AGE SENSITIVITY TO BARBITURATES AND ITS DEPENDENCE ON THE DEVELOPMENT OF THE HYPOPHYSEAL- ADRENAL SYSTEM

I. V. Markova

Department of Pharmacology (Head-V. M. Karasik, Active Member of the AMN SSSR) of the Leningrad Institute of Pediatric Medicine
(Presented by V. N. Chernigovskii, Active Member of the AMN SSSR)
Translated from Byulleten' Eksperimental'noi Biologii i Meditsiny, Vol. 50, No. 10, pp. 87-91, October, 1960
Original article submitted December 22, 1959

According to the literature [5] and to our own data [2,3], newborn animals and those at early stages of postnatal development are considerably more sensitive than mature animals to narcotics. Since this greater sensitivity of very young animals has been observed with the use of both medinal [barbital sodium] and ether, narcotics which are not broken down in the organism, it cannot, as some authors believe [5], be held to depend on the rate of the drug's detoxication in the organism.

According to the literary data [11, 14], extirpation of the adrenal glands prolongs and deepens narcosis in rats and mice. This prolongation is due to the higher barbiturate content in the brain of adrenalectomized animals [14].

Because the function of the hypophyseal-adrenal system is insufficient during the early stages of postnatal development in rats and other animals [7,9,12], these very young animals can be compared with mature animals from which the adrenohypophysis has been removed.

This led us to propose that the the higher sensitivity of newborn animals to barbiturates might be due to their underdeveloped hypophyseal-adrenal system. This hypothesis could be confirmed by experimentally determining whether the removal of the adrenal glands from baby rats (in which the hypophyseal-adrenal system is not yet fully developed) would have a lesser effect on the duration of the narcotic influence than in the case of mature rats.

According to the data of Hungarian authors [12], the hypothalamus does not begin its regulation of the hypophyseal-adrenal system function in rats until the animals are two weeks old; therefore, one can propose that, from this time on, gluco-corticoids, hitherto not secreted in physiologically adequate quantities, increase the resistance of the vascular wall, thereby inhibiting penetration of the barbiturates into the central nervous system and, in this way, decreasing their narcotic effect. The greater permeability of the blood-brain barrier in newborn rats is attested to in the works of L. S. Shtern [6], Bakay [8], et al.

In this work, we determined: 1) the effect of adrenalectomy on the narcotic effect of hexenal and 2) the content of medinal in the brain of rats of various ages.

EXPERIMENTAL METHODS

The first series of experiments were carried out in 75 rats from three age groups (adult males, six-week-old males and females and month-old males and females from the same nursery). Adrenalectomy was performed from the dorsal aspect. To keep the animals alive, we administered cortisone (Roussel) before the operation in a dose of 30-50 γ/1 g of animal weight. After the operation, the animals were fed only water and a 0.9% sodium chloride solution. After 1-5 days, we again determined the duration of the lateral (reclining) posture induced in the rats by hexenal, intraperitoneally or subcutaneously injected in a dose of 80-150 γ/g and compared this with the duration of the lateral posture in the control rats. The individual animals were usually given the barbiturate injections only once every 5 days. The observation period was limited to 2-3 weeks by the fact that most of the adrenalectomized animals died within this period.

A second series of experiments was performed on 250 rats from one nursery. In the three-week-old or younger age groups, we used both male and female rats in the experiment; the older rats used were all females. The rats of each age group (except for the mature age group) were all used in the experiment the same day. Every rat was intraperitoneally injected with a uniform dose of medinal (200 γ/g each). Medinal was chosen for two reasons: one, because it does not break down in the organism, and two, because the medinal dose inducing the lateral posture in mature rats can be tolerated by newborn rats. The rats were decapitated at fixed time intervals (10, 20, 40, 60 and 150 min) after the medinal injection, and the medinal content of the brain was determined according to the method described by

TABLE 1. Medinal Content in Brain of Experimental Rats of Different Ages after Its Intraperitoneal Injection

