepatic alcohol dehydrogenase (258). Since the latter can be increased by chronic administration of ethanol (149, 196, 205, 237, 379) it might account for the faster disappearance of tolbutamide (197). Similar considerations might apply to pentobarbital and meprobamate.

Moreover, the fact that the rate of metabolism of these drugs *in vivo* is reduced by the simultaneous administration of ethanol (274) cannot be taken as evidence of competition for the MMFO system. Ethanol was found to inhibit the hydroxylation of amphetamine acutely *in vivo* (51), but the disagreement about induction of amphetamine hydroxylation by phenobarbital, noted above, and the failure to demonstrate this hydroxylation with hepatic microsomes *in vitro* (69), raise doubts that the ethanol effect is exerted on the microsomal system.

In summary the bulk of evidence presently available suggests that ethanol is not metabolized to a significant extent by liver microsomal enzyme systems, and that mutual induction of this system is probably not an important factor in cross-tolerance between ethanol and other psychoactive drugs.

E. Functional tolerance

1. INTRODUCTION. Most of the extant literature makes a distinction between acute and chronic tolerance. This suggests that, regardless of mechanism, tolerance can be developed at different rates. Indeed, under appropriate conditions tolerance can occur within hours or even minutes.

The term *acute* has at least two possible meanings. One concept of acute tolerance is that which occurs *after* one exposure to the drug. This is usually determined by two tests on the same group of animals, or by group comparisons before and after one treatment. Acute tolerance is also used to indicate changes in sensitivity to a drug *within* the duration of one continuous drug exposure. The two meanings perhaps can be considered conceptually distinct. The former is clearly on a continuum of tolerance after n exposures to the drug, where n can range from 1 to infinity. The fundamental distinction to be made, then, is between intrasessional adaptation (acute tolerance) and intersessional adaptation

(chronic tolerance). The empirical and mechanistic relationship between them must be investigated in greater depth.

The methods of study of functional tolerance are quite similar in the case of acute and chronic tolerance. Since the study of acute tolerance requires that at least two comparisons between drug concentration and effect be made within a single drug session, it is more difficult to do than parallel chronic studies. Usually in the latter type, a single comparison between drug concentration and effect is made either at the same point of maximum effect or at the same fixed time after drug administration. The data to be discussed below suggest that much could be learned by applying the acute study approach to chronic study designs.

2. ACUTE TOLERANCE. Mellanby (266) was the first to report that the degree of impairment was greater at a given ethanol concentration in the rising portion of the blood alcohol curve than at the same concentration in the descending part of the curve. This has been confirmed repeatedly, with a wide range of physiological and behavioral measures of alcohol effect (8, 35, 126, 357). Harger and Forney (144) have pointed out that Mellanby measured ethanol levels in venous blood from the arm. During the rising phase of the alcohol curve, *i.e.*, while absorption and distribution are still proceeding, skin and muscle equilibrate with the blood much less rapidly than the brain does (for references, see 184). The venous blood in the limbs at this stage has a lower alcohol concentration than arterial blood or brain. They believe, therefore, that the "Mellanby phenomenon" results from this imbalance of alcohol distribution rather than from acute tolerance.

The same objection could also be raised against the observations (34, 76, 187, 242, 284) that a greater degree of functional disturbance is produced at a given blood ethanol concentration when that concentration is reached rapidly, than when it is reached slowly. By infusing the same total dose of ethanol intravenously at different rates, Gostomzyk *et al.* (133) showed that the arteriovenous concentration difference in the limbs is greater, the more rapid the infusion.

An acute tolerance to glutethimide has been reported (55) on the basis of evidence similar to that originally advanced by Mellanby in relation to ethanol. Smooth tracking eye movements returned at a drug concentration in venous blood which was higher than that at which they had been suppressed. With a small dose, the effect was small or even absent by the time the peak blood concentration was reached. The same objections raised by Harger and Forney (144) about the early work with ethanol apply equally to this study. However, the drug was given by mouth and was probably absorbed relatively slowly. In view of the alcohol distribution studies by Gostomzyk *et al.* (133), it seems unlikely that there was a substantial difference between concentrations in brain and in venous blood under these conditions, but it is impossible to be certain.

However, this criticism does not apply to certain other experiments. Ethanol-induced diuresis, resulting from inhibition of the secretion of vasopressin, occurred only while the concentration of ethanol in the blood was rising, and disappeared when the level was kept high but steady by repeated small doses (77). Similarly, Mirsky *et al.* (273) made repeated intravenous injections of ethanol

into hepatectomized rabbits, in which metabolism was virtually abolished, so that the blood alcohol level could be kept practically constant for a long time. They found that ear-drop and nystagmus gradually disappeared despite the continuing alcohol level. A new injection produced a new plateau, with return of the signs of intoxication followed by gradual disappearance again, despite blood alcohol concentrations of up to 400 mg/100 ml. Heidelmann *et al.* (150) have reported that the cutaneous vasodilatation produced by ethanol is also seen only during the rising part of the blood alcohol curve.

Evidence of acute tolerance to paraldehyde, thiopental, pentobarbital, and trichlorethanol as well as to ethanol was found by Maynert and Klingman (257). They administered 4 or 5 different dose levels of each drug to groups of dogs, in a random order, at intervals of 1 to 2 weeks. The plasma concentration of the drug was measured in jugular venous blood; at least in the case of ethanol, this is known to come rapidly into equilibrium with the brain (184). The results for all 5 drugs were consistent in showing that as the initial dose was increased, the plasma concentration at the time of disappearance of ataxia increased significantly until a dose producing clinical anesthesia (5 median ataxic doses) was reached. Above this level further increases in dose were not accompanied by increases in plasma concentration at the termination of drug effect. The data indicated that the observed tolerance developed quite rapidly. For some of the drugs a significant increase in tolerance resulted from an elevation in dose which increased the total duration of measured effect by only 5 or 10 min. In addition, it was found that the maximum amount of tolerance which could be produced acutely to trichlorethanol developed within 40 min and that a substantial increase in the duration of drug effect by administration of supplementary doses did not result in a higher level of tolerance than when a single large dose was given.

Comparable results were obtained by Brodie *et al.* (27), and Dundee *et al.* (72) in studies of acute tolerance to thiopental in man. When the drug was administered for short periods there was a positive correlation between size of induction dose and plasma level of thiopental at the time of awakening. As noted by Maynert and Klingman (257) these findings seem consistent with their own and with the suggestion that acute adaptation to the effects of the drug develops rapidly and in direct proportion either to the peak concentration reached in the brain or to the intensity of the depression caused by the drug. They quite correctly add, however, that existing data do not permit one to choose between these two possibilities. Future investigations of this problem will presumably include in their design some provision for distinguishing between the effects of intensity and duration of exposure of the target tissue in the development of acute tolerance. This question is discussed further in relation to chronic tolerance (section II E 3c).

Loomis and West (239), by studying the effect of ethanol on human performance in a simulated driving test, concluded that there was no evidence of a "Mellanby phenomenon" in relation to the objective measures of performance, but that the subjective effects in some cases were perceived to be less intense

after 3 to 5 hr at a maintained blood alcohol level. However, examination of their data indicates that the blood alcohol level was relatively stable in 36 of 40 separate trials, and in 14 of these (such as in their fig. 1), there was some indication of improved performance; this is practically the same proportion as that reporting subjective improvement. Difficulties arise in the interpretation of human studies of this type because it is almost impossible to control the level of motivation. As the authors point out, the measured performance is subject to the influence of sleepiness and loss of interest, which are common after-effects of alcohol. When the subjects know that errors of performance in a simulated task cannot harm them, it is difficult to be certain how much of the later effect represents the direct action of the drug.

Subjective ratings are possibly less affected by changes in motivation, and it is therefore interesting that data based on such ratings also indicate the development of acute tolerance to ethanol (127). Intensity of subjective effects at the time of the maximum blood alcohol concentration was plotted against that concentration for each subject, in separate experiments involving low and high doses of ethanol. The regression line relating peak concentration to effect was different in the two experiments; less effect was noted at the same blood alcohol level in the high-dose experiment. Since more time was required to reach maximum level in the high-dose experiment, the findings are consistent with the idea that there was more opportunity for acute tolerance to develop under these conditions.

The most direct proof of acute tolerance, however, is provided by correlation of effects with actual concentration in the brain. Rats were trained to perform the treadmill test (121) and were then tested at various times after the injection of various doses of ethanol. Each animal was given a single 2-min test under ethanol, and immediately afterward was decapitated and the brain removed for analysis of ethanol concentration. Animals tested during the first 10 min after injection showed significantly worse performance at a given brain alcohol level than those tested at 30 or 60 min (226). The data are strongly indicative of the rapid development of acute tolerance.

It has been reported very recently (385) that acute tolerance to ethanol in the rat confers acute tolerance to hexobarbital at the same time. The threshold dose of hexobarbital required to produce 1-sec suppression of EEG bursts was reduced by ethanol, the extent of the reduction in threshold being directly proportional to the concentration of ethanol in the blood. But the reduction in threshold was consistently greater at a given alcohol level on the descending limb of the blood ethanol curve than at the same level on the ascending limb. Further, the duration of sleep produced by the threshold dose was considerably greater in the latter case. The significance of this observation will be discussed in section IV C.

In an experiment purporting to investigate acute tolerance to LSD-25, Freedman *et al.* (113) compared the lever-pressing performance of three groups of rats on a fixed ratio schedule of reinforcement during a 32-min test period following: 1) the last of three 130 μg/kg doses, given 1 hr apart; 2) a single dose of 130 μg/kg; and 3) a single dose of 390 μg/kg. The measure of drug effect employed

was the number of bar presses during the test period, expressed as a percentage of the mean number during the four saline control sessions preceding the first dose of LSD-25. Analysis of the data revealed that animals in the first group were significantly less impaired than those in the third, but did not differ significantly from those in the second. Given that the half-life of LSD-25 in the rat is about 80 min (25), a substantial proportion of the first two doses administered to group 1 would probably have been cleared from the body at the time of the test. Consequently the results do not warrant the authors' contention that they show partial tolerance and that this can develop rapidly.

We are not aware of any other published experiments directed explicitly toward an examination of the development of acute tolerance to the behavioral effects of other hallucinogens, cannabis, stimulants of the amphetamine type, or major or minor tranquilizers.

3. CHRONIC TOLERANCE. (*a*) *Speed of production.* It is commonly believed, especially in clinical circles, that tolerance develops only after long exposure to ethanol, barbiturates, or other drugs. This idea was perhaps most clearly enunciated by Jellinek (174) in relation to ethanol, but is implicit in the design of many experimental studies of tolerance to ethanol, barbiturates and other drugs (25, 157, 286, 301) in which measurements were made only at long intervals or after long periods of treatment. It is therefore of interest to note the times actually required.

Tolerance to ethanol has been observed after 2 to 3 weeks in people (1, 164, 268, 395), dogs (245), rabbits (2) and rats (225, 390). Still others have found it to develop within a few days (153), while one group (78) reported it after only one day, but did not present the actual experimental data on which the claim is based.

Tolerance to so-called short- and intermediate-acting barbiturates such as pentobarbital and amobarbital has been observed to begin in dogs in from 2 to 7 days (56, 97, 135, 138, 361), and in cats (171) and rabbits (103, 124, 138, 254) in about the same period. It has been quite consistently reported to occur after 1 to 5 days in rats (9, 10, 138, 277, 288, 383) and mice (156, 319, 320). In man it has been found to begin in 3 days, reaching a maximum in from 5 to 8.5 days (18). Tolerance to long-acting barbiturates (barbital and phenobarbital) has been noted in mice in from 4 to 14 days (115) and in rats (383), people (157) and dogs (215) after 35 to 60 days. In man, EEG signs of tolerance to meprobamate were noted after 1 week of treatment (63) and increased up to the 3rd week (23). However, in most of these studies tolerance was demonstrated only as a reduction in sleeping time or in intensity of effects, and it is impossible to decide whether increased drug metabolism or nervous system adaptation was involved.

The matter is easily settled, however. Actual measurements have shown higher drug concentrations in brain or blood for a given degree of effect, in tolerant subjects as compared to controls (31, 74, 115, 156, 198, 207a, 225, 310, 400).

A diminished behavioral response to chlorpromazine has been produced in rats and dogs in 14 to 15 days (159, 255, 387) and mice exhibit tolerance to behavioral

and physiological effects of promazine after 1 to 4 weeks of chronic administration (218, 368). Since metabolic tolerance does not appear to play a significant role with these drugs, the times presumably correspond to those required for central adaptive processes.

Reports concerning tolerance to amphetamines are quite contradictory. Some investigators have found no tolerance to the effects on locomotor activity in mice (265, 369), while others have found a disappearance of this effect with methamphetamine in about 10 days (373). In the rat, tolerance to the effects of *d*-amphetamine on operant behavior was found to develop in 7 to 30 days (331, 333), and on the anorexigenic effect in roughly the same time (145, 369) but not to the effects on spontaneous motility (333, 369). In man, tolerance to the subjective effects on mood and appetite has been detected in less than 2 weeks of daily administration (316).

Tolerance to LSD-25 develops rapidly in man and other species. It has been observed in rats after 5 to 7 daily administrations (6, 113, 352), in rabbits after 4 days (125) and in man in from 3 to 13 days (40, 165, 166). Similarily, rats exhibit tolerance to bromolysergic acid, mescaline and psilocybin in from 2 to 7 days (6, 352) and people show a diminished response to psilocybin in from 1 to 2 weeks (166).

Despite the range of times noted, it is clear that tolerance can develop rapidly. However, other drugs with actions closely similar to some of those mentioned above may give rise to no tolerance at all. Perhaps the most striking instance is cocaine. Early studies (70, 71, 363) indicated that daily administration of cocaine to man, monkey, dog or rat resulted in an increase rather than a decrease in sensitivity to cocaine and to ephedrine. This has been confirmed by later investigations (140), and is in agreement with the reported lack of tolerance to the effects of cocaine on forced swimming activity or on a discrimination task motivated by thirst (212).

It has also been reported that no tolerance develops to the effects of phenmetrazine or of methylphenidate (265) on spontaneous behavior in mice, yet this appears to be contradicted by the numerous clinical reports of large increases in self-administered dosage, with daily ingestion of as much as 1750 mg of phenmetrazine or 200 mg of methylphenidate (191). So far no satisfactory explanation of this discrepancy has been proposed. As noted above for amphetamine, tolerance does not develop equally for all effects. Perhaps the effect of these drugs on spontaneous movement in the rodent is an inappropriate index of those drug actions underlying self-administration by man. This point will be considered further in relation to models of tolerance and dependence.

(*b*) *Extent of tolerance.* The degree of tolerance produced by chronic drug treatment varies not only with the method of investigation, but even more strikingly with the type of drug used. It is much less for ethanol, barbiturates, other sedatives and tranquilizers than for opiates. In contrast, tolerance to amphetamines, LSD and THC can be very great, and the subject may become quite refractory. This probably reflects differences in the basic mechanisms of tolerance and should give rise to a different type of alteration in the dose-response curve (fig.

1). Even if, as in the case of ethanol and pentobarbital, there is a parallel shift of the dose-effect curve to the higher end of the dose scale (10, 225), there is some difficulty in assessing the degree of tolerance. This can be expressed either as the vertical separation of the two regression lines (*i.e.*, the difference in effect at a given dose), or the horizontal separation (*i.e.*, the difference in dose required to produce a given effect). The percentage change can vary considerably, depending on which method of comparison and what points on the curves are selected (fig. 1).

On theoretical grounds, when dealing with a horizontal displacement of the curve one should express tolerance in terms of horizontal separation at a fixed

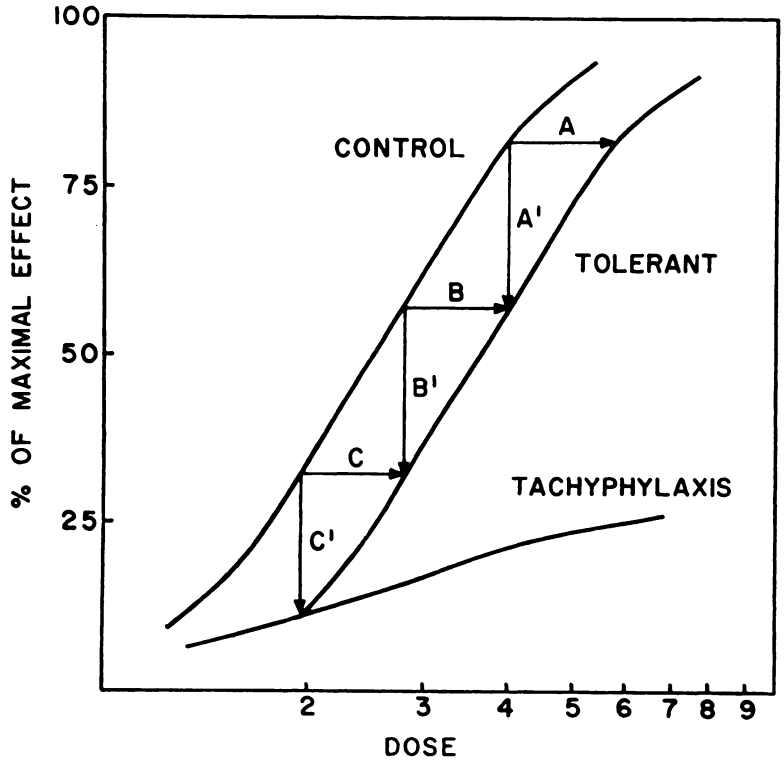

FIG. 1. Diagrammatic illustration of possible changes in dose-response curves associated with tolerance. The lower curve ("tachyphylaxis") is consistent with depletion of a response mechanism. The middle curve ("tolerant") indicates a parallel shift of the dose-response curve consistent with *functional tolerance*. If tolerance is expressed as the increase in dose required to produce a given effect, then the horizontal shift as measured at A, B and C is the same, about 0.16 log units in each case. When tolerance is expressed as decrease in effect at a given dose, this corresponds to reductions of 30%, 44% and 65% at A', B' and C' respectively. A single measurement of the reduction at C', coinciding with the point of intersection of the "tolerant" and "tachyphylaxis" curves, would obviously give no indication that two fundamentally different mechanisms might be involved. The use of a single test dose at an unknown point in the dose-response curve may explain many of the apparent differences in tolerance to the same drug found by different investigators.

reference point on the response axis. In many studies, however, the shape of the dose-response curve is unknown. With some processes the curves are clearly not simple in either shape or sign (67, 186, 261, 397). In such cases the assessment of degree of tolerance is rendered more difficult. If dose-response curves are not known or given, the only figure that can be extracted from the data is the change of effect at a given dose. It should be recognized, however, that the change may be markedly influenced by the initial value of baseline measures.

In the case of ethanol, only two studies (225, 286) give complete dose-response curves. In both, the increase in dose required for half-maximal effect was of the order of 30 to 50%. One full dose-response study with pentobarbital (10), indicated increases of about 40% for female rats and 12% for males. For other drugs, there is a remarkable deficiency of such complete studies. The types of data available are illustrated in table 1.

(*c*) *Effect of drug dose.* Drug dose can affect both the degree of tolerance and the speed of its production. Most studies of tolerance have been designed primarily to demonstrate only its presence or absence; parametric studies of the influence of drug dose are clearly needed. An important variable in such studies is the route of administration. The level in the blood or brain produced by a given dose of drug varies considerably with different routes (17, 50, 184). Because of secondary effects on absorption and distribution, the level in the brain may not be strictly proportional to the dose. For example the blood ethanol curve following gastric administration of the same dose of ethanol differs substantially depending on the concentration of the ethanol solution, because of effects on gastric emptying (187).

Study of the effect of dose on rate of tolerance development is complicated by the time of exposure, since a larger dose almost inevitably increases both the maximum concentration and the effective duration of drug exposure. It is difficult to design an experiment to separate these factors, if the drug is not one to which a specific antagonist exists. One possible approach might be to vary the duration of effect for a given dose by means of hemodialysis, forced diuresis, change of urinary pH, or induction of drug metabolism by agents devoid of action on the nervous system. If successful, this would permit a quantitative assessment of the relative contributions of dose and duration.

Partial results in a single species make it clear that tolerance to ethanol develops more rapidly with higher doses. Rats trained on the treadmill test developed maximum tolerance within 19 to 22 days, on a schedule of increasing daily dosage rising gradually from 3 to 9 g/kg during the whole period (225). Other rats performing the same test developed the same maximum tolerance in 13 to 16 days when given 6 g/kg/day from the outset (223).

Aston (9) has examined the relationship between dose of pentobarbital and magnitude of tolerance development. Groups of rats were injected with one of five different dose levels of the drug ranging from 25 to 45 mg/kg, and 24 hr later were given a second dose of 40 mg/kg. All five groups showed a significantly shorter mean sleeping time on the second day than that of a group given 40 mg/kg without pretreatment. Moreover, the degree of tolerance was found to be directly proportional to the size of the tolerance-inducing (first) dose.

