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Original article

Investigations of cerebrospinal fluid in Borna disease virus seropositive psychiatric patients

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Summary – Borna disease virus (BDV) appears to cause meningoencephalitis and schizophreniform psychosis in sporadic cases according to earlier cerebrospinal fluid (CSF) inoculation experiments (Rott *et al*, 1991). However, CSF parameters in BDV seropositive psychiatric patients proved nearly all normal; only the most sensitive CSF/serum index I-BDV for intrathecally produced BDV specific IgG was pathologic in 10.5–29.0% (according to different methodological limits) of patients. An increase in sensitivity was attempted to detect specific IgG in CSF in a part of the cases by concentration. Concentration procedure does not significantly increase methodological bias according to a statistical analysis of the results. Our findings support the hypothesis that BDV may cause or contribute to the pathogenesis of a diagnostically broad pattern of psychiatric syndromes. The occurence of a spectrum of diagnoses is expected from non-specificity of psychiatric symptoms in other infectious diseases of the brain as well as from results in experimental Borna disease (BD) in animals, when a majority of the animals showed rather unspecific symptomatology due to slight, preferentially limbic encephalitis. Slight deficiencies from an earlier BDV infection could explain continuing symptoms in a part of the cases. Recurrences years after infection are well known in experimental and natural BD in animals. It remains open, whether this mechanism could play a more prominent role in a form of "symptomatic" cyclothymia and "symptomatic" schizophrenia, although the results of CSF investigations are more clear in BDV seropositive patients with major psychoses.

cerebrospinal fluid antibodies / Borna disease virus

INTRODUCTION

Borna disease (BD) is the most frequent meningoencephalitis of horses and sheep in Germany caused by a strongly neurotropic virus (Borna disease virus, BDV), which has only recently been characterized as containing a negative single-stranded RNA of 8.5 or 10.5 kb (De'La Torre *et al*, 1990; Lipkin *et al*, 1990; Vande Woude *et al*, 1990; Richt *et al*, 1991; Thierer *et al*, 1992; Cubbitt *et al*, 1994). BDV shares properties with slow (long incubation period) as well as conventional viruses (physical properties, replication, RNA) (Rott *et al*, 1991; Richt *et al*, 1993). Formerly, BDV was assumed nonpathogenic for humans, the natural BD usually having a fatal course (Heinig, 1969; Danner, 1982). In low primates a latent course was observed, most diseased animals showing only behavioral abnormalities, few animals showing neurological symptoms (Sprankel *et al*, 1978). The occurence of BDV serum antibodies has thus been found in psychiatric patients with affective psychoses but not in controls (Amsterdam *et al*, 1985; Rott *et al*, 1985; Kao *et al*, 1993). But a significant increase in prevalence of BDV serum antibodies has also been reported and replicated in unselected psychiatric (PS) patients with various diagnoses (6.8%; n = 1,003 and 5.9%, n = 2,377) compared to surgical (SU) controls (3.5%; n = 569) as well as in unselected neurological (NL) patients (4.86%; n = 1,791) (Bechter *et al*, 1987; 1992a). The increase of seroprevalence rates in NL and PS patients came mainly from an increased prevalence in the young age groups up to 50 years of age. Respective BDV seroprevalence rates in the young age groups were: PS 6.02% (n = 1,312), NL 3.7% (n = 653), SU 2.2% (n = 276). We interpreted these findings as follows: in older

