Irregular presence of abnormal prion protein in appendix in variant Creutzfeldt-Jakob disease

We have investigated the presence of disease related prion protein (PrP$^{\text{Sc}}$) in appendix samples obtained at necropsy from four neuropathologically confirmed cases of vCJD, Creutzfeldt-Jakob disease (vCJD). PrP$^{\text{Sc}}$ was detected in only one vCJD appendix, at a level lower than that found in a diagnostic tonsil biopsy sample obtained from the same patient. The single positive appendix, but not the other samples, also showed abnormal prion protein immunohistochemistry. The finding that appendix samples from three of four cases of vCJD are devoid of detectable PrP$^{\text{Sc}}$ questions the utility of screening appendectomy tissues to estimate the prevalence of pre-clinical vCJD infection within the UK population.

The appearance of a novel human prion disease, variant Creutzfeldt-Jakob disease (vCJD), in the United Kingdom from 1995 onwards, and the experimental confirmation that this is caused by the same prion strain as that causing BSE in cattle, has raised the possibility that a major epidemic of vCJD will occur in the United Kingdom and other countries as a result of dietary or other exposure to BSE prions. The pathogenesis of vCJD differs significantly from that of other forms of CJD. Disease associated prion protein (PrP$^{\text{Sc}}$) is readily detectable in lymphoreticular tissues in vCJD and not in classic CJD. High levels of PrP$^{\text{Sc}}$ are uniformly found in the central nervous system and lymphoreticular system of vCJD patients. The highest levels of PrP$^{\text{Sc}}$ seen outside the central nervous system in vCJD are in tonsils (about 10% of that found in brain) and tonsil biopsy is used for ante-mortem diagnosis of vCJD. To date, positive prion protein immunohistochemistry has been reported in only a single appendix sample, although, importantly, this was removed from the patient before the onset of overt features of vCJD. While the stage at which lymphoreticular infection occurs in vCJD is unknown, PrP$^{\text{Sc}}$ accumulation is detectable in the lymphoreticular system in natural sheep scrapie and in experimental rodent models of scrapie at a very early stage of the incubation period, long before the clinical phase of the disease. Based upon these data it has been suggested that large scale screening of surgical tonsillectomy and appendectomy tissues for PrP$^{\text{Sc}}$ could provide early warning of a high level of vCJD prion infection and several such studies are in progress.

Recently we reported our concern after finding that PrP$^{\text{Sc}}$ was undetectable in appendix samples obtained at necropsy from two neuropathologically confirmed vCJD cases. While we were not able to examine these samples using immunohistochemical methods, we now have had the opportunity to investigate appendices from two further vCJD cases by both high sensitivity western blotting and immunohistochemistry.

Methods

Tissue samples

Tissues were collected at necropsy with consent of relatives from two patients with clinical presentations consistent with vCJD. Definite diagnoses of vCJD were confirmed by neuropathological examination and the demonstration of type 4 PrP$^{\text{Sc}}$ in tonsill. Appendix samples from these vCJD cases, and appendices from single neuropathologically confirmed cases of either sporadic CJD or inherited prion disease (144 base-pair insertion), were divided and prepared as either 10% homogenates in phosphate buffered saline (PBS) or fixed in 10% formal saline.

Immunohistochemistry

Tonsil tissues were collected at necropsy with consent of relatives from two patients with vCJD. PrP$^{\text{Sc}}$ was detected in only one vCJD appendix, at a level lower than that found in a diagnostic tonsil biopsy sample obtained from the same patient. The single positive appendix, but not the other samples, also showed abnormal prion protein immunohistochemistry. The finding that appendix samples from three of four cases of vCJD are devoid of detectable PrP$^{\text{Sc}}$ questions the utility of screening appendectomy tissues to estimate the prevalence of pre-clinical vCJD infection within the UK population.