TABLE 1

Development of tolerance to psychotropic drugs in various species

Drug	Species	Time of Measurement	Extent of Tolerance	Reference
			%	
Barbiturate	Dog	6 weeks	43–57	215
	Dog	2 days	50	97
	Dog	4–7 days	60–100	135
	Dog	5–7 days	43–64	138
	Guinea pig	4 weeks	60	32
	Mouse	5–6 days	50	156
	Mouse	5 days	23–27	320
	Mouse	4–14 days	150–327*	115
	Rabbit	2–3 days	50	103
	Rabbit	4–7 days	27–35	254
	Rabbit	3–5 days	36–47	138
	Rabbit	3 days	67	124
	Rat	2–3 days	100	288
	Rat	2 days	43–50	277
	Rat	5 days	15–57	138
	Rat	1–3 days	11–40	9
	Rat	1 day	26–55	10
	Rat	4 days	21–22	383
	Rat	35 days	46	383
	Man	80 days	"Virtually complete"	165
	Man	30–70 days	100	211
Major tranquilizers	Dog	15 days	53	387
	Mouse	2–6 days	60	218
	Mouse	1–4 weeks	60–81	368
	Rat	12 weeks	62	25
	Rat	14 days	"Almost complete"	159
	Rat	15 days	80	256
Minor tranquilizers	Cat	21–50 days	60	401
	Rat	35 days	44	301
	Rat	5 days	—	155
	Rat	14 days	42–71	128
	Rat	9–35 days	58–67	255
	Mouse	6–11 days	50–75	38, 39
Stimulants	Rat	1–21 days	19	333
	Rat	13–30 days	75	331
	Man	14 days	Complete	316
Hallucinogens	Rabbit	4 days	54	125
	Rat	7–8 days	100	113
	Rat	7 days	83	352
	Rat	2–7 days	70–100	6
	Man	7–21 days	0–87	162
	Man	6–13 days	Complete	166
	Man	14 days	33–100	315, 316

* In this experiment, tolerance was determined as the change in drug dose or concentration of drug in the serum which provided 50% protection against a standard electroconvulsive stimulus. In all other cases cited, tolerance is given as the reduction in effect produced by a given dose, so that the maximum tolerance cannot exceed 100%.

In experiments involving the hypothermic action of promazine the size of dose used in chronic treatment was found to be directly related to both the degree (218) and rate (368) of development of tolerance. We are unaware of any systematic exploration of the relationship between size of dose and development of tolerance to other drugs. However, there is an as yet unexplained observation that tolerance to meprobamate developed in patients receiving less than 60 mg/kg daily, but not in those receiving 80 mg/kg or more (23). Since blood levels of meprobamate were not measured, it is possible that the latter group had cumulative build-up of drug concentration which masked the development of tolerance.

(*d*) *Duration and carry-over.* It is important to know not only the degree of change of sensitivity to a drug, but also the duration of that change. If tolerance endures from one drug exposure to the next, it should be demonstrable at the earliest possible measurements during succeeding drug exposures. By the use of techniques which demonstrate effects promptly after drug administration (such as time taken to lose the righting reflex, or earliest effect on the treadmill test) it is indeed possible to detect tolerance at the earliest times measured within a session (135, 138, 225). This suggests that tolerance does endure between sessions. Although the rate of disappearance of tolerance has been studied (11, 225) the factors influencing its duration have not been explored systematically. Ideally the rate of decline of tolerance should be examined as a function of speed of acquisition, level of tolerance attained, duration at that level, and history of previous drug exposure. Such studies should take into account the possible effect of the test doses themselves upon this decline.

Another aspect of tolerance, not usually considered, is one which we shall designate *carry-over*. It is the feature which connects two discrete episodes of tolerance. If carry-over occurs, it should be demonstrable as an alteration in the rate of subsequent reacquisition of tolerance in successive exposures to the drug. This could take two forms. The first might be an increase in the rate of development of acute tolerance in successive exposures to a drug. The second might be a more rapid development of maximal tolerance in successive cycles of chronic exposure separated by intervals in which drug sensitivity has returned completely to normal.

One report has suggested a more rapid induction of tolerance to tremorine during a second cycle of chronic treatment than during the first (59); unfortunately no data were provided to permit independent assessment. In the case of ethanol, the rate of development of *acute* tolerance in rats was found to increase from session to session until maximal adaptation was achieved (226). In addition, tolerance was reacquired more rapidly on each of three successive cycles of *chronic* treatment and recovery in rats (190). This carry-over appears to be analogous to that noted in experiments with meperidine (151) and has been found to persist when cycles of ethanol treatment were separated by alcohol-free periods as long as 3 months. The apparent retention of tolerance by intermittent drinkers from one drinking bout to another, even when these are separated by rather long intervals, might be explained by such a carry-over of ability to re-evoke tolerance rapidly in each bout.

(*e*) *Generalization of tolerance.* In most of the work mentioned above, tolerance has been studied in relation to a single behavioral test or physiological measure. However, a few studies have involved several different indices. Goldberg (126) used a battery of psychomotor, sensory and physiological measures, but he was comparing alcoholic patients with moderate drinkers and non-drinkers, rather than the same subjects before and after several weeks of heavy drinking. Isbell *et al.* (164) and Mendelson (268) and his collaborators did carry out such before-and-after studies on the same subjects, with a wide variety of tests. However, in both cases the subjects were alcoholics or drug addicts who had been abstinent for a few months by virtue of confinement to a hospital or prison. In view of present knowledge concerning carry-over effect, the conclusions of these studies may not be completely applicable to all instances of tolerance.

If tolerance represents a fundamental change in the effect of the drug on the organism, rather than a highly specific functional compensation for individual types of impairment, one might predict that tolerance should develop simultaneously to all the central effects produced by a specified dose of the drug. Animal experiments do not appear to bear out this prediction consistently. For example, rats may develop tolerance to the anorexiant effect of amphetamine (145, 369) and to its effects on timing behavior (333), but not to its effects on general unconditioned activity. Repeated administration of *d*-amphetamine increased the rate of responding in a shock-avoidance situation for as long as the drug was given, whereas tolerance developed to its effects on two food-reinforced schedules in which it had initially reduced the frequency of food reward (331).

Similarly, rats chronically treated with oxazepam developed tolerance to its response-decreasing effect on the unpunished component of a multiple schedule of reinforcement, but not to its response-increasing or disinhibiting effect on the punished component of the same schedule (250). Sometimes, however, the difference between effects on different parameters is only one of rate. For example, tolerance developed more rapidly to the overt behavioral impairment produced by ethanol in monkeys than to its effects on the EEG (153).

In other instances, tolerance has been shown to generalize. When rats were made tolerant to ethanol during repeated training sessions on the treadmill test, they were found to be tolerant on a circular maze test also, even though no transfer of learning could be demonstrated between these tests (224). The reason for the apparent discrepancies among the results of different studies with respect to generalization of tolerance is not entirely clear. In some cases it may reflect the methods of estimation of tolerance. For example in a study of direct and cross tolerance between LSD and amphetamine in man (315), tolerance was considered to be present when a particular drug effect was significantly less after the period of chronic treatment than it had been on the first test. On this basis, tolerance to LSD was demonstrable only with respect to its effects on pupillary dilatation, subjective psychic effects, and clinical rating of behavioral disturbance. However, if tolerance were defined as the disappearance of significant difference between post-treatment tests and placebo trials, with respect to measures in which such difference had been present in pretreatment trials, then body temperature, pulse

rate and other measures would also have revealed tolerance. There is need for more work in this area, to follow in detail in the same subjects the kinetics of acquisition and loss of tolerance, by several different measures based on different components of behavior or of nervous system function.

(*f*) *Tolerance and learning.* A distinction is commonly made between physiological tolerance and psychological or "learned" tolerance. The implication appears to be that the first involves a compensatory or homeostatic change in the neurons affected by the drug, which renders them less sensitive to it, while the second depends upon the acquisition of new skills or functions to replace those which remain impaired. Chen (37) reported an experiment which appeared to justify this distinction. Rats were trained to criterion in a circular maze, and tested under ethanol to provide an initial measure of impairment of performance. They were then divided into two groups and given further training sessions on 3 successive days. During these sessions, one group received a dose of ethanol shortly before the training, and the other group received the same dose immediately after it. On the 4th day both groups were retested under ethanol, and the second measure of impairment was compared with the first. The group trained under ethanol showed significant tolerance, while the other group did not. The results were interpreted as evidence of learning, rather than of physiological adaptation of the nervous system to ethanol.

These results have been verified (190, 224) with both the circular maze and the treadmill test. However, when the experiment was carried on for a longer time, the difference between groups proved to be only one of rate (190). Three groups of animals were used for the alcohol-training portion of the experiment. They were all tested initially under ethanol to establish baseline measures of sensitivity. Then one group (ES) received an injection of ethanol immediately before training and saline after, the second (SE) received saline before and ethanol after, and the third (SS) received saline before and after. They received a cycle of 3 training days, followed by retest under ethanol on the 4th day. The ES animals were found to have developed significant tolerance. This cycle was repeated without interruption for a total of 16 test days. After the second cycle the SE group was also significantly more tolerant than the SS. The ES group reached maximal tolerance after 3 cycles, the SE group after 5, while the SS group continued unchanged through 11 cycles. Daily intubation with ethanol, 6 g/kg given at the end of each training or test session, was then started; it did not increase further the tolerance of the ES or SE groups, but it rapidly brought the SS group up to the same level of tolerance as the others.

An apparently similar phenomenon has been noted in relation to barbiturate tolerance (106, 109) in man. Tolerance developed more rapidly to the drug effects on competitive performance tests motivated by rewards (analogous to the ES groups) than to those on unstimulated functions such as hours of sleep (analogous to the SE group).

From this work it appears that the "learned tolerance" is essentially the same as "physiological tolerance," except that it is acquired somewhat more rapidly. In other experiments, it has been found to show the same carry-over effect as described above in section II E 3 d, and to bear the same relationship to physical

dependence, to be discussed in Part III (224). For this reason, we propose the term *behaviorally augmented tolerance*, in place of "learned tolerance," to emphasize the similarity to physiological tolerance rather than the minor difference.

A major argument in support of the distinction between physiological and psychological tolerance is based on the demonstration of state-dependent learning in relation to ethanol, pentobarbital and other psychoactive drugs (132, 175, 294–297, 322, 356). An animal which learns to perform a task while under the influence of one of these drugs may subsequently perform the task much better in the drug state than in the drug-free state. The interpretation is that the action of the drug modifies the subject's perception of both internal and external stimuli, and that these modified cues constitute the specific constellation or "gestalt" in which the task is learned. Consequently the task is not performed properly until that constellation is produced again by administration of the drug.

The observations have been greatly refined by Kubena and Barry (217), who were able to show that a task learned under the influence of small doses of alcohol could also be performed under equivalent doses of drugs with similar effects (pentobarbital or chlordiazepoxide), but not under the influence of drugs with different pharmacological actions. "Learned tolerance" to ethanol (37) can be interpreted as arising from the ability of the subject, which had previously learned a task such as the treadmill test in the drug-free state, to relearn it in relation to the new constellation of drug-related stimuli. The tolerance curve would be seen simply as a new learning curve. Cross-tolerance between alcohol and barbiturates would depend upon the resemblance (as perceived by the subject) of one drug state to the other (217). Physiological tolerance would be seen as a separate process involving cellular adaptation, and relating to basic physiological processes involved in consciousness and survival, rather than in learned performance.

As already noted, however, the only difference detectable between "learned tolerance" and "physiological tolerance" is one of rate. In addition, the demonstration that an animal can acquire control of heart rate and other "autonomic" functions by instrumental learning (68, 270, 271) makes it difficult to accept a differentiation between learning and basic neurophysiological processes. It seems more economical to regard drug-dependent learning as simply one specific manifestation of functional tolerance.

4. CROSS-TOLERANCE. It has long been recognized that cross-tolerance can exist between various depressant drugs, such as ethanol and barbiturates (111, 189) or ethanol and other hypnotics and sedatives (338). Alcoholics require a higher alveolar concentration of halothane to induce anesthesia (142). Instances of cross-tolerance among hallucinogens have already been noted (14, 166, 209, 275, 315, 398).

However, not all instances of cross-tolerance can be explained on the same basis. Just as tolerance may be either dispositional, functional or mixed, so may cross-tolerance. The extensive literature on induction of drug-metabolizing enzymes (47) provides many examples of dispositional cross-tolerance. Examples of proven functional cross-tolerance are not nearly so numerous (189). Perhaps some of the apparent contradictions in the literature might be resolved if drug

concentrations in blood or brain were measured in more studies. For example, several reports indicate cross-tolerance between some barbiturates (110, 138, 154), and lack of cross-tolerance between others (56, 308). The discrepancy probably can be explained by reference to the duration of treatment and the time of measurement of drug effect, since metabolic tolerance to barbiturates develops more rapidly than functional tolerance.

III. DEPENDENCE

A. Definition

Just as in the case of tolerance, so also has it been traditional to distinguish between "physical" dependence and "psychic" or "psychological" dependence on alcohol and other mood-modifying drugs. The distinction is embodied in the definition of drug dependence by the Expert Committee on Addiction-Producing Drugs of the World Health Organization (75). In their formulation, dependence is in effect operationally defined as a state of discomfort produced by withdrawal of a drug from a subject who has been chronically or repeatedly exposed to it, and alleviated by renewed administration of that drug or another with similar pharmacological actions. The discomfort may consist of a non-specific and ill-defined dissatisfaction giving rise to a desire (ranging from a mild wish to intense craving) for the perceived effects of the drug; this is called psychological dependence, and may persist with varying intensity for a long time after drug withdrawal. The discomfort may also include a more specific set of physiological disturbances, of varying intensity, and related in a fairly characteristic way to the dosage and pharmacological actions of the drug; this is called physiological dependence, and is usually confined to the first few days or weeks after drug withdrawal.

This concept of dependence places principal emphasis upon the pharmacological actions of the drug, with little or no attention to the factors initiating the drug use. A second model is based upon the concept that drug-taking is initiated by events occurring within the organism (*e.g.*, "anxiety"). Psychological dependence is seen as the maintenance of drug-taking behavior by the continued need to experience the drug effects upon these internal events. In the third and most recent model, psychological dependence is viewed as an example of operant conditioning in which drug-taking is an operant response which may be reinforced by the pharmacological effects of the drug.

These various concepts of dependence imply a dichotomy between mind and body which, as Oswald *et al.* (293) have observed, will become increasingly untenable as the physiological and biochemical bases of psychological function become better known. For the present, however, the distinction is an operational one because there is no experimental evidence of the type noted in section II E 3 f to relate the two types of dependence to each other. They will therefore be considered separately in the following sections.

B. Psychological dependence

Many investigators have approached the problem of drug dependence with a preconception that the essential role of psychoactive drugs in dependent people

is that of reduction of anxiety or internal conflict, and have attempted to create "drug dependence" in experimental animals by exposing them to situations involving noxious stimuli or "stress." The classic work of this type is that of Masserman and Yum (253), who reported that cats consuming ethanol (mixed with milk to mask its aversive taste) showed a reduction in the severity of pathological behavior evoked by a conflict situation, in which an attempt to obtain food caused the animal to receive a blast of air in its face. Since then, many investigators have used other techniques, such as electric shock, strong noise, or conditioned avoidance of electrical shock to induce states of "stress" with the expectation that this would increase consumption of solutions of ethanol which were available to the animals.

Many of these studies have been reviewed by Lester (232) and by Schuster and Thompson (332). It is perhaps sufficient for our purposes to summarize Lester's comments: while these studies did indicate that stressed animals increased their intake of alcohol solution, there is no evidence that most of them consumed enough to obtain a significant pharmacological effect, that the increased consumption was correlated in a consistent manner with the timing of exposure to stress, or that the increase persisted in a manner suggestive of psychological dependence. The failure of these approaches was often implicity assumed to be attributable to deficiencies in the procedures rather than in the model (232).

A clearly different result was obtained when the drinking of ethanol solution was made the instrumental response by which a rat could prevent electric shock (396). By this method, rats were readily induced to maintain a high daily intake of ethanol, with clear signs of intoxication. However, this did not prove to be related specifically to shock avoidance, because similar results were obtained when alcohol consumption was made the instrumental response for obtaining food (203, 299, 339). Further, the pharmacological effect of the ingested ethanol was not in itself reinforcing, because when the rats could obtain food by drinking either ethanol in saccharin solution or saccharin solution alone, they chose the latter (203). Therefore these experimental approaches were successful in producing drinking behavior, but not alcohol dependence.

A different approach, also related to food intake, was devised by Lester (231). Food-deprived rats, trained to bar-press to obtain small pellets of food at intervals of about 1 to 1.5 min, consume an astonishing quantity of water together with the pellets (99). Lester found that they would consume a dilute alcohol solution in the same way, to the point of gross intoxication, and would persist in this behavior for many weeks of trials. This has been confirmed by others (98). However, an operant response by the rat proved to be unnecessary, because "free" presentation of pellets at fixed intervals was just as effective in promoting ethanol ingestion (98, 203). Further, when saccharin solution was available as an alternative the rats did not drink alcohol (203). Finally, any preference for ethanol over water in these conditions appears to be a consequence of the caloric value of the ethanol, rather than of its intoxicant properties (112). The same consideration seems to apply also to ethanol consumption by rats with lesions in the ventromedial hypothalamus (249) which produced hyperphagia and obesity. These

animals became obese, to the same extent as similarly lesioned animals consuming an ordinary diet. There was merely partial replacement of calories from solid foods by calories from alcohol. Again, there was no evidence of intoxication, or of any reinforcement of intake by the pharmacological effect of ethanol.

This brief survey is meant only to point out the main shortcomings of the various experimental approaches based on the concept that psychological dependence on drugs has its origins exclusively in internal events within the organism. The only model which is based upon the pharmacological effect of the drug as a primary determinant of both the genesis and the maintenance of drug-taking behavior is the operant model employing the intravenous self-administration technique (392) originally developed for studies on opiate dependence (for references and descriptions see 60, 365). As a rule, the animal is first trained to press a bar in order to obtain food. It is then enabled to self-administer a drug solution through an indwelling venous cannula by pressing another bar which activates an injection pump. The effect of the drug is the sole reinforcer, and the experimenter can study the effects of various environmental and physiological manipulations on the pattern of self-administration by the animal.

Most of this literature has been reviewed elsewhere (332, 365, 366, 399). It is sufficient for the present purposes to mention the principal conclusions to date. The first is that different classes of drugs differ in their ability to act as primary reinforcers. For example opiates, cocaine, and amphetamines are highly effective, barbiturates and ethanol moderately effective, and mescaline and chlorpromazine relatively ineffective. The second is that not all animals of the same species will respond to the drug with continued self-administration (62). A third conclusion is that the effective drugs behave in the same way as other types of reinforcer, with respect to the effects of experimental variables such as size, frequency and schedule of reinforcement. Finally, when continuous reinforcement is available, some animals will develop patterns of drug intake which are comparable to those of drug-dependent people, leading to severe intoxication, physical illness and gross withdrawal reactions.

One further approach, which was initiated without any theoretical rationale, is the repeated administration of minute amounts of ethanol solution by cannula directly into the lateral ventricles of the brain. Myers (280) reported that such treatment caused a lasting increase in the preference for ethanol over water. Again, the amounts of ethanol ingested were not large, and there was no evidence that the animals became intoxicated. Others have been unable to find any effect of this treatment on alcohol consumption by monkeys (213) or dogs (178). The effect in rats was attributed to biochemical alterations in serotoninergic structures in the walls of the ventricular system, because *p*-chlorophenylalanine abolished the increased oral intake of ethanol (281). However, Nachman *et al.* (282) have pointed out that the effect of *p*-chlorophenylalanine may have been only to produce a non-specific aversive reaction similar to that produced by any noxious stimulus paired with eating or drinking. The significance of the phenomena associated with intraventricular infusion of ethanol remains unclear.

C. Physical dependence

1. INTRODUCTION. Of the drugs discussed in this review, only the so-called general depressants (ethanol, barbiturates, other hypnotics and minor tranquilizers) give rise to clearly recognized withdrawal syndromes, both in people and in experimental animals. There is less certainty concerning phenothiazine tranquilizers, and amphetamines and other stimulants. Symptoms appearing on drug withdrawal have been reported for both these groups, but considerable controversy (some of it apparently semantic) surrounds the interpretation. There is, at present, general agreement that no identifiable withdrawal reactions follow the use of LSD, other hallucinogens or cannabis.

2. SIGNS AND SYMPTOMS of withdrawal of depressants in man are so well known that they require little description here. Excellent descriptions of the alcohol withdrawal syndrome are to be found in the human experimental studies by Isbell *et al.* (164) and Mendelson and his colleagues (268), and in the reviews of clinical experience by Victor and Adams (378) and Victor (377). It is sufficient to point out here that the various clinical pictures fall on a continuum of increasing severity, from the mildest picture of tremulousness, sleeplessness and irritability, increasing through hallucinatory states and seizures, to the severest type, delirium tremens. All of these states are characterized by varying degrees of hyperexcitability and hyperactivity of all portions of the nervous system—central, peripheral, somatic, and autonomic. Some of the auditory hallucinations have actually been attributed to misinterpretation of sounds generated in the middle ear by twitching of the stapedius, tensor tympani and tensor veli palatini muscles (324). In general, the signs and symptoms are the opposite to those characterizing the picture of acute intoxication.