age groups (age 50 years and more), BDV serum antibodies appear to be mainly an incidental finding, but in a considerable part of young PS and NL patients this may have a causal meaning. This view has been corroborated by the finding that in the younger BDV seropositive NL patients, otherwise unexplained cases of lymphocytic meningoencephalitis and further unexplained neurological syndromes which are possibly related to BDV, occured statistically more frequent than in pair-matched (for age and sex) BDV seronegative neurological controls (Bechter *et al*, 1992a). Furthermore in three BDV seropositive patients (2 neurological patients with lymphocytic meningoencephalitis, 1 psychiatric patient with acute relapse of schizophrenia), BDV was demonstrated in CSF (Rott *et al*, 1991). Therefore increased evidence exists, that a human BD in the form of a neuropsychiatric disorder might exist, which may be more important than previously thought. BDV specific serum antibodies have now been found in countries (USA, Germany, Japan, Africa) in which such investigations have been performed in humans (Amsterdam *et al*, 1985; Rott *et al*, 1985; 1991; Bechter *et al*, 1987; 1992a; Bode *et al*, 1988; 1992; Fu *et al*, 1993). However, a major difficulty is determining in what cases the psychiatric disorder can be causally related to BDV, due to several reasons: i) BDV IgG serum antibodies show no peak in acute phases of the disease (-Narayan *et al*, 1983) and the antibody titers can be extremely low also in acute disease in horses (Lange *et al*, 1987; Herzog *et al*, unpublished); ii) When screening investigations for viral antibodies are undertaken one can expect a mixture of incident and (possibly) relevant cases. The question remains, how to focus on the cases of interest; iii) Psychiatric symptomatology due to any etiologies is widely non-specific, especially in low grade encephalitis. So in rare cases, in which known viruses lead to slowly progressive encephalitis, with increasing severity of the encephalitis a broad pattern of psychiatric diagnoses from initial phobias to final schizophrenic psychosis can be attributed to one patient in a few weeks (Ullmann and Kühn, 1988). The situation is complicated in that, if a viral encephalitis only rarely developes to an advanced state, as is hypothetically possible for BD in humans based upon analogies to experimental BD in animal; iv) In known latent and slow virus encephalitis no pathological parameters in cerebrospinal fluid (CSF) are usually found and direct demonstration of the virus in CSF is generally difficult (Kennedy and Johnson, 1987).

In summary, it is difficult to establish possible BDV related neuropsychiatric disorders in humans, since histopathological investigations are unsuitable for ethical reasons. A recent CSF/serum index on specific immune globulins, originally developed to differentiate active from inactive cases of tertiary syphilis (Prange *et al*, 1983; Prange and Ritter, 1986) has proven a most sensitive parameter for CSF diagnosis in other encephalitis, for example the beginning of HIV encephalitis (Ackermann *et al*, 1986; Lüer *et al*, 1988; Felgenhauer, 1991). By introducing this parameter in BD research, we earlier reported a few cases with increased BDV specific IgG in CSF (Bechter *et al*, 1989). Although in animals with acute BD an increase of cells, total protein (Hiepe, 1960) and in a part of cases, specific antibodies as oligoclonal bands (in concentrated CSF) have been found in CSF (Ludwig and Thein, 1977; Ludwig *et al*, 1977), it should be stated that the species investigated suffered from an acute encephalitis with severe neurological symptoms. But CSF investigation may have reached its limitations when slight or protracted encephalitis occurs. In conclusion, increased BDV specific antibodies in CSF (according to increased I-BDV) could well be the only pathologic sign in hypothetic slight human BD and might indicate a causal relationship to the respective neuropsychiatric disorders. Here we report on the complete results of CSF investigations in BDV seropositive psychiatric patients collected over the last 6 years.

SUBJECTS AND METHODS

Subjects

Informed consent was obtained from the BDV-seropositive patients and in cases of reduced legal responsibility, additionally from the legal representatives. CSF was taken by lumbar puncture and simultaneously serum from a forearm vein. All treatment was continued unchanged, many patients received drugs, mainly neuroleptics and antidepressants. Lacking clear criteria for selection of the patients, we preferred somewhat more acute major psychoses, which were regarded as most possibly linked to a hypothetic BD from analogies to symptoms in BD of animals and from earlier studies, the patients being under 50 years of age, because an increase of BDV seroprevalence rates has been found mainly in these younger age groups (Bechter *et al*, 1992a). Diagnoses were made according to ICD-10 (WHO, German translation, 1991) by consensus conferences based on knowledge of the charts and the patients. Because of the limited total number statistical evaluation

with respect to diagnostic subclasses was performed after grouping the patients to main diagnostic groups according to ICD-10 (F0x.x, F1x.x, ..., F9x.x).

Statistical methods

Correlational analysis was performed with Spearman rank correlation, significance levels of differences were tested with Fisher's exact test (two-sided).