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The highest levels of PrP$^{\text{Sc}}$ seen outside the central nervous system in vCJD are in tonsils (about 10% of that found in brain) and tonsil biopsy is used for ante-mortem diagnosis of vCJD. To date, positive prion protein immunohistochemistry has been reported in only a single appendix sample, although, importantly, this was removed from the patient before the onset of overt features of vCJD. While the stage at which lymphoreticular infection occurs in vCJD is unknown, PrP$^{\text{Sc}}$ accumulation is detectable in the lymphoreticular system in natural sheep scrapie and in experimental rodent models of scrapie at a very early stage of the incubation period, long before the clinical phase of the disease. Based upon these data it has been suggested that large scale screening of surgical tonsillectomy and appendectomy tissues for PrP$^{\text{Sc}}$ could provide early warning of a high level of vCJD prion infection and several such studies are in progress.

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Immunohistochemistry

Tonsil tissue was fixed in 10% buffered formal saline and inactivation of prion infectivity was accomplished by incubation in 98% formic acid for one hour. After further washing for 24 hours in 10% buffered formal saline, tissue samples were processed and paraffin wax embedded. Sections were cut at a nominal thickness of 4 µm, treated with 98% formic acid for five minutes and then boiled in EDTA-TRIS-citrate buffer pH 7.8 for 20 minutes. Immunohistochemical staining was performed with anti-PrP monoclonal antibody 12F10 on a Ventana automated immunohistochemical staining machine using a basic diaminobenzidine detection system according to the manufacturers instructions (Ventana Medical Systems, Tucson, Arizona).

Detection of PrP$^{\text{Sc}}$

Sodium phosphotungstic acid precipitation of PrP$^{\text{Sc}}$ from 0.5 ml 10% tissue homogenates and western blotting using high sensitivity enhanced chemiluminescence was performed as described previously.

Results

Recently we reported that appendix samples obtained at necropsy from two neuropathologically confirmed vCJD cases contained undetectable levels of PrP$^{\text{Sc}}$. We have now examined appendix samples from two further neuropathologically confirmed vCJD cases and have detected PrP$^{\text{Sc}}$ in only one vCJD appendix (fig 1A). The level of PrP$^{\text{Sc}}$ present in this appendix was compared directly with the level of PrP$^{\text{Sc}}$ present in a diagnostic tonsil biopsy sample obtained from the same patient. After proteinase K treatment of equivalent aliquots (20 µl) of 10% appendix homogenate or 10% tonsil biopsy homogenate, we observed clear detection of PrP$^{\text{Sc}}$ in biopsy tonsil homogenate, but not in appendix homogenate (fig 1B). Similarly, PrP$^{\text{Sc}}$ was readily detectable in necropsy tonsil obtained from the vCJD patient with PrP$^{\text{Sc}}$ negative appendix (fig 1C). The background single immunoreactive band seen in appendix is also present in a lymphatic follicle of the appendix (anti-PrP monoclonal antibody 12F10). The immunostaining pattern is similar to that reported in tonsils in vCJD patients and suggests deposition mainly in dendritic cells.
seen in normal tonsil and is attributable to weak cross-reactivity of the secondary antibody with an immunoglobulin fragment. While this band is consistently observed after high sensitivity enhanced chemiluminescence of total lymphoepithelial homogenate, it is not recovered after sodium phosphotungstic acid precipitation. We determined that the level of PrP<sup>Sc</sup> present in the brain of the vCJD patient with PrP<sup>Sc</sup>-positive appendix patient is approximately 15-fold lower than the maximum level we have observed in vCJD brain (fig 1D). Based upon these findings we estimate that biopsy tonsil and appendix, contain levels of PrP<sup>Sc</sup> of about 4% and about 0.5%, respectively, of that found in the brain of the same vCJD patient (see legend to fig 1).

Importantly, we were able to correlate the detection of PrP<sup>Sc</sup> by western blotting in vCJD appendix with the detection of abnormal prion protein staining by immunohistochemistry. Abnormal prion protein deposits were clearly seen on sections from the PrP<sup>Sc</sup>-positive vCJD appendix (fig 1E), while prion protein staining by immunohistochemistry was unremarkable in sections from the PrP<sup>Sc</sup>-negative vCJD appendix or on sections of appendix from the sporadic CJD or inherited prion disease cases (data not shown).

**Discussion**

Our findings, together with our previously reported inability to detect PrP<sup>Sc</sup> in two other vCJD appendixes, indicate that appendix does not reliably report vCJD infection even at the end stage of the disease. This observation must be considered when estimating the possible prevalence of vCJD based upon the analysis of archival appendectomy tissues. Only one of four appendixes in vCJD may contain detectable levels of PrP<sup>Sc</sup>, surgical instruments used for appendicectomy should remain a cause of concern for potential iatrogenic transmission of vCJD prions.

