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BRIEF COMMUNICATION

Lack of Tolerance Development to the Dipsogenic Actions of Barbital[1]

ROGER P. MAICKEL AND GREG J. MALONEY[2]

Department of Pharmacology, Medical Sciences Program, Indiana University, Bloomington, IN 47401

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MAICKEL, R. P. AND G. J. MALONEY. *Lack of tolerance development to the dipsogenic actions of barbital.* PHARMAC. BIOCHEM. BEHAV. 2(3) 431–434, 1974. – Chronic dosage of rats with barbital produced a dipsogenic state with overhydration. No tolerance could be seen to this effect in fourteen days; on withdrawal of drugs, the water intake of the rats fell to below normal levels, on a weight basis. When daily intake of fluid was restricted during the period of barbital dosage, overhydration did not occur and drinking during the withdrawal phase was increased.

Barbital Drug induced drinking Tolerance

SCHMIDT and co-workers [11–19] have studied the effects of a variety of barbiturates on water consumption induced in rats by various means. In general, the compounds had a dipsogenic action, a finding corroborated by us for a variety of depressant drugs [7]. In addition, this laboratory recently reported an exhaustive study of factors involved in the dipsogenic actions of barbital [6].

Schmidt, *et al.* [16,18] administered phenobarbital to deprived rats daily for 30 days and measured water intake; no indication of tolerance to the dipsogenic effect of the drug was observed. Upon withdrawal of the drug from these animals, and also upon withdrawal of a group given drug for 14 days, the rats consumed less water than a saline control group for several days. This was later interpreted as the development of a hypersensitivity to the dipsogenic effect after a 15 day phenobarbital treatment and a 10 day withdrawal [19]. As evidence of hypersensitivity the authors cited the fact that the response to the second phenobarbital treatment, that is, after 15 days treatment and 10 days withdrawal, was greater than the initial response to the same dose.

The present paper reports on the effects of chronic daily dosage of barbital on the fluid intake of rats with restricted or unlimited access to water.

MATERIALS AND METHOD

Adult (280–290 g) male, Sprague-Dawley rats (Murphy Breeding Laboratories, Plainfield, Indiana) were used in all experiments. The animals were maintained on tap water and Purina Rat Chow ad lib for at least 1 week after arrival in the laboratory. All solutions for injection were made in glass distilled water such that a dose volume of 0.1 ml per 100 g body weight contained the desired dose; all injections were given intraperitoneally, 15 min prior to placing the animals in the drinking cages.

The procedures used to measure deprivation-induced water consumption were basically those of Gerald and Maickel [5]. Rats were placed in cages, 7 × 7 × 14 in., identical to the home cages of the animals. The cages were suspended in individual compartments of a sound-proofed box with uniform fluorescent lighting. Constant circulation of air by blowers maintained uniform temperature in the compartments; the noise level of blowers served as a white noise. Each cage contained a drinking tube connected to an external 50 ml buret filled with distilled water at the start of each test run and stoppered. As the animal consumed water, the change in volume was measured visually to the nearest 0.1 ml. The front door of each compartment was equipped with an eye-piece lens to permit visual observation of the rats without disturbing their behavioral performance. For at least 1 week prior to testing drug effects, rats were deprived of water daily for 23 hr prior to testing, then placed in the drinking cages and allowed to drink for 1 hr. Food was available ad lib in the home cages but was not available in the drinking cages. In the restricted intake studies, rats were given only a fixed volume of water, 14.0 ml.

Drug trials were started only after the animals demonstrated stable baselines (less than 5% daily variation) of

