Pharmacology Biochemistry and Behavior, Vol. 2, pp. 523–529. ANKHO International Inc., 1974. Printed in the U.S.A.

Dissociation Between Physical Dependence and Volitional Ethanol Consumption: Role of Multiple Withdrawal Episodes[1,2]

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(Received 3 April 1974)

HUNTER, B. E., D. W. WALKER AND J. N. RILEY. *Dissociation between physical dependence and volitional ethanol consumption: role of multiple withdrawal episodes.* PHARMAC. BIOCHEM. BEHAV. 2(4) 523–529, 1974. – The experiment examined the effects of single and multiple episodes of forced administration of a liquid diet containing ethanol on subsequent volitional ethanol consumption. Rats were subjected to a series of 3 sequences of forced liquid diet consumption lasting 20, 50 and 50 days. One group (AD) received a liquid diet with 35–42% of the calories in the form of ethanol. Another group (SD) received identical diets except sucrose was isocalorically substituted for ethanol. Following each sequence a free-choice test was given in which the rats were allowed to choose between an alcohol diet, a sucrose diet and water. After 20 days of alcohol consumption, rats in the AD group rejected the alcohol diet, despite the occurrence of severe withdrawal symptoms including tail-stiffening, ataxia, tremors and hyperreactivity. During subsequent preference tests, a substantial, but transient, increase in alcohol self-selection was observed. It was concluded that rats may learn the association between alcohol and relief of withdrawal symptoms, but a number of withdrawal episodes are required.

Alcohol Ethanol Alcohol dependence Drug withdrawal Alcohol self-selection Ethanol preference

ETHANOL dependence and alcoholism have traditionally been subdivided into two categories: psychic or psychological dependence and physiological dependence. Physiological dependence is rather easily defined as a discomfort resulting from a set of physiological disturbances following alcohol removal, which can be related to the length of alcohol administration and the pharmacological characteristics of alcohol [10]. On the other hand, psychological dependence is often referred to ambiguously and definitions may include both motivational properties of alcohol-seeking behavior as well as primary psychological alcohol-organismic interactions [15]. Kalant, *et al.* [10], for example, have defined psychological dependence as "a nonspecific dissatisfaction giving rise to a desire, ranging from a mild wish to intense craving, for the perceived effects of the drug" (p. 250). If, however, psychological dependence is limited to behaviorally observable, excessive, physically-addictive alcohol consumption, then two questions are raised: (1) what variables influence and/or contribute to the development of psychological dependence and (2) what role does physiological dependence play in the development of psychological dependence?

With respect to alcoholism, physiological dependence has been considered to influence the maintenance of addictive drinking as well as the reinitiation of alcohol-seeking behavior after a period of abstinence [15]. For example, it has generally been assumed that in the physically dependent individual, drug consumption is maintained to avoid the occurrence of withdrawal symptoms [25]. However, the nature of the behavioral and physiological mechanisms involved in the association between alcohol and relief of withdrawal symptoms, together with the effects of this association on subsequent alcohol-seeking behavior have apparently not been investigated. Furthermore, Mello [15] has questioned the validity of this assumption, based on evidence obtained in human alcoholics and laboratory animals. Alcoholics, allowed free access to alcohol for extended periods, show a tendency to separate drinking periods into discrete episodes, separated by periods of self-imposed abstinence often involving several days. In this situation, in which withdrawal symptoms were often observed during periods of abstinence, the alcoholics did not invariably respond by consuming alcohol [14]. A comparable cyclicity of alcohol consumption has

[1] Supported by the Veterans Administration Project No. MRIS 9183 and by PHS Grant AA00200 from NIH-NIAAA to D.W.W. B.E.H. is a predoctoral fellow supported by training grant MH 10320 awarded to the Center for Neurobiological Sciences, University of Florida. Requests for reprints should be addressed to Don W. Walker, Veterans Administration Hospital, Gainesville, Florida, U.S.A.

[2] We thank Patricia Burnett, Larry Ezell and Dot Robinson for expert technical assistance. We also thank Gale Hunter for preparation of the illustrations.

been observed in monkeys trained to administer ethanol via intravenous infusion [3].

Although a number of pharmacological models of ethanol dependence have been developed [15], there has been little examination of the role of physiological dependence, induced by forced administration of alcohol, on subsequent motivated alcohol consumption. The present experiment was designed to examine the question of whether rats, rendered physiologically dependent during single or multiple episodes in which they were restricted to consumption of a liquid diet containing ethanol [9], would volitionally consume an ethanol solution to avoid the occurrence of withdrawal symptoms.

METHOD

Animals

Thirteen male Long-Evans hooded rats, approximately 60 days old, weighing 140–160 g, were used. The rats were housed individually in stainless steel cages. Each cage was equipped with brackets to which 3 calibrated bottles (Wahmann, Co.) could be attached. The rats were housed in a colony room with a 12 hr light–dark cycle.

