Absence of Brain Antibodies in Patients with Schizophrenia*


In recent years the recognition of autoimmune processes has led to important advances in our understanding of certain diseases of hitherto uncertain causation (Mackay and Burnet, 1962). Burch (1964) postulated an autoimmune origin for schizophrenia on theoretical grounds from a study of the age- and sex-specific rates of first admissions to psychiatric hospitals. Heath and Krupp (1967) used immunofluorescence to study sections of brain and serum of schizophrenic patients; they found that globulin, presumably antibody, had become attached to nuclei of certain cells of the brain in vivo, and this antibody was also present in the serum. The claims that autoimmunity is concerned in schizophrenia have been cautiously appraised (Brit. med. J., 1967; Lancet, 1967). The present study describes an unsuccessful attempt to demonstrate antibodies to brain cells in schizophrenia.

Materials and Methods

Patients

We examined the brains of three patients with chronic schizophrenia and tested the serum of 15 patients with acute schizophrenia (eight males and five females) and 40 males with chronic schizophrenia.

The patients with acute schizophrenia were under the care of the University Department of Psychiatry (B. D. and I. H. J.) in short-stay beds; the symptoms had been severe, and in most cases were present for only a few weeks or months before admission. The patients with chronic schizophrenia had been in a Victorian mental hospital for at least 10 years. Our criteria for diagnosis of schizophrenia were as follows: the establishment, either from a clear description in the clinical notes or from interview, of passivity feelings or a primary delusion. When neither of these symptoms was present a patient who showed at least four of the following five symptoms was diagnosed as a case of schizophrenia: (1) presence of delusions of elaboration; (2) presence of schizophrenic thought disorder; (3) presence of hallucinations; (4) flexibilitas cerea, catatonic episodes, or stereotypies; (5) ideas of reference.

In the Venables (1957) Activity Withdrawal Scale the patients with chronic schizophrenia showed less than the normal amount of activity. We excluded patients with brain damage or mental deficiency and those who had previously had a leucotomy. No patient was receiving drugs.

Sera from 53 healthy subjects matched for age and sex with the patients were used as controls.

Tissues and Antiserum

Brains were obtained from three patients with chronic schizophrenia (who had died with cancer not affecting the nervous system) and one from a control without schizophrenia. These were dissected within seven hours of death and tested within 24 hours of death. Cubes of tissue 2 mm. in size were excised from the septal region, caudate nucleus, cerebellum, and cortex as described by Heath and Krupp (1967), and were snap-frozen either in a bath of solidified carbon dioxide and isopentane or on a strip of filter paper in a closed glass tube in a bath of solidified carbon dioxide and ethanol. The tissue was kept at -70° C. until sectioned at 4 μ. in a cryostat at -20° C. Two sections were prepared: one was air-dried and the other was fixed for 30 seconds in 95% ethanol in a bath of solidified carbon dioxide and isopentane. Other tissues used were rat liver and human peripheral blood smears fixed in ethanol (Smalley et al., 1968), unfixed thyroid gland from a patient with thyrotoxicosis of blood group O, and unfixed rat stomach (Holborow et al., 1959; De Boer et al., 1965).

The brains of the three patients studied and the thymus glands of two were examined microscopically after routine histological processing.

Antihuman immunoglobulins G and A were prepared by injecting rabbits with 2.5 mg. of immunoglobulin in 0.5 ml. of saline and 1 ml. of Freund's complete adjuvant: 0.1 ml. was injected intraperitoneally and 0.9 ml. subcutaneously into the flanks. The rabbits were injected 30 days later with the same amount of globulin in Freund's incomplete adjuvant and bled two weeks later. The immunoglobulin G fraction of rabbit serum containing antibody was isolated and conjugated with fluorescein isothiocyanate (Wood et al., 1965). Rabbit antihuman immunoglobulin M conjugated with fluorescein (Batch No. F 211D) was donated by Australian Hoechst Ltd.

Immunofluorescence Tests

In testing for globulins bound in vivo to the brain of schizophrenic patients frozen sections of brain were washed in six changes of phosphate-buffered saline pH 7.3 incubated at 37° C. for 30 minutes with antihuman globulin conjugated with fluorescein, washed in two changes of phosphate-buffered saline and mounted in 10% phosphate-buffered saline in glycerol. The various preparations were examined under a Leitz microscope fitted with a source of ultraviolet light. Light of two wavelengths was used: one was obtained by a BG 12 transmission filter and OG 1 suppression filter, and the other by a UG 1 transmission filter and a K 430 suppression filter. A BG 38 filter eliminated red light.

The same immunofluorescence procedure was used when testing for antibody in serum except that tissue was incubated with serum at 37° C. for 30 minutes and washed in two changes of phosphate-buffered saline before applying antihuman globulin.

Results

Brains of Schizophrenic Patients.—None of the three brains obtained from patients with chronic schizophrenia showed globulin bound to brain cells in any of the regions examined.
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Summary
Recent discussions on autoimmune as a cause of schizophrenia led us to examine by immunofluorescence three brains from patients with chronic schizophrenia and serum from 53 patients with acute or chronic schizophrenia and 53 normal controls. In contrast to Heath et al. (1967) we could not show that globulin became bound in vivo to nuclei of brain cells of schizophrenic patients nor could we show that the incidence of serum antibody against brain cell nuclei was greater in schizophrenic patients than in controls.

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REFERENCES
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