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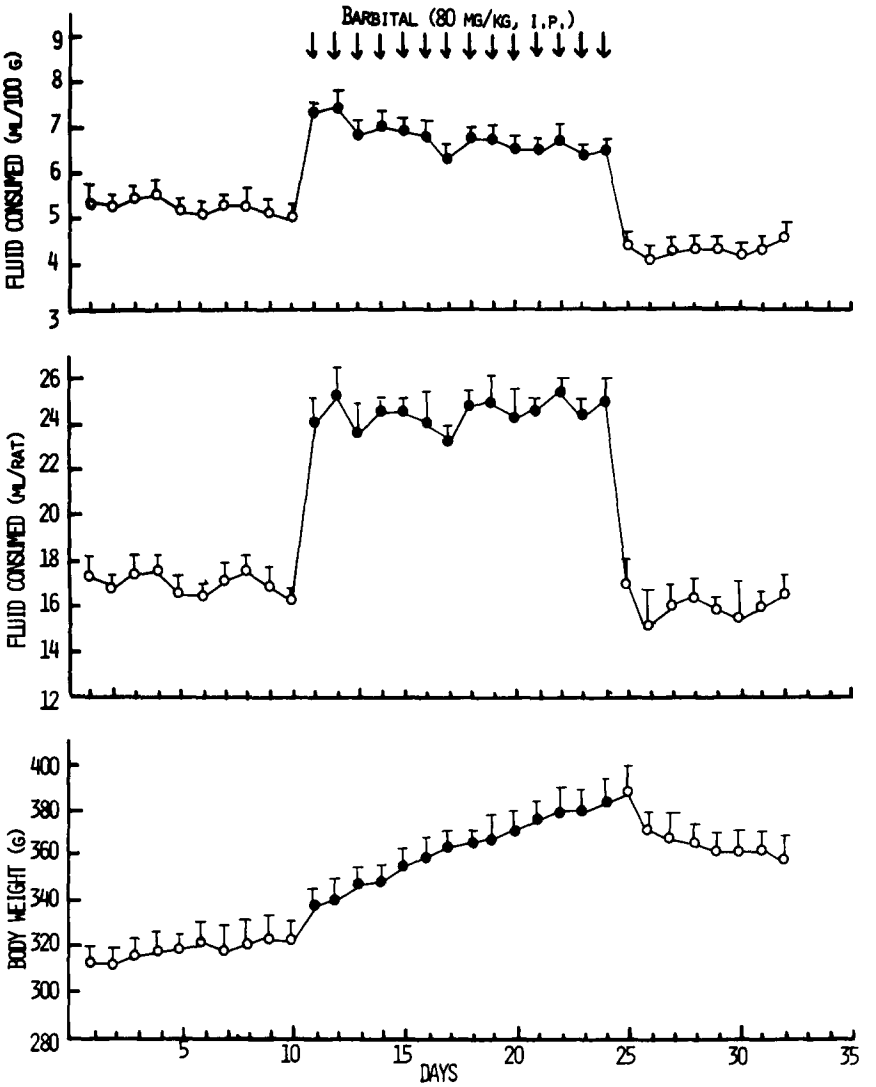

FIG. 1. Effects of barbital on fluid consumption-unrestricted intake. Each symbol is the mean of values obtained from 8 rats as described in Materials and Method. Vertical bars indicate 1 S.D. Solid circles indicate days of dosage with barbital (80 mg/kg, i.p.); open circles are placebo dosage.

water intake. The schedule for drug studies was arranged so that the rats were run daily. Water intake was recorded at 15, 30, and 60 min of the consummatory sessions, the greatest proportion of drinking occurred in the first 15 min in all cases.

Barbital sodium was used in all experiments, dosages are reported as weight of barbital administered in mg/kg body weight. Groups of 8 rats were run in each test system; data are reported as mean ± S.D.

RESULTS

Effect of Chronic Daily Dosage of Barbital on Fluid Consumption of Rats with Unlimited Volume Available

As shown in Fig. 1, chronic barbital (80 mg/kg, i.p.) increased daily fluid consumption above baseline for the duration of drug administration. There is a trend suggesting development of tolerance to the dipsogenic effect over the 14 days of drug treatment, seen when consummatory volume was plotted as ml/100 g body weight. The animals gained an average of 100 g of body weight during the course of this experiment. Substitution of saline for barbital resulted in a decrease in fluid consumption on the second day after drug withdrawal with a return to normal on the third and subsequent days. A weight loss of 18 g was also observed during this withdrawal period. Of particular interest is the fact that administration of a single dose of barbital on the ninth day after drug withdrawal (Day 33) gave a normal dipsogenic response (Table 1).

Effect of Chronic Daily Dosage of Barbital on Fluid Consumption of Rats with Limited Volume Available

Figure 2 shows the effect of limiting fluid intake during the drug treatment period of 14 ml/rat/day. Here, overdrinking and the weight gain during the drug period are

TABLE 1

DIPSOGENIC EFFECTS OF BARBITAL BEFORE AND AFTER CHRONIC TREATMENT

Day of Schedule	Drug	Volume Consumed total ml	Volume Consumed ml/100 g
1–10	Saline	16.9 ± 1.0*	5.3 ± 0.4*
11–24	Barbital	24.5 ± 1.7*	6.7 ± 0.6*
25–32	Saline	16.0 ± 1.5*	4.3 ± 0.4*
33	Barbital	23.2 ± 1.8†	6.4 ± 0.7†

Data are presented as volume consumed by 8 animals. Barbital (80 mg/kg, i.p.) was given as described in Materials and Methods.
*mean ± S.D. of 8 animals per day for number of days indicated
†mean ± S.D. of 8 animals on a single day

prevented with the result that now there is no withdrawal decrease in fluid intake nor weight loss. The period of withdrawal shows a significantly greater fluid intake by the animals.