Liquid Diets

Details of the preparation, composition and nutritional adequacy of the liquid diets have been presented previously [24]. Briefly, the diet was prepared from a 63.3% (v/v) stock solution (prepared from 95% ethanol and distilled water) mixed with Metrecal Shape (Mead Johnson Co.) and contained 35–42% of the calories in the form of ethanol. The ethanol concentration was 8.1–10.4% (v/v) with the diet providing approximately 1.3 KCal/ml. Sucrose was isocalorically substituted for ethanol in control diets. Both the alcohol and sucrose diets were additionally fortified with Vitamin Diet Fortification Mixture, 0.3 g/100 ml of diet, and Salt Mixture XIV, 0.5 g/100 ml of diet (Nutritional Biochemicals Corporation). The diets were prepared fresh daily and administered in calibrated bottles (Wahmann Co.).

Procedure

Prior to the liquid diet treatment period, a 5 day preference test was given to each rat using a modified 3-bottle choice technique [17]. Three solutions were used: (1) a liquid diet containing 42% of the calories in the form of ethanol (10.4% v/v), an identical diet with sucrose isocalorically substituted for ethanol, and (3) tap water. All preference tests were conducted in the animal's home cage and the positions of the bottles were rotated daily to prevent the development of a position habit [17].

Following the initial preference test, the rats were divided into two groups: an alcohol diet group (AD, n = 8) and a sucrose group (SD, n = 5), matched as closely as possible for weight and ethanol consumption during the preference pretest. Both groups were then subjected to a series of 3 sequences of diet administration, in which the liquid diets served as the sole source of calories and fluids. The AD group received liquid diets containing ethanol. The percentage of calories in the form of ethanol was gradually increased during each sequence from 35 to 42%. The SD group received the sucrose liquid diets according to a modified pair-feeding procedure in which each rat in the SD group received the mean quantity of liquid diet consumed by the AD group.

Sequence 1 – The rats were reduced to 75% of ad lib weight by restricting pelleted laboratory food consumption to 5 g/day for 8 days. The groups were then placed on their respective diets for 20 days. On Day 21 of this sequence a 3-choice preference test was given using the same solutions as before. The consumption of alcohol diet, sucrose diet and water was recorded at hourly intervals for the first 8 hr and the rats were frequently observed for behavioral symptoms of physical dependence [9].

Sequence 2 – The rats were allowed food and water ad lib for 4 days prior to the initiation of this sequence. They were then restricted to 5 g lab chow/day for 4 days before receiving their respective diets for 7 weeks. Once each week during the first 5 weeks the diets were removed from the AD and SD groups during morning maintenance for a period of 3 hr (a total of 5 trials). This procedure was used to allow the rats to experience the ameliorative effects of the alcohol diet on withdrawal symptoms. The 3 hr interval was chosen since we have previously observed the appearance of withdrawal symptoms 2–4 hr following alcohol abstinence using similar procedures [9]. On Day 50 of this sequence a 2-day preference test was initiated using identical procedures.

Sequence 3 – The rats were allowed food and water ad lib for 7 days and were then restricted to 5 g chow/day for 8 days. Both groups then received their respective diets continuously for a final 7 weeks. On Day 50 of this sequence a final 3 day preference test was initiated.

During periods of liquid diet administration the bottles containing sucrose or alcohol diets were placed in the middle positions on the home cage. On the first day of each of the 3 preference tests, the bottle containing water was placed in the middle position and the sucrose or alcohol diet bottles were randomly placed on either side. On subsequent days during the 3 preference tests the bottles were rotated in a random fashion.

RESULTS

The mean body weights of the AD and SD groups and ethanol consumption during the 3 sequences of the experiment are shown in Fig. 1. The rats gained weight during each sequence and showed no evidence of malnutrition. Mean daily ethanol consumption during the forced administration sequences ranged from 10–14 g/kg and appeared to decline during the latter portions of Sequences 2 and 3. The rats did not appear to increase total caloric consumption markedly during the course of the experiment despite an increase in body weight of approximately 100%. Thus, the effective dose/body weight of ethanol declined while the absolute level of ethanol intake remained the same. A similar phenomenon has been observed during prolonged administration of aqueous ethanol solutions [2].

Preference Pretest

During the initial pretest the rats unequivocally chose the sucrose diets over the alcohol diets as shown in the first panel of Fig. 2. The rats consumed approximately 96% of their calories from the sucrose diets. Mean daily intake of ethanol diet during the 5 day pretest was 2.4 ml (0.76 g/kg; SEM: 0.13).