**Cytokine profiles in HIV seropositive patients with tuberculous meningitis**

The immunological response in pulmonary and pleural tuberculosis has been extensively studied. However, the response in tuberculous meningitis has not been well documented. In pulmonary disease, exposure to tuberculous antigens results in a T cell and natural killer cell and cytokine response, elaborating various cytokines, mainly of Th helper type 1 (Th1) origin. Stimulated macrophages elaborate tumour necrosis factor (TNF)α, interleukin (IL) 12, and IL 1, promoting further recruitment and activation of T lymphocytes and macrophages. TNFα correlates with disease severity and may contribute to tissue necrosis; however, TNFα has also contributed to survival in mouse studies. Transforming growth factor β (Th3 cytokine) suppresses macrophage activation. IL 2 may be beneficial in promoting an immune response in HIV seropositive patients. Th1 and Th2 cytokine responses have been observed in cerebrospinal fluid (CSF) of HIV seronegative patients with tuberculosis meningitis.

Tuberculous meningitis is diagnosed on clinical and CSF examination. CSF analysis is based on exclusion of viral, acute bacterial, and other causes of aseptic meningitis. Disease severity was assessed according to the Medical Research Council stages 1 to 3. HIV ELISA was done on all patients. CSF samples were subjected to microscopy, culture, and glucose and protein analysis. Venereal Disease Research Laboratory test, fluorescent treponemal antibody analysis, cryptococcal antigen analysis, viral studies, cistercerus ELISA, CD4 counts, and determination of concentrations of adenosine deaminase (ADA), CSF IgG, and albumin. For cytokine assays, CSF was centrifuged at 3000 g, and supernatant was aliquoted and stored at −70°C. TNFα, interferon (IFN) γ, and IL 10 concentrations were measured by ELISA kits (Genzyme Diagnostics, Cambridge, Massachusetts, USA) with detection limits of 3 pg/ml, 3 pg/ml, and 5 pg/ml, respectively.

Data were summarised as medians and ranges. Non-parametric Wilcoxon rank sum tests were used to compare HIV seropositive groups with HIV seronegative groups, tuberculous meningitis severity groups, and groups derived according to the blood brain barrier index for cytokine concentrations. Spearman’s rank correlation was used to derive correlations of cytokine concentration, ADA concentrations, and CD4 counts in CSF.

There were 27 patients: 18 (67%) women and 9 (33%) men. Seventeen were HIV seropositive and 10 HIV seronegative. The average interval between onset of symptoms and the first clinical assessment was 17 days (range 5–90 days) in 18 patients where this was recorded. The mean (SD) age was 26.8 (11.6) years. There was one patient aged 10 and one aged 60, and the rest were between 25 and 40. The cytokine concentrations were not analysed according to age, as this would make the categories too small and of little value. The IgG index was calculated for 23 patients. There was no significant difference between the HIV seropositive and HIV seronegative groups for ADA (p = 0.4) and CD4 counts (p = 0.19) in CSF and cytokine concentrations (table 1).

Ten patients (37%) were classified as having grade 1 tuberculosis meningitis (59%) had grade 2 and one (4%) grade 3, which for analysis was considered to be grade 2. Table 1 summarises the cytokine concentrations for patients in stages 1 and 2.

Patients with stage 2 disease had significantly stronger Th1 responses. There was no difference in the IL 10 concentrations. The two patients with stage 2 disease who died had very high IFN γ concentrations, both greater than 2048 pg/ml.

10 IL concentrations were moderately positively correlated with IFN γ concentrations (r = 0.53). The correlation coefficients were 0.40 for IFN γ, 0.33 for TNFα, and 0.22 for IL 10. Correlation coefficients between ADA and cytokine concentrations were 0.34 for IFN γ, 0.47 for TNFα, and 0.22 for IL 10. Cytokine concentrations correlated poorly with CD4 counts in CSF.