Virtually identical reactions have been described in relation to withdrawal of barbiturates, glutethimide, meprobamate, chlordiazepoxide, and a variety of other hypnotics and minor tranquilizers (23, 83, 85–87, 106, 143, 161, 173, 342, 394, 400). It is noteworthy that tybamate (N-butylmeprobamate) does not produce withdrawal symptoms even after chronic administration in high dosage (46, 100, 344); this will be commented on in the next section.

The picture appears to be essentially the same in all species tested. Tremor, convulsions, and peculiar behavior suggestive of responses to hallucinatory stimuli have been observed in dogs (83, 95, 108), cats (90, 171) and monkeys (81). In mice a similar picture, with gross tremor, rigidity and convulsions, has been produced (114) and the threshold for electroshock and pentylenetetrazole seizures is below the normal level in mice (38, 39, 264, 362), and cats (171). Similar results have been obtained in the rat (53, 80). There appears to be some difference between species with respect to the duration of treatment required for production of the full picture. Rats made maximally tolerant to ethanol (225) showed, on withdrawal, only hyperirritability on being handled or receiving mild electroshock to the feet (122). Spontaneous convulsions did not occur until the animals had been kept on ethanol for 6 to 8 weeks before withdrawal (80). In contrast, mice developed seizures after treatment lasting only 9 days or less (114, 131).

Several reports have described withdrawal reactions after termination of prolonged high-dose treatment with phenothiazine tranquilizers (28, 119, 180, 267). Among the most prominent symptoms were nausea and vomiting, diarrhea, sweating, tension and restlessness. They began less promptly, and lasted somewhat longer, than those noted above in relation to withdrawal of depressants. It has been suggested (16, 346) that these symptoms result from withdrawal, not of the phenothiazines, but of the anticholinergic drugs which are frequently given for control of parkinsonian symptoms produced by phenothiazine therapy. However, this suggestion appears to be invalidated by the experimental study of Gallant *et al.* (119), in which no antiparkinsonian medication was given. The prominence of vomiting as a symptom would be consistent with the suggestion of a rebound phenomenon after cessation of the anti-emetic activity of the phenothiazines. However perphenazine, which is an effective anti-emetic, did not lead to vomiting on withdrawal whereas thioridazine did (141). This finding leaves in question the mechanism of the withdrawal symptoms and their relation to the acute actions of phenothiazines.

Withdrawal of cocaine, phenmetrazine and other stimulants is said to give rise only to fatigue and depression in man (191), and to negligible behavioral modification in other species (212). However, marked increase in appetite has been observed after the abrupt withdrawal of amphetamine in man (214) and rat (369). This increase cannot be attributed simply to the food deprivation resulting from the anorexiant action of the drug, since starvation is usually followed by impairment of appetite during the early period of refeeding. It therefore appears to represent a true withdrawal reaction analogous to the hyperirritability seen on withdrawal of depressants (227). On this basis it is difficult to see why the profound sleepiness and depression observed in man should not also be considered a withdrawal rebound phenomenon. Johnston *et al.* (177) have reported a significant decrease in the minimum alveolar concentration of halothane required for anesthesia in dogs previously given amphetamine for 7 days; this would be quite consistent with the suggestion of a rebound effect though it might also be explainable on the basis of catecholamine depletion by amphetamine. The matter cannot yet be considered settled.

A further phenomenon which has attracted considerable attention in relation to withdrawal reactions after drugs of several different types is *disturbance in the phases of sleep*. Ethanol, in a dose of 1 g/kg given during the evening, has been found to decrease the fraction of the first few hours of sleep which is spent in the phase of rapid eye movements (REM sleep) associated with dream activity (136). With this dose of ethanol, the reduction in REM sleep during the first half of the night was offset by an increase during the second half (210, 402). However, when a dose nearly twice as large was used (210) the amount of REM sleep remained low through the whole night. It was suggested that prolonged suppression of REM sleep might produce anxiety and irritability, which the alcoholic attempted to treat by ingestion of more alcohol, and which might also contribute to the symptoms of the withdrawal reaction.

A puzzling aspect, however, is that similar suppression of REM sleep is ap-

parently caused by amphetamines and other drugs (192, 292) which give rise to different types of clinical picture on drug withdrawal. In addition, Johnson *et al.* (176) studied the pattern of REM and non-REM sleep in alcoholics at the end of a drinking bout and during withdrawal, and observed that the change in clinical state correlated with a composite index of "goodness of sleep" rather than with the amount of REM sleep. They concluded that the sleep pattern was a reflection, rather than a cause, of the clinical state. These findings must still be regarded as providing a basis for extensive investigation, rather than as the basis of a strong plausible hypothesis.

3. MEASUREMENT OF DEPENDENCE has been attempted mainly by visual observation of overt withdrawal signs and assessment of their severity by experienced observers. For example, in withdrawal reactions occurring after chronic ingestion of barbiturates and ethanol, tremor, bizarre behavior, muscular rigidity, and even the electroencephalographic effects of photic stimulation, have been rated on arbitrary scales of intensity (83, 86, 107, 131, 195). Limitations in the interpretation of such quasi-quantitative measures have been discussed above in relation to tolerance. Relatively few investigators have used true quantitative measures such as weight loss, body temperature change, food and water consumption, number of struggle responses and number of convulsions (86, 90, 330, 353).

The measurement of thresholds for electroshock and pentylenetetrazole seizures in mice (38, 39, 86, 264, 362) and cats (171) has already been mentioned, as well as the threshold for production of startle response in rats by administration of electric shock to the feet (122). One clinical study of human alcoholics undergoing withdrawal (340) used a battery of objective measurements of continuous variables, including photoelectric measurement of hand tremor and electrical conductance of the skin. More detailed studies of the kinetics and mechanisms of development of physical dependence will probably require the development of additional sensitive quantitative techniques.

4. RELATION TO DRUG LOAD. The schedule of drug intake or administration used to produce these pictures has been quite varied. In the human experiments with ethanol, Isbell *et al.* (164) gave an average total daily dose of roughly 4 to 5.5 g/kg for 7 to 87 days, while Mendelson and his collaborators (268) gave approximately the same dosage for about 3 weeks. In experiments with dogs, Essig and Lam (95) gave roughly the equivalent of 76 g of absolute ethanol daily over a period of 54 days. Since the weights of the dogs are not given, one can offer only a guess that the dosage was of the order of 7 to 8 g/kg/day. No blood alcohol concentrations are given, but the animals were grossly intoxicated throughout the ethanol treatment period. In Freund's study (114) the mice consumed an average of 12 to 13 g/kg/day for 4 to 5 days. Ellis and Pick (81) used 4 to 8 g/kg/day in divided doses, and terminated the treatment when fine tremor of the monkeys' fingers appeared before each dose on 2 consecutive days; this happened in 10 to 18 days. Gibbins *et al.* (122) used a schedule of incremental dosage rising in 1 g steps from 3 g/kg to 7 g/kg over a 19-day period. With a constant dose of 6 g/kg/day, McQuarrie and Fingl (264) found the fall in electroconvulsive threshold after

ethanol withdrawal to be related to the duration of the preceding period of intoxication.

The same observation has been made with respect to withdrawal hypersensitivity to pentylenetetrazol seizures in cats after pentobarbital treatment (171). A careful study of human volunteers taking pentobarbital and secobarbital (108, 109, 165) showed that no withdrawal symptoms occurred in subjects receiving less than 0.4 g daily (roughly 5 to 6 mg/kg); with increasing dosage up to a maximum of 2.2 g daily, the frequency and severity of withdrawal symptoms was proportional to the dose. In rats permitted to drink only during a 30-min period every 24 hr, chronic treatment with phenobarbital produced a dose-dependent increase in fluid intake; drug withdrawal resulted in a sharp decrease in drinking, the magnitude and duration of which were also related to the preceding drug dosage (330). It is a reasonable conjecture that the speed and intensity of development of physical dependence on other drugs will also be found to vary with the dosage. However, no one has yet studied the relationship as systematically as Andrews and Himmelsbach (5) have done for morphine.

5. RELATION TO TOLERANCE. Experimental studies in man (164, 268) showed that during sustained high intake of ethanol the behavioral impairment gradually became less marked, unless the dose was suddenly raised (268). At the same time, withdrawal signs began to appear in the intervals between doses, though major signs did not occur in most cases until alcohol intake was curtailed or stopped. Similarly, in animal studies with ethanol (81, 95) and meprobamate (83) signs of withdrawal began to appear in the longer intervals between doses, suggesting that the duration of effect of each dose was diminishing.

These observations indicated that tolerance and physical dependence were developing in a roughly parallel fashion. The relation was demonstrated quantitatively (122) in rats treated chronically with ethanol. Startle thresholds were measured either 30 min after the preceding dose of ethanol, when the blood alcohol concentration was high, or 23 hr after, when the level was negligible. The results showed that tolerance and dependence developed in parallel, over the same period as that in which tolerance had been found to develop in the treadmill test (225). Less complete but essentially similar observations have been reported, in relation to concurrent development of tolerance and physical dependence on pentobarbital (171), meprobamate and phenaglycodol (38, 39, 362). Moreover, the recommended method for withdrawal of barbiturates without precipitation of a serious abstinence reaction (20, 88) consists essentially of a titration of the patient's tolerance by giving just enough drug to produce minimal intoxication, then gradually reducing the dose over many days. This demonstrates that a dose which just matches the degree of acquired tolerance also meets the requirements of the physical dependence.

Another parallel between tolerance and physical dependence is suggested by experiments (84, 85) on barbital withdrawal seizures. As noted in section II E 3 d, tolerance develops more rapidly in successive cycles of ethanol treatment (190). Withdrawal seizures were also more numerous and severe in animals exposed to a second period of barbital treatment after recovery from the first (84, 85). A sim-

ilar increase in severity and promptness of development of ethanol withdrawal reactions occurs during repeated cycles in monkeys (81). Rats which have previously experienced physical dependence and withdrawal from barbital will again develop physical dependence on meprobamate at a lower dosage than pharmacologically naive animals (289).

As already noted, however, the two processes do not necessarily continue to develop in parallel at all stages. Even though no further tolerance develops on continued administration of ethanol (225) or barbiturates (10, 307) to rats, prolonged administration is required to produce a maximal withdrawal reaction with spontaneous convulsions (53, 80, 86). This suggests that physical dependence on these drugs may have two components, one of which develops in parallel with tolerance while the other develops slowly during prolonged drug treatment. This suggestion is consistent with the observation (264) that the severity of alcohol withdrawal reactions in mice depended upon both the intensity and the duration of alcohol treatment.

6. CROSS-DEPENDENCE. As we have already noted, extensive cross-tolerance occurs among drugs with similar actions, regardless of their chemical structures. In view of the parallels between the development of tolerance and that of physical dependence, it is not surprising that transfer of dependence occurs among the same drugs.

For example, ethanol has been found partially effective in the prevention of barbiturate withdrawal phenomena (111, 289) and severe ethanol withdrawal reactions such as grand mal seizures are effectively prevented by a variety of depressant drugs including barbiturates (94), chlordiazepoxide (340), paraldehyde and chloral hydrate (106, 181, 377). Barbital withdrawal convulsions in rats were prevented by chlordiazepoxide or meprobamate but not by morphine or chlorpromazine (289). It is also of interest that the seizures were prevented by a barbiturate anticonvulsant agent, primidone (289), but not by diphenylhydantoin (89).

The chemical non-specificity of drugs in the "general depressant" group, and the ease with which one can substitute for another, are the basis for a drug screening procedure for liability to produce dependence (61). Dogs are made dependent upon sodium barbital and new drugs are tested for their ability to postpone a withdrawal reaction when substituted for the barbital. This is exactly analogous to the procedure for testing opiate-type dependence liability in morphine-dependent monkeys (338).

IV. MECHANISMS OF TOLERANCE AND PHYSICAL DEPENDENCE

A. Hypothetical models

Although a number of general theories or hypotheses have been advanced to explain the development of tolerance to and physical dependence on centrally acting drugs, they have tended to give selective emphasis to one or other component of the total picture. It is desirable to begin with a list of all the phenomena which a suitably comprehensive hypothesis must explain. These include the following:

i) graded magnitude of tolerance to a given drug, and difference in maximum degree of tolerance which can be developed to different drugs
ii) time course of acquisition and loss of tolerance, in relation to differences in drug dosage
iii) the appearance of abstinence signs and symptoms which are basically the reverse of those characterizing the acute drug action
iv) time course of acquisition and loss of physical dependence relative to that of tolerance
v) cross-tolerance to other drugs with similar actions, regardless of chemical similarity or dissimilarity, and substitution or cross-dependence on these drugs
vi) the modification of rate of development of tolerance and dependence by behavioral or environmental manipulation
vii) carry-over from one cycle of tolerance to another, and other analogies between tolerance and learning.

All hypotheses proposed so far are variants of the concept of a homeostatic response to the effects of administered drugs. Some self-regulatory process is assumed to adapt to the drug effects in such a manner as to restore normal function in the continued presence of the drug. On drug withdrawal, the erstwhile adaptive process then constitutes an uncompensated disturbance in its own right, giving rise to the withdrawal reaction. The exact formulation of each hypothesis probably reflects the specific research orientation of its originators.

Goldstein and Goldstein (129, 130) proposed an enzyme induction theory based on the idea that ethanol, or any other drug producing similar dependence, inhibits the synthesis of some substance essential for neuronal function, and that the reduction in concentration of this substance leads to derepression of the enzymes which synthesize it, thus causing tolerance. On withdrawal of the drug, excessive synthesis of the substance would lead to withdrawal over-activity.

Collier (43, 44) proposed a variant of this idea, in which the induced synthetic system is that which produces receptor sites for a neurotransmitter substance. Interference by the drug with either the release or action of the transmitter would, in this scheme, induce a synthesis of new receptors until the rate of combination between the receptors and the reduced concentration of transmitter molecules was restored to normal, *i.e.*, tolerance resulted. On withdrawal of the drug, the increased probability of combination between a normal concentration of transmitter substance and the increased concentration of receptors would lead to overstimulation of the cell bearing the receptors.

This suggestion is a refinement of the disuse or *denervation supersensitivity hypothesis* (45, 171, 172), which is also based on the premise that drugs such as ethanol, barbiturates or opiates block the release or action of some neurotransmitter, leading to oversensitivity of the receptors for that substance. Though this concept originally derived from observations of denervation supersensitivity in structures innervated by peripheral autonomic fibers, Friedman *et al.* (116) have recently shown that a similar phenomenon can be produced by anticholinergic blockade at cholinergic synapses in the central regulatory mechanism for body temperature.

An alternative hypothesis relating to neurotransmitters (298) is that the drug may impair the release of neurotransmitter, leading to an increase in its concentration within the nerve ending (54). When the concentration becomes high enough, even though the fraction released per impulse is reduced by the drug, the absolute amount released would become normal and function would return to normal even in the presence of drug (*i.e.*, tolerance). On removal of the drug, uninhibited release of the large stock of transmitter would cause over-activity in the postsynaptic cells (52). However, an argument against this hypothesis is the fact that ethanol (186) or pentobarbital (261), in concentrations which inhibited release of acetylcholine from brain slices *in vitro*, did not significantly raise the content of bound acetylcholine in the slices. Presumably the level of acetylcholine in the nerve endings provides feedback control of the rate of synthesis.

Another hypothetical model of tolerance and dependence which has gained some interest is based on the concept of *redundancy* (252a). It is suggested that multiple neuronal pathways can subserve the same physiological function, but that one of these is normally the principal active one while the others are inoperant or "redundant." If the principal pathway is functionally impaired by a drug, the "redundant" pathways become operant, provided they are more resistant to drug impairment than the major pathway. This would produce tolerance. On removal of the drug, all pathways would be operant and the end-function would therefore be overstimulated, giving rise to a withdrawal reaction. This model is in some ways closer to the original concept of "learned tolerance" than of cellular adaptation. To that extent it is subject to the criticisms already raised against the separation of the two. However, the hypothesis can easily be rephrased to include alternate response mechanisms within the same neuron. It then becomes, in effect, indistinguishable from the other hypotheses mentioned.

While all of these hypotheses are attractive, they cannot be tested until specific enzymes, substances, neurotransmitters or neuronal pathways are proposed as the targets for the initial action of the drugs in question. These can then be examined systematically with respect to their relation to tolerance and dependence. If one or more biochemical measures are found consistently to change in parallel with the development of tolerance, both in time and in degree, and to disappear progressively with the disappearance of signs of dependence, then these changes may be classed as "biochemical correlates" of tolerance and dependence (321). It will still be necessary to determine, by experimental manipulation, whether they are causes of tolerance, effects of it, or parallel results of some other more fundamental change.

B. Specific cellular mechanisms

1. PHYSIOLOGICAL. At a physiological level no pathway, nucleus, or functional division within the nervous system has been specifically implicated in the production of tolerance or physical dependence to any of the drugs considered in this review. Wikler *et al.* (394) suggested that hypersynchrony and seizure activity in the EEG during withdrawal from secobarbital could mean either that there was a rebound hyperexcitability of cortical neurons in general, or that the cortex recovered from the effects of the drug earlier than diencephalic structures did. The

latter possibility would, it was suggested, be analogous to the situation resulting from destructive lesions of the reticular activating system. To some extent this is supported by the finding that direct electrical excitation of the cortex by subconvulsive stimuli, perhaps serving a function analogous to activation by the reticular formation, could prevent spontaneous convulsions during barbiturate withdrawal (96).

However, this would still not identify the site of initiation of seizure discharge. Available evidence suggests a major role for subcortical structures. The signs of hyperexcitability which characterize the development of dependence during chronic treatment with ethanol, barbiturates, meprobamate and other sedatives, appear in all parts of the brain. Spontaneous electrical seizure activity in the EEG appeared simultaneously and symmetrically in all cortical leads (91, 93, 143), so that it could not be attributed to a single epileptogenic focus in the cortex. Moreover, barbiturate withdrawal seizures appeared, even though in a somewhat attenuated form, in decorticated (84) and decerebellated (85) animals. These findings are quite consistent with the observation that direct local excitability of acutely deafferented cortical slabs was only minimally increased during barbiturate withdrawal (343).

These findings are all consistent with the suggestion that the electrical seizure activity begins subcortically, and is propagated to and amplified by the cortex, leading to grand mal convulsions only in the most appropriate conditions for impulse spread. Typical epileptic spike and slow wave patterns have been observed in interictal EEGs taken during barbiturate or alcohol withdrawal (334, 394, 395) and photic stimulation has evoked myoclonic twitches or grand mal seizures during withdrawal from ethanol (195, 376), barbiturates (400), meprobamate (23) and bromisovalum (143). Since the spike-and-wave pattern could be produced in the thalamus of normal animals by a combination of photic stimulation and pentylenetetrazole, and appeared a few milliseconds later in the cortex, Gastaut (120) suggested that it is a normal thalamic response which reaches the cortex only when the threshold for stimulus propagation is reduced.

The evidence still does not reveal whether the subcortical hyperactivity is of diffuse or localized origin. The concentration of corticosterone in rat plasma, which is low during chronic barbital intoxication, rises sharply during withdrawal (230). This finding is parallelled by a sharp fall in circulating eosinophiles in man (108). While these changes suggest a sudden rise in the release of ACTH, they do not necessarily indicate a primary overactivity of the hypothalamus and hypophysis, since they might be a response to the stress of convulsions. However, the concurrent finding of increased urinary specific gravity during alcohol withdrawal (341) is at least suggestive of a rebound hypersecretion of vasopressin, even in the absence of convulsions. Tremor, nystagmus, and hyperactivity of tendon reflexes, which are prominent during these withdrawal reactions (164, 269, 394), as well as the changes in critical flicker fusion frequency or intensity thresholds (157, 393) are suggestive of hyperactivity of those brainstem neurons which facilitate transmission in peripheral synapses.

Comparably detailed neurophysiological analyses of the mechanisms of toler-

ance and physical dependence do not appear to have been carried out with the other groups of drugs covered in this review. However, there is some evidence that chronic administration of phenothiazine tranquilizers leads to a compensatory increase in sympathetic nervous activity. Some of the manifestations are peripheral, in the form of raised blood pressure, and disappearance of the phenothiazine inhibition of pressor responses to epinephrine, norepinephrine and yohimbine (219). Others are central, such as enhancement of the stereotyped behavior induced by amphetamine and apomorphine, and prevention of tetrabenazine-induced hypothermia (328).

In summary, chronic administration of many drugs appears to lead to adaptive changes in the activity of neurophysiological mechanisms, of such direction and intensity as to compensate functionally for the drug action (tolerance) and to give rise to overshoot when the drug is withdrawn (physical dependence). However, it is not yet possible to identify a primary site in the initiation of the adaptive changes.

