ROLE OF THE POSTERIOR HYPOTHALAMIC NUCLEI IN DEVELOPMENT OF EXPERIMENTAL ALLERGIC ENCEPHALOMYELITIS AND POSTDIPHTHERITIC POLYNEURITIS

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Electrolytic destruction of the posterior hypothalamic nuclei, or pharmacological blocking of these nuclei with chlorpromazine and reserpine was carried out on guinea pigs poisoned with sublethal doses of diphtheria toxin or immunized with an encephalitogenic mixture. Blocking of the posterior hypothalamic nuclei delayed the development of paralysis in diphtheria and allergic encephalomyelitis if the animals retained adequate osmoregulatory reflexes to hydration and if their production of antibodies against nerve tissue was sharply inhibited.

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The development of several autoallergic diseases, including experimental nephroso-nephritis, experimental allergic encephalomyelitis (EAE), postdiphtheritic polyneuritis (PDP), and rejection of skin homografts are preceded by changes in the reflex regulation of water and salt metabolism and by an increase in the antibody-producing power of the body. These changes are connected with increased activity of the posterior hypothalamic centers which regulate water and salt metabolism and also immunogenesis [4, 6, 12]. Electrolytic destruction or chemical blocking of the posterior hypothalamic nuclei prevents disturbance of the osmoregulatory reflexes and inhibits the development of allergic reactions of the delayed type during, for example, rejection of recent skin homografts [6, 3].

The object of the present investigation was to study the effect of disturbance of the functions of the posterior hypothalamic nuclei on the development of EAE and PDP.

EXPERIMENTAL METHOD

Experiments were carried out on guinea pigs of both sexes weighing 300±20 g. Postdiphtheritic paralysis was produced by Frick's method [10] by injecting sublethal doses of diphtheria toxin, namely 0.5 MLD/300 g body weight (1 MLD for a guinea pig = 0.003 ml of the liquid toxin). EAE was produced by subcutaneous injection of 0.025 mg of encephalitogenic mixture (spinal cord homogenate with Freund's adjuvant, 1:1.5) into each plantar pad twice on alternate days. Electrolytic destruction of the posterior hypothalamic nuclei was performed with a direct current of 1 mA, 30 sec in duration, by means of a stereotoxic apparatus using the coordinates of Blobel and co-workers [7]. The criterion of successful destruction of the posterior nuclei was increased excretion of sodium during the first 2 h after the operation [12]. This function test accurately reflects the effectiveness of destruction of the posterior hypothalamic centers controlling water and salt metabolism and immunogenesis [6]. Pharmacological blocking of the adrenergic structures of the posterior hypothalamus was produced by injection of chlorpromazine or reserpine in doses abolishing the abnormal salt excretion by the kidneys of the experimental animals (daily dose of chlorpromazine 7.5 mg/kg, of reserpine 0.25 mg/kg). The state of function of the hypothalamo-hypophyseo-adrenal system (HHAS) was judged from the reflex liberation of the hormones vasopressin and aldosterone, which can easily be tested by measuring changes in the salt-excreting ability of the kidneys [2, 9]. For

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TABLE 1. Effect of Blocking of Posterior Hypothalamic Nuclei on Development of Postdiphtheritic Polyneuritis and Experimental Allergic Encephalomyelitis

Series of experiments	Nature of expt. disease	No. of animals used in expt.	No. of animals developing disease	No. of animals dying	Time of appearance of paresis (in days)	Time of death (in days)	Sodium excretion after 60 min (in % initial)		Antibodies (on logarithmic scale) CFT		Antibodies (on logarithmic scale) Coombs' test	
Healthy							25	218± 12,9	22	0,68± 0,12	17	1± 0,3
Control	PDP	50	43	37	15,9± 0,8	19,3± 1,1	15	68± 9,7 $P<0,001$	12	4,08± 0,29 $P<0,001$	12	5,25± 0,26 $P<0,001$
							4*	242,0± 24,3 $P>0,1$				
	EAE	40	31	26	14,1± 0,39	16,9± 1,0	15	22,7± 2 $P<0,001$	15	3,3± 0,41 $P<0,001$	15	5,33± 0,19 $P<0,001$
Electrolytic destruction of posterior nuclei	PDP	26	10	8	20,6± 0,9 $P<0,05$	26,4± 1,7 $P<0,05$	26	381± 24,0 $P<0,001$	17	1,06± 0,31 $P>0,2$	15	1,4± 0,34 $P>0,5$
	EAE	15	13	12	20,7± 0,98 $P<0,05$	27,3± 2,1 $P<0,05$	17	198,9± 2,8 $P>0,1$	17	1,18± 0,28 $P>0,2$	17	1,8± 0,39 $P>0,05$
Blocking of posterior nuclei with chloropromazine	PDP	50	32	24	20,4± 1,11 $P<0,05$	25,6± 1,5 $P<0,05$	13	375,5± 53,0 $P>0,01$	13	1,5± 0,36 $P<0,05$	10	2,2± 0,15 $P<0,001$
							3†	32,4± 6,6 $P<0,001$				
	EAE	40	26	19	16,6± 0,5 $P<0,05$	22,5± 1,58 $P<0,05$	13	494,6± 133,0 $P>0,01$	18	0,85± 0,9 $P>0,2$	20	2,4± 0,13 $P<0,001$
							4†	55,9± 6,4 $P<0,001$				
Blocking of posterior nuclei with reserpine	PDP	50	31	20	19,8± 1,11 $P<0,05$	25,0± 1,99 $P<0,05$	9	553,3± 81,2 $P<0,01$	15	0,8± 0,16 $P>0,2$	18	3,8± 0,3 $P<0,001$
							3†	60,2± 3,4 $P<0,001$				
	EAE	40	24	18	17,9± 0,71 $P<0,05$	21,3± 1,11 $P<0,05$	10	385,9± 76,0 $P>0,05$	15	1,7± 0,29 $P<0,001$	15	2,53± 0,23 $P<0,001$
							3†	44,6± 5,2 $P<0,001$				