Age of rats	Amount of medinal (in γ/100 mg of dry brain weight)				
	Time after intraperitoneal medinal injection				
	10 min	20 min	40 min	60 min	150 min
Newborn	117.4±4.4	184.2±8.4	165.6±2.4	167.2±11.6	170.2±11.3
1 week	168.5±10.8	171.4±7.7	195.3±13.3	253.3±22.4	229.4±32.5
2 weeks	49.6±3.1	130.2±7.7	142.6±7.1	107.5±4.6	90.2±4.4
3 weeks	91.3±1.17	108.4±3.9	109.9±5.7	111.9±4.5	132.7±1.7
4 weeks	60.1±6.5	114.5±3.2	98.0±5.4	103.4±2.3	87.4±4.2
1 1/2 mos	38.2±6.7	128.8±4.3	147.6±9.0	94.5±2.1	71.2±3.7
Adult	66.4±8.7	74.5±6.8	112.6±9.1	99.3±10.0	66.5±8.9

Goldschmidt and co-workers [13]. Five animals were sacrificed for each determination of the medinal content of the brain. In the experiments with newborn animals, the brains of two rats were used for each determination. The medinal was extracted from the cerebral hemispheres (barbiturates are known to disperse rather evenly in the brain [16]). Since the water content of the brain varies in rats of different ages [10], one would expect different quantities of medinal to be extracted. However, the addition of medinal to and its subsequent extraction from brain homogenates taken from newborn and adult rats showed that 80% of the medinal added was extracted.

EXPERIMENTAL RESULTS

Fig. 1 illustrates the data from the experiments with extirpation of the adrenal glands, showing the change in the ratio of the average duration of the lateral posture in the experimental rats (M_1) to that in the control (M_2). It is evident from the graph that removal of the adrenal glands sharply prolonged hexenal narcosis in the case of adult males; 14 days after the operation, the duration of narcosis was $6\frac{1}{2}$ times longer than the control. According to the literary data [11, 19], it has been observed to become 20-360 times longer. In six-week-old rats, extirpation of the adrenal glands had less influence on the duration of the lateral posture, and this influence was more marked in the males than in the females. In month-old females, only a slight (statistically unreliable) prolongation of the lateral posture was observed in some of the experiments (on the 5th and 15th days) after adrenalectomy. In month-old males, no changes in the duration of the lateral posture were observed after extirpation of the adrenal glands.

Table 1 and Fig. 2 illustrate the medinal content in the brains of rats of different ages. The data cited indicate that the greatest amounts of medinal were found in the brains of newborn and week-old rats. The medinal content in the brains of rats two or more weeks old was smaller, and by the 3rd-4th weeks approximated that found in adult rats. In almost all the age groups, the maximal content of medinal in the brain was observed 20-40 min after its injection. The week-old group of rats were, however, an exception, as the

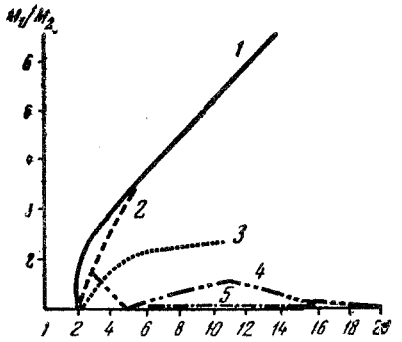

Fig. 1. Change in duration of the lateral posture induced by hexenal in adrenalectomized rats of different ages. 1) Adult males; 2) six-week old males; 3) six-week old females; 4) month-old females; 5) month-old males.

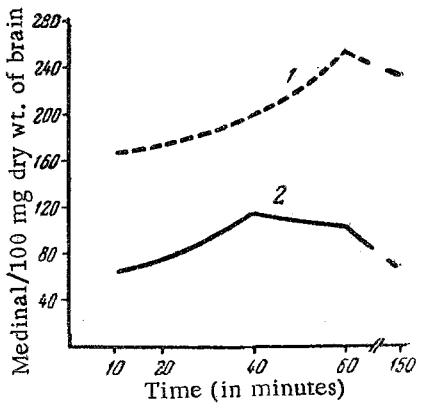

Fig. 2. Medinal content in brains of experimental rats after intraperitoneal injection of 200γ medinal per 1 g of weight. 1) Week-old rats; 2) adults.