Routine parameters in CSF were evaluated in the laboratory of the Kliniken Günzburg (normal values in parentheses): cells were counted in the Fuchs-Rosenthal chamber (0–5 cells/mm^3); glucose was determined enzymatically (40–75 mg/dl), total proteins by coomassie blue reaction (–50 mg/dl) and colour visually evaluated.

Antibody determination

Antibodies were determined by indirect immunofluorescence assay as described earlier (Herzog and Rott, 1980) under blind conditions; positive (from 1:5) specimen were repeatedly investigated. If CSF specimen showed no antibodies, the probe was concentrated; CSF and serum probes of one patient were always assayed in the same batch.

Determination of proteins

Concentrations of albumin and IgG were determined in unconcentrated CSF and in serum by rate nephelometry (Arrey Protein System, Beckman Instruments GmbH, Munich, Germany) with detection limits of 6.2 mg/l for albumin and 11.1 mg/l for IgG. Blood brain barrier functions and the amount of locally synthesized IgG were evaluated by analyzing the protein profiles on the updated Reiber graph (Reiber and Felgenhauer, 1987). After isoelectric focusing in thin-layer polyacrylamide gels (Ampholine PAG-plates, pH 3.5–9.5) silverstained oligoclonal bands were qualitatively determined by visual evaluation (Olsson *et al*, 1984; Lubahn and Silverman, 1984; Reiber, 1988a, b). BDV specific antibody index (I-BDV): to determine if possible BDV antibodies in CSF exceeded the amount expected from passive filtration through the blood-brain-barrier, the index I-BDV was calculated following the basic approach for analyzing the fraction of antibodies produced intrathecally (Felgenhauer, 1982; Reiber, 1988b):

$$\text{I-BDV} = \frac{\text{BDV-specific IgG CSF: total IgG CSF}}{\text{BDV-specific IgG-serum: total IgG-serum}}$$

Theoretically any value of the index > 1 is pathologic; because of methodological uncertainties a value > 1.5 is usually considered pathologic (Felgenhauer, 1991); our methodology appears more weak, because CSF was concentrated in some of the cases and titers were calculated. So we decided to take two borderline values: > 2 as being possibly pathologic and > 4 as definitely pathologic.

RESULTS

In an open clinical study BDV serum antibodies were determined in diagnostically unselected psychiatric patients. A total of about 6,000 serum investigations were performed over six years (Bechter *et al*, 1987; 1992a; unpublished). In a total of 38 younger (up to age 50 years) BDV seropositive patients consecutively collected, additional CSF investigations were performed. These patients were newly admitted psychiatric inpatients, aged 35.4 (19–50) years, 17 of them male, 21 female. BDV serum antibodies were positive with titers from 1:5–1:320. Results from parallel attempts to directly demonstrate virus in CSF were already reported earlier (Rott *et al*, 1991).

According to the selection criteria a spectrum of psychiatric diagnoses was found in the patients investigated (see table I); in every case of alcoholism an additional psychiatric disorder (in most cases organic personality disorder or earlier personality disorder not linked to alcoholism or an additional affective disorder) was diagnosed.

Routine parameters (cells, colour, glucose, total proteins) in CSF were normal in all cases. Evaluation of blood brain barrier function and IgG content in CSF by the Reiber graph revealed in three cases a very slight or borderline proportional blood brain barrier dysfunction (all 3 with affective disorders; 1 of the patients was an earlier NL patient, see Bechter *et al*, 1992a), another of these cases showed native BDV specific antibodies in CSF with I-BDV = 22.8. Autochtonous IgG production according to the evaluation on the Reiber graph was not found in any patient. In two cases (1 borderline personality, 1 affective disorder) we found oligoclonal bands in CSF, both did not show any other CSF pathologies. BDV serum antibodies were dominated by low titer values of about 1:10 (see fig 1) similar to their distribution in the screening group (Bechter *et al*, 1992a). Elevation of I-BDV > 4 was found in 10 cases, in 1 case > 2 < 4, that is an elevated I-BDV according to our definition in 26–29% of the cases investigated. In five cases BDV antibodies were found in unconcentrated CSF (in 4 of them I-BDV > 4, in 1 case I-BDV > 2 < 4), that is in 10.5% (13%) of the cases. The mean values, standard deviations, variation coefficients and minimum-maximum values

Table I. Distribution of cases (n = 38) with normal and elevated BDV specific IgG in CSF (I-BDV) in relation to diagnostic subgroups (according to F-groups, ICD-10).