2. BIOCHEMICAL. Biochemical studies have not yet advanced our understanding of specific mechanisms in the adaptive process. Most of the biochemical mechanisms examined are common to many or all parts of the nervous system, and some of them to other tissues as well. Therefore it is difficult to determine the nature of the connection between biochemical changes and changes in CNS activity during drug administration and withdrawal.

Active transport of sodium and potassium ions across the cell membrane is inhibited by ethanol (167, 184, 188) and other general depressants, including nitrous oxide (134) and cyclopropane (4), in the brain and other tissues. The ion transport depends upon activity of the sodium-potassium-stimulated adenosine triphosphatase (ATPase) located in the cell membrane. The activity of this ATPase is reduced to varying degrees by ethanol (58, 167, 184, 314, 359), other general depressants (167, 169), barbiturates (58) chlorpromazine and antihistaminics (179). Since active transport is essential for the maintenance of a normal resting potential across the cell membrane, inhibition of it would be expected to reduce the ability of the cell to maintain a high frequency of response.

In animals made tolerant to ethanol, the rate of active transport of cations in brain tissue, and the activity of Na, K-dependent adenosine triphosphatase in brain, were found to be significantly increased when measured in the absence of ethanol (168). This change reverted to normal by 2 weeks after withdrawal of ethanol. In the same study, active transport by erythrocytes from hospitalized alcoholic patients was found to be higher than in healthy controls. Similar studies have yet to be carried out with chronic administration of barbiturates and other drugs. Moreover, this work does not yet provide any basis for differentiation between cerebral cortex and other parts of the brain, with respect to the degree of adaptive change.

A large body of evidence indicates that depressant drugs reduce the release of acetylcholine from axon terminals in the brain (for references see 186). The drugs include ethanol, pentobarbital, ether, and other general anesthetics, and the methods of study have included measurement of whole brain content of acetyl-

choline, release into superfusate over the surface of the intact cortex, and release from cortical slices *in vitro*. The findings suggest that any drug or condition which decreases neuronal activity also inhibits the release of acetylcholine, but they do not prove a cause-and-effect relation. Smyth and Beck (351) reported that chronic intake of ethanol solutions by rats was accompanied by a fall in the concentration of coenzyme A in the brain, and later by decreases in acetylcholine levels and in the activity of the enzyme systems for biosynthesis and degradation of acetylcholine in brain. These findings were cited in support of the hypothesis (252) that ingestion of ethanol leads to impairment of acetylcholine synthesis and release, and that this in turn leads to a compensatory fall in cholinesterase activity and induction of acetylcholine receptors as proposed by Collier (43).

However, this hypothesis would not explain the finding that brain slices from ethanol-tolerant animals released acetylcholine *in vitro* at the same rate as control slices in the absence of ethanol, but no longer showed inhibition by ethanol (186). All the findings would be consistent with the hypothesis that ethanol, by decreasing neuronal activity through some other mechanism, decreases both the energy metabolism and the acetylcholine release in the brain. With chronically reduced neuronal activity, the relevant enzymatic pathways might decrease as a result of feed-back control, until the development of tolerance caused a return to normal neuronal activity (Smyth and Beck did not show that their animals had actually become tolerant to ethanol). Gage (118), Okada (290) and Inoue and Frank (158) have presented evidence that the direct action of ethanol on nerve endings in isolated nerve-muscle preparations leads to an *increase* in the amount of acetylcholine released per minute. If this is so, it is even more probable that the effects on brain noted above are secondary to a reduction of total activity by some other means.

The same consideration applies to effects of ethanol, barbiturates and other anesthetic agents on oxidative metabolism of brain tissue, formation and release of catecholamines, *gamma*-aminobutyric acid and other possible neurotransmitters. Acute drug effects on these processes have been reviewed elsewhere (57, 167, 260, 303, 389). The available evidence does not permit any conclusion as to whether these effects are causes or manifestations of the drug-induced changes in neuronal activity. The apparent similarity of effects of chronic treatment with such diverse drugs as barbital, ethanol, methamphetamine and various hallucinogens on oxygen uptake, lactate production or brain levels of ATP or ammonia (229, 373, 374), and the lack of correlation between EEG activity and brain phosphorylase activity during chronic administration of meprobamate (182) suggests that many of the metabolic changes are indirect consequences, many steps removed from the primary actions of the drugs.

Similar conclusions arise from the evidence which is available to date, concerning the effects of chronic drug treatment on the turnover of various neurotransmitters. For example, the turnover of catecholamines has been reported to increase in rat brain during chronic treatment with morphine (42) and in mouse brain (152) and rat adrenal medulla after chronic administration of amphetamine, methamphetamine, reserpine or 6-hydroxydopamine (247). Choline acetyltrans-

ferase activity in cerebral cortex, midbrain and optic lobe of young chickens was increased after chronic administration of reserpine, but also after methamphetamine (246). As pointed out by Mandell and Morgan (247), the similarity in effects of such diverse drugs on the same transmitter systems makes it probable that these effects are indirect, and not specific adaptive changes to primary actions of the drugs.

A final difficulty in the search for specific cellular mechanisms is that acute effects are demonstrable in many types of cells (183) and tolerance has been reported in non-neural tissues, in which the processes related to impulse propagation and transmission are not necessarily present. Some of these, such as development of tolerance and withdrawal phenomena in tissue cultures of iris epithelium (243) or spleen (347) exposed to alcohols, or fibroblasts (326) or tumor cells (48) exposed to morphine, are questionable on methodological grounds. Since the criterion of tolerance is successful growth of the culture in drug concentrations which were initially inhibitory, there is no way of distinguishing between acquired tolerance and genetic selection of those cells which had a higher initial tolerance. On the other hand, the recovery of contraction in an isolated frog heart during continued perfusion with an ethanol solution which originally arrested it (306) must be accepted as valid evidence of acute tolerance. Here, however, it is not yet clear whether the alcohol effect and the tolerance are occurring in neural elements at the pacemaker, or on the myocardium.

In the case of amphetamine an entirely different cellular mechanism of tolerance has been proposed. Metabolism of amphetamine in several species gives rise to varying but substantial amounts of *p*-hydroxynorephedrine (13, 26, 49, 350). This has been found to accumulate in sympathetic nerve endings in heart and brain, and to be selectively released by *d*-amphetamine in preference to norepinephrine (26, 49). It has been suggested that the *p*-hydroxynorephedrine acts as a false transmitter in place of norepinephrine, thus giving rise to tachyphylaxis. The accumulation of it during chronic treatment would then presumably give rise to refractoriness to amphetamine, which normally acts *via* release of norepinephrine. This refractoriness, or extended tachyphylaxis, would be quite different in character from tolerance of the barbiturate or ethanol type, which involves a compensatory change counter to the drug effect.

An alternative hypothesis to account for tachyphylaxis has been proposed recently (304, 305). The combination of drug and receptor can, according to this model, give rise not only to a pharmacological effect but also to a conformational change in the receptor with relatively long relaxation time. After dissociation of the drug-receptor complex, the altered receptor would have either a reduced affinity for the drug, or a reduced ability to initiate the characteristic response. This hypothesis offers an attractive explanation for tachyphylaxis with drugs which are presumed to have specific receptors. However, it cannot be readily linked to acute tolerance to drugs such as ethanol, barbiturates or general anesthetics, which do not have specific receptor sites (337).

It is difficult to see how the proposed tachyphylactic model of amphetamine "tolerance" could account for withdrawal symptoms of the type noted earlier.

Indeed, Brodie *et al.* (26) report that administration of *p*-hydroxynorephedrine decreased the peripheral cardiovascular effects of amphetamine, but not the behavioral stimulant and anorexigenic effects which are presumably of central origin. It has recently been reported that chronic administration of amphetamine is actually followed by an *increase* in the rate of synthesis and turnover of catecholamines in mouse brain (152), presumably as a response to the increased release of catecholamines brought about by amphetamine. This seems to imply continued action of amphetamine, so that tolerance would have to depend upon some counteracting adaptive change and not upon a tachyphylactic effect such as that observed in peripheral sympathetic nerves (176). No reports of such adaptive changes have yet appeared.

In summary, the various cellular processes which have been examined do not yet permit identification of the fundamental alterations related to the development of functional tolerance and physical dependence, though a number of promising developments have occurred. The various hypothetical models outlined in the preceding section have not been particularly helpful in directing research toward more specific mechanisms. It is useful, therefore, to develop another formulation or model to account for quantitative and temporal relations independently of specific cellular processes. In enzymology, the development of mathematical and kinetic analyses helped greatly to orient the gathering of data in a manner which later permitted a purposeful investigation of mechanisms. This might be a suitable goal at the present stage of the study of tolerance and dependence.

C. Mathematical or kinetic formulations

One of the features mentioned in section IV A which should be accounted for by a comprehensive model of tolerance is the relation between acute and chronic tolerance. In the various studies cited in section II E 2, the magnitude of acute tolerance to ethanol, barbiturates and other general depressants has been very similar to that observed in chronic studies. Also, some evidence of adaptive response, in the form of an opposite effect when the drug level falls, is seen after single doses as well as after withdrawal of chronic treatment (8, 264). A single model which could account for both acute and chronic processes on the same basis would be logically the most economical one to explore.

As a basis for such a model, one may begin with the assumption that the measured drug effect at any time t depends upon $C_t - C_{thr}$, *i.e.*, the difference between drug concentration at time t and the threshold concentration for drug effect. The value of C_{thr} on the first exposure to a drug is equivalent to initial tolerance, and presumably reflects such factors as species, genetic constitution, metabolism and environmental influences which are beyond the scope of this review. The drug effect to which we refer here is the effect at cellular level. In the intact organism this effect will produce different degrees of functional impairment depending upon the behavioral state of the organism. The relationship between tolerance and drug effect might be indicated as

$$S_a = f[(C_t - C_{thr}), (B)]$$

where S_a indicates the stimulus to adaptation or tolerance and B is some parameter of the behavioral state.

A number of findings are consistent with this concept. A greater increase in tolerance to chlorpromazine (160), amphetamine (331), and ethanol (190, 224) in rats, and to morphine in rats (202) and people (102), was found when the subjects were challenged by a task or a painful stimulus. Further, in the absence of tasks or pain stimulus, no tolerance at all developed with the same doses of morphine (202). Wahlström (384) found much more rapid development of tolerance to hexobarbital when it was administered during the normal waking hours of the rat than when the same dose was given during its normal sleep period. These findings suggest that the same degree of drug-induced reduction of cellular capabilities (such as excitability or transmitter release) constitutes different degrees of impairment according to the varying functional demands placed upon the system. This in turn would lead to different degrees of adaptation.

It was noted earlier (section II B 3) that very little increase in LD50 has been found in animals made tolerant to ethanol or barbiturates. One possible explanation was offered, based on the idea that doses used to induce tolerance were too small. An alternative suggestion is based on the role of functional demand in defining the stimulus to adaptation generated by a drug effect. If the animal is anesthetized by the given dose, other functional demands on it are reduced to a minimum, except for basic autonomic reflexes. Under these conditions, it is conceivable that little or no stimulus to tolerance is generated during the period of unconsciousness, and that such stimulus occurs only while the animal is entering or recovering from the period of drug action. This would be consistent with the observed relation between acute tolerance to thiopental (27, 72) and total duration of the anesthesia. The effect of increasing dosage on the stimulus to adaptation may therefore be self-limiting.

The concept of functional impairment as the effective stimulus to tolerance makes many instances of cross-tolerance between chemically dissimilar drugs quite easily comprehensible (section II E 4). If their end-results upon the function of a neuronal sequence are the same, even if their cellular actions are exerted at different points in the sequence, they should give rise to cross-tolerance. The earlier their point of convergence in the final effector pathway, the greater should be the degree of cross-tolerance. On the other hand, the same concentration of a given drug may affect several different integrated functions to different degrees (244), presumably because these functions depend to different extents upon the cellular capabilities affected by the drug. This would provide an explanation for differential rates of development of tolerance to different effects of the same drug, as noted earlier in relation to generalization of tolerance.

On the basis of this model, the development of acute tolerance can be represented graphically. A line connecting the point on the drug concentration curve at which the effect first appears, with the corresponding point at which it disappears (fig. 2), represents the rise of C_{thr} due to the adaptive processes causing acute tolerance. The changing value of $C_t - C_{thr}$ should then parallel the changing value of observed drug effect, accounting for the more rapid decay of drug effect than of drug concentration (8, 127). The recent report (385) of crossed acute

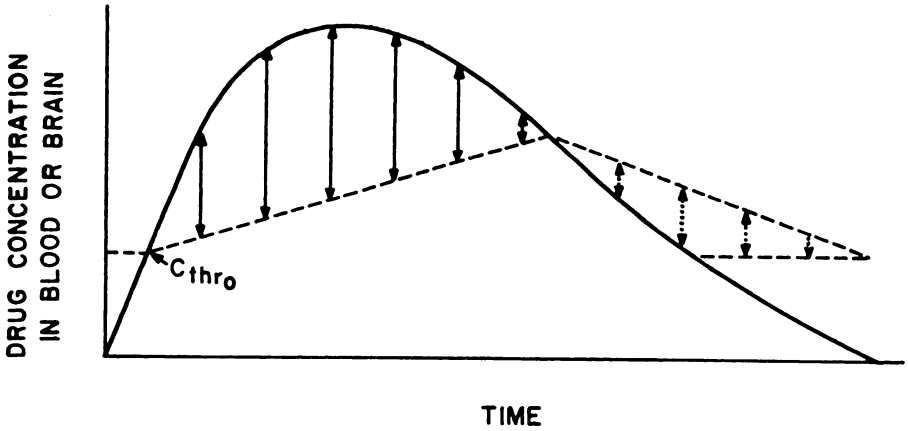

FIG. 2. Hypothetical model for acute tolerance and "hang-over." Solid line indicates curve of drug concentration *versus* time. The initial threshold concentration with respect to a specific drug effect (C_{thr_o}) is shown at both the extreme left and right of the concentration curve. Once the concentration exceeds C_{thr_o} an adaptive change results in a rise in threshold; when the concentration falls below the new threshold, a back-adaptation begins. Solid arrows indicate the magnitude of drug effect; broken arrows indicate the severity of "hang-over" or acute withdrawal reaction. Although the changes in C_{thr} are shown as linear, it seems most probable that they are not actually so and that they have finite lag times.

tolerance between ethanol and hexobarbital, together with the finding of a strictly additive acute interaction between them, lends support to the concept that the drug effect, rather than the drug itself, is the stimulus to the adaptive change.

To account for the development of chronic tolerance on the same model, we must postulate either a duration of the adaptive change which is greater than the interval between successive drug exposures, or a progressive increase in steepness of the C_{thr} line, or both (fig. 3). In the first instance, C_{thr} would begin at a progressively higher point on each succeeding drug exposure, and would continue to rise during each exposure until it reached a maximum determined by some inherent limit in the adaptive capacity of the organism. This would result in a constant decrease in the value of $C_t - C_{thr}$ at all values of t, thus producing a shift in the dose-response or concentration-response curve.

The second possibility, an increase in steepness of the C_{thr} line during succeeding drug exposures, may be designated *carry-over*. This would require an additional factor C in the equation defining the stimulus to adaptation

$$S_a = f[(C_t - C_{thr}), (B), (C)]$$

which allows for the influence of the previous drug history of the individual subject. Increased steepness of the C_{thr} line would also shift the dose-response curve, but the decrement in $C_t - C_{thr}$ within a given drug exposure would not be the same at each value of t. Most published studies of tolerance do not provide data adequate to distinguish between these possibilities, because drug response is usually measured at a fixed time after drug administration, rather than at re-

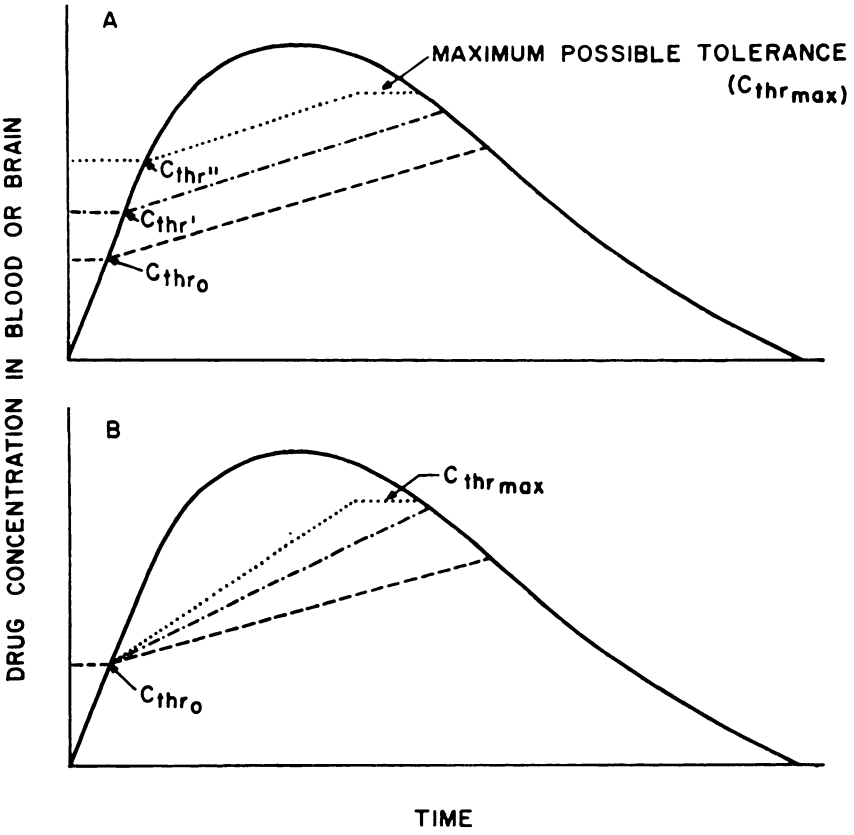

FIG. 3. Diagrammatic representation of possible kinetic features of chronic functional tolerance. The solid curves represent the concentration of drug *versus* time. Broken lines represent the threshold as explained in figure 2. In A, C_{thr} is shown as rising at the same rate within each drug exposure, but starting from a progressively higher level (C_{thr_o}, $C_{thr'}$, $C_{thr''}$, *etc.*) on successive occasions because of prolonged duration of threshold change each time. In B, the threshold is shown as starting from the same initial value (C_{thr_o}) each time, but rising more rapidly on each successive drug exposure ("carry-over"). A third possibility would be a combination of both prolonged duration and carry-over.

peated short intervals. Even the observed changes in time of onset of drug action (189, 358, 383) do not permit definitive answers because the endpoint is again a single observation. Future studies might profitably be designed with this question in mind.

As C_{thr} rises, either within an individual drug exposure or from one exposure to the next, the value of $C_t - C_{thr}$ for any given drug concentration will decrease and the stimulus to adaptation should also diminish. This is consistent with the observed curvature of the line representing acquisition of tolerance (190), indicative of an asymptotic approach to the theoretical maximum.

Carry-over effects from one cycle of tolerance to another are, for the purposes of this type of analysis, treated in exactly the same way as carry-over from one

drug exposure to another within the same cycle. They should be recognizable as an increase in steepness of the line representing C_{thr} *versus* time, starting from the same initial value but rising more rapidly to the theoretical maximum. This relationship, and its resemblance to carry-over in learning, together with the observations on behavioral augmentation of tolerance, suggest that the acquisition of tolerance generally should be amenable to the same types of biochemical and neurophysiological manipulations as are currently applied to the study of memory fixation (15, 259), and opiate dependence (371, 372).

Presumably the adaptive process underlying the tolerance involves synthesis of a protein or some other cell constituent, but it is not necessary for our present purposes to identify the substance. It is sufficient to recognize that the adaptive mechanism has a finite rate of induction, and a characteristic rate of decay when the administration of drug is stopped.

The likelihood of occurrence of an observable withdrawal reaction is then dependent upon the rate of decay of this adaptive mechanism and the rate of elimination of the drug from its sites of action, whether by metabolism, excretion, or competitive displacement by specific antagonists. So far, this latter possibility applies only to the opiates. Kalinowsky (193) pointed out clearly that the speed of onset and the severity of convulsions during withdrawal of ethanol, barbital, paraldehyde and other hypnotics are directly related to the speed of elimination of the drugs. Schutz (335, 336) expressed the relationship in the form $dD/dt > dC/dt$, where D represents the drug action, C the adaptive or counterprocess, and t is time. When both are decreasing, D at a greater rate than C, a withdrawal reaction is probable. Schutz suggested that the same expression, with reversal of the signs, would also serve to describe the acquisition of tolerance. However, evidence from the study of enzyme induction shows that induction and decay of a cell constituent need not, and seldom do, follow the same kinetics.

It is therefore better to say that the likelihood of a withdrawal reaction is expressed by the ratio $t_{1/2a}/t_{1/2d}$ where $t_{1/2a}$ is the half-life of the adaptive mechanism and $t_{1/2d}$ is the half-life of the active drug and its active metabolites if any. The actual magnitude or intensity of the reaction is proportional to $C_{thr} - C_t$. As tolerance develops and C_{thr} rises, a point may be reached at which even a substantial drug concentration C_t is insufficient to produce the usual effect, and the counterprocess prevails (fig. 3). This has been described clearly in several studies (110, 164, 268).