TABLE 2. Lateral Posture in Rats of Different Ages with Intraperitoneal Injection of Medinal

Age of rats	Ratio of number of rats in lateral posture (numerator) to number of rats used in expt. (denominator)				
	Time after medinal injection				
	10 min	20 min	40 min	60 min	150 min
Newborn	8/8	10/10	10/10	10, 10	10/10
1 week	4/4	8/8	8/8	8/8	4/4
2 weeks	4/5	5/5	5/5	4/5	5/5
3 weeks	0/5	4/5	5/5	4/5	5/5
4 weeks	0/5	2/5	5/5	5/5	5/5
1 1/2 mos	0/5	0/5	4/5	5/5	5/5
Adult	0/5	2/5	10/5	5/5	3/5

medinal content of their brains continued to increase for one hour.

The speed with which the rats went into the lateral posture corresponded to the rate of medinal accumulation in the brain. Table 2 gives the number of animals in each group found in the lateral posture. Up to the age of two weeks, as Table 2 shows, the lateral posture developed very quickly (within 10 min) and continued throughout the $2\frac{1}{2}$ hours of observation. The lateral posture was slower to occur in rats aged three weeks or more, and narcosis ceased in some of the adult rats towards the end of the observation period.

According to the literary data, the hypophyseal-adrenal system does not mature in rats until the animal is 2-3 weeks old [7,9]. This opinion was based on tests: decrease in the ascorbic acid content of the adrenal glands or in the blood eosinophils in response to stimulations inducing a condition of stress. The results of our experiments, however, indicate that the function of the hypophyseal-adrenal system is still insufficient in rats a month old, as at this age, removal of the adrenal glands had hardly any influence on the duration of the lateral position induced in the rats by hexenal. The reason for the variance of our observations with the literary data may be that the various systems and organs of the body acquire the ability to react to secreted corticoids at different stages in the animal's development, the peripheral organs (the hematopoietic organs and the adrenal glands) acquiring this ability earlier than does the central nervous system.

Now, how can the maturation of the hypophyseal-adrenal system affect the reactivity of the developing organism to narcotics, in particular to barbiturates?

Gluco-corticoids are known to raise the tonus of the central nervous system. This is evinced by the increase in the "electroconvulsive threshold" of experimental animals and in the greater excitability of the central nervous system in people who have taken cortisone or ACTH for a long time [19]. Therefore, because of the deficiency of gluco-corticoids in very young animals, it is possible that the tonus of their central nervous system is comparatively low, and this too would promote barbiturate narcosis.

Gluco-corticoids are also known to strengthen the resistance of the vascular walls [4, 17], and a deficiency of these substances could increase the penetration of various substances, barbiturates included, into the central nervous system.

The fact that we found a greater amount of medinal in the brains of the very young experimental rats confirms the hypothesis of its greater penetration into the brain tissue. The medinal content is lower when the hypophyseal-adrenal system of the rats is mature [12]. At about this time, the sensitivity of the young rats to medinal becomes equal to that of adult rats [2,3]. Also at this time, ACTH injections begin to have an abbreviating effect on barbiturate narcosis [1].

Hexenal is known to penetrate the central nervous system more strongly and quickly than medinal [15]. It is only natural, therefore, that hexenal penetration is not affected as soon as that of medinal by the increase in vascular wall resistance. A considerably greater degree of development is required to inhibit hexenal penetration into the brain of a growing animal.