DISCUSSION

From a consideration of the data presented in this paper, it seems likely that the earlier work of Schmidt and coworkers [16, 18, 19] suffers from several defects. First, no recognition was made of the large weight gain during the period of the study. It is possible that the increased intake attributed to hypersensitivity after a period of drug trials [19] may merely reflect the fact that heavier animals consume more fluid. When fluid consumption by our animals is expressed in ml consumed/100 g body weight, the daily intake decreases slightly over the drug period of 14 days. A transient weight loss and decrease in fluid consumption are present during the withdrawal period as previously reported by others [14,15].

Schmidt *et al.* [12,15] also considered the chronic overhydration of the animals during the drug period as inconse-

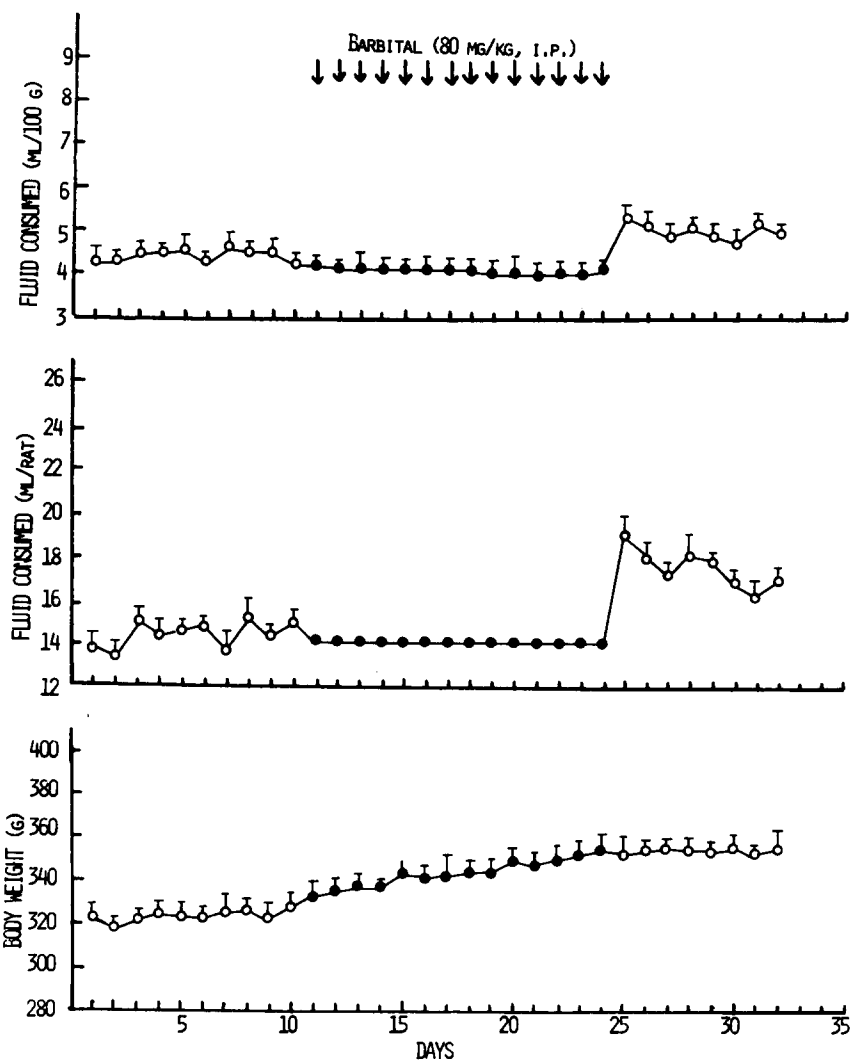

FIG. 2. Effects of barbital on fluid consumption-restricted intake. Each symbol is the mean of values obtained from 8 rats as described in Materials and Method. Vertical bars indicate 1 S.D.; no S.D. is seen for total fluid consumed on Days 11–24 when fluid was restricted. Solid circles indicate days of dosage with barbital (80 mg/kg, i.p.); open circles are placebo dosage.

quential to the characteristics of the withdrawal state. In the present study, when overhydration was prevented, the decrease in fluid consumption and body weight during withdrawal did not occur. The increase in fluid intake over baseline on the first day of withdrawal may reflect the presence of barbital in the animal since the drug is not appreciably metabolized and is excreted slowly [6]. It is unlikely, however, that this drug carryover would explain the increased consumption on subsequent days.

It is reasonable, in light of the changes in neurosecretory material in the neurohypophyseal system during overhydration [1], that prolonged overhydration in itself could lead to a decrease in fluid consumption upon removal of the stimulus for overdrinking. The animal is in a state of positive water balance and will restore his fluid balance by consuming less fluid. The post-drug weight loss appears directly related to the decrease in fluid consumption since the animals did not lose weight if they were prevented from overdrinking prior to withdrawal. The dipsogenic response during the post-drug treatment period by the limited-volume animals may reflect an alteration in fluid balance systems induced by the drug treatment.