Diagnosis (ICD-10)	*I-BDV normal*	*I-BDV elevated (> 4)*	*I-BDV > 4* (BDV-ab's in native CSF)*
Schizophrenia (F 20.x)	7	5	2
Affective disorders (F 30-33)	12	2	2
Alcoholism (F 10.x)	6	2	0
Personality disorders (F 60-62)	3	1	0
Sum	28	10	4

* All values actually being > 10.

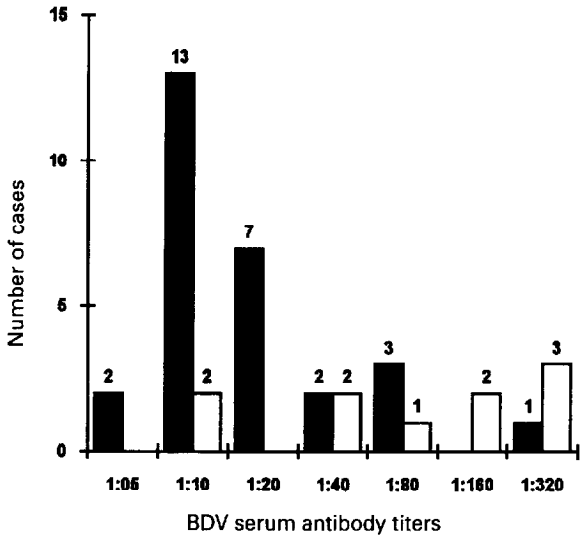

Fig 1. Frequency of cases with normal (≤ 4) and increased (> 4) I-BDV values related to the respective BDV serum antibody titers; n = 38. ■ Number of cases with normal I-BDV at the respective serum titers; □ number of cases with increased I-BDV at the respective serum titers.

Table II. Frequency of cases with normal (≤ 4) and increased (> 4) I-BDV in relation to BDV serum antibody titers.

	I-BDV ≤ 4	*I-BDV > 4*
BDV serum antibody titers 1:5–1:20	22	2
BDV serum antibody titers 1:40–1:320	6	8

p = 0.0186.

Table III. Frequency of cases with normal (≤ 4) and increased (> 4) I-BDV in relation to concentration procedure of CSF.

CSF	*I-BDV ≤ 4*	*I-BDV > 4*
Unconcentrated (c = 1)	14	4
Concentrated (c > 1)	14	6

p = 0.719; not significant.

were for I-BDV 5.6; 17.24; 3.05; 0–101.5; and for serum antibody titers 62.6; 96.99; 1.55; 5–320. For relationships between diagnosis and I-BDV values see table I.

Increased I-BDV values were found from serum titers of 1:10 and more. Native BDV specific IgG in CSF was only found from serum titers of 1:80 (–1:320; n = 4); I-BDV mean values were clearly higher in these cases (33.6; 2.8–101.5) than in the cases, in which elevated I-BDV was found only after concentration of CSF (7.8; 4.2–14.6). The I-BDV values correlate positively with serum titers (r = 0.64; n = 38; $p < 0.001$; see fig 1). Considering exclusively the total of cases with BDV serum antibody titers of 1:40 and above, 64% of them showed increased I-BDV. Increased I-BDV is more frequent in cases with higher BDV serum antibody titers ($p < 0.01$) (see table II).

Depending on the available quantum, CSF was concentrated if BDV antibodies were not detected in native CSF (n = 20): mean values, SD and range of concentration factor are in cases with normal I-BDV (n = 14); 14.1; 11.9; 0–55; and in cases with increased I-BDV (n = 6); 14.5; 8.3; 0–30. Increased I-BDV values do not correlate with concentration factor (r = 0.24; not significant; see table III).