Yugoslav researchers have recently published an observation [18] to the effect that the hypophyseal-adrenal system of newborn animals, because of the transition to life under new conditions, is in a state of hyperfunction and therefore cannot react to additional stress. Our data do not favor this hypothesis. If the hypophyseal-adrenal ystem of newborn animals were in a state of hyperfunction, their sensitivity to narcotics should be less than that of adults; in the latter, injections of ACTH, which intensifies the function of the adrenal cortex, diminish the narcotic effect of barbiturates [19]. Newborn animals, however, are extraordinarily sensitive to narcotics [2, 3, 5]. If the hypophyseal-adrenal system were in a state of hyperfunction, extirpation of the adrenal glands from infant animals would acutely intensify and prolong narcosis. Even in animals a month

old, however, adrenalectomy does not affect the narcotic action of barbiturates.

SUMMARY

Removal of the adrenals in adult rats markedly prolongs the lateral posture induced by the intraperitoneal injection of hexenal; in one-month-old rats, the removal of the adrenals exerts no such effect. After the intraperitoneal injection of 200 γ of medinal per g of body weight, spectrography revealed a high medinal content in the brains of the newly born and one-week-old rats; in two-week-old rats, the content of medinal in the brain decreases, and after 3 – 4 weeks, it reaches the level observed in adult rats. The greater penetration of medinal into the brain of very young rats is believed to be due to the underdevelopment of the hypothalamohypophyseal-adrenal system. These data shed light on the earlier investigations: after three weeks of life the sensitivity of rats to medinal becomes equal to the tolerance of the adult animals, and from the same age, injections of ACTH begin to shorten the duration of barbiturate narcosis.

LITERATURE CITED

1. I. V. Markova, Byull. Éksp. Biol. Med. 47, 3, 57 (1959). *
2. I. V. Markova, in: Transactions of the IX Convention of the All-Union Society of Physiologists, Biochemists and Pharmacologists [in Russian] (1959), Vol. 2, p. 172.
3. I. V. Markova, Fiziol. Zhur. 68, 12, 1485 (1959).
4. G. L. Mednik, Problemy Endokrinol. i Gormonoterap. 3, 2, 40 (1957).
5. T. N. Reshtnikova, Author's Abstract of Candidate's Dissertation: The Features of Barbiturates' Effect on Young Animals [in Russian] (Tashkent, 1956).
6. L. S. Shtern, in: The Blood-Brain Barrier [in Russian] (Moscow, 1935).
7. I. A. Eskin, Pat. Fiziol. i Éksp. Terap. 1, 4, 10 (1957).
8. L. Bakay, in: Blood-Brain Barrier (Springfield, Illinois, USA, 1956)
9. H. W. Deane, in Collection: Adrenal Function in Infants and Children. A. symposium N. J. (London, 1954) No. 4.
10. H. H. Donaldson, J. Compar. Neurology and Psychol. 26, 2, 119 (1910).
11. F. Eichholtz, R. Hotovv, E. Collischonn and H. Kauer, Arch. Exp. Path. and Pharmakol. 207, 5, 397 (1949).
12. E. Endröczi, J. Szalay and K. Lissak, Endokrinologie 34, 5 – 6, 331 (1957).
13. S. Goldschmidt, W. Lamprecht and E. Helmreich, Z. f. Physiol. Chemie 292, 3 – 5, 125 (1953).
14. A. Komiya and K. Shibata, J. Pharm. and Exp. Therap. 117, 1, 68 (1956).
15. L. C. Mark, J. J. Burns, L. Brand, C. I. Campomanes, N. Trousof, E. M. Papper and B. B. Brodie, J. Pharm. and Exp. Therap. 123, 1, 70 (1958).
16. L. C. Mark, J. J. Burns, C. I. Campomanes, S. H. Ngai, N. Trousof, E. M. Papper and B. B. Brodie, J. Pharm. and Exp. Therap. 119, 1, 35 (1957).
17. V. Menkin, Am. J. Physiol. 129, 3, 691 (1940).
18. K. Milkovic and S. Milkovic, Endokrinologie, 37, 5 – 6, 301 (1959).
19. D. M. Woodbury, Pharmacological Reviews 110, 2, 275 (1958).

* Original Russian pagination. See C. B. translation.