Accidental transmission of Creutzfeldt-Jakob disease by dural cadaveric grafts

Juan F Martínez-Lage, Máximo Poza, Joaquín Sola, Jose G Tortosa, Paul Brown, Larisa Cervenáková, Juan A Esteban, Andrés Mendoza

Abstract

Four patients who received dural grafts of cadaveric origin in the course of posterior fossa procedures subsequently developed Creutzfeldt-Jakob disease (CJD). The interval from dural placement to clinical onset of CJD ranged from 16 months to nine years. Initial clinical presentation consisted of cerebellar symptoms, with dementia and myoclonus developing in later stages of the disease. EEGs showed diffuse slowing that evolved to a periodic activity pattern. CT and MRI were unremarkable in the early stages but pronounced cerebral and cerebellar atrophy with widened sulci and collections of fluid over the convexities were seen in the late stages of disease. The diagnosis was histologically proved by brain biopsy in all four cases. Molecular genetic analysis showed that the four patients were homozygous for methionine at codon 129 of the PrP gene. From this experience, and from six previous descriptions of this occurrence in the literature, it is manifest that awareness of the means of iatrogenic transmission of CJD, and the adoption of preventive measures, constitute the only effective way to stop the spread of CJD among patients who have neurosurgery.

(*J Neurol Neurosurg Psychiatry* 1994;57:1091-1094)

Creutzfeldt-Jakob disease (CJD) is a progressive fatal form of dementia that is caused by an as yet undefined transmissible agent.^{1,2} The triad of dementia, myoclonus, and triphasic periodic activity on EEG is virtually diagnostic of the disease.¹ In its spontaneous form, CJD is mainly a disorder of elderly people. The estimated prevalence in Europe and in the United States is about 0.5-1 cases per million yearly.^{1,3} For persons younger than 30 years, the estimated incidence is less than one case per 200 million.⁴ When the disease occurs in a young patient, especially after treatment with hormones of cadaveric origin or after surgical procedures, iatrogenic CJD must be considered as a possibility.

In 1988, Thadani *et al*⁵ published the first case of CJD in a patient treated with a cadaveric dura mater graft. At that time we had already experienced two instances of dura mater related CJD, although we were uncertain about the part played by the dural graft.

The recent occurrence of two further cases of CJD in recipients of dural implants has prompted us to review our experience with these four iatrogenic cases of CJD.

Clinical material and methods

We reviewed the medical records of the four patients who developed CJD after posterior fossa operations in which dural closure was achieved with a graft of cadaveric origin (Lyodura; B Braun, Melsungen AG, Germany). Pertinent clinical data, including surgical antecedents and family history of dementia were analysed, as were the results of EEG, CT, MRI, and those of the brain biopsies.

Characterisation of codon 129 of the PrP gene was performed on DNA extracted from anticoagulated blood or paraffin embedded brain tissue. The fragment comprising codons 108 to 253 was amplified by the polymerase chain reaction with *Taq* polymerase (Perkin Elmer, Norwalk CT, USA) and two synthetic primers: NC (sense) (5'TACTGAGCGGCC GCCAACATGAAGCACATGGCTGGT3') and SS (antisense) (5'TACTGAGTCGAC CCTTCCTCATCCCACATATCAGG3'). The amplified fragment was digested with the restriction enzyme *Mae* II (Boehringer Mannheim, Indianapolis IN, USA) and then analysed by electrophoresis in a horizontal 3% Metaphor agarose gel (FMC bioproducts, Rockland ME, USA) in Tris/borate/EDTA buffer at 80V for three hours. DNA was stained with ethidium bromide.