DISCUSSION

CSF investigations in encephalitis of conventional viruses can fail to show pathologic parameters in well documented single cases (Körber and Huffmann, 1991; Felgenhauer, 1991), depending on the severity of encephalitis and the distance to CSF space communicating with ventricles and basal cisternas (Felgenhauer, 1991). A failure to detect pathologic findings is frequent in slow virus encephalitis (Kennedy and Johnson, 1987; Lüer *et al*, 1988). BDV, sharing properties of conventional and slow viruses, causes meningoencephalitis in animals strikingly differing in severity and course depending on the species, route of infection, genetic and immunological factors (Danner, 1982; Carbone *et al*, 1987; Morales *et al*, 1988; Herzog *et al*,

1991). A form of a human BD with mostly slight courses has been hypothesized from the results of earlier epidemiological investigations on the prevalence of BDV serum antibodies (Bechter *et al*, 1992a). A most sensitive specific antibody index (I-BDV) applied here analogously to the results in known virus encephalitis (Ackermann *et al*, 1986; Lüer *et al*, 1988; Felgenhauer, 1991) should indicate, if elevated, autochtonous BDV antibody production in the intrathecal space and by this active BDV encephalitis. In a sample of selected young BDV seropositive psychiatric inpatients, the age groups of interest (see introduction), we found an increased I-BDV in 26–29% of the patients. These findings may be considered as a specific although slight sign of BDV encephalitis.

These results can be discussed from a methodological point of view: in approximately one half of the cases with increased I-BDV, CSF has been concentrated, a procedure which could be a source of error. Increased I-BDV values correlate positively with high BDV serum antibody titers. This correlation might indicate a systematic bias due to concentration of the portion of BDV serum antibodies filtrated passively through the blood brain barrier. But in that case one would, for other reasons, expect a tendency against the observed correlation: because I-BDV values are calculated from specific IgG and total IgG from both CSF and serum (see Methods) and the ratio of the filtrated portion of total and specific IgG is therefore not affected, a false positive correlation of I-BDV with serum values would only be possible due to methodological faults of the concentration procedure, which should be distributed at random, occuring with similar frequency in cases with low serum antibody titers; of which did not occur. Furthermore, the proportion of cases in which increased I-BDV was found, does not significantly differ between cases in which CSF was concentrated, compared to unconcentrated (table III), and increased I-BDV values do not correlate with concentration factor. A second explanation for the positive correlation between BDV serum antibody titers and I-BDV values would be a more frequent causal or pathogenic meaning of BDV serum antibodies in cases with high humoral antibodies. But humoral antibodies do not play a role in the pathogenesis of BD (Rott *et al*, 1991) and no correlation between higher serum antibody titers and certain diagnoses or more severe illnesses has been found up to now (Bechter *et al*, 1992a). Nevertheless, here we found higher I-BDV values in patients with major psychoses than with other psychiatric disorders, so this explanation may be possible to some extent. A third explanation would be an increased likelihood to disclose an intrathecal humoral immune response in cases with a generally stronger production of humoral antibodies. Detection limits in CSF are probably bound to the overall strength of humoral immune response in blood as CSF. Because IgG is generally much lower in CSF than in serum (about 1:500 filtrated), the detection limits could be mirrored as a common correlation. The third explanation seems a likely one because it is the most simple, nevertheless the second one should be considered.

To be quite sure to be unbiased due to concentration procedure, we might exclusively consider cases in which BDV antibodies were found in native CSF, so a portion of 13% ($n = 5$) with increased I-BDV > 2 remain. A further source of error could arise from calculating the index by titer values in the case of index values ≤ 4 (Reiber, 1988a). Now considering only cases with native BDV antibodies in CSF as I-BDV values above 4, there remains a minimum of 10.5% ($n = 4$) of cases with an increased I-BDV. Summarizing the results, from the epidemiologic data about 60% of relevant cases could be expected in the sample of young psychiatric patients investigated here, but we had no clear criteria for selecting cases of interest for CSF investigation other than age. In the selected sample we found in 10.5–29% (according to different methodological limits) direct evidence for BDV encephalitis in the form of intrathecally produced BDV specific IgG. IgM antibodies were not investigated because in BDV infected animals it is rarely detected.