At the same time, the magnitude of $C_{thr} - C_t$ can be viewed as the effective stimulus to de-adaptation, just as $C_t - C_{thr}$ was the initial stimulus to tolerance. It has been found, for example, that repeated production of electroshock convulsions in rats give rise to an adaptive rise in seizure threshold (92). On this basis, a severe withdrawal reaction should also be a relatively short-lasting one, because the de-adaptation should proceed more rapidly. This should also be testable experimentally.

It has been noted (11, 12, 225, 277, 355) that rats which have become tolerant to ethanol, pentobarbital or hexobarbital during chronic treatment develop hypersensitivity to the drug some time after withdrawal. This phenomenon has

been attributed to a fall in the rate of microsomal drug metabolism to subnormal values (355), but the reported fall does not parallel the course of the change in sleeping time. It seems more plausible, therefore, in the light of the present model, to regard this as the negative equivalent of the previous withdrawal reaction, reflecting a disparity between the $t_{1/2}$ of the de-adaptive process and that of the withdrawal reaction which stimulated it. This should be easy to examine experimentally by studying concentration-response rather than dose-response relations during the hypersensitivity phase.

A final implication of the above formulation is that withdrawal reactions should become more severe if $t_{1/2a}/t_{1/2d}$ and $C_{thr} - C_t$ are increased artificially by speeding the process of drug removal, as by hemodialysis, forced diuresis, or some similar technique. By this means it should be possible to provoke or exaggerate withdrawal reactions from drugs such as methadone, chlorpromazine, amphetamine or tetrahydrocannabinol, which are cleared slowly in man. This would help to decide whether the tolerance to amphetamine, for example, is really based on a tachyphylactic mechanism as suggested by some, or really involves an adaptive countermechanism in the same manner as the other drugs.

Conversely, drugs which are metabolized or eliminated too rapidly may not give rise to CNS adaptation and withdrawal reactions because the duration of the effective period of $C_t - C_{thr}$ is too short to cause much change in C_{thr}. Thus, no appreciable value of $C_{thr} - C_t$ would be obtained during withdrawal. For example, tybamate has a much shorter $t_{1/2}$ than meprobamate (46, 344). Therefore a dose of tybamate which produces the same degree of peak effect as a dose of meprobamate lasts for a much shorter time. This may explain why tybamate was found to give rise neither to tolerance nor to withdrawal symptoms (46). In this case, a slowing of drug metabolism by some appropriate inhibitor should cause tybamate to give rise to both tolerance and physical dependence. This can easily be tested. The situation seems quite comparable to that of short-acting barbiturates which do not give rise to true functional tolerance unless given in repeated doses throughout the day, so as to maintain an effective concentration in the blood and brain for a sufficiently large part of each day (138, 171).

The model presented above permits quantitative description of a number of phenomena not readily identified or dealt with otherwise, and permits predictions which can be tested experimentally. Its maximum benefits, however, will be obtained only when a computer program employing successive approximations permits a more precise mathematical statement of the function describing S_a. The nature of this function should then provide more selective clues concerning the underlying biological processes to be examined.

REFERENCES

1. Adams, A. E. and Hubach, H.: Hirnelektrische Korrelate der Wirkungen zentral dämpfender chemischer Substanzen im normalen EEG des Erwachsenen. Deut. Z. Nervenheilk. 181: 71–92, 1960.
2. Ahlquist, R. P. and Dille, J. M.: Reactions of alcohol tolerant rabbits to pentobarbital, evipal, ether, amidopyrine and Metrazol. J. Pharmacol. Exp. Ther. 70: 301–308, 1940.
3. Allan, F. D. and Swinyard, C. A.: Evaluation of tissue tolerance to ethyl alcohol by alterations in electroshock seizure threshold in rats. Anat. Rec. 103: 419, 1949.

4. ANDERSON, N. B.: Synergistic effect of cyclopropane and epinephrine on sodium transport in toad bladder. Anesthesiology **28:** 438-445, 1967.
5. ANDREWS, H. L. AND HIMMELSBACH, C. K.: Relation of the intensity of the morphine abstinence syndrome to dosage. J. Pharmacol. Exp. Ther. **81:** 288-293, 1944.
6. APPEL, J. B. AND FREEDMAN, D. X.: Tolerance and cross-tolerance among psychotomimetic drugs. Psychopharmacologia **13:** 267-274, 1968.
7. ARVOLA, A., SAMMALISTO, L. AND WALLGREN, H.: A test for level of alcohol intoxication in the rat. Quart. J. Stud. Alc. **19:** 563-572, 1958.
8. ASCHAN, G., BERGSTEDT, M., GOLDBERG, L. AND LAURELL, L.: Positional nystagmus in man during and after alcohol intoxication. Quart. J. Stud. Alc. **17:** 381-405, 1956.
9. ASTON, R.: Quantitative aspects of tolerance and post-tolerance hypersensitivity to pentobarbital in the rat. J. Pharmacol. Exp. Ther. **150:** 253-258, 1965.
10. ASTON, R.: Acute tolerance indices for pentobarbital in male and female rats. J. Pharmacol. Exp. Ther. **152:** 350-353, 1966.
11. ASTON, R.: Latent hypersensitivity to pentobarbital in the rat. Proc. Soc. Exp. Biol. Med. **121:** 623-626, 1966.
12. ASTON, R. AND HIBBELN, P.: Induced hypersensitivity to barbital in the female rat. Science **157:** 1463-1464, 1967.
13. AXELROD, J.: Amphetamine: metabolism, physiological disposition, and its effects on catecholamine storage. *In* International Symposium on Amphetamines and Related Compounds, ed. by E. Costa and S. Garattini, pp. 207-216, Raven Press, New York, 1970.
14. BALESTRIERI, A. AND FONTANARI, D.: Acquired and crossed tolerance to mescaline, LSD-25, and BOL-148. Arch. Gen. Psychiat. (Chicago) **1:** 279-282, 1959.
15. BARONDES, S. H.: Effect of inhibitors of cerebral protein synthesis on "long-term" memory in mice. *In* Psychopharmacology, a Review of Progress 1957-1967, ed. by D. H. Efron, pp. 905-908, U. S. Govt. Printing Office, Washington, D. C., 1968.
16. BATTEGAY, R.: Drug dependence as a criterion for differentiation of psychotropic drugs. Compr. Psychiat. **7:** 501-509, 1966.
17. BECKETT, A. H., TAYLOR, J. F. AND KOUROUNAKIS, P.: The absorption, distribution and excretion of pentazocine in man after oral and intravenous administration. J. Pharm. Pharmacol. **22:** 123-128, 1970.
18. BELLEVILLE, R. E. AND FRASER, H. F.: Tolerance to some effects of barbiturates. J. Pharmacol. Exp. Ther. **120:** 469-474, 1957.
19. BENAKIS, A. AND THOMASSET, M.: Metabolism of amphetamines and their interaction with barbiturates and SKF-525A. *In* International Symposium on Amphetamines and Related Compounds, ed. by E. Costa and S. Garattini, pp. 153-164, Raven Press, New York, 1970.
20. BLACHLY, P. H.: Procedure for withdrawal of barbiturates. Amer. J. Psychiat. **120:** 894-895, 1964.
21. BLACK, M. B., WOODS, J. H. AND DOMINO, E. F.: Some effects of (—)-Δ^9-trans-tetrahydrocannabinol and other cannabis derivatives on schedule-controlled behavior. Pharmacologist **12:** 258, 1970.
22. BOISSIER, J. R., TARDY, J. AND DIVERRES, J.-C.: Une nouvelle méthode simple pour explorer l'action "tranquilisante": le test de la cheminée. Med. Exp. **3:** 81-84, 1960.
23. BOKONJIC, N. AND TROJABORG, W.: The effect of meprobamate on the electroencephalogram during treatment, intoxication, and after abrupt withdrawal. Electroencephalogr. Clin. Neurophysiol. **12:** 177-184, 1960.
24. BOREN, J. J.: The study of drugs with operant techniques. *In* Operant Behaviour: Areas of Research and Application, ed. by W. K. Konig, pp. 531-564, Appleton-Century-Crofts, New York, 1966.
25. BOYD, E. M.: Chlorpromazine tolerance and physical dependence. J. Pharmacol. Exp. Ther. **128:** 75-78, 1960.
26. BRODIE, B. B., CHO, A. K. AND GESSA, G. L.: Possible role of *p*-hydroxynorephedrine in the depletion of norepinephrine induced by *d*-amphetamine and in tolerance to this drug. *In* International Symposium on Amphetamine and Related Compounds, ed. by E. Costa and S. Garattini, pp. 217-230, Raven Press, New York, 1970.
27. BRODIE, B. B., MARK, L. C., LIEF, P. A., BURNSTEIN, E. AND PAPPER, E. M.: Acute tolerance to thipoental. J. Pharmacol. Exp. Ther. **102:** 215-218, 1951.
28. BROOKS, G. W.: Withdrawal from neuroleptic drugs. Amer. J. Psychiat. **115:** 931-932, 1959.
29. BÜCH, H., GRUND, W., BUZELLO, W. AND RUMMEL, W.: Narkotische Wirksamkeit und Gewebsverteilung der optischen Antipoden des Pentobarbitals bei der Ratte. Biochem. Pharmacol. **18:** 1005-1009, 1969.
30. BUSH, M. T. AND SANDERS, E.: Metabolic fate of drugs: barbiturates and closely related compounds. Annu. Rev. Pharmacol. **7:** 57-76, 1967.
31. BUTLER, T. C., MAHAFFEE, C. AND WADDELL, W. J.: Phenobarbital: studies of elimination, accumulation, tolerance and dosage schedules. J. Pharmacol. Exp. Ther. **111:** 425-435, 1954.
32. CARMICHAEL, E. B. AND THOMPSON, W. D.: Effect of repeated administration of Delvinal sodium [5-ethyl-5-(1-methyl-1-butenyl) barbituric acid] to guinea pigs. Proc. Soc. Exp. Biol. Med. **46:** 233-235, 1941.
33. CARRO-CIAMPI, G. AND BIGNAMI, G.: Effects of scopolamine on shuttle-box avoidance and go–no go discrimination: response-stimulus relationships, pretreatment baselines, and repeated exposure to drug. Psychopharmacologia **13:** 89-105, 1968.
34. CASPERS, H.: Die Beeinflussung der corticalen Krampferregbarkeit durch das aufsteigende Reticulärsystem des Hirnstammes. II. Narkosewirkung. Z. gesamte Exp. Med. **129:** 582-600, 1958.
35. CASPERS, H. AND ABELE, G.: Hirnelektrische Untersuchungen zur Frage der quantitativen Beziehungen zwischen Blutalkoholgehalt und Alkoholeffekt. Deut. Z. Gesamte Gerichtl. Med. **45:** 492-509, 1956.
36. CHARNEY, N. H. AND REYNOLDS, G. S.: Tolerance to the behavioral effects of scopolamine in rats. Psychopharmacologia **11:** 379-387, 1967.
37. CHEN, C.-S.: A study of the alcohol-tolerance effect and an introduction of a new behavioural technique. Psychopharmacologia **12:** 433-440, 1968.

38. Chin, L. and Swinyard, E. A.: Tolerance and withdrawal hyperexcitability induced in mice by chronic administration of phenaglycodal. Proc. Soc. Exp. Biol. Med. **97**: 251-254, 1958.
39. Chin, L. A. and Swinyard, E. A.: Pentylenetetrazol seizure threshold in meprobamate- and phenaglycodol-treated mice. J. Amer. Pharm. Ass. **48**: 6-8, 1959.
40. Cholden, L. S., Kurland, A. and Savage, C.: Clinical reactions and tolerance to LSD in chronic schizophrenia. J. Nerv. Ment. Dis. **122**: 211-221, 1955.
41. Ciotti, M. M. and Kaplan, N. O.: DPN determination by alcohol dehydrogenase (ADH). *In* Methods in Enzymology, ed. by S. P. Colowick and N. O. Kaplan, vol. III, pp. 891-892, Academic Press, New York, 1957.
42. Clouet, D. H. and Ratner, M.: Biosynthesis of catecholamines in brains of morphine-treated rats. Pharmacologist **12**: 210, 1970.
43. Collier, H. O. J.: A general theory of the genesis of drug dependence by induction of receptors. Nature (London) **205**: 181-182, 1965.
44. Collier, H. O. J.: Tolerance, physical dependence and receptors. A theory of the genesis of tolerance and physical dependence through drug-induced changes in the number of receptors. Advan. Drug Res. **3**: 171-188, 1966.
45. Collier, H. O. J.: Supersensitivity and dependence. Nature (London) **220**: 228-231, 1968.
46. Colmore, J. P. and Moore, J. D.: Lack of dependence and withdrawal symptoms in healthy volunteers given high doses of tybamate. J. Clin. Pharmacol. J. New Drugs **7**: 319-323, 1967.
47. Conney, A. H.: Pharmacological implications of microsomal enzyme induction. Pharmacol. Rev. **19**: 317-366, 1967.
48. Corssen, G. and Skora, I. A.: "Addiction" reactions in cultured human cells. J. Amer. Med. Ass. **187**: 328-332, 1964.
49. Costa, E. and Groppetti, A.: Biosynthesis and storage of catecholamines in tissues of rats injected with various doses of *d*-amphetamine. *In* International Symposium on Amphetamines and Related Compounds, ed. by E. Costa and S. Garattini, pp. 231-255, Raven Press, New York, 1970.
50. Crawford, J. S.: Speculation: the significance of varying the mode of injection of a drug. With special reference to the brain and the placenta. Brit. J. Anaesth. **38**: 628-640, 1966.
51. Creaven, P. J., Barbee, T. and Roach, M. K.: The interaction of ethanol and amphetamine metabolism. J. Pharm. Pharmacol. **22**: 828-831, 1970.
52. Crossland, J. 1957: Cited by M. H. Wulff, 1959 (400).
53. Crossland, H. and Leonard, B. E.: Barbiturate withdrawal convulsions in the rat. Biochem. Pharmacol. suppl. **12**: 103, 1963.
54. Crossland, J. and Merrick, A. J.: The effect of anaesthesia on the acetylcholine content of brain. J. Physiol. (London) **125**: 56-66, 1954.
55. Curry, S. H. and Norris, H.: Acute tolerance to a sedative in man. Brit. J. Pharmacol. **38**: 450P-451P, 1970.
56. Dallemagne, M. J.: L'accoutumance expérimentale a l'Evipan et au Numal. Arch. Int. Pharmacodyn. Thér. **65**: 52-62, 1941.
57. Decsi, L.: Biochemical effects of drugs acting on the central nervous system. Fortschr. Arzneimittelforschung **8**: 53-194, 1965.
58. Decsi, L. and Rodnight, R.: The phosvitin kinase enzyme of cerebral microsomes. J. Neurochem. **12**: 791-796, 1965.
59. Decsi, L. Várszegi, M. and Méhes, G.: Tolerance to tremorine. Acta Physiol. Acad. Sci. Hung. **18**: 353-356, 1961.
60. Deneau, G. A.: Psychogenic dependence in monkeys. *In* Scientific Basis of Drug Dependence, ed. by H. Steinberg, pp. 199-207, Churchill, London, 1969.
61. Deneau, G. A., Klima, A. and Wilson, M.: Evaluation of sedative-hypnotic agents for barbiturate-like physiological dependence capacity in the dog. Reported to Committee on Problems of Drug Dependence, NRC-NAS, Indianapolis, Indiana, 1968.
62. Deneau, G. A., Yanagita, T. and Seevers, M. H.: Self-administration of psychoactive substances by the monkey. A measure of psychological dependence. Psychopharmacologia **16**: 30-48, 1969.
63. DeRomanis, F. and Liberati, F.: Modificazioni eegrafiche da psicofarmaci. Revisione critica. Contributo personale: qualificazione pratica dell'eegramma da somministrazione cronica ed in rapporto a miglioramento clinico in pazienti psichiatrici. Riv. Neurol. **35**: 96-138, 1965.
64. DeSalva, S. J.: Tolerance development to anticonvulsive drugs. J. Pharmacol. Exp. Ther. **116**: 15-16, 1956.
65. Devenyi, P. and Wilson, M.: Barbiturate abuse and addiction and their relationship to alcohol and alcoholism. Can. Med. Ass. J. **104**: 215-218, 1971.
66. Dewey, W. L., Harris, L. S., Howes, J. F. and Kennedy, J. S.: Pharmacological effects of some active constituents of marihuana. Pharmacologist **11**: 372, 1969.
67. Dews, P. B.: Studies on behavior. I. Differential sensitivity to pentobarbital of pecking performance in pigeons depending on the schedule of reward. J. Pharmacol. Exp. Ther. **113**: 393-401, 1955.
68. DiCara, L. V. and Miller, N. E.: Instrumental learning of systolic blood pressure responses by curarized rats: dissociation of cardiac and vascular changes. Psychosom. Med. **30**: 489-494, 1968.
69. Dingell, J. V. and Bass, A. D.: Inhibition of the hepatic metabolism of amphetamine by desipramine. Biochem. Pharmacol. **18**: 1535-1538, 1969.
70. Downs, A. W. and Eddy, N. B.: The effect of repeated doses of cocaine on the dog. J. Pharmacol. Exp. Ther. **46**: 195-198, 1932.
71. Downs, A. W. and Eddy, N. B.: The effect of repeated doses of cocaine on the rat. J. Pharmacol. Exp. Ther. **46**: 199-200, 1932.
72. Dundee, J. W., Price, H. L. and Dripps, R. D.: Acute tolerance to thiopentone in man. Brit. J. Anaesth. **28**: 344-352, 1956.

73. DUNHAM, N. W. AND MIYA, T. S.: A note on a simple apparatus for detecting neurological deficit in rats and mice. J. Amer. Pharm. Ass. 46: 208-209, 1957.
74. EBERT, A. G., YIM, G. K. W. AND MIYA, T. S.: Distribution and metabolism of barbital-^{14}C in tolerant and intolerant rats. Biochem. Pharmacol. 13: 1267-1274, 1964.
75. EDDY, N. B., HALBACH, H., ISBELL, H. AND SEEVERS, M. H.: Drug dependence: its significance and characteristics. World Health Organ. Bull. 32: 721-733, 1965.
76. EGGLETON, M. G.: The effect of alcohol on the central nervous system. Brit. J. Psychol. 32: 52-61, 1941.
77. EGGLETON, M. G.: The diuretic action of alcohol in man. J. Physiol. (London) 101: 172-191, 1942.
78. EICKHOLT, T. H., SCHILLACI, L. J. AND SEARCY, S. A.: Possible ethanol-induced tolerance in rats. J. Pharm. Sci. 56: 275-277, 1967.
79. EIDELBERG, E. AND WOOLEY, D. F.: Effects of ethyl alcohol upon spinal cord neurons. Arch. Int. Pharmacodyn. Thér. 185: 388-396, 1969.
80. ELLIS, F. W. AND PICK, J. R.: Dose- and time-dependent relationships in ethanol-induced withdrawal reactions. Fed. Proc. 30: 568, 1971.
81. ELLIS, F. W. AND PICK, J. R.: Experimentally induced ethanol dependence in Rhesus monkeys. J. Pharmacol. Exp. Ther. 175: 88-93, 1970.
82. EMMERSON, J. L. AND MIYA, T. S.: The metabolism and excretion of S^{35}-chlorpromazine by the rat. J. Pharmacol. Exp. Ther. 137: 148-155, 1962.
83. ESSIG, C. F.: Withdrawal convulsions in dogs following chronic meprobamate intoxication. Arch. Neurol. Psychiat. (Chicago) 80: 414-417, 1958.
84. ESSIG, C. F.: Convulsive and sham rage behaviors in decorticated dogs during barbiturate withdrawal. Arch. Neurol. 7: 471-475, 1962.
85. ESSIG, C. F.: Barbiturate withdrawal convulsions in decerebellate dogs. Int. J. Neuropharmacol. 3: 453-456, 1964.
86. ESSIG, C. F.: Barbiturate withdrawal in white rats. Int. J. Neuropharmacol. 5: 103-107, 1966.
87. ESSIG, C. F.: Increased water consumption following forced drinking of alcohol in rats. Psychopharmacologia 12: 333-337, 1968.
88. ESSIG, C. F.: Barbiturate dependence. *In* Drug Dependence, ed. by R. T. Harris, W. M. McIsaac and C. R. Schuster, Jr., chapt. 11, Univ. Texas Press, Austin, 1970.
89. ESSIG, C. F. AND CARTER, W. W.: Failure of diphenylhydantoin in preventing barbiturate withdrawal convulsions in the dog. Neurology 12: 481-484, 1962.
90. ESSIG, C. F. AND FLANARY, H. G.: Convulsions in cats following withdrawal of barbital sodium. Exp. Neurol. 1: 529-533, 1959.
91. ESSIG, C. F. AND FLANARY, H. G.: Convulsive aspects of barbital sodium withdrawal in the cat. Exp. Neurol. 3: 149-159, 1961.
92. ESSIG, C. F. AND FLANARY, H. G.: The importance of the convulsion in occurrence and rate of development of electroconvulsive threshold elevation. Exp. Neurol. 14: 448-452, 1966.
93. ESSIG, C. F. AND FRASER, H. F.: Electroencephalographic changes in man during use and withdrawal of barbiturates in moderate dosage. Electroencephalogr. Clin. Neurophysiol. 10: 649-656, 1958.
94. ESSIG, C. F., JONES, B. E. AND LAM, R. C.: The effect of pentobarbital on alcohol withdrawal in dogs. Arch. Neurol. 20: 554-558, 1969.
95. ESSIG, C. F. AND LAM, R. C.: Convulsions and hallucinatory behavior following alcohol withdrawal in the dog. Arch. Neurol. 18: 626-632, 1968.
96. ESSIG, C. F. AND WIKLER, A.: Prevention of barbiturate withdrawal convulsions in cats by cerebral electrostimulations. Fed. Proc. 15: 59, 1956.
97. ETTINGER, G. H.: The duration of anaesthesia produced in the dog by the repeated administration of Dial and Nembutal. J. Pharmacol. Exp. Ther. 63: 82-87, 1938.
98. EVERETT, P. B. AND KING, R. A.: Schedule-induced alcohol ingestion. Psychonom. Sci. 18: 278-279, 1970.
99. FALK, J. L.: Production of polydipsia in normal rats by an intermittent food schedule. Science 133: 195-196, 1961.
100. FELDMAN, H. S. AND MULINOS, M. G.: Lack of addiction from high doses of tybamate. J. Clin. Pharmacol. J. New Drugs 6: 354-360, 1966.
101. FERGUSON, H. C.: Effect of red cedar chip bedding on hexobarbital and pentobarbital sleep time. J. Pharm. Sci. 55: 1142-1143, 1966.
102. FERGUSON, R. K. AND MITCHELL, C. L.: Pain as a factor in the development of tolerance to morphine analgesia in man. Clin. Pharmacol. Ther. 10: 372-382, 1969.
103. FITCH, R. H.: An experimental study of tolerance to barbiturates. J. Pharmacol. Exp. Ther. 39: 266-267, 1930.
104. FLEMING, R. AND STOTZ, E.: Experimental studies in alcoholism. I. The alcohol content of the blood and cerebrospinal fluid following oral administration in chronic alcoholism and the psychoses. Arch. Neurol. Psychiat. (Chicago) 33: 492-506, 1935.
105. FLEMING, R. AND STOTZ, E.: Experimental studies in alcoholism. II. The alcohol content of the blood and cerebrospinal fluid following intravenous administration of alcohol in chronic alcoholism and the psychoses. Arch. Neurol. Psychiat. (Chicago) 35: 117-125, 1936.
106. FRASER, H. F.: Tolerance to and physical dependence on opiates, barbiturates and alcohol. Annu. Rev. Med. 8: 427-440, 1957.
107. FRASER, H. F. AND ISBELL, H.: Abstinence syndrome in dogs after chronic barbiturate medication. J. Pharmacol. Exp. Ther. 112: 261-267, 1954.
108. FRASER, H. F., ISBELL, H., EISENMAN, A. J., WIKLER, A. AND PESCOR, F. T.: Chronic barbiturate intoxication. Further studies. Arch. Intern. Med. 94: 34-41, 1954.