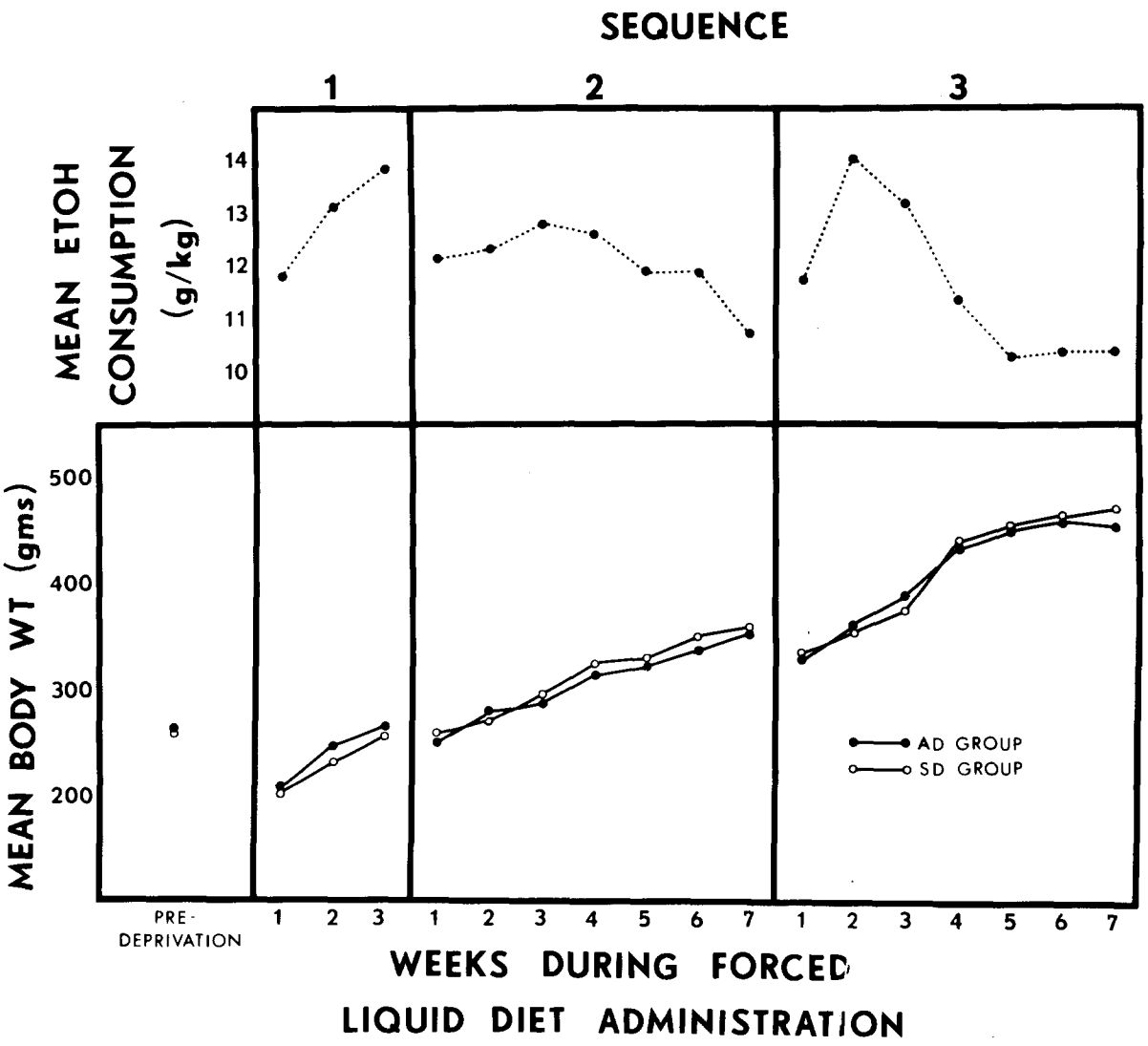

FIG. 1. Mean body weight and ethanol consumption during each week of the 3 liquid diet forced administration sequences in the alcohol diet group (AD) and sucrose diet group (SD).

Preference Test #1:

After 20 days of consumption of the ethanol diets, the rats in the AD group failed to consume the ethanol diets in the free-choice situation, despite the occurrence of severe alcohol withdrawal symptoms. Mean intake of alcohol and sucrose diets for the 2 groups is shown in the second panel of Fig. 2. The withdrawal symptoms were identical to those previously observed in rats using this technique [9]. Initially the symptoms consisted of tail stiffening, piloerection and broad-based gait, beginning 3–4 hr after the initiation of the preference test. As the motor components of the withdrawal sequence increased in severity, the rats became grossly inactive. The inactivity progressed, coincident with the development of the most severe withdrawal symptoms including rigidity, tail arching, mild tremors, fasciculations of axial musculature, and extreme hyperreactivity. These symptoms normally reached their greatest intensity 8–10 hr after the initiation of the preference test. No spontaneous convulsions were observed. This constellation of symptoms was not observed in rats of the SD group. Finally the rats in the AD group also showed evidence of anorexia as can be seen by comparing the intake of sucrose diets in Fig. 2.