Our detection of oligoclonal bands in two cases might be incidental since similar frequency has been reported in normal controls (Roos *et al*, 1985). The finding of slight or borderline proportional blood brain barrier dysfunction according to the Reiber graph in three cases, in one of them paralleled by an increased I-BDV, might be considered as questionable or incidental. Because of the lack of an adequate control group and of the heterogeneity of diagnoses we have chosen relatively high borderline values for CSF proteins and so we might have failed to detect slight protein increase regarding the total group. Protein increase has been reported in a considerable part of general psychiatric patients but the results remained conflicting (Roos *et al*, 1985; Bauer and Kornhuber, 1987; Pitts *et al*, 1990; Samuelson *et al*, 1994). Many factors can significantly influence the concentration of CSF proteins and the source of such variability is hard to define (Reiber, 1993). One of these factors is the quantum of CSF taken (we took

about 12 ml). Furthermore, our patients were not drug-free; this could have affected CSF protein concentrations (Smith, 1958; Denzel, 1959; Simpson and Cooper, 1966; Preskorn *et al*, 1982). But neither a blood CSF barrier dysfunction nor undetected slight protein increase in the total group would confound I-BDV values in principle, the measurement of our special interest (see Felgenhauer, 1991), and there is no indication that drugs could influence BDV-specific IgG or produce a cross-reactive protein.

One might wonder about the negative finding of oligoclonal bands in cases with increased I-BDV; in known encephalitis higher index values (> 10) are regularly paralleled by oligoclonal bands (Felgenhauer, personal communication). The number of cases here with native BDV antibodies is small and CSF was not concentrated for determination of oligoclonal bands. Even in peracute BD in animals with high BDV antibody titers in serum as CSF oligoclonal bands have only been found after concentration of CSF (Ludwig and Thein, 1977; Ludwig *et al*, 1977). So our negative findings might be explained by the overall low antigenicity of BDV: the serum antibody titers in BD are uniquely low and play no role in the T-cell mediated pathogenesis of BD (Rott *et al*, 1991). In Western blot assay only few bands appear (maximum 2 bands in the same animals respectively in the same human individuals; only 3 bands have been observed; Vande Woude *et al*, 1990; Rott *et al*, 1991; Bode *et al*, 1992; Kao *et al*, 1993). The negative finding of oligoclonal bands in cases with increased I-BDV on the other hand argues rather against the possibility that increased I-BDVs are only due to a non-specific polyclonal activation. In this context one might mention the recent observation of a shift of the T-cell memory from a specific to a auto-immune one after the acute phase in experimental BD (Rott *et al*, 1993). Auto-immune measures have repeatedly been reported to be pathologic in different psychiatric disorders (Müller *et al*, 1991; Schott *et al*, 1992). However, we have not investigated auto-immune parameters in our patients up to now.

What about a possible relationship between diagnosis and increased I-BDV values? The course of a hypothetic human BDV encephalitis could be acute, subacute, chronic or chronically-recurrent, with or without slight defect, according to the results of BD in animals (Sprankel *et al*, 1978; Narayan *et al*, 1983). The height of BDV serum antibody titers appears unsuitable for classifying the stage or the course of BD (see *introduction*). The time point of the assumed BDV infection in our human sample might differ significantly between cases. So our only idea for selecting possibly relevant cases was an acute psychiatric disease, but we do not know, if we were successful in selecting relevant cases. What seems true is, that in incidental cases we can expect no increase of I-BDV. Considering only the four cases, in which increased I-BDVwas found in unconcentrated CSF: 2 patients suffered from paranoid schizophrenia; 2 patients from affective disorders; one of them showing comorbidity with paranoid personality. It appears that especially these patients could present relevant cases caused by a slight human BD, because methodological failures due to concentration procedure are excluded and the I-BDV values were more than 4-fold higher than in concentrated cases. One of these cases did not only show the highest I-BDV value, but BDV was also isolated from CSF (patient T; in Rott *et al*, 1991). Considering the cases in which CSF was concentrated: 2 cases with chronic schizophrenia, 3 cases of personality disorders in comorbidity with alcoholism and 1 case of developmental brain abnormalities with paranoid psychosis are to be added. In all these cases an organic factor might be discussed, because personality disorders might have preceded alcoholism or BDV encephalitis might have contributed to comorbidity (see also Bechter *et al*, 1994).