A direct central action upon neurons involved in the regulation of water balance may cause the dipsogenic effect by depression of inhibiting centers. Animals with lesions in the septal area and in the anterior hypothalamus show an increased fluid consumption [2,10]. These drug effects could be due to chemical lesions via a depression of the septal area or hypothalamus. Interference with the hypothalamus neurohypophyseal system may also be involved in the dipsogenic action. Barbiturates are known to release ADH from the posterior pituitary [3]. The depression of osmoreceptor cells in the CNS, possibly those near the ADH secreting neurons, may alter an animals preception of his actual state of hydration and cause him to continue drinking longer than usual. The consummatory act itself may become pleasurable if the drug has depressed inhibitory brain areas.

In the rat satiation precedes restoration of the osmotic imbalance and comes about via impulses from nerves in the oral cavity traveling along cranial nerves to brain regions involved in fluid consumption [4]. If drugs were to interfere with the transmission of information as to the nature and amount of the fluid the rat is consuming, the animal might ingest larger amounts or cease earlier depending on the nature of the interference. Evidence for the involvement of the peripheral nervous system [5] suggests that depression of these neurons may interfere with the regulation of fluid intake. Another peripheral action could be drug interaction with the renin-angiotensin system. The release of ADH by barbiturates could also be a manifestation of the mediation of angiotensin since angiotensin itself releases ADH [8].

REFERENCES

1. Andersson, B. The effect of long term periods of continuous hydration on the neurosecretory material in the hypothalamus of the dog. *Endocrinology* **15:** 332–338, 1957.
2. Blass, E. M. and D. Hanson. Primary hyperdipsia in the rat following septal lesions. *J. comp. physiol. Psychol.* **70:** 87–93, 1970.
3. Bodo, R. L. and K. F. Prescott. The antidiuretic action of barbiturates and the mechanism involved in this action. *Pharmac. exp. Ther.* **229:** 223–233, 1945.
4. Deutsch, J. A. Hunger and thirst. In: *Physiological Psychology*, edited by J. A. Deutsch and D. Deutsch. Homewood, Ill.: Dorsey Press, 1966.
5. Gerald, M. C. and R. P. Maickel. Evidence for peripheral cholinergic components in thirst-induced water consumption. *Int. J. Neuropharm.* **8:** 337–346, 1969.
6. Maickel, R. P. and G. J. Maloney. Effect of barbital on deprivation-induced water consumption by rats. *Physiol. Behav.* **8:** 1175–1178, 1972.
7. Maickel, R. P. and G. J. Maloney. Effects of various depressant drugs on deprivation-induced water consumption. *Neuropharmac.* **12:** 777–782, 1973.
8. Malvin, R. Possible role of the renin angiotensin system in regulation of antidiuretic hormone secretion. *Fedn Proc.* **30:** 1387–1394, 1971.
9. Maynert, E. W. and H. B. van Dyke. The metabolism of barbiturates. *Pharmac. Rev.* **1:** 217–242, 1949.
10. Richter, C. F. and J. F. Eckert. Further evidence for the primary of polydipsia in diabetes insipidus. *Am. J. Physiol.* **113:** 578–581, 1935.
11. Schmidt, H., Jr. Pentobarbital facilitation of water ingestion in the albino rat. *J. comp. physiol. Psychol.* **51:** 26–28, 1958.
12. Schmidt, H., Jr. Variations of water ingestion: The response to barbiturates. *Prog. Brain Res.* **16:** 263–284, 1965.
13. Schmidt, H., Jr. Alterations of central thirst mechanisms by drugs. *Ann. N.Y. Acad. Sci.* **157:** 962–976, 1969.
14. Schmidt, H., Jr. and L. Dry. Comparison of phenobarbital and pentobarbital actions upon water ingestion. *J. comp. physiol. Psychol.* **56:** 179–182, 1963.
15. Schmidt, H., Jr. and L. Dry. Dose and time as variables in barbiturate action upon water ingestion. *Am. J. Physiol.* **204:** 817–820, 1963.
16. Schmidt, H. and K. Kleinman. Effects of chronic administration and withdrawal of barbiturates upon drinking in the rat. *Archs int. Pharmacodyn. Ther.* **151:** 142–149, 1964.
17. Schmidt, H., Jr. and S. J. Moak. Some drug effects influencing barbiturate facilitation of water ingestion. *Am. J. Physiol.* **196:** 307–310, 1959.
18. Schmidt, H., K. Kleinman and T. Douthitt. Enhancement of the drinking response to phenobarbital after a previous course of barbiturates. *Physiol. Behav.* **2:** 265–271, 1967.
19. Schmidt, H., K. Skinner and K. Kleinman. Phenobarbital withdrawal weight loss is in part independent of ingestion. *Archs int. Pharmacodyn. Ther.* **160:** 241–247, 1966.