Preference Test #2:

An increase in alcohol self-selection was observed in rats from the AD group during the second preference test, which returned to control levels on the second day. As shown in the third panel of Fig. 2, mean alcohol diet consumption for the AD group was 18.0 ml (4.1 g/kg; SEM: 1.1) while mean alcohol consumption for the SD group was 1.9 g/kg (SEM: 0.7).

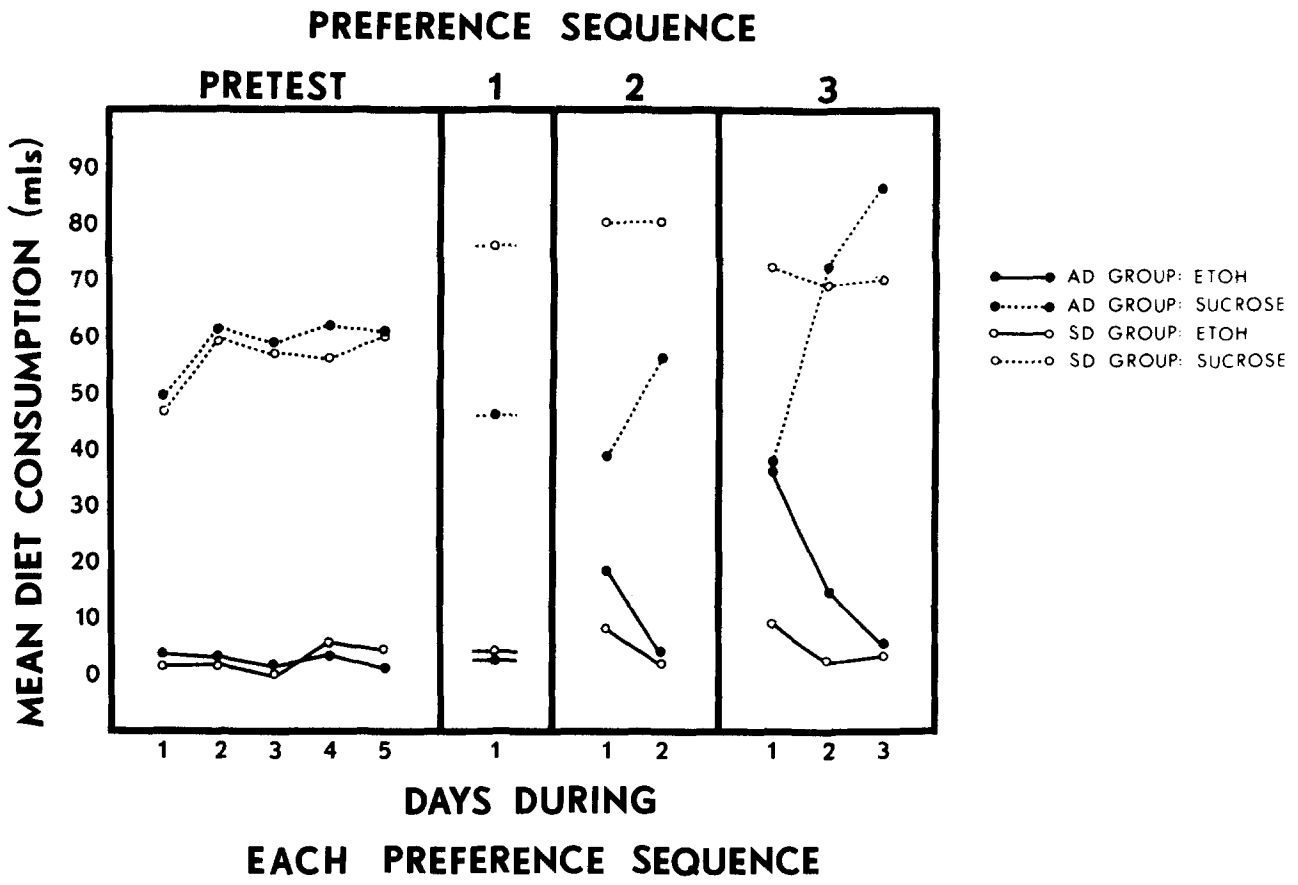

FIG. 2. Mean intake of alcohol and sucrose solutions during pretest and 3 free-choice tests in the alcohol diet group (AD) and sucrose diet group (SD).

