THE STUDY OF THE ACTIVITY OF ANTIBRAIN ANTIBODIES IN THE SERA OF PATIENTS WITH PSYCHOSES TOWARD THE MITOCHONDRIA AND OTHER FRACTIONS OF BRAIN TISSUE EXTRACT

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The results of previous investigations [1-7] showed that the serum of patients with psychoses contains complement-fixing antibodies reacting specifically with saline extracts from the brain tissue of man and animals, or only of certain species of animals. A positive reaction with brain antigens was discovered much more frequently in such patients than in other persons [1,3,4,7]. In these investigations, saline extracts from brain tissues obtained by repeated freezing and thawing followed by centrifugation (to remove intact cells) were used as test antigens. Such antigens consisted of protein-containing extracts in which cell fragments were present. It was interesting to discover which part of such an antigen – the structural or the nonstructural part – was most active against antibrain antibodies in the serum of patients with psychoses. Such an investigation would fill to some extent the gap in our knowledge of the composition and distribution of the brain antigen. In addition, the study of the immunological properties of the organoids of the cell, and especially of the mitochondria, where the enzyme activity of the cell is concentrated, would be of great importance to the elucidation of the mechanism of action of organ-specific antibodies on the tissue.

The object of the present investigation was to study the antigenic activity of the mitochondria and fractions of a saline extract of rat's brain obtained by ultracentrifugation. The rat's brain was chosen as test object because of its high activity as a test antigen for detecting antibrain antibodies in the serum of patients with psychoses [2-7].

EXPERIMENTAL METHOD

A 20% suspension of rat's brain in physiological saline with phosphate buffer (pH 7.2-7.4) was frozen three times with dry ice and subsequently thawed. The suspension thus obtained was centrifuged for 45 min at 6000 rpm in an angular centrifuge (model MOM, Hungary). The supernatant fluid part, the original saline extract, was further centrifuged for 1 h at 45,000 rpm (105,000-110,000 g) in the PHYWE ultracentrifuge. The residue, subsequently called conventionally the microsomes, was homogenized in buffer solution by means of a Potter's homogenizer. The mitochondria were obtained by the method of Fonyo and Somogyi [10], slightly modified by the staff of the Laboratory of Biohistochemistry, Institute of the Brain, Academy of Medical Sciences of the USSR. The protein content in all the antigens was determined by Lowry's method [11]. Before the experiment, the antigens were diluted so that the protein concentration in the original extract and in the supernatant fluid after ultracentrifugation was 0.332 mg/ml; because of their considerable anticomplementary activity, the mitochondria and microsomes were used in a dilution anticipated as being one-eighth and one-half the protein content, respectively.

The investigation was carried out by the complement fixation method in the cold with a dose of complement amounting to 170% of the titer determined by the warm method. The reaction was read after complete hemolysis had developed in the control.

EXPERIMENTAL RESULTS

The results of the investigation of the sera of several patients* with a series of antigenic preparations from rat's brain tissue and extracts of rat's liver (control of specificity of the reaction with brain) are given in Table 1.

*Twelve patients were admitted to a hospital with a diagnosis of schizophrenia or were suspected of having this disease, in one patient a diagnosis of traumatic psychosis was made, and in one patient – alcoholic psychosis.

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TABLE 1. Activity of Antibrain Antibodies in Serum of Patients with Psychoses toward Mitochondria and Fractions of Saline Extract Obtained by Ultracentrifugation

Serum of patients	Reaction with antigenic preparations from				
	Brain				Liver
	I	II	III	IV	
R-v	8610	8500	8510	∅	0000
S-na	8551	8651	8641	∅	0000
I-v	8720	8610	8600	∅	0000
L-v	8800	8100	8100	∅	0000
K-v	8720	8521	8521	7421	0000
M-na	8400	4100	8621	5542	0000
B-va	4410	5210	8721	6543	0000
B-a	8875	8887	∅	8887	0000
A-na	8830	5410	∅	8663	0000
M-va	8864	8865	∅	8886	0000
G-v	8710	8620	∅	∅	0000
K-ov	8610	∅	∅	8640	0000
V-v	8532	∅	∅	8864	0000
K-k	8864	5210	∅	8642	0000

TABLE 2. Results of Investigation of Immune Antibrain Sera with Extracts of Rat's Brain Tissue and Its Fractions

Serum No.	Reaction with antigenic preparations from				
	Brain				Liver
	I	II	III	IV	
1294	8844	4100	8644	8654	4000
1417	8887	1000	8886	8887	0000
1418	8621	2000	8522	8754	2200
1420	6321	1000	6421	7432	0000
1440	8763	2000	8621	8862	0000

These particular sera were used because of their marked complement-fixing activity in relation to saline extracts of rat's brain.

It is clear from Table 1 that the sera of the patient reacted both with the original saline extract and with its fraction obtained by ultracentrifugation, and also with the mitochondria.

It should be noted that some sera reacted with the water-soluble fraction less strongly than with the remaining preparation.

In a similar way, a series of immune antibrain sera obtained from rabbits in response to immunization with rat's brain homogenate in buffered physiological saline (seven intraperitoneal injections each of 2 ml of 10% homogenate at intervals of 3-4 days between injections) were investigated with all the above-mentioned antigenic preparations. Before the experiment, the serum was absorbed with rat's liver homogenate, stored in 40% formalin, and diluted with physiological saline 1:8. For absorption, one volume of homogenate (the residue obtained as a result of repeated rinsing to remove formalin) was treated with one volume of serum in a dilution 1:10.

It is clear from Table 2 that the immune sera gave a clear reaction with the original saline extract, the residue obtained as a result of ultracentrifugation of this extract, and with the mitochondria, but at the same time they proved practically inactive in relation to the soluble fraction of the saline extract.

The results of this investigation thus showed that antibodies to heterologous brain in the serum of certain patients may react both with antigens fixed to cell structures and also with antigens passing into the saline extract. This shows that the antigens leading to the appearance of antibodies in these cases were substances immunologically similar in all the antigenic preparations which were investigated. On the basis of these results, however, it cannot be concluded that these antigens are identically immunogenic for different lesions of the nervous system. On the contrary, it may be postulated that, in some cases, a selective immune reaction develops to certain biochemical and morphological structures of brain tissue cells. This hypothesis is based on data showing the distinctive immunological character of the individual subcellular structures of the tissues [8,9], and also on the results of the investigation of immune antibrain sera described in this communication, which demonstrated that immunization of animals with brain tissue homogenate may lead to the formation of antibodies against antigens of the mitochondria and the residual fraction of the saline extract, but not against the soluble antigens.

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of the first issue of this year.