A broad pattern of psychiatric diagnoses is seen in any known single etiology, refered to as non-specificity. Such has been found in identified genetic causes of psychiatric disorders (Propping, 1983); one might argue, that this principle is true also for other etiologies taking effect only slowly. Non-specificity of psychiatric symptomatology (Bonhoeffer, 1917) and etiological heterogeneity of certain distinct psychiatric disorders (Buchsbaum and Rieder, 1979) appear as two sides of one coin. Nevertheless, one might expect some related specificity of symptomatology from preferential involvement of certain brain regions, *ie* the limbic system in BD (Seifried and Spatz, 1930; Danner, 1982). The limbic system is likely to be in particular involved in the pathogenesis of major psychoses (Weinberger, 1986; Bogerts, 1991; Beckmann and Jakob, 1991; Gross *et al*, 1989; Royston and Lewis, 1993). The significance, however, of the limbic system for schizophrenia is not specific nor exclusive (Weinberger, 1986). From these points of view one might suspect that a slight human BD could cause non-specific symptoms resembling a personality disorder shifting to more specific "symptomatic" major psychosis with increasing severity of limbic involvement (see also Conrad,

1972; Huber, 1972; Ullmann and Kühn, 1988; Gross *et al*, 1989) and being modulated by independent preexistent factors. In conclusion, a pattern of different psychiatric disorders fitting with some organic alteration might well be related to an acute, subacute or chronic BDV encephalitis of several degrees. An earlier finding, that psychiatric patients with schizophrenia as "character disorders" frequently show neurological soft signs, might be of some interest in this context (Quitkin *et al*, 1976). BDV could be one of several etiologies for a spectrum of psychiatric disturbances possibly with a preponderance of "symptomatic" schizophrenic and cyclothymic disorders, because in these diagnostic subgroups increased I-BDV has been more clearly found than in others. The development of certain psychiatric disorders due to BDV-encephalitis might depend on genetic factors, because in rats a strong influence of genetic factors on the development of BDV encephalitis as well as on the symptomatology, at different pathogenetic levels, has been demonstrated (Herzog *et al*, 1991).

Our view of a pathogenic meaning of BDV for human psychiatric disorders is further corroborated by environmental investigations (Bechter *et al*, 1992b,c); increased prevalence of cases with slight brain atrophy in BDV seropositive patients compared to seronegative controls (Bechter *et al*, 1994). Brain atrophy is found after BD in some animals depending on severity of the encephalitis.

CONCLUSIONS

Earlier several authors suggested undetected viral causes of psychiatric disorders, especially of schizophrenia (Torrey and Peterson, 1976; Libikova, 1983; Kurstak, 1991; Roos *et al*, 1985; Bechter and Herzog, 1990; Bechter, in press). Recently it has been shown in known encephalitis that a solitary increase of specific antibodies in CSF can most sensitively indicate CNS involvement by an infectious agent (Ackermann *et al*, 1986; Lüer *et al*, 1988; Felgenhauer, 1991). If the disease is not progressive, such slight CSF pathology can be the only pathologic finding. Innoculation experiments from CSF are generally difficult and not conclusive in each case in clinical practice. In this study we found increased I-BDV values (the proposed measure for defining intrathecal production of BDV specific IgG) in 10.5%–29% of BDV seropositive psychiatric patients depending on different methodological limits. This should indicate active BDV encephalitis in these cases, be it acute, subacute or chronic regarding its course. Because the immunological reaction appears generally weak in BD, CSF was concentrated in a part of the cases to increase sensitivity. This could increase methodological bias. A statistical analysis rather supports that this procedure did not confound the data and one therefore might consider 29% of the cases investigated as showing signs of an active, though slight, BDV encephalitis. Such interpretation is further supported by the parallel demonstration of BDV specific antigen in CSF as a strong increase of I-BDV in one patient with schizophrenia. BDV encephalitis might lead to different psychiatric disorders, appearing as "symptomatic" schizophrenic and cyclothymic psychosis but also as personality disorders, which may occur in comorbidity with alcoholism. Different clinical pictures might develop under the influence of genetic factors, analogous to results in experimental BD in animals, and possible unknown pre-existing factors.

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