However, there was great variability in alcohol consumption among rats in the AD group which was generally reflected in variability in the appearance of withdrawal symptoms. Since spontaneous convulsions were not observed during the initial preference test, the rats were tested for susceptibility to audiogenic convulsions [9] 9 hr after the initiation of the second preference test. A shaking of keys (3–5 seconds) near each home cage resulted in tonic-clonic convulsions in 5 rats from the AD group. Some of the convulsions were very severe, ending in respiratory arrest and death in 3 AD rats. A comparison of alcohol consumption during the pre-convulsive period (9 hr) revealed that rats which had convulsions consumed an average of 3.6 ml of alcohol diet (0.86 g/kg; SEM: 0.18). The 3 non-convulsing AD rats consumed 11.0 ml (2.5 g/kg; SEM: 0.80). These results indicate that alcohol consumption in these rats may have prevented the development of susceptibility to audiogenic convulsions. However, mild withdrawal symptoms (tail stiffening, broad-based gait, tremors) were observed in these rats, so that the dose of alcohol consumed was not sufficient to prevent withdrawal entirely. Finally since 3 rats died during convulsions, the 24 hr consumption values for the AD group reported in the third panel of Fig. 2 are based on 5 rats.

Preference Test #3:

An additional 2 rats from the AD group died during Sequence 3. These rats were found in postures, which have previously been characterized in rodents, as occurring as a result of death from alcohol withdrawal convulsions [5]. The amount of alcohol diet consumed by these rats during the time period preceding their death, indicated that these rats may have undergone spontaneous withdrawal episodes. We have previously observed spontaneous abstinence phenomena in both mice [6] and rats [9] using the liquid diet technique and have recently concluded (Hunter, Walker, Riley and Freund, in preparation) that this phenomenon may be etiologically similar to that previously reported in man [14] and primates [3]. Thus, the results from the AD group in Fig. 1 (Sequence 3) and Fig. 2 (Preference Test #3) are based on the surviving 3 AD rats.

As shown in Fig. 2, a marked increase in alcohol consumption in the AD group occurred during the final preference sequence. This increase was again only transient, but declined more slowly; reaching control levels by the third day. Mean ethanol diet consumption for the AD group on the first day was 36.0 ml (6.6 g/kg; SEM: 2.2) while the SD group consumed 9.2 ml (1.6 g/kg; SEM: 0.5).