It is postulated that in HIV infection a predominant Th2 response accounts for extra-pulmonary disease. This study does not favour a predominance of either Th1 or Th2 in the disease process. It is possible that a Th0 response, which is a non-differential response seen soon in immune activation, was seen in our patients, as they were examined untreated and relatively early in the disease. Other investigators have also documented this phenomenon. The positive correlation between IFN γ and IL 10 suggests that these were produced concurrently. This may reflect a control mechanism regulating Th1 and Th2 responses.

There was no difference in cytokine and ADA concentrations and CD4 counts between HIV seropositive and HIV seronegative patients. It is known that the clinical response to antituberculous treatment in both groups is similar. Perhaps this similarity correlates with similar immune responses in both groups. The size of each group is small and a type 1 statistical error has to be considered.

**Table 1 Differences between HIV seropositive and HIV seronegative groups and tuberculous meningitis severity**

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>HIV positive</th>
<th>HIV negative</th>
<th>Stage 1</th>
<th>Stage 2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Median</td>
<td>Range</td>
<td>Median</td>
<td>Range</td>
</tr>
<tr>
<td>IFN γ</td>
<td>569.9</td>
<td>16.0–2048</td>
<td>890.6</td>
<td>0–2048</td>
</tr>
<tr>
<td>TNFα</td>
<td>1.6</td>
<td>0–0.675</td>
<td>9.8</td>
<td>0–309.3</td>
</tr>
<tr>
<td>IL 10</td>
<td>24.6</td>
<td>0–127.9</td>
<td>17.3</td>
<td>0–296.3</td>
</tr>
</tbody>
</table>

IFN, interferon; IL, interleukin; TNF, tumour necrosis factor.

References

Further studies to confirm our findings would be of value.

The significantly greater TNF-α and IFN-γ concentrations in the severe group of tuberculous meningitis is confirmed by other studies and suggests that disease severity results mainly from the immune response rather than the organism itself.

The lack of correlation between CD4 and cytokine concentrations may be explained by the fact that there are other sources of cytokines in the CSF, namely macrophages and natural killer cells. Concentrations of ADA, which are derived from lymphocytes, are consistent with other reports, where they were found to be correlated with cytokine concentrations.

There was no correlation between the IgG index and cytokine concentrations, suggesting that the blood brain barrier did not significantly influence concentrations. Unfortunately, corresponding serum concentrations were not available. This would have been valuable. This is the first study correlating CSF cytokine responses to severity of tuberculous meningitis and comparing HIV positive with HIV negative groups. Further studies should be done to confirm these findings, perhaps to define their relevance to complications and to explore the possibility of IL-2 treatment in HIV positive patients.

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VB Patel, Al Bhigjee, PLA Bill
Division of Neurology, Nelson R Mandela School of Medicine, University of Natal, Durban, South Africa

CA Connolly
Biostatistics, Medical Research Council, Durban, South Africa

Correspondence to: Dr VB Patel, Department of Neurology, Ward A3, Wentworth Hospital, Private Bag Jacobs, Durban 4026, South Africa; patelv@nsc.ac.za

References

Festination as the leading symptom of late onset idiopathic aqueductal stenosis
Late onset idiopathic aqueductal stenosis (IAS) may be manifest clinically either by headaches or by hydrocephalic symptoms such as gait disturbance, urinary urge, and other symptoms. In particular, there was no rigidity or tremor, parasitis, or impairment of joint position sense. The Middlesex elderly assessment of mental state and Wechsler memory scales showed no significant abnormality. Magnetic resonance imaging showed a lateral and third ventricular hydrocephalus with small aqueduct and normal fourth ventricle. Serial lumbar punctures, which showed pressures from 10–14 cm H₂O, had no beneficial or adverse effects. Late onset IAS was diagnosed. The patient underwent ventriculoperitoneal shunting with a medium pressure valve and an infusion device. Postoperatively, she made a slow but steady recovery. Two months after shunting her postural stability and balance had improved considerably. She no longer walked with a stooped posture and her speed of walking had improved significantly. She is now able to walk around the house with the assistance of a cane.