109. Fraser, H. F., Isbell, H., Wikler, A., Belleville, R. E., Essig, C. F. and Hill, H. E.: Minimum dose of barbiturates required to produce physical dependence. Fed. Proc. **15**: 423, 1956.
110. Fraser, H., Wikler, A., Essig, C. F. and Isbell, H.: Degree of physical dependence induced by secobarbital or pentobarbital. J. Amer. Med. Ass. **166**: 126–129, 1958.
111. Fraser, H. F., Wikler, A., Isbell, H. and Johnson, N. K.: Partial equivalence of chronic alcohol and barbiturate intoxications. Quart. J. Stud. Alc. **18**: 541–551, 1957.
112. Freed, E. X. and Lester, D.: Schedule-induced consumption of ethanol: calories or chemotherapy? Physiol. Behav. **5**: 555–560, 1970.
113. Freedman, D. X., Appel, J. B., Hartman, F. R. and Molliver, M. E.: Tolerance to behavioral effects of LSD-25 in rat. J. Pharmacol. Exp. Ther. **143**: 309–313, 1964.
114. Freund, G.: Alcohol withdrawal syndrome in mice. Arch. Neurol. **21**: 315–320, 1969.
115. Frey, H.-H. and Kampmann, E.: Tolerance to anticonvulsant drugs. Acta Pharmacol. Toxicol. **22**: 159–171, 1965.
116. Friedman, M. J., Jaffe, J. H. and Sharpless, S. K.: Central nervous system supersensitivity to pilocarpine after withdrawal of chronically administered scopolamine. J. Pharmacol. Exp. Ther. **167**: 45–55, 1969.
117. Furness, F. N. and Dews, P. B. (eds.): Techniques for the study of behavioral effects of drugs. Ann. N. Y. Acad. Sci. **65**: 249–355, 1956.
118. Gage, P. W.: The effect of methyl, ethyl and n-propyl alcohol on neuromuscular transmission in the rat. J. Pharmacol. Exp. Ther. **150**: 236–243, 1965.
119. Gallant, D. M., Edwards, C. G., Bishop, M. P. and Galbraith, G. C.: Withdrawal symptoms after abrupt cessation of antipsychotic compounds: clinical confirmation in chronic schizophrenics. Amer. J. Psychiat. **121**: 491–493, 1964.
120. Gastaut, H.: Combined photic and Metrazol activation of brain. Electroencephalogr. Clin. Neurophysiol. **2**: 249–261, 1950.
121. Gibbins, R. J., Kalant, H. and LeBlanc, A. E.: A technique for accurate measurement of moderate degrees of alcohol intoxication in small animals. J. Pharmacol. Exp. Ther. **159**: 236–242, 1968.
122. Gibbins, R. J., Kalant, H., LeBlanc, A. E. and Clark, J. W.: The effects of chronic administration of ethanol on startle thresholds in rats. Psychopharmacologia **19**: 95–104, 1971.
123. Gillespie, A. M.: A spectrofluorometric study of selected hallucinogens. Anal. Lett. **2**: 605–622, 1969.
124. Gluckman, M. I. and Gruber, C. M.: Development of tolerance and cross-tolerance to mephobarbital. Proc. Soc. Exp. Biol. Med. **79**: 87–88, 1952.
125. Goggerty, J. H. and Dille, J. M.: Tolerance to the pyretogenic effects of lysergic acid diethylamide. J. Pharmacol. Exp. Ther. **116**: 450–452, 1956.
126. Goldberg, L.: Quantitative studies on alcohol tolerance in man. The influence of ethyl alcohol on sensory, motor and psychological functions referred to blood alcohol in normal and habituated individuals. Acta Physiol. Scand. **5**: suppl. 16, 1–128, 1943.
127. Goldberg, L.: Behavioral and physiological effects of alcohol in man. Psychosom. Med. **28**: 570–595, 1966.
128. Goldberg, M. E., Manian, A. A. and Efron, D. H.: A comparative study of certain pharmacologic responses following acute and chronic administration of chlordiazepoxide. Life Sci. **6**: 481–491, 1967.
129. Goldstein, A. and Goldstein, D. B.: Enzyme expansion theory of drug tolerance and physical dependence. *In* Addictive States, ed. by A. Wikler, Ass. Res. Nerv. Ment. Dis. Res. Publ. **46**: 265–267, Williams & Wilkins, Baltimore, 1968.
130. Goldstein, D. B. and Goldstein, A.: Possible role of enzyme inhibition and repression in drug tolerance and addiction. Biochem. Pharmacol. **8**: 48, 1961.
131. Goldstein, D. B. and Pal, N.: Alcohol dependence produced in mice by inhalation of ethanol: grading the withdrawal reaction. Science, **172**: 288–290, 1971.
132. Goodwin, D. W., Powell, B., Bremer, D., Hoine, H. and Stern, J.: Alcohol and recall: state-dependent effects in man. Science **163**: 1358–1360, 1969.
133. Gostomzyk, J. G., Dilger, B. and Dilger, K.: Untersuchungen über die arteriovenöse Differenz der Alkoholkonzentration im Blut und ihre Beziehung zum Alkoholgehalt des Gehirns. Z. Klin. Chem. **7**: 162–166, 1969.
134. Gottlieb, S. F. and Savran, S. V.: Nitrous oxide inhibition of sodium transport. Anesthesiology **28**: 324–326, 1967.
135. Green, M. W. and Koppanyi, T.: Studies on barbiturates. XXVII. Tolerance and cross tolerance to barbiturates. Anesthesiology **5**: 329–340, 1944.
136. Gresham, S. C., Webb, W. B. and Williams, R. L.: Alcohol and caffeine: effect on inferred visual dreaming. Science **140**: 1226–1227, 1963.
137. Groppetti, A. and Costa, E.: Factors affecting the rate of disappearance of amphetamine in rats. Int. J. Neuropharmacol. **8**: 209–215, 1969.
138. Gruber, C. M. and Keyser, G. F.: A study on the development of tolerance and cross tolerance to barbiturates in experimental animals. J. Pharmacol. Exp. Ther. **86**: 186–196, 1946.
139. Gudzinowicz, B. J.: Gas Chromatographic Analysis of Drugs and Pesticides. Marcel Dekker, New York, 1967.
140. Gunne, L. M. and Jonsson, J.: Effects of cocaine administration on brain, adrenal and urinary adrenaline and noradrenaline in rats. Psychopharmacologia **6**: 125–129, 1964.
141. Haden, P.: Gastrointestinal disturbances associated with withdrawal of ataractic drugs. Can. Med. Ass. J. **91**: 974–975, 1964.
142. Han, Y. H.: Why do chronic alcoholics require more anesthesia? Anesthesiology **30**: 341–342, 1969. Further details in Med. Post (Toronto), November 19, 1968.
143. Harenko, A. and Huhmar, E.: EEG changes in chronic bromisovalum poisoning. Ann. Med. Intern. Fenn. **56**: 79–85, 1967.

144. HARGER, R. N. AND FORNEY, R. B.: The aliphatic alcohols. *In* Progress in Chemical Toxicology, ed. by A. Stolman, vol. 1, pp. 53-134, Academic Press, New York, 1963.
145. HARRISON, J. W. E., AMBRUS, C. M. AND AMBRUS, J. L.: Tolerance of rats toward amphetamine and methamphetamine. J. Amer. Pharm. Ass. **41**: 539-541, 1952.
146. HART, L. G. AND FOUTS, J. R.: Effects of acute and chronic DDT administration on hepatic microsomal drug metabolism in the rat. Proc. Soc. Exp. Biol. Med. **114**: 388-392, 1963.
147. HART, L. G. AND FOUTS, J. R.: Further studies on the stimulation of hepatic microsomal drug-metabolizing enzymes by DDT and its analogs. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol. **249**: 486-500, 1965.
148. HART, L. G., SHULTICE, R. W. AND FOUTS, J. R.: Stimulatory effects of chlordane on hepatic microsomal drug metabolism in the rat. Toxicol. Appl. Pharmacol. **5**: 371-386, 1963.
149. HAWKINS, R. D., KALANT, H. AND KHANNA, J. M.: Effects of chronic intake of ethanol on rate of ethanol metabolism. Can. J. Physiol. Pharmacol. **44**: 241-257, 1966.
150. HEIDELMANN, G., PETZOLD, H. AND TASCHEN, B.: Untersuchung über die Nikotin- und Alkoholwirkung auf die akrale Arteriolenfunktion. Deut. Arch. Klin. Med. **199**: 431, 1952. (Abst. in Quart. J. Stud. Alc. **14**: 642-643 1953.)
151. HIMMELSBACH, C. K.: Further studies of the addiction liability of Demerol (1-methyl-4-phenyl-piperidine-4-carboxylic acid ethyl ester hydrochloride). J. Pharmacol. Exp. Ther. **79**: 5-9, 1943.
152. HITZEMANN, R. J. AND LOH, H. H.: The role of adrenergic mechanisms in amphetamine tolerance. Reported to Committee on Problems of Drug Dependence NRC-NAS, Toronto, Canada, 1971.
153. HOGANS, A. F., MORENO, O. M. AND BRODIE, D. A.: Effects of ethyl alcohol on EEG and avoidance behavior of chronic electrode monkeys. Amer. J. Physiol. **201**: 434-436, 1961.
154. HOLCK, H. G. O., RIEDESEL, C. C. AND ROBIDOUX, F. A.: Studies on tolerance and cross-tolerance to Noctal (propallylonal; isopropyl-beta-bromallyl barbituric acid). J. Amer. Pharm. Ass. (Sci. Ed.) **39**: 630-637, 1950.
155. HOOGLAND, D. R., MIYA, T. S. AND BOUSQUET, W. F.: Metabolism and tolerance studies with chlordiazepoxide-2-^{14}C in the rat. Toxicol. Appl. Pharmacol. **9**: 116-123, 1966.
156. HUBBARD, T. F. AND GOLDBAUM, L. R.: The mechanism of tolerance to thiopental in mice. J. Pharmacol. Exp. Ther. **97**: 488-491, 1949.
157. IDESTRÖM, C.-M.: Flicker-fusion in chronic barbiturate usage. A quantitative study in the pathophysiology of drug addiction. Acta Psychiat. Neurol. Scand., suppl. **91**: 1-93, 1954.
158. INOUE, F. AND FRANK, G. B.: Effects of ethyl alcohol on excitability and on neuromuscular transmission in frog skeletal muscle. Brit. J. Pharmacol. Chemother. **30**: 186-193, 1967.
159. IRWIN, S.: The actions of drugs on psychomotor activity. Can. Rev. Biol. **20**: 143-154, 1961.
160. IRWIN, S.: Influence of external factors and arousal mechanisms on the rate of drug tolerance development. Arch. Int. Pharmacodyn. Thér. **142**: 152-162, 1963.
161. ISBELL, H.: Addiction to barbiturates and the barbiturate abstinence syndrome. Ann. Intern. Med. **33**: 108-121, 1950.
162. ISBELL, H., BELLEVILLE, R. E., FRASER, H. F., WILKER, A. AND LOGAN, C. R.: Studies on lysergic acid diethylamide (LSD-25). Arch. Neurol. Psychiat. (Chicago) **76**: 468-478, 1956.
163. ISBELL, H. AND FRASER, H. F.: Addiction to analgesics and barbiturates. Pharmacol. Rev. **2**: 355-397, 1950.
164. ISBELL, H., FRASER, H. F., WIKLER, A., BELLEVILLE, R. E. AND EISENMAN, A. J.: An experimental study of the etiology of "rum fits" and delirium tremens. Quart. J. Stud. Alc. **16**: 1-33, 1955.
165. ISBELL, H., WIKLER, A., BELLEVILLE, R. E., EISIG, C. F. AND HILL, H. E.: Minimal dose of barbiturates required to produce physical dependence. Fed. Proc. **15**: 423, 1956.
166. ISBELL, H., WOLBACH, A. B., WIKLER, A. AND MINER, E. J.: Cross tolerance between LSD and psilocybin. Psychopharmacologia **2**: 147-159, 1961.
167. ISRAEL, Y.: Cellular effects of alcohol. A review. Quart. J. Stud. Alc. **31**: 293-316, 1970.
168. ISRAEL, Y., KALANT, H., LEBLANC, A. E., BERNSTEIN, J. C. AND SALAZAR, I.: Changes in cation transport and (Na+K)-activated adenosine triphosphatase produced by chronic administration of ethanol. J. Pharmacol. Exp. Ther. **174**: 330-336, 1970.
169. ISRAEL, Y. AND SALAZAR, I.: Inhibition of brain microsomal adenosine triphosphatases by general depressants. Arch. Biochem. Biophys. **122**: 310-317, 1967.
170. ISSELBACHER, K. J. AND CARTER, E. A.: Ethanol oxidation by liver microsomes: evidence against a separate and distinct enzyme system. Biochem. Biophys. Res. Commun. **39**: 530-537, 1970.
171. JAFFE, J. H. AND SHARPLESS, S. K.: The rapid development of physical dependence on barbiturates. J. Pharmacol. Exp. Ther. **150**: 140-145, 1965.
172. JAFFE, J. H. AND SHARPLESS, S. K.: Pharmacological denervation supersensitivity in the central nervous system: a theory of physical dependence. *In* Addictive States, ed. by A. Wikler, Ass. Res. Nerv. Ment. Dis. Res. Publ. **46**: 226-246, Williams & Wilkins, Baltimore, 1968.
173. JAMES, I. P.: Drug-withdrawal psychoses. Amer. J. Psychiat. **119**: 880-881, 1963.
174. JELLINEK, E. M.: The Disease Concept of Alcoholism. Hillhouse Press, New Haven, 1960.
175. JOHN, E. R.: State-dependent learning. *In* Mechanisms of Memory, pp. 67-91, Academic Press, New York, 1967.
176. JOHNSON, L. C., BURDICK, J. A. AND SMITH, J.: Sleep during alcohol intake and withdrawal in the chronic alcoholic. Arch. Gen. Psychiat. **22**: 406-418, 1970.
177. JOHNSTON, R. R., WAY, W. L. AND MILLER, R. D.: Alteration in anesthetic requirement with amphetamines. American Society of Anesthesiologists Meeting, Abst., October 1970.
178. JONES, B. J. AND ROBERTS, D. J.: The quantitative measurement of motor incoordination in naive mice using an accelerating rotarod. J. Pharm. Pharmacol. **20**: 302-304, 1968.