Mean ethanol consumption for the AD group was mis-

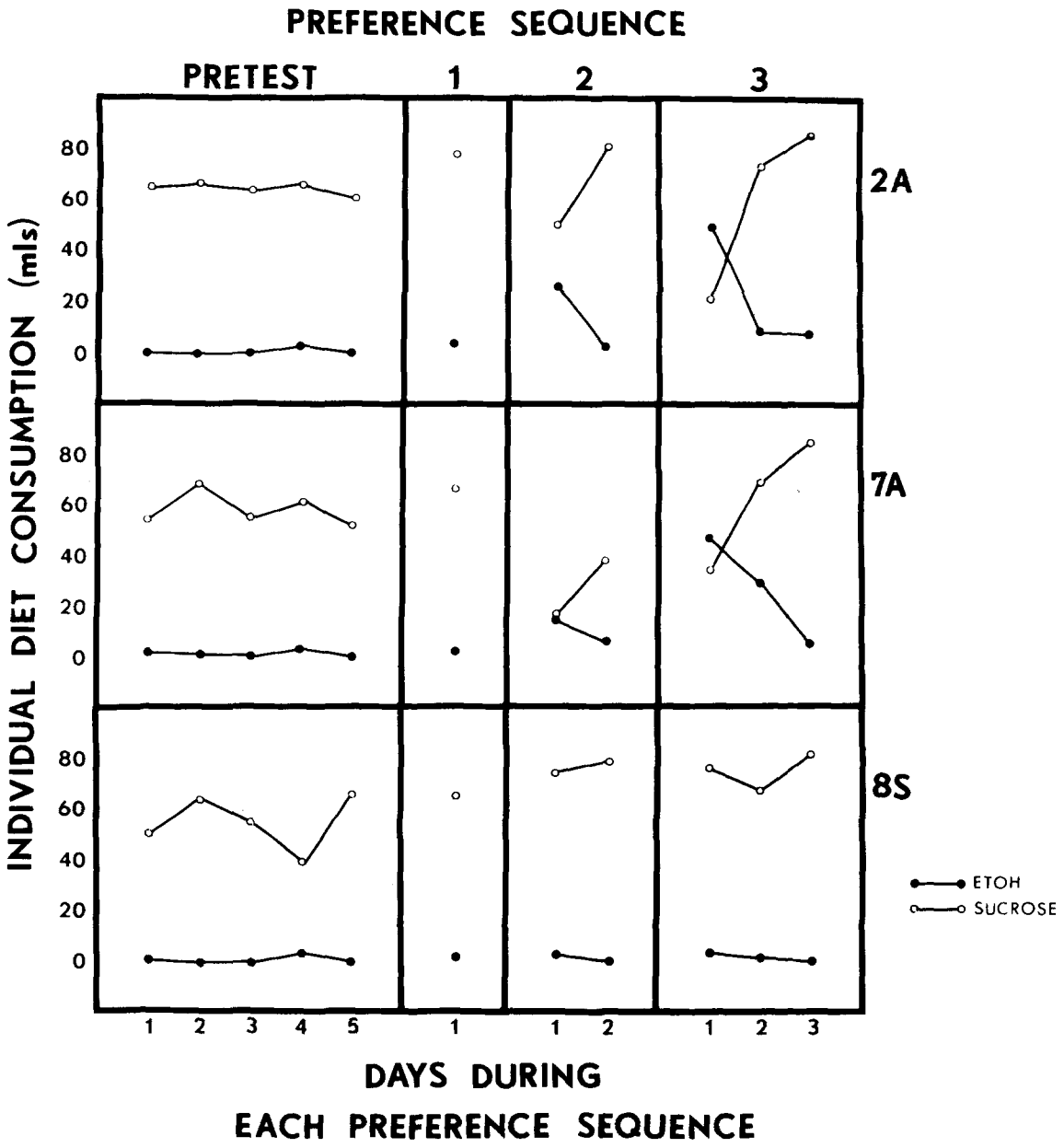

FIG. 3. Individual preference data from 2 animals in the AD group (2A and 7A) and 1 animal from the SD group (8S) during pretest and 3 free-choice tests.

leading, because only 2 of 3 AD rats showed a pattern of alcohol consumption that differed from previous preference sequences. Individual consumption data from these 2 rats, 2A and 7A, during all preference tests are shown in Fig. 3. Consumption data from a representative member of the SD group are also shown in Fig. 3. As shown in Fig. 3, 2A and 7A actually consumed more alcohol diet than sucrose diet on the first day of the final preference sequence. The dose of ethanol consumed was substantial, 8.8 and 8.9 g/kg respectively, and appeared to be pharmacologically significant, since these rats maintained behavioral intoxication during the first day. On the second day, when alcohol consumption began to decline, only mild withdrawal symptoms were observed.

The consumption of water during free-choice tests was remarkably low. Mean daily water consumption for both groups during pretests was 3.4 ml (1.3 ml/100g). A slight increase in water consumption was observed during the latter preference tests. For example, mean water consumption during the final preference sequence for both groups was 14.1 ml (3.0 ml/100g). Water consumption did not appear to be effected by the experimental treatments and the slight increase observed during the latter preference sequences may have reflected alterations in the fluid

requirements of older, heavier animals.

DISCUSSION

The results of the present experiment indicate that alcohol self-selection, in the face of fluids competing for taste and calories, can be dissociated from physiological dependence as indicated by withdrawal symptoms. Rats, made physically dependent as a result of the consumption of ethanol liquid diets for 20 days, failed to consume ethanol in a free-choice test despite the occurrence of severe withdrawal symptoms: results consistent to those previously reported in mice [4]. However, after a series of episodes of forced administration of ethanol diets, a substantial, but transient, increase in ethanol self-selection was observed. These results suggest the possibility that laboratory rodents may be capable of learning the association between alcohol and relief of withdrawal symptoms, thereby maintaining intoxication to avoid withdrawal illness. These conclusions must be interpreted cautiously as a result of several methodological considerations.

The increase in alcohol self-selection, observed after several withdrawal episodes, could be attributed to prolonged alcohol exposure, since a control group, receiving equal doses of ethanol and no withdrawal experience, was not used. Such a control group would be impossible to assemble using the liquid diet technique, in view of spontaneous withdrawal phenomena. Although we cannot unequivocally dismiss this possible interpretation of the results, it seems unlikely that prolonged alcohol exposure could have accounted for the increase in volitional alcohol consumption. Previous experiments, using forced administration of aqueous ethanol solutions, have failed to observe a significant change in volitional ethanol consumption after long-term exposure [3, 16, 20, 21]. Such free-choice tests are crucially dependent upon the solutions used. Thus, when the choice is offered between aqueous alcohol solutions and water, animals have appeared to consume pharmacologically significant volumes of alcohol. For example, Veale [23] has reported that rats, subjected to forced administration of alcohol solutions, which were gradually increased in concentration over an 11 month period, subsequently consumed substantial quantities of alcohol in a free-choice situation in which water was the alternative solution. However, in every experiment in which a fluid alternative, competing for taste and calories, has been offered, laboratory rodents have consistently discontinued alcohol self-selection [12,13]. Thus, despite numerous experimental attempts, it has not yet been unambiguously demonstrated that prolonged alcohol consumption per se, results in significant alterations in motivated alcohol-seeking behavior [19].