*Animals did not develop disease.

†Animals developed disease at the same time as controls (with intact hypothalamus).

this purpose, the dynamics of sodium excretion by the experimental animals was investigated on the 5th, 12th, and 20th days after injection of sublethal doses of diphtheria toxin or encephalitogenic mixture, against the background of water loading. Water was given through a tube (5 ml/100 g body weight). Every 30 min after water loading for 1.5 h the sodium excretion was measured. In animals with normal excitability of the HHAS, the response to water loading was inhibition of the liberation of vasopressin and an increase in the minute sodium excretion, but if the excitability of the HHAS was abnormal, sodium excretion was considerably reduced, presumably because of liberation of an excessive quantity of mineralocorticoids [2, 5, 11, 14]. The intensity of the immunologic reactions was assessed from the production of circulating (antimyelin, complement-fixing) antibodies and of incomplete hemagglutinins. Antibodies were determined by the complement fixation test (CFT) in the cold and by the Coombs' test in the usual manner [1, 8].

EXPERIMENTAL RESULTS

In the experiments of series I the character of the osmoregulatory reflexes and antibody production were investigated in animals with an intact hypothalamus but with PDP and EAE. The results (Table 1) show that the development of EAE and PDP was preceded by the appearance of a "paradoxical" salt-excretory reflex: on the 4th-5th day after injection of diphtheria toxin or the encephalitogenic mixture, and long before the appearance of clinical manifestations of the disease, water loading was followed by a decrease in sodium excretion instead of the usual increase. Changes in excitability of the hypothalamic centers regulating water and salt metabolism as a rule were combined with increased ability of antibody production. In animals with an intact hypothalamus, antibodies were found in the preparalytic period in high titers (Table 1). Abnormal osmoregulatory reflexes persisted until paralysis developed, but the quantity of antibodies circulating in the blood stream fell considerably toward this time. This was due to their fixation in the damaged nerve tissue.

Changes in function of the HHAS thus precede the appearance of antibodies against nerve tissue in the blood during the development of EAE and PDP. Paradoxical excitability of the HHAS was due mainly to changes in the function of the adrenergic nuclei, which are located in the posterior hypothalamus [5, 13].

To study more closely the pathogenetic role of these brain centers in the mechanism of development of autoallergic diseases, in the experiments of series II osmoregulatory reflexes and antibody synthesis were studied in animals following electrolytic destruction or pharmacological blocking (with chlorpromazine and reserpine) of the posterior hypothalamic nuclei. In guinea pigs with disturbed function of the posterior hypothalamic nuclei, the characteristic "paradoxical" reflex change in sodium excretion was not observed: on the 4th-5th day after injection of sublethal doses of diphtheria toxin or encephalitogenic mixture, all the animals showed an increase in the sodium excretion in the urine, i.e., the character of the reaction was the same as in healthy animals (Table 1). Destruction of the posterior nuclei or administration of sympatholytics inhibited antibody production to roughly the same degree in animals immunized with encephalitogenic mixture as in animals poisoned with sublethal doses of diphtheria toxin.

Restoration of the normal osmoregulatory reflex and inhibition of antibody synthesis in the animals with disturbed function of the posterior hypothalamic nuclei were accompanied by the later development of paralysis in both EAE and PDP, at about the same times. Destruction of the posterior hypothalamic nuclei prevented the development of PDP in some of the animals after injection of sublethal doses of diphtheria toxin, but after immunization with encephalitogenic mixture, this effect was less marked. Systematic administration of the sympatholytic drugs lowered both the morbidity and mortality rates among the experimental animals from EAE and PDP about equally.

Electrolytic destruction of the adrenergic nuclei of the posterior hypothalamus or blocking of their activity by adrenelytic drugs thus appreciably delayed the development of autoallergic demyelinating diseases of the nervous system provided that there were no severe pathological changes in the level of HHAS function. In that case, the synthesis of antibodies against nerve tissue was considerably depressed, as a result of which the development of paralysis in diphtheria and EAE was appreciably inhibited.

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