Discussion
Festination was the leading symptom of late onset IAS in both patients reported here. Lack of awareness of this association may cause diagnostic delay and may be related to functional disturbance of the third ventricle. The anatomical substrates of festination have not been elucidated but it is most likely related to functional disturbance of the thalamus or brainstem locomotor centres. This assumption would also explain the reversibility of festination after third ventriculostomy and shunting. In summary, this is the first report of the occurrence of festination as the leading symptom of IAS. As shown, festination may be misinterpreted and be considered psychogenic in an elderly patient who otherwise is suffering only mild hydrocephalic symptoms.
Myoclonic movement disorder associated with microdeletion of chromosome 22q11

With a prevalence of approximately 1:4000 interstitial chromosome 22q11 deletion within the DiGeorge syndrome critical region is the commonest chromosome microdeletion syndrome. The better known clinical features of this disorder are cardiac abnormalities, short stature, palatal abnormalities or velopharyngeal insufficiency, renal abnormalities, hypocalcaemia, psychotic symptoms, learning difficulties, and developmental delay.

There is wide variability in this clinical spectrum and many case reports draw attention to new clinical features that have been published. Alongside the larger studies of 22q11 cohorts these have proved useful in delineating this particular syndrome.

Case report

We present a family where the proband at 3 years of age exhibited the typical facial features of deletion of chromosome 22q11 (fig 1a) of low set laterally rotated ears, small mouth and mandible, short philtrum, and short palpebral fissures, as well as developmental delay. His height was on the fourth centile and he also had pectus carinatum. His mother (fig 1b) also had learning difficulties and delayed motor milestones as a child and subsequently developed an unusual movement disorder, which was first noted as a tremor at age 5 and was brought to medical attention in her teens. The movement disorder consisted of longstanding pronounced myoclonus of the head, trunk, and limbs, which is worsened by intention and exacerbated by startle. She had slow tongue movements. Eye movements were normal. Extensive neurological investigations were normal: caeruloplasmin, molecular testing for Huntington’s disease, myotonic dystrophy, mitochondrial mutations, muscle biopsy, electromyography, and magnetic resonance imaging of the brain. Echocardiography and renal ultrasound in both mother and son were also normal. Although standard cytogenetic testing showed a normal karyotype, fluorescence in situ hybridisation analysis with the chromosome 22q11 deletion.

Discussion

In the European collaborative study presenting data on 558 patients with deletions within the DiGeorge syndrome critical region showed that both mother and son had a deletion in this region.

Figure 1  [a] Proband with facial features of 22q11 microdeletion syndrome. [b] Mother of proband.

References

Phenobarbitone induced gingival hyperplasia

Among the long term adverse effects of anticonvulsants, gum hyperplasia is cosmetically disturbing and can give rise to complications such as bleeding and gingivitis. Long term use and high doses of phenytoin sodium (diphenylhydantoin) have been implicated in the occurrence of gingival hyperplasia. Serum phenytoin level in these patients are high. Rarely other drugs such as primidone, sodium valproate, nifedipine, and cyclosporin have also been implicated.

Case report

A 26 year old man had been suffering from hot water reflex epilepsy and primary generalised tonic-clonic seizures since he was 12 years of age. The seizures were poorly controlled because of his inability to avoid taking hot baths, inadequate dosage of anticonvulsants, and poor compliance. Since 1987, he had been taking phenobarbitone, 60 to 90 mg/day. In 1988, phenytoin (100 mg twice daily) was added for six months but was stopped because there was no benefit. In 1989, carbamazepine (200 mg twice daily) was added but was discontinued by the patient in 1996 for unspecified reasons. From 1996 onwards he had been receiving only phenobarbitone 120 mg/day. He was referred to us for progressive gum hyperplasia with bleeding from the gums, both spontaneously and with tooth brushing, since 1996. There was no history suggesting any haematological malignancy, and he was not on any other drug treatment.

On examination, he had gingival hyperplasia involving both the upper and the lower jaws (fig 1). There was occasional bleeding from the hyperplastic gum tissue. General physical examination did not reveal any pallor, hepatosplenomegaly, or lymphadenopathy and neurological evaluation was unremarkable.

Haematological investigations, including haemoglobin, white blood count, erythrocyte sedimentation rate, and peripheral smear, were all normal. Blood glucose, renal and liver function tests, serum electrolytes, and urine analysis were normal. Scalp electroencephalography and cranial computed tomography were normal. Serum concentrations of phenobarbitone, phenytoin, and carbamazepine were measured. Phenobarbitone was within the therapeutic range (11.2 µg/ml) while the other two drugs were not detectable.