179. JUDAH, J. D. AND AHMED, K.: Inhibitors of transport and cation activated ATP-ases. J. Cell. Comp. Physiol. **64:** 355–361, 1964.
180. JUDAH, L. N., JOSEPHS, Z. M. AND MURPHREE, O. D.: Results of simultaneous abrupt withdrawal of ataraxics in 500 chronic patients. Amer. J. Psychiat. **118:** 156–158, 1961.
181. KAIM, S. C., KLETT, C. J. AND ROTHFELD, B.: Treatment of the acute alcohol withdrawal state: a comparison of four drugs. Amer. J. Psychiat. **125:** 1640-1646, 1969.
182. KAJTOR, F., VERES, O. AND KOCSAR, L.: Effect of protracted meprobamate treatment on the bioelectric and phosphorylase activity of the dog brain. Acta Med. Acad. Sci. Hung. **18:** 373–384, 1962.
183. KALANT, H.: Cellular effects of alcohols. *In* Alcohol and Alcoholism, ed. by R. E. Popham, pp. 22–32, Univ. Toronto Press, Toronto, 1970.
184. KALANT, H.: Absorption, distribution and elimination of alcohols. Effect on biological membranes. *In* The Biology of Alcoholism, vol. 1, Physiology and Biochemistry, ed. by B. Kissin and M. M. Begleiter, chapt. 1, Plenum, New York, 1971.
185. KALANT, H. AND CZAJA, C.: The effect of repeated alcoholic intoxication on adrenal ascorbic acid and cholesterol in the rat. Can. J. Biochem. Physiol. **40:** 975–981, 1962.
186. KALANT, H. AND GROSE, W.: Effects of ethanol and pentobarbital on release of acetylcholine from cerebral cortex slices. J. Pharmacol. Exp. Ther. **158:** 386–393, 1967.
187. KALANT, H., HAWKINS, R. D. AND CZAJA, C.: Effect of acute alcohol intoxication on steroid output of rat adrenals *in vitro*. Amer. J. Physiol. **204:** 849–855, 1963.
188. KALANT, H. AND ISRAEL, Y.: Effects of ethanol on active transport of cations. *In* Biochemical Factors in Alcoholism, ed. by R. E. Maickel, pp. 25–37, Pergamon Press, Oxford, 1967.
189. KALANT, H., KHANNA, J. M. AND MARSHMAN, J.: Effect of chronic intake of ethanol on pentobarbital metabolism. J. Pharmacol. Exp. Ther. **175:** 318–324, 1970.
190. KALANT, H., LEBLANC, A. E. AND GIBBINS, R. J.: Pharmacological and behavioral variables in the development of alcohol tolerance. Reported to Committee on Problems of Drug Dependence, NAS-NRC, Toronto, Canada, 1971.
191. KALANT, O. J.: The Amphetamines. Toxicity and Addiction. Univ. Toronto Press, Toronto, 1966.
192. KALES, A.: UCLA Interdepartmental Conference. Drug dependency, investigation of stimulants and depressants. Ann. Intern. Med. **70:** 591–614, 1969.
193. KALINOWSKY, L. B.: Convulsions in nonepileptic patients on withdrawal of barbiturates, alcohol and other drugs. Arch. Neurol. Psychiat. (Chicago) **48:** 946–956, 1942.
194. KANANEN, G., LEON, M. AND SUNSHINE, I.: A novel method for the detection of drugs. Presented at Fifth International Meeting of Forensic Science, Toronto, 1969.
195. KARPATI, G., LLOYD-SMITH, D. L., GIGUERE, R. AND GLOOR, P.: Graded photic hyperexcitability and cerebral seizures in alcohol withdrawal syndromes. Electroencephalogr. Clin. Neurophysiol. **15:** 1051, 1963.
196. KATER, R. M. H., CARULLI, N. AND IBER, F. L.: Differences in the rate of ethanol metabolism in recently drinking alcoholic and non-drinking subjects. Amer. J. Clin. Nutr. **22:** 1608–1617, 1969.
197. KATER, R. M. H., ZEIVE, P., TOBON, F., ROGGIN, G. AND IBER, F. L.: Accelerated metabolism of drugs in alcoholics. Gastroenterology **56:** 412, 1969.
198. KATO, R.: Development of tolerance to meprobamate: tentative enzymatic interpretation. Neuro-psychopharmacol. Proc. Meet. Coll. Int. Neuro-psychopharmacol. **2:** 57–61, 1960.
199. KATO, R.: Analysis and differentiation of the mechanism in the development of drug tolerance. Jap. J. Pharmacol. **17:** 499–508, 1967.
200. KATO, R. AND CHIESARA, E.: Increase of pentobarbitone metabolism induced in rats pretreated with some centrally acting compounds. Brit. J. Pharmacol. **18:** 29-38, 1962.
201. KATO, R. AND VASSANELLI, P.: Induction of increased meprobamate metabolism in rats pretreated with some neurotropic drugs. Biochem. Pharmacol. **11:** 779–794, 1962.
202. KAYAN, S., WOODS, L. A. AND MITCHELL, C. L.: Experience as a factor in the development of tolerance to the analgesic effect of morphine. European J. Pharmacol. **6:** 333–339, 1969.
203. KEEHN, J. D.: "Voluntary" consumption of alcohol by rats. Quart. J. Stud. Alc. **30:** 320–329, 1969.
204. KHANNA, J. M. AND KALANT, H.: Effects of inhibitors and inducers of drug metabolism on *in vivo* ethanol metabolism. Biochem. Pharmacol. **19:** 2033–2041, 1970.
205. KHANNA, J. M., KALANT, H. AND BUSTOS, G.: Effects of chronic intake of ethanol on rate of ethanol metabolism. II. Influence of sex and of schedule of ethanol administration. Can. J. Physiol. Pharmacol. **45:** 777–785, 1967.
206. KHANNA, J. M., KALANT, H. AND LIN, G.: Metabolism of ethanol by rat liver microsomal enzymes. Biochem. Pharmacol. **19:** 2493–2499, 1970.
207. KHANNA, J. M., KALANT, H. AND LIN, G.: The effect of carbon tetrachloride on ethanol metabolism in the rat. Biochem. Pharmacol., in press, 1971.
207a. KINOSHITA, Y., OSHIKA, H. AND NAKAI, K.: Neuronal adaptation to barbiturates. Jap. J. Pharmacol. **17:** 326–327, 1967.
208. KLAASSEN, C. D.: Ethanol metabolism in rats after microsomal metabolizing enzyme induction. Proc. Soc. Exp. Biol. Med. **132:** 1099–1102, 1969.
209. KNOLL, J.: Psychotomimetic effects of amphetamines. *In* International Symposium on Amphetamines and Related Compounds, ed. by E. Costa and S. Garattini, pp. 761–780, Raven Press, New York, 1970.
210. KNOWLES, J. B., LAVERTY, S. G. AND KUECHLER, H. A.: Effects of alcohol on REM sleep. Quart. J. Stud. Alc. **29:** 342–349, 1968.
211. KORNETSKY, C. H.: Psychological effects of chronic barbiturate intoxication. Arch. Neurol. Psychiat. (Chicago) **65:** 557–567, 1951.

212. KOSMAN, M. E. AND UNNA, K. R.: Effects of chronic administration of the amphetamines and other stimulants on behaviour. Clin. Pharmacol. Ther. **9**: 240–254, 1968.
213. KOZ, G. AND MENDELSON, J. H.: Effects of intraventricular ethanol infusion on free choice alcohol consumption by monkeys. *In* Biochemical Factors in Alcoholism, ed. by R. P. Maickel, pp. 17–24, Pergamon Press, Oxford, 1967.
214. KRAMER, J. C., FISCHMAN, V. S. AND LITTLEFIELD, D. C.: Amphetamine abuse. Pattern and effects of high doses taken intravenously. J. Amer. Med. Ass. **201**: 305–309, 1967.
215. KRAUTWALD, A. AND OETTEL, H.: Wirkung und Verhalten von Veronal und Luminal bei chronischer Zufuhr. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Path. **186**: 498–512, 1937.
216. KROP, S. AND GOLD, H.: Comparative study of several barbiturates with observations on irreversible neurological disturbances. J. Pharmacol. Exp. Ther. **88**: 260–267, 1946.
217. KUBENA, R. K. AND BARRY, H., III.: Generalization by rats of alcohol and atropine stimulus characteristics to other drugs. Psychopharmacologia **15**: 196–206, 1969.
218. LAGERSPETZ, K.: The induction of physiological tolerance to promazine in mice. II. The development of induced tolerance. Ann. Med. Exp. Biol. Fenn. **41**: 214–219, 1963.
219. LANG, W., BROWN, M., GERSHON, S., SLETTEN, I. AND KOROL, B.: Effects of chronic chlorpromazine administration upon the blood pressure responses to autonomic drugs in conscious dogs. Arch. Int. Pharmacodyn. Thér. **162**: 330–344, 1966.
220. LAUFFER, S., SCHMID, E. AND WEIST, F.: Dünnschichtchromatographische Trennung und spektrophotofluorimetrischer Nachweis psychotroper Pharmaka. Arzneimittel-Forschung **19**: 1965–1971, 1969.
221. LEAF, G. AND ZATMAN, L. J.: A study of the conditions under which methanol may exert a toxic hazard in industry. Brit. J. Ind. Med. **9**: 19–31, 1952.
222. LEBLANC, A. E.: Microdetermination of alcohol in blood by gas liquid chromatography. Can. J. Physiol. Pharmacol. **46**: 665–667, 1968.
223. LEBLANC, A. E.: Methodological studies on the measurement of ethanol intoxication and acquired tolerance in rats. M.Sc. Thesis, University of Toronto, 1968.
224. LEBLANC, A. E., GIBBINS, R. J. AND KALANT, H.: Training under the influence of ethanol as a factor in the development of tolerance and physical dependence in the rat. Reported to Committee on Problems of Drug Dependence, NAS-NRC, Washington, D. C., 1970.
225. LEBLANC, A. E., KALANT, H. AND GIBBINS, R. J.: Acquisition and loss of tolerance to ethanol by the rat. J. Pharmacol. Exp. Ther. **168**: 244–250, 1969.
226. LEBLANC, A. E., KALANT, H. AND GIBBINS, R. J.: Unpublished observations.
227. LEBLANC, A. E., KALANT, H. AND KALANT, O. J.: The psychopharmacology of amphetamine dependence. Appl. Ther. **12**: 30–34, 1970.
228. LEHMAN, A. J., SCHWERMA, H. AND RICHARDS, E.: Isopropyl alcohol. Acquired tolerance in dogs, rate of disappearance from the blood stream in various species and effects on successive generation of rats. J. Pharmacol. Exp. Ther. **85**: 61–69, 1945.
229. LEONARD, B. E.: The effect of the chronic administration of barbitone sodium on labile compounds in the rat brain. Biochem. Pharmacol. **15**: 255–262, 1966.
230. LEONARD, B. E.: The effect of the chronic administration of barbitone sodium on pituitary-adrenal function in the rat. Biochem. Pharmacol. **15**: 263–268, 1966.
231. LESTER, D.: Self-maintenance of intoxication in the rat. Quart. J. Stud. Alc. **22**: 223–231, 1961.
232. LESTER, D.: Self-selection of alcohol by animals, human variation and the etiology of alcoholism. Quart. J. Stud. Alc. **27**: 395–438, 1966.
233. LÉVY, J.: Contribution à l'étude de l'accoutumance expérimentale aux poisons. II. Alcoolisme expérimental. Fixation de l'alcool sur les tissus de l'organisme accoutumé à cette substance. Bull. Soc. Chim. Biol. **17**: 27–46, 1935.
234. LÉVY, J.: Contribution à l'étude de l'accoutumance expérimentale aux poisons. III. Alcoolisme expérimental. L'accoutumance à l'alcool peut-elle être considérée comme une conséquence de l'hyposensibilité cellulaire? Bull. Soc. Chim. Biol. **17**: 47–59, 1935b.
235. LEWANDER, T.: Influence of various psychoactive drugs on the *in vivo* metabolism of *d*-amphetamine in the rat. European J. Pharmacol. **6**: 38–44, 1969.
236. LIEBER, C. S.: Metabolic derangement induced by ethanol. Annu. Rev. Med. **18**: 35–54, 1967.
237. LIEBER, C. S. AND DE CARLI, L. M.: Ethanol oxidation by hepatic microsomes: adaptive increase after ethanol feeding. Science **162**: 917–918, 1968.
238. LIEBER, C. S. and DE CARLI, L. M.: Hepatic microsomal ethanol-oxidizing system. *In vitro* characteristics and adaptive properties *in vivo*. J. Biol. Chem. **245**: 2505–2512, 1970.
239. LOOMIS, T. A. AND WEST, T. C.: The influence of alcohol on automobile driving ability. An experimental study for the evaluation of certain medicolegal aspects. Quart. J. Stud. Alc. **19**: 30–46, 1958.
240. LUNDQUIST, F.: Enzymatic pathways of ethanol metabolism. *In* International Encyclopedia of Pharmacology and Therapeutics, Section 20: Alcohols and Derivatives, vol. 1, ed. by J. Trémolières, chapt. 4, Pergamon Press, Oxford, 1970.
241. LUNDQUIST, F.: The metabolism of ethanol. *In* Biological Basis of Alcoholism, ed. by Y. Israel and J. Mardones, Chapt. 1, Wiley, New York, in press, 1971.
242. MACLEOD, L. D.: The controlled administration of alcohol to experimental animals. Brit. J. Addict. **45**: 112–124, 1948.
243. MAEDA, M.: On the addiction of cultures *in vitro* of the iris epithelium to ethyl alcohol and its abstinence phenomena in the tissue. Folia Pharmacol. Jap. **25**: 11–23, 1938.

244. MAICKEL, R. P., COX, R. H., JR., MILLER, F. P., SEGAL, D. S. AND RUSSELL, R. W.: Correlation of brain level of drugs with behavioral effects. J. Pharmacol. Exp. Ther. **165**: 216–224, 1969.
245. MALMÉJAC, J. AND PLANE, P.: Étude expérimentale sur l'influence nerveuse général de faibles doses d'alcool. Bull. Acad. Nat. Méd. (Paris) **140**: 38–42, 1956.
246. MANDELL, A. J.: Drug induced alterations in brain biosynthetic enzyme activity—a model for adaptation to the environment by the central nervous system. *In* Biochemistry of Brain and Behavior, ed. by R. E. Bowman and S. Tata, pp. 97–121, Plenum Press, New York, 1970.
247. MANDELL, A. J. AND MORGAN, M.: Amphetamine induced increase in tyrosine hydroxylase activity. Nature (London) **227**: 75–76, 1970.
248. MAPOTHER, E.: Aetiology of alcoholism. Proc. Roy. Soc. Med. **21**: 1346–1351, 1928.
249. MARFAING-JALLAT, P., LARUE, C. AND LEMAGNEN, J.: Alcohol intake in hypothalamic hyperphagic rats. Physiol. Behav. **5**: 345–351, 1970.
250. MARGULES, D. L. AND STEIN, L.: Increase of "antianxiety" activity and tolerance of behavioral depression during chronic administration of oxazepam. Psychopharmacologia **13**: 74–80, 1968.
251. MARSHMAN, J.: Unpublished observations.
252. MARTIN, G. J.: A concept of the etiology of alcoholism. Exp. Med. Surg. **23**: 315–319, 1965.
252a. MARTIN, W. R.: A homeostatic and redundancy theory of tolerance to and dependence on narcotic analgesics. *In* The Addictive States, ed. by A. Wikler, Ass. Res. Nerv. Ment. Dis. Res. Publ. **46**: 206–225, 1968.
253. MASSERMAN, J. H. AND YUM, K. S.: An analysis of the influence of alcohol on experimental neuroses in cats. Psychosom. Med. **8**: 36–52, 1946.
254. MASUDA, M., BUDDE, R. N. AND DILLE, J. M.: An investigation of acquired tolerance to certain short-acting barbiturates. J. Amer. Pharm. Ass. **27**: 830–836, 1938.
255. MATSUKI, K. AND IWAMOTO, T.: Development of tolerance to tranquilizers. Jap. J. Pharmacol. **16**: 191–197, 1966.
256. MATSUKI, K. AND IWAMOTO, T.: Development of tolerance to chlorpromazine in the rat. Jap. J. Pharmacol. **18**: 274–277, 1968.
257. MAYNERT, E. W. AND KLINGMAN, G. I.: Acute tolerance to intravenous anesthetics in dogs. J. Pharmacol. Exp. Ther. **128**: 192–200, 1960.
258. MCDANIEL, H. G., PODGAINY, H. AND BRESSLER, R.: The metabolism of tolbutamide in rat liver. J. Pharmacol Exp. Ther. **167**: 91–97, 1969.
259. MCGAUGH, J. L. AND PETRINOVICH, L. F.: Effects of drugs on learning and memory. Intern. Rev. Neurobiol. **8**: 139–196, 1965.
260. MCILWAIN, H.: Biochemical comment on barbiturates, tranquilizers and ethanol. *In* Experimental Approaches to the Study of Drug Dependence, ed. by H. Kalant and R. D. Hawkins, pp. 76–99, Univ. Toronto Press, Toronto, 1969.
261. MCLENNAN, H. AND ELLIOTT, K. A. C.: Effects of convulsant and narcotic drugs on acetylcholine synthesis. J. Pharmacol. Exp. Ther. **103**: 35–43, 1951.
262. MCMILLAN, D. E., HARRIS, L. S., FRANKENHEIM, J. M. AND KENNEDY, J. S.: 1-Δ^9-trans-tetrahydrocannabinol in pigeons; tolerance to the behavioral effects. Science **169**: 501–503, 1970.
263. MCMILLAN, D. E., HARRIS, L. S., TURK, R. F. AND KENNEDY, J. S.: Development of marked behavioral tolerance to 1-Δ^9-tetrahydrocannabinol (Δ^9-THC) and cross tolerance to 1-Δ^8-tetrahydrocannabinol (Δ^8-THC) in the pigeon. Pharmacologist **12**: 258, 1970.
264. MCQUARRIE, D. G. AND FINGL, E.: Effects of single doses and chronic administration of ethanol on experimental seizures in mice. J. Pharmacol. Exp. Ther. **124**: 264–271, 1958.
265. MEIER, R., GROSS, F., AND TRIPOD, J.: Ritalin, eine neuartige synthetische Verbindung mit spezifischer zentralerregenden Wirkungskomponente. Klin. Wochnschr. **32**: 445–450, 1954.
266. MELLANBY, E.: Alcohol: its absorption into and disappearance from the blood under different conditions. Special Report Series No. 31, Medical Research Committee, London, 1919.
267. MELNYK, W. T., WORTHINGTON, A. G. AND LAVERTY, S. G.: Abrupt withdrawal of chlorpromazine and thioridazine from schizophrenic in-patients. Can. Psychiat. Ass. J. **11**: 410–413, 1966.
268. MENDELSON, J. H. (ed.): Experimentally induced chronic intoxication and withdrawal in alcoholics. Quart. J. Stud. Alc. suppl. **2**: 1964.
269. MENDELSON, J. H. AND LADOU, J.: Psychophysiological findings: experimentally induced chronic intoxication and withdrawal in alcoholics. Quart. J. Stud. Alc. suppl. **2**: 14–39, 1964.
270. MILLER, N. E. AND DICARA, L. V.: Instrumental learning of heart-rate changes in curarized rats: shaping, and specificity to discriminative stimulus. J. Comp. Physiol. Psychol. **63**: 12–19, 1967.
271. MILLER, N. E. AND DICARA, L. V.: Instrumental learning of urine formation by rats: changes in renal blood flow. Amer. J. Physiol. **215**: 677–683, 1968.
272. MILNER, G.: Modified confinement motor activity test for use in mice. J. Pharm. Sci. **57**: 1900–1902, 1968.
273. MIRSKY, I. A., PIKER, P., ROSENBAUM, M. AND LEDERER, H.: "Adaptation" of the central nervous system to varying concentrations of alcohol in the blood. Quart. J. Stud. Alc. **2**: 35–45, 1941.
274. MISRA, P. S., LEFEVRE, A., RUBIN, E. AND LIEBER, C. S.: Effect of ethanol ingestion on ethanol, meprobamate and pentobarbital metabolism. Gastroenterology **58**: 308, 1970.
275. MISSERE, G. AND TONINI, G.: Contributo sperimentale al problema della possibile assuefazione crociata tra farmaci psicotici di diversa struttura. Boll. Soc. Ital. Biol. Sper. **35**: 1119–1120, 1959.
276. MISTILIS, S. P. AND BIRCHALL, A.: Induction of alcohol dehydrogenase in the rat. Nature (London) **223**: 199–200 1969.
277. MOIR, W. M.: The influence of age and sex on repeated administration of sodium pentobarbital to albino rats. J. Pharmacol. Exp. Ther. **59**: 68–85, 1937.

278. MOLINENGO, L.: Azione di alcuni depressivi del SNC sulla coordinazione motoria del ratto. Arch. Ital. Sci. Farmacol. **14:** 288-291, 1964.
279. MORETON, J. E. AND DAVIS, W. M.: Effects of Δ^9-tetrahydrocannabinol on locomotor activity and on phases of sleep. Pharmacologist **12:** 258, 1970.
280. MYERS, R. D.: Alcohol consumption in rats: effects of intracranial injections of ethanol. Science **142:** 240-241, 1963.
281. MYERS, R. D. AND VEALE, W. L.: Alcohol preference in the rat: reduction following depletion of brain serotonin. Science **160:** 1469-1471, 1968.
282. NACHMAN, M., LESTER, D. AND LEMAGNEN, J.: Alcohol aversion in the rat: behavioral assessment of noxious drug effects. Science **168:** 1244-1246, 1970.
283. NEWMAN, H. W.: Acute Alcoholic Intoxication. Stanford Univ. Press, Stanford, 1941.
284. NEWMAN, H. AND ABRAMSON, M.: Relation of alcohol concentration to intoxication. Proc. Soc. Exp. Biol. Med. **48:** 509-513, 1941.
285. NEWMAN, H. AND CARD, J.: Duration of acquired tolerance to ethyl alcohol. J. Pharmacol. Exp. Ther. **59:** 249-252, 1937.
286. NEWMAN, H. W. AND LEHMAN, A. J.: Nature of acquired tolerance to alcohol. J. Pharmacol. Exp. Ther. **62:** 301-306, 1938.
287. NEWMAN, H. W., WILSON, R. N. L. AND NEWMAN, E. J.: Direct determination of maximal daily metabolism of alcohol. Science **116:** 328-329, 1952.
288. NICHOLAS, J. S. AND BARRON, D. H.: The use of sodium Amytal in the production of anesthesia in the rat. J. Pharmacol. Exp. Ther. **46:** 125-129, 1932.
289. NORTON, P. R. E.: The effects of drugs on barbiturate withdrawal convulsions in the rat. J. Pharm. Pharmacol. **22:** 763-766, 1970.
290. OKADA, K.: Effects of alcohols and acetone on the neuromuscular junction of frog. Jap. J. Physiol. **17:** 245-261, 1967.
291. ORME-JOHNSON, W. H. AND ZIEGLER, D. M.: Alcohol mixed function oxidase activity of mammalian liver microsomes. Biochem. Biophys. Res. Commun. **21:** 78-82, 1965.
292. OSWALD, I.: Effects on sleep of amphetamine and its derivatives. *In* International Symposium on Amphetamines and Related Compounds, ed. by E. Costa and S. Garattini, pp. 865-888, Raven Press, New York, 1970.
293. OSWALD, I., EVANS, J. I. AND LEWIS, S. A.: Addictive drugs cause suppression of paradoxical sleep with withdrawal rebound. *In* Scientific Basis of Drug Dependence, ed. by H. Steinberg, pp. 243-257, Churchill, London, 1969.
294. OVERTON, D. A.: State-dependent or "dissociated" learning produced with pentobarbital. J. Comp. Physiol. Psychol. **57:** 3-12, 1964.
295. OVERTON, D. A.: State-dependent learning produced by depressant and atropine-like drugs. Psychopharmacologia **10:** 6-31, 1966.
296. OVERTON, D. A.: Differential responding in a three choice maze controlled by three drug states. Psychopharmacologia **11:** 376-378, 1967.
297. OVERTON, D. A.: Dissociated learning in drug states (state dependent learning). *In* Psychopharmacology. A Review of Progress 1957-1967, ed. by D. Efron, pp. 918-930, Public Health Service Publ. No. 1836, U. S. Govt. Printing Office, Washington, D. C., 1968.
298. PATON, W. D. M.: A pharmacological approach to drug dependence and drug tolerance. *In* Scientific Basis of Drug Dependence, ed. by H. Steinberg, pp. 31-47, Churchill, London, 1969.
299. PERSENSKY, J. J., SENTER, R. J. AND JONES, R. B.: Induced alcohol consumption through positive reinforcement. Psychonom. Sci. **11:** 109-110, 1968.
300. PETERS, J. M.: Factors affecting caffeine toxicity. A review of the literature. J. Clin. Pharmacol. J. New Drugs **7:** 131-141, 1967.
301. PHILLIPS, B. M., MIYA, T. S. AND YIM, G. K. W.: Studies on the mechanism of meprobamate tolerance in the rat. J. Pharmacol. Exp. Ther. **135:** 223-229, 1962.
302. PRINGSHEIM, J.: Chemische Untersuchungen über das Wesen der Alkoholtoleranz. Biochem. Z. **12:** 143-192, 1908.
303. QUASTEL, J. H.: Effects of drugs on metabolism of the brain *in vitro*. Brit. Med. Bull. **21:** 49-56, 1965.
304. RANG, H. P. AND RITTER, J. M.: A new kind of drug antagonism: evidence that agonists cause a molecular change in acetylcholine receptors. Mol. Pharmacol. **5:** 394-411, 1969.
305. RANG, H. P. AND RITTER, J. M.: On the mechanism of desensitization at cholinergic receptors. Mol. Pharmacol. **6:** 357-382, 1970.
306. RANSOM, F.: Acquired tolerance for alcohol in the frog's heart. J. Physiol. (London) **53:** 141-146, 1919.
307. REMMER, H.: Drug tolerance. *In* Ciba Foundation Symposium on Enzymes and Drug Action, ed. by J. L. Mongar and A. V. S. de Reuck, pp. 276-298, Churchill, London, 1962.
308. REMMER, H.: Die Ursache der Gewohnung an oxydable Barbiturate. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol. **244:** 311-333, 1963.
309. REMMER, H. AND SIEGERT, M.: Beschleunigung des Abbaus und Adaptation des ZNS während der Gewöhnung an Barbiturate. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol. **240:** 22-23, 1960.
310. REMMER, H., SIEGERT, M., NITZE, H. R. AND KIRSTEN, I.: Die Gewöhnung an langwirkende Barbiturate. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol. **243:** 468-478, 1962.
311. RICHELLE, M.: A note on behavioral tolerance to meprobamate. J. Exp. Anal. Behav. **8:** 45-46, 1965.
312. RICHELLE, M. AND DJAHANGUIRI, B.: Effet d'un traitement prolongé au chlordiazepoxide sur un conditionnement temporel chez le rat. Psychopharmacologia **5:** 106-114, 1964.