Most experiments concerned with alcohol self-selection, have utilized aqueous ethanol solutions in spite of the aversive taste properties [19]. The rationale for this procedure has been that alcohol should be consumed because of its pharmacological properties and not as a function of increased palatability resulting from adulteration with masking agents [19]. However, free-choice tests in which the aversive taste of ethanol is masked, may remain meaningful, if a more palatable, isocaloric solution is used as an alternative. In the present experiment the sucrose diets were unequivocally chosen over the alcohol diets during pretests, the rats consuming 96% of their calories from the sucrose diets. Furthermore we have found that rats allowed free access to sucrose diets, gain weight at rates higher than controls allowed laboratory chow and water ad lib (unpublished observations). Thus it appears that rats in the present experiment consumed the alcohol diets during free-choice tests as a result of the pharmacological properties of these diets and not because of palatability factors.

A final difficulty, inherent in all free-choice tests, is that an increase in preference for a given solution, may reflect an increase in the attractiveness of that solution or a decrease in the attractiveness of the alternative solution. Learned taste aversions have been the subject of a great deal of experimental attention [7,22]. Rats, in particular, rapidly learn to avoid a solution which is associated with illness. Such effects are dependent upon the temporal contiguity between the presentation of solutions and the induction of illness. In this regard, it has also been reported that rats show an increased preference for solutions which are paired with recovery from an illness [8]. It is apparent that both an approach response to the alcohol diet or a learned avoidance of the sucrose diet may have interacted in the increase in alcohol self-selection observed during the final preference sequence.

The results of the present experiment strongly support the conclusion that if laboratory rodents are to learn the association between alcohol and relief of withdrawal symptoms, a number of withdrawal episodes are required. Rats unequivocally rejected the alcohol diets during the first preference test, choosing instead to consume the more palatable sucrose diets. Even during the second preference sequence, a majority of rats in the AD group continued to reject the alcohol diets, despite severe withdrawal episodes including audiogenic convulsions leading to death. However, during the final preference test, 2 of the 3 remaining AD rats substantially increased their volitional ethanol consumption. Although only a small number of AD rats survived until the last preference sequence (as a result of withdrawal convulsions), the results, nevertheless appear to be quite powerful. The 2 AD rats initially consumed 1.4% of their calories from ethanol diets during pretests, while 65% of the calories were consumed from this diet during the first day of the final preference sequence. Furthermore, the dosage of ethanol consumed was approximately 9 g/kg, sufficient to maintain self-intoxication despite the presence of a more palatable alternative solution. These results support the conclusion that rats can learn the association between alcohol and relief of withdrawal symptoms.

The transient nature of the increase in volitional ethanol consumption suggests two interpretations. For example, it is conceivable that further withdrawal episodes may have resulted in a more prolonged alteration in alcohol preference. During the second free-choice test, a small increase in alcohol consumption was observed in the AD group, which returned to control levels on the second day. During the final preference sequence, the increase in alcohol consumption was not only more pronounced, but also more prolonged, returning to control levels on the third day. On the other hand, it is also conceivable that avoidance of withdrawal symptoms may not exert a powerful, prolonged influence on motivated alcohol-seeking behavior.

The role of physiological dependence in the development of psychological dependence remains to be elucidated. Several factors appear to be of importance. For example, Branchey *et al.* [1] have shown that rats, formerly made physically dependent on ethanol, reacquire signs of physical dependence more rapidly than control animals. In

this study [1] a significant dependence liability was demonstrated even though a 2-week abstinence interval separated the initial induction of ethanol dependence and tests of dependence reacquisition. The duration of this dependence liability is unknown, although similar reports on the reacquisition of alcohol tolerance have demonstrated effects persisting for as long as 3 months [10,11]. These results, together with those of the present experiment, indicate that physiological dependence may exert a more rapid effect on psychological dependence, after reinitiation of alcohol consumption following a period of abstinence. The role of physiological dependence in the reinitiation of alcohol consumption following a period of abstinence, however, remains to be investigated. Myers, *et al.* [18] found that monkeys did not increase volitional ethanol consumption following a period of alcohol intubation sufficient to produce physical dependence, when the free-choice test was conducted after an abstinence period of two days. A final factor that must be accounted for in any formulation concerning physiological dependence and motivated alcohol consumption is the observed cyclicity in alcohol drinking. Mello [14] reported that self-imposed abstinence periods in alcoholics, were often, but not invariably, observed coincident with gastritis. Such a peripheral mechanism could also account for spontaneous withdrawal episodes observed using the liquid diet technique, but could not explain abstinence phenomena found in monkeys trained to self-administer alcohol via intravenous infusion [3]. Thus it appears reasonable to assume that alcohol may possess some pharmacological property whereby continued alcohol consumption (during a given drinking sequence) may become more aversive than alcohol abstinence and accompanying withdrawal discomfort.