Discussion

This patient with longstanding epilepsy probably had phenobarbitone induced gingival hyperplasia. He had used phenytoin in 1988 for only six months without developing this symptom, and it only appeared later in 1996 when he was on phenobarbitone alone. After that time it ran a progressive course. Primidone, the metabolites of which contain phenylthylmalonamide and phenobarbitone, has also been reported to be a rare cause of gingival hyperplasia. However, it is difficult to state which of the two components contributes to the hyperplasia.

Phenytoin sodium is known to be the most common cause of gingival hyperplasia. A minor degree of hyperplasia occurs quite commonly and this generally causes only cosmetic problems, especially for women. However, when phenytoin is used for prolonged periods in high dosage a severe degree of gingival overgrowth occurs. In such cases the hyperplastic gingiva bleed with minimal trauma or spontaneously, and may sometimes become secondarily infected.

The mechanism of anticonvulsant induced gingival hyperplasia is not known. It is believed that these drugs cause tissue collagen proliferation but the reason for this is unknown. It has been speculated that a high phenytoin concentration in the pituitary and adrenal glands may be related to hirsutism and gingival hyperplasia. However, there is at present no evidence that high concentrations of phenytoin and these tissues produce changes in their secretion rate, or indeed that these alterations cause hirsutism and gingival hyperplasia.

Antiepileptic drug levels are generally high in cases of gingival hyperplasia. However, in our case the phenobarbitone level was within normal therapeutic limits, while phenytoin and carbamazepine were undetectable.

Treatment consists of stopping the offending drug and providing supplements of folic acid and ascorbic acid. If regression does not occur, reconstructive surgery of the hyperplastic gingival tissue is advised. Our patient has been switched to sodium valproate, and reconstructive cosmetic surgery is planned.

In conclusion, we report a case of phenobarbitone induced gingival hyperplasia because of its rarity. The mechanism of this side effect remains unclear. Accurate drug history, thorough investigations, and punctilious reporting of such cases may help us gain a better understanding of the condition.

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S Sinha, V Kamath, G R Arunodaya, A B Taly

Department of Neurology, National Institute of Mental Health and Neurosciences (NIMHANS), Bangalore 560 029, India.

Correspondence to: Professor Arun B Taly; abtaly@yahoo.com

References


Hashimoto’s encephalopathy mimicking Creutzfeldt-Jakob disease: brain biopsy findings

A previous report in this journal described seven cases of Hashimoto’s encephalopathy (HE) clinically resembling Creutzfeldt-Jakob disease (CJD). Brain biopsies in such cases are rare and have suggested “vasculitis”. We contribute a report of rapidly progressive dementia in a patient undergoing brain biopsy before the diagnosis of HE was established, showing features suggesting early spongiform change but with inflammation.

A 57 year old woman was taken to a local hospital following a generalised seizure. She was discharged that night after negative cranial computed tomography and cerebrospinal fluid (CSF) analysis. Within a few days she was noted by family members to be acting strangely and hallucinating. Her doctor found her to be somnolent and rigid without focal neurological findings. Magnetic resonance imaging of the brain showed a questionable increase in gadolinium contrast uptake in a 7 mm area of the left medial frontal cortex. An electroencephalogram (EEG) showed bihemispheric slowing without epileptiform activity. Despite an extensive inpatient evaluation (including biochemical, haematological, endocrine, infectious, autoimmune, and toxic analyses), no cause for the encephalopathy could be found. The patient was then referred to our institution for brain biopsy and further care.

Samples of left frontal cortex showed light microscopic evidence of rare vacuoles abutting neurons, suggesting early spongiform...
change (fig 1). Gliarial fibrillary acid protein staining showed moderate gliosis. A few perivascular lymphoid cells and macrophages were present, with microglia scattered throughout the parenchyma; there was no evidence of true vasculitis (inflammation and fibrinoid necrosis of arterial vessels) or of microglial nodules. Because prion disease could not be excluded historically and because the clinical suspicion of CJD remained high, a tissue block was sent to the National Prion Disease Reference Laboratory (NPDRL).