Patients

CASE 1

On 22 November 1983, a 17 year old boy received a Lyodura graft after the removal of a benign cerebellar astrocytoma. Sixteen months later, the patient was readmitted to hospital complaining of blurred vision, dysarthria, and unsteady gait. The patient was conscious and had pallor in both optic discs, left dysmetria, and an ataxic gait. CT showed slight ventricular enlargement. Results of analysis of CSF were normal. After two weeks, the patient was demented and exhibited generalised myoclonic jerks. An EEG showed diffuse slow activity and periodic paroxysms of spike waves bilaterally. A right frontal biopsy was performed that confirmed CJD. A second CT showed pronounced generalised brain atrophy. The patient died in 1986, 21 months after the diagnosis of CJD.

Regional Service of Neurosurgery, Virgen de Arrixaca University Hospital, Murcia, Spain

J F Martínez-Lage
M Poza
J G Tortosa
J A Esteban
A Mendoza

Section of Neuropathology, Virgen de Arrixaca University Hospital, Murcia, Spain
J Sola

Laboratory of Central Nervous System Studies, National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, Maryland, USA
P Brown
L Cervenáková

Correspondence to: Dr Juan F Martínez-Lage, Regional Service of Neurosurgery, Virgen de Arrixaca University Hospital, E-30120 Murcia, Spain.

Received 25 October 1993 and in revised form 26 January 1994. Accepted for publication 21 February 1994

CASE 2

A 53 year old woman was given a posterior fossa decompression, which included a Lyodura graft, for treatment of Chiari malformation and syringomyelia, on 24 January, 1984. Forty three months later she was readmitted with the complaints of dizziness, dysarthria, oscillopsia, and somnolence. The patient was conscious, hypotonic, and had an ataxic gait. CT showed faint bilateral calcifications in the basal ganglia. An EEG showed generalised slowing of electrical activity. The CSF was normal. Two weeks after admission the patient had myoclonus and dementia. A repeat EEG showed periodic activity, but not the classical triphasic waves. A cerebral biopsy showed the findings of CJD. A second CT showed asymmetry of the lateral ventricles and enlarged sulci. The patient died in 1989, 25 months after clinical onset.

CASE 3

A 10 year old boy⁶ was first treated with a ventriculoperitoneal shunt, and on 18 April 1983 underwent removal of a grade 2 cerebellar astrocytoma, which included dural closure by a Lyodura graft. The child was subsequently treated by cobalt therapy to the posterior fossa. During the postoperative period he required a blood transfusion. Seventy nine months after operation, the patient developed slurred speech, dizziness, and unsteady gait. He had pale optic discs, bilateral dysmetria and hyperreflexia, dysarthria, and gait ataxia. CT and MRI were unremarkable. An EEG showed a pattern of diffuse slowing. The CSF contained 165 mg/dl protein. The boy exhibited aggressive behaviour and two weeks later, was demented, mute, and had a decorticate posture and myoclonic jerks. A second EEG showed typical periodic triphasic waves. On 23 December 1991 a brain biopsy confirmed the diagnosis of CJD. He died in 1992, three months after clinical onset.

CASE 4

A 25 year old man, diagnosed as having a Chiari malformation and syringomyelia, was given a posterior fossa decompression on 5 December 1983. The procedure ended with the placement of a Lyodura graft. One hundred and five months later the patient complained of dizziness, unsteadiness, and dysarthria. Clinical examination showed pale

optic discs, nystagmus, hyperreflexia, and an unstable gait and stance. Subsequent clinical evolution was very rapid with hallucinations, aggressiveness, and then mutism and decorticate posturing with myoclonic movements in all four limbs. An EEG showed some periodic activity. Cerebral CT and MRI showed mild cerebral atrophy. Brain biopsy, performed on 13 January 1993, was consistent with CJD. In January 1994, the patient was still in hospital.

The table summarises the relevant clinical features of the four patients. None had undergone prior surgical procedures, had received hormones obtained from cadaveric sources, or had a family history of CJD or of other illnesses causing dementia. All four patients were operated on at our hospital between April 1983 and January 1984.

Molecular genetic analysis

Paraffin embedded brain tissue was available from cases 1, 2, and 3, and