REFERENCES

1. Branchey, M., G. Rauscher and B. Kissin. Modifications in the response to alcohol following the establishment of physical dependence. *Psychopharmacologia (Berlin)* **22:** 314–322, 1971.
2. Cicero, T. J. and B. R. Smithloff. Alcohol oral self-administration in rats: Attempts to elicit excessive intake and dependence. In: *Alcohol Intoxication and Withdrawal: Experimental Studies,* edited by M. M. Gross. New York: Plenum Press, 1973, pp. 213–214.
3. Deneau, G., T. Yanagita and M. H. Seevers. Self-administration of psychoactive substances by the monkey. *Psychopharmacologia (Berlin)* **16:** 30–48, 1969.
4. Freund, G. Alcohol withdrawal syndrome in mice. *Archs. Neurol.* **21:** 315–320, 1969.
5. Freund, G. Alcohol, barbiturate, and bromide withdrawal syndrome in mice. In: *Recent Advances in Studies of Alcoholism,* edited by N. K. Mello and J. H. Mendelson. Washington, D.C.: U. S. Government Printing Office, 1971, pp. 453–471.
6. Freund, G. and D. W. Walker. Impairment of avoidance learning by prolonged ethanol consumption in mice. *J. Pharmac. exp. Ther.* **179:** 284–292, 1971.
7. Garcia, J., D. S. Kimeldorf and R. A. Koelling. Conditioned aversion to saccharin resulting from exposure to gamma radiation. *Science* **122:** 157–158, 1955.
8. Green, K. F. and J. Garcia. Recuperation from illness: Flavor enhancement for rats. *Science* **173:** 749–751, 1971.
9. Hunter, B. E., C. A. Boast, D. W. Walker and S. F. Zornetzer. Alcohol withdrawal syndrome in rats: Neural and behavioral correlates. *Pharmac. Biochem. Behav.* **1:** 719–725, 1973.
10. Kalant, H., A. E. Leblanc and R. J. Gibbins. Tolerance to, and dependence on, ethanol. In: *Biological Basis of Alcoholism,* edited by Y. Israel and J. Mardones. New York: John Wiley and Sons, 1971, pp. 235–269.
11. Leblanc, A. E., H. Kalant and R. J. Gibbins. Acquisition and loss of tolerance to ethanol by the rat. *J. Pharmac. exp. Ther.* **168:** 244–250, 1969.
12. Lester, D. and L. A. Greenberg. Nutrition and the etiology of alcoholism: The effect of sucrose, fat and saccharin on the self-selection of alcohol by rats. *Q Jl Stud. Alc.* **13:** 553–560, 1952.
13. Mardones, J., N. Segovia-Rigeulme, A. Hederra and F. Alcainno. Effect of some self-selection conditions on voluntary alcohol intake of rats. *Q Jl Stud. Alc.* **16:** 445–437, 1955.
14. Mello, N. K. Behavioral studies on alcoholism. In: *The Biology of Alcoholism: Vol II, Physiology and Behavior,* edited by B. Kissin and H. Begleiter. New York: Plenum Press, 1972, pp. 219–291.
15. Mello, N. K. A review of methods to induce alcohol addiction in animals. *Pharmac. Biochem. Behav.* **1:** 89–101, 1973.
16. Myers, R. D. Changes in learning, extinction, and fluid preferences as a function of chronic alcohol consumption in rats. *J. comp. physiol. Psychol.* **54:** 510–516, 1961.
17. Myers, R. D. and R. B. Holman. A procedure for eliminating position habit in preference-aversion tests for ethanol and other fluids. *Psychon. Sci.* **6:** 235–236, 1966.
18. Myers, R. D., W. P. Stoltman and G. E. Martin. Effects of ethanol dependence induced artificially in the rhesus monkey on the subsequent preference for ethyl alcohol. *Physiol. Behav.* **9:** 43–48, 1972.
19. Myers, R. D. and W. L. Veale. The determinants of alcohol preference in animals. In: *The Biology of Alcoholism, Vol II, Physiology and Behavior,* edited by B. Kissin and H. Begleiter. New York: Plenum Press, 1972, pp. 131–168.
20. Richter, C. P. Alcohol, beer and wines as foods. *Q. Jl Stud. Alc.* **14:** 525–539, 1953.
21. Rick, J. T. and C. W. M. Wilson. Alcohol preference in the rat: Its relationship to total fluid consumption. *Q. Jl Stud. Alc.* **27:** 447–458, 1966.
22. Rozin, P. Central or peripheral mediation of learning with long CS-US interval. *J. comp. physiol. Psychol.* **67:** 421–429, 1969.
23. Veale, W. L. Ethanol selection in the rat following forced acclimation. *Pharmac. Biochem. Behav.* **1:** 233–235, 1973.
24. Walker, D. W. and G. Freund. Impairment of shuttlebox avoidance learning following prolonged alcohol consumption in rats. *Physiol. Behav.* **7:** 773–778, 1971.
25. Wikler, A. On the nature of addiction and habituation. *Br. J. Addict.* **57:** 73–79, 1961.