Subsequently the patient’s condition deteriorated. She became wheelchair bound, akinetic, and mute, with startle myoclonus and prominent frontal release signs. CSF analysis showed increased protein (890 mg/l, reference range < 450 mg/l)). A sample was sent to be tested for the 14–3–3 protein, which is a CNS protein detected in spinal fluid (named after the chemical formula for one of its seven distinct isofoms and two phosphorylated forms) and has been shown recently to have a high sensitivity and specificity (96% and 99%, respectively) for the diagnosis of CJD. The finding of the NPDRL that of the EEG has recently been added to the clinical criteria for the "probable" sporadic form of the prion disease. 14 Magnetic resonance imaging repeated with gadolinium and diffusion weighted sequences showed only postictal changes. Repeat EEG showed continued bihemispheric slowing with the addition of triphasic waves. The patient had a family history of hypothyroidism but at the time of admission was euthyroid (thyroid stimulating hormone 2.89 mU/l, reference range 0.5–5 mU/l; thyroxine 65.79 pmol/l, reference range 64.5–141 pmol/l; triiodothyronine 1.386 pmol/l, reference range 1.078–2.464 pmol/l). Nevertheless, in the absence of a clear cut cause and in view of prior reports of the clinical overlap between CJD and HE, we measured thyroid peroxidase antibodies, which were increased (79 IU/ml, reference range 0–20 IU/ml). An ultrasound of the thyroid showed several hypoechogenic areas, fine needle biopsies of which were non-diagnostic. The fact that many euthyroid middle aged women can be shown to have mildly increased thyroid peroxidase antibodies inevitably gave rise to speculation that the finding was incidental. However, in the absence of another obvious cause for the rapidly evolving dementia, and giving due weight to the findings in previous studies that the majority of reported cases were euthyroid, we began steroid treatment for a presumptive diagnosis of HE. 14 Within 24 hours the patient had visible neurological improvement and within one week she was mobile and conversant, with reversal of most of her cognitive and motor deficits. Shortly thereafter, the NPDRL reported that the tissue did not show evidence of abnormal prion protein. The CSF did not contain the 14–3–3 protein. The patient was discharged on a slow taper of steroids. Six weeks later she was seen in the outpatient clinic and was considered fully recovered by her husband and her doctors.

Rapidly progressive encephalopathy with myoclonus, extrapyramidal rigidity, and akinetic mutism suggest the diagnosis of prion disease such as CJD. However, the differential diagnosis is wide and includes infections of the CNS, toxins, vitamin B12 deficiency, hyperthyroidism, autoimmune or idiopathic cerebrovascularitis, paraneoplastic encephalitides, epileptic pseudodementia, and other rapidly progressive dementias, especially familial forms of Alzheimer disease. HE, a disorder of presumed autoimmune origin in which patients present with altered levels of consciousness, seizures, and dyskinesias, 5,6,7,8 extrapyramidal rigidity, has recently been added to this list. 14 Patients with HE are usually euthyroid, with increased antithyroid antibodies often discovered only after the diagnosis of encephalopathy. 14

Although presentation with a seizure is unusual in CJD, our patient’s other clinical features of rapid cognitive deterioration, myoclonus, and extrapyramidal rigidity were highly suggestive of a diagnosis of CJD. The initial laboratory evaluation, the lack of convincing imaging findings, and the EEG were also consistent with early prion disease. In fact, according to current organizational criteria, the clinical and EEG features of this case alone already indicated a diagnosis of “probable” CJD. 14 Furthermore, the presence of vacuoles on the brain biopsy was very suggestive of the spongiform change (due to swelling of neuritic processes) in early CJD, a diagnostic possibility additionally evoked by the moderate gliosis. Although the absence of pathognomonic amyloidoid (“kuru”) plaques on a limited biopsy does not exclude a diagnosis of “definite” CJD in a suspected patient, we felt that the features of the case justified immunooassay for prion protein in the tissue and CSF. The presence of perivascular mononuclear cells, though sparse, is not typical for CJD, and may, in the end, be the greatest supportive evidence of encephalitis as the true cause of the symptoms. The changes are reminiscent of those illustrated by Nolte et al., although we strongly disagree with their term “vascularitis”, instead preferring to reserve that term for true necrotising arteritis, which implies a different pathological process. Unlike their patient in whom the inflammation was confined to basal leptomeninges in the setting of prior subarachnoid haemorrhage, ours involves parenchymal vessels, with permeation of the cortex by microglia, suggesting a more credulable substrate for the seizures and cognitive deficits. In conclusion, we recommend that patients who, according to World Health Organization criteria, have clinical and EEG features of probable CJD be considered for empirical treatment with steroids for HE, pending laboratory results for antithyroid antibodies as well as for 14–3–3 protein. Furthermore, we present additional evidence that HE is an encephalitic process, likely autoimmune given the dramatic response to anti-inflammatory treatment.