313. Roach, M. K., Reese, W. N., Jr. and Creaven, P. J.: Ethanol oxidation in the microsomal fraction of rat liver. Biochem. Biophys. Res. Commun. 36: 596–602, 1969.
314. Rodnight, R.: The effect of chemical agents on the turnover of the bound phosphate associated with the sodium and potassium ion-stimulated adenosine triphosphatase in ox brain microsomes. Biochem. J. 120: 1–13, 1970.
315. Rosenberg, D. E., Isbell, H., Miner, E. J. and Logan, C. R.: The effect of N,N-dimethyltryptamine in human subjects tolerant to lysergic acid diethylamide. Psychopharmacologia 5: 217–227, 1964.
316. Rosenberg, D. E., Wolbach, A. B., Jr., Miner, E. J. and Isbell, H.: Observations on direct and cross tolerance with LSD and *d*-amphetamine in man. Psychopharmacologia 5: 1–15, 1963.
317. Rubin, A., Tephly, T. R. and Mannering, G. J.: Kinetics of drug metabolism by hepatic microsomes. Biochem. Pharmacol. 13: 1007–1016, 1964.
318. Rubin, E., Hutterer, F. and Lieber, C. S.: Ethanol increases hepatic smooth endoplasmic reticulum and drug-metabolizing enzymes. Science 159: 1469–1470, 1968.
319. Rümke, C. L.: The influence of drugs on the duration of hexobarbital and hydroxydione narcosis in mice. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol. 244: 519–530, 1963.
320. Rümke, C. L., Van Strik, R., DeJonge, H. and Delver, A.: Experiments on the duration of hexobarbital narcosis in mice. Arch. Int. Pharmacodyn. Thér. 146: 10–26, 1963.
321. Russell, R. W.: Behavioral effects of psychoactive drugs. *In* Experimental Approaches to the Study of Drug Dependence, ed. by H. Kalant and R. D. Hawkins, pp. 1–33, Univ. Toronto Press, Toronto, 1969.
322. Ryback, R. S.: State-dependent or "dissociated" learning with alcohol in the goldfish. Quart. J. Stud. Alc. 30: 598–608, 1969.
323. Santesson, C. G.: Kurze Betrachtungen über Toleranz, Giftsucht und Abstinenzsymptome. Skand. Arch. Physiol. 25: 29–36, 1911.
324. Saravay, S. M. and Pardes, H.: Auditory elementary hallucinations in alcohol withdrawal psychosis. Arch. Gen. Psychiat. 16: 652–658, 1967.
325. Sardesai, V. M.: Biochemical and Clinical Aspects of Alcohol Metabolism. Thomas, Springfield, Ill., 1969.
326. Sasaki, M.: Studies on the phenomena of morphine abstinence of cultures *in vitro* of fibroblasts, and on the curative effect of morphine and its derivatives on them. Arch. Exp. Zellforsch. 21: 289–307, 1938.
327. Sauerland, E. K., Mizuno, N. and Harper, R. M.: Presynaptic depolarization of trigeminal cutaneous afferent fibers induced by ethanol. Exp. Neurol. 27: 476–489, 1970.
328. Schelkunov, E. L.: Adrenergic effect of chronic administration of neuroleptics. Nature (London) 214: 1210–1212, 1967.
329. Schild, H. O.: A pharmacological approach to drug receptors. *In* Importance of Fundamental Principles in Drug Evaluation, ed. by D. H. Tedeschi and R. E. Tedeschi, pp. 257–276, Raven Press, New York, 1968.
330. Schmidt, H., Jr. and Kleinman, K. M.: Effect of chronic administration and withdrawal of barbiturates upon drinking in the rat. Arch. Int. Pharmacodyn. Thér. 151: 142–148, 1964.
331. Schuster, C. R., Dockens, W. S. and Woods, J. H.: Behavioural variables affecting the development of amphetamine tolerance. Psychopharmacologia 9: 170–182, 1966.
332. Schuster, C. R. and Thompson, T.: Self administration of and behavioral dependence on drugs. Annu. Rev. Pharmacol. 9: 483–502, 1969.
333. Schuster, C. R. and Zimmerman, J.: Timing behaviour during prolonged treatment with *dl*-amphetamine. J. Exp. Anal. Behav. 4: 327–330, 1961.
334. Schuster, D. B.: The EEG in sedative withdrawal. A case report. Electroencephalogr. Clin. Neurophysiol. 5: 607–610, 1953.
335. Schutz, F.: Mechanism of drug addition and drug tolerance. Nature (London) 148: 725, 1941.
336. Schutz, F.: Serum choline esterase in barbiturate addiction and epilepsy. Quart. J. Exp. Physiol. Cog. Med. Sci. 33: 35–52, 1944.
337. Seeman, P.: Membrane stabilization by drugs: tranquilizers, steroids, and anesthetics. Int. Rev. Neurobiol. 9: 145–221, 1966.
338. Seevers, M. H. and Deneau, G. A.: Physiological aspects of tolerance and physical dependence. *In* Physiological Pharmacology, ed. by W. S. Root and F. G. Hofmann, vol. 1, pp. 565–640, Academic Press, New York, 1963.
339. Senter, R. J., Smith, F. W. and Lewin, S.: Ethanol ingestion as an operant response. Psychonom. Sci. 8: 291–292, 1967.
340. Sereny, G. and Kalant, H.: Comparative clinical evaluation of chlordiazepoxide and promazine in treatment of alcohol withdrawal syndrome. Brit. Med. J. 1: 92–97, 1965.
341. Sereny, G., Rapoport, A. and Husdan, H.: The effect of alcohol withdrawal on electrolyte and acid-base balance. Metabolism 15: 896–904, 1966.
342. Shagass, C., Azima, H. and Sangowicz, J.: Effect of meprobamate in sustained high dosage on the electroencephalogram and sedation threshold. Electroencephalogr. Clin. Neurophysiol. 11: 275–283, 1959.
343. Sharpless, S. K. and Jaffe, J. H.: The electrical excitability of isolated cortex during barbiturate withdrawal. J. Pharmacol. Exp. Ther. 15: 321–329, 1966.
344. Shelton, J. and Hollister, L. E.: Simulated abuse of tybamate in man. Failure to demonstrate withdrawal reactions. J. Amer. Med. Ass. 199: 338–340, 1967.
345. Siegel, M., Ellison, T., Silverman, A. G. and Okun, R.: Tissue distribution of *dl*-^{3}H-amphetamine HCl in tolerant and nontolerant cats. Proc. West. Pharmacol. Soc. 1: 90–94, 1968.
346. Simpson, G. M., Amin, M. and Kunz, E.: Withdrawal effects of phenothiazines. Compr. Psychiat. 6: 347–351, 1965.
347. Singer, E. and Hoder, F.: Wirkung von Alkoholen auf Gewebskulturen. Arch. Exp. Zellforsch. 8: 447–451, 1929.

348. SMITH, D. E.: Physical *vs.* psychological dependence and tolerance in high-dose methamphetamine abuse. Clin. Toxicol. **2:** 99–103, 1969.

349. SMITH, M. E. AND NEWMAN, H. W.: Rate of ethanol metabolism in fed and fasting animals. J. Biol. Chem. **234:** 1544–1549, 1959.

350. SMITH, R. L. AND DRING, L. G.: Patterns of metabolism of β-phenyl-isopropylamines in man and other species. *In* International Symposium on Amphetamines and Related Compounds, ed. by E. Costa and S. Garattini, pp. 121–139, Raven Press, New York, 1970.

351. SMYTH, R. D. AND BECK, H.: The effect of time and concentration of ethanol administration on brain acetylcholine metabolism. Arch. Int. Pharmacodyn. Thér. **182:** 295–299, 1969.

352. SMYTHIES, J. R., SYKES, E. A. AND LORD, C. P.: Structure-activity relationship studies on mescaline: II. Tolerance and cross tolerance between mescaline and its analogues in the rat. Psychopharmacologia **9:** 434–446, 1966.

353. STANTON, E. J.: Addiction and tolerance to barbiturates? The effects of daily administration and abrupt withdrawal of phenobarbital-sodium and pentobarbital-sodium in albino rat. J. Pharmacol. Exp. Ther. **57:** 245–252 1936.

354. STEINBERG, H., DEREUCK, A. V. S. AND KNIGHT, J. (eds.): Animal Behaviour and Drug Action. Churchill, London, 1964.

355. STEVENSON, I. H. AND TURNBULL, M. J.: Hepatic drug-metabolising enzyme activity and duration of hexobarbitone anaesthesia in barbitone-dependent and withdrawn rats. Biochem. Pharmacol. **17:** 2297–2305, 1968.

356. STEWART, J.: Differential responses based on the physiological consequences of pharmacological agents. Psychopharmacologia **3:** 132–138, 1962.

357. STORY, J. L., EIDELBERG, E. AND FRENCH, J. D.: Electrographic changes induced in cats by ethanol intoxication. Arch. Neurol. **5:** 565–570, 1961.

358. STUMPF, C. and CHIARI, I.: Echte Gewöhnung an Hexobarbital. Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol. **251:** 275–287, 1965.

359. SUN, A. Y. AND SAMORAJSKI, T.: Effects of ethanol on the activity of adenosine triphosphatase and acetylcholinesterase in synaptosomes isolated from guinea-pig brain. J. Neurochem. **17:** 1365–1372, 1970.

360. SVENSSON, T. and THIEME, G.: An investigation of a new instrument to measure motor activity of small animals. Psychopharmacologia **14:** 157–163, 1969.

361. SWANSON, E. E., WEAVER, M. M. AND CHEN, K. K.: Repeated administration of Amytal. Amer. J. Med. Sci. **193:** 246–251, 1937.

362. SWINYARD, E. A., CHIN, L. AND FINGL, E.: Withdrawal hyperexcitability following chronic administration of meprobamate to mice. Science **125:** 739–741, 1957.

363. TATUM, A. L. AND SEEVERS, M. H.: Experimental cocaine addiction. J. Pharmacol. Exp. Ther. **36:** 401–410, 1929.

364. TEPHLY, T. R., TINELLI, F. AND WATKINS, W. D.: Alcohol metabolism: role of microsomal oxidation *in vivo*. Science **166:** 627–628, 1969.

365. THOMPSON, T. AND PICKENS, R.: Drug self-administration and conditioning. *In* Scientific Basis of Drug Dependence, ed. by H. Steinberg, pp. 177–198, Churchill, London, 1969.

366. THOMPSON, T. AND PICKENS, R.: Behavioral variables influencing drug self-administration. *In* Drug Dependence, ed. by R. T. Harris, W. M. McIsaac and C. R. Schuster, pp. 143–157. Univ. Texas Press, Austin, 1970.

367. TIMAR, M., LICURICI, V. AND LAZARESCU, M.: Incorporation du barbital C^{14} chez les rats dans les conditions de l'accoutumance à certains barbituriques. Med. Pharmacol. Exp. **14:** 24–30, 1966.

368. TIRRI, R.: Induced tolerance to promazine in mice as a physiological adaptation. Ann. Acad. Sci. Fenn. Ser. A, IV Biol. **103:** 1–54, 1966.

369. TORMEY, J. AND LASAGNA, L.: Relation of thyroid function to acute and chronic effects of amphetamine in the rat. J. Pharmacol. Exp. Ther. **128:** 201–209, 1960.

370. TROSHINA, A. E.: O mekhanizmakh privykaniya organisma k alkogolyu (On the mechanism of habituation of the organism to alcohol). Sb. Nauch. Tr. Ryazan. Med. Inst. **4:** 1, 1957. (Abst. in Quart. J. Stud. Alc. **20:**783–784, 1959.)

371. TURNBULL, M. J. AND STEVENSON, I. H.: Tolerance experiments with barbitone-dependent rats. J. Pharm. Pharmacol. **20:** 884–885, 1968.

372. UNGAR, G. AND COHEN, M.: Induction of morphine tolerance by materials extracted from brain of tolerant animals. Int. J. Neuropharmacol. **5:** 183–192, 1966.

373. UTENA, H.: Behavioral aberrations in methamphetamine-intoxicated animals and chemical correlates in the brain. Progr. Brain Res. **21:** 192–207, 1966.

374. UTENA, H., TAKANO, S., YUASA, S., SCHIMIZU, T., KATO, T. AND FUNATOGAWA, S.: Behavioral abnormalities in animals and metabolic changes in the brain. Brain Nerve (Japan) **13:** 687–695, 1961.

375. VESELL, E. S.: Factors altering the responsiveness of mice to hexobarbital. Pharmacology **1:** 81–97, 1968.

376. VICTOR, M.: The pathophysiology of alcoholic epilepsy. *In* The Addictive States, ed. by A. Wikler, Ass. Res. Nerv. Ment. Dis. Res. Publ. **46:** 431–454, Williams & Wilkins, Baltimore, 1968.

377. VICTOR, M.: The treatment of alcoholism. *In* International Encyclopedia of Pharmacology and Therapeutics, Section 20: Alcohol and Derivatives, ed. by J. Trémolières, Vol. 2, pp. 413–444, Pergamon, Oxford, 1970.

378. VICTOR, M. AND ADAMS, R. D.: The effect of alcohol on the nervous system. *In* Metabolic and Toxic Diseases of the Nervous System, Ass. Res. Nerv. Ment. Dis. Res. Publ. **32:** 526–573, 1953.

379. VIDELA, L. AND ISRAEL, Y.: Factors that modify the metabolism of ethanol in rat liver and adaptive changes produced by its chronic administration. Biochem. J. **118:** 275–281, 1970.

380. VON WARTBURG, J. P.: Metabolism of alcohol in alcoholics and normals and mechanism of action of alcohol dehydrogenase. *In* Biology of Alcoholism, ed. by B. Kissin and H. Begleiter, vol. 1, Biochemistry, chapt. 2, Plenum, New York, 1971.

381. Von Wartburg, J. P. and Papenberg, J.: Biochemical and enzymatic changes induced by chronic ethanol intake. *In* International Encyclopedia of Pharmacology and Therapeutics, Section 20: Alcohols and Derivatives, vol. 2, ed. by J. Trémolières, pp. 301–343, Pergamon, Oxford, 1970.
382. Votava, Z., Horvath, M. and Vinar, O.: Psychopharmacological Methods. Pergamon Press, London, 1963.
383. Wahlström, G.: Hexobarbital (enhexymalum NFN) sleeping times and EEG threshold doses as measurements of tolerance to barbiturates in the rat. Acta Pharmacol. Toxicol. **26**: 64–80, 1968.
384. Wahlström, G.: Differences in tolerance to hexobarbital (enhexymalum NFN) after barbital (diemalum NFN) pretreatment during activity or rest. Acta Pharmacol. Toxicol. **26**: 92–104, 1968.
385. Wahlström, G. and Widerlöv, E.: Interaction and acute cross tolerance between ethanol and hexobarbitone in the rat. J. Pharm. Pharmacol. **23**: 58–60, 1971.
386. Wallace, J. E. and Ladd, S. L.: The determination of drugs in biological specimens as an employee screening procedure. Ind. Med. Surg. **39**: 412–419, 1970.
387. Waller, M. B.: Effects of chronically administered chlorpromazine on multiple-schedule performance. J. Exp. Anal. Behav. **4**: 351–359, 1961.
388. Wallgren, H.: Absorption, diffusion, distribution and elimination of ethanol. Effect on biological membranes. *In* International Encyclopedia of Pharmacology and Therapeutics, Section 20: Alcohols and Derivatives, vol. 1, ed. by J. Trémolières, pp. 161–193, Pergamon, Oxford, 1970.
389. Wallgren, H. and Barry, H., III: Actions of Alcohol. Elsevier Publ. Co., Amsterdam, 1970.
390. Wallgren, H. and Lindbohm, R.: Adaptation to ethanol in rats with special reference to brain tissue respiration. Biochem. Pharmacol. **8**: 423–424, 1961.
391. Wattenberg, L. W. and Leong, J. L.: Effects of phenothiazines on protective systems against polycyclic hydrocarbons. Cancer Res. **25**: 365–370, 1965.
392. Weeks, J. R.: Experimental morphine addiction: method for automatic intravenous injections in unrestrained rats. Science **138**: 143–144, 1962.
393. Weiss, A. D., Victor, M., Mendelson, J. H. and LaDou, J.: Experimentally induced chronic intoxication and withdrawal in alcoholics. Part 6. Critical flicker fusion studies. Quart. J. Stud. Alc., suppl. **2**: 87–95, 1964.
394. Wikler, A., Fraser, H. F., Isbell, H. and Pescor, F. T.: Electroencephalograms during cycles of addiction to barbiturates in man. Electroencephalogr. Clin. Neurophysiol. **7**: 1–13, 1955.
395. Wikler, A., Pescor, F. T., Fraser, H. F. and Isbell, H.: Electroencephalographic changes associated with chronic alcoholic intoxication and alcohol abstinence syndrome. Amer. J. Psychiat. **113**: 106–114, 1956.
396. Williams, D. R. and Teitelbaum, P.: Control of drinking behavior by means of an operant conditioning technique. Science **124**: 1294–1296, 1956.
397. Winters, W. D. and Wallach, M. B.: Drug induced states of CNS excitation: A theory of hallucinosis. *In* Psychotomimetic drugs, ed. by D. H. Efron, pp. 193–228, Raven Press, New York, 1970.
398. Wolbach, A. B., Jr., Isbell, H. and Miner, E. J.: Cross tolerance between mescaline and LSD-25 with a comparison of the mescaline and LSD reactions. Psychopharmacologia **3**: 1–14, 1962.
399. Woods, J. H. and Schuster, C. R., Jr.: Regulation of drug self-administration. *In* Drug Dependence, ed. by R. T. Harris, W. M. McIsaac and C. R. Schuster, Jr., pp. 158–169, Univ. Texas Press, Austin, 1970.
400. Wulff, M. H.: The barbiturate withdrawal syndrome. A clinical and electroencephalographic study. Electroencephalogr. Clin. Neurophysiol. suppl. **14**: 1–173, 1959.
401. Xhenseval, B. and Richelle, M.: Behavioural effects of a long-term treatment with meprobamate in cats. Int. J. Neuropharmacol. **4**: 1–12, 1965.
402. Yules, R. B., Freedman, D. X. and Chandler, K. A.: The effect of ethyl alcohol on man's electroencephalographic sleep cycle. Electroencephalogr. Clin. Neurophysiol. **20**: 109–111, 1966.