C P Doherty, M Schlossmacher, N Torres, E Bromfield, M A Samuels
Department of Neurology, Partners Neurology, Brigham and Women’s Hospital, Massachusetts General Hospital and Harvard Medical School, Boston, Massachusetts 02115, USA

R Folkert
Department of Pathology (Neuropathology)

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Correspondence to: Dr C P Doherty; cpdoherty@partners.org

Long term tolerability of high dose ergoline derived dopamine agonist therapy for the treatment of Parkinson’s disease

During the past decade new direct acting dopamine agonists (DAs) have become widely deployed for the treatment of the early stages of Parkinson’s disease (PD), because randomised controlled trials (RCT) have demonstrated that their use is associated with a reduced incidence of motor fluctuations and dyskinasias compared to levodopa therapy. 1,2,3,4,5,6,7,8,9,10,11,12 However, published randomised controlled trials of DAs with levodopa have tended to focus on the development of motor fluctuations and dyskinasias as primary end points. 1,2,3,4,5,6,7,8,9,10,11,12 Curiously these studies have also shown that the effect of levodopa on motor function or activities of daily living (measured by Unified Parkinson’s Disease Rating Scale (UPDRS) parts II and III) was superior to that obtained by the DAs, irrespective of whether the latter were administered as monotherapy or with open label levodopa supplement.

Our understanding of the reasons behind why the DAs were consistently less effective than levodopa on the UPDRS is limited. Particularly as these RCTs were performed double blind. Two plausible explanations are: (1) the UPDRS does not capture the whole picture; for example, the DAs and levodopa may have differential effects on mood; or (2) the doses (therapeutic levels) deployed in these studies were set so that the DAs were slightly suboptimal compared to levodopa, but this difference was not apparent to the blinded clinicians assessing the trial patients.
The latter notion encouraged us to examine the dose of the DAs used to treat PD in routine clinical practice. Thus we systematically reviewed the database of all the PD patients under our care in order to identify those receiving DA therapy (cabergoline, pergolide, pramipexole, or ropinirole) above their maximum recommended therapeutic daily dose (respectively 6 mg, 5 mg, 4.5 mg, and 24 mg). This was probably because of our greater duration of therapeutic experience and pergolide therapy.

We identified 18 patients fulfilling these criteria. All were either on cabergoline (n = 11) or pergolide (n = 7), which are ergoline derived dopamine agonists. This represents about 23% and 27% of PD patients in current clinical practice. However, we do not know whether this capacity is confined to ergoline derived dopamine agonists and pergolide therapy in Parkinson’s disease.

Table 1 illustrates the clinical characteristics of these patients and shows that cabergoline and pergolide are tolerable, on average follow up periods of 2.3 and 2.5 years respectively, at dosages well above those recommended in their data sheets. Indeed it is clear from these data that the therapeutic windows of the ergoline derived dopamine agonists are at least twice that presently acknowledged.

Consequently, we speculate that the ergoline derived dopamine agonists (cabergoline and pergolide) may not have been deployed to their full therapeutic potential, either in the major RCTs comparing DAs with levodopa or in current clinical practice. However, we do not know whether this capacity is confined to the ergoline derived DAs. As the direct acting DAs are now considered to be first line therapy for younger people with Parkinson’s disease, we would be very interested to hear of other neurologists’ experiences of using these drugs for prolonged periods above their recommended doses. There is a case for further studies involving high dose dopamine agonist treatment for people with PD.

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Phenobarbitone induced gingival hyperplasia

S Sinha, V Kamath, G R Arunodaya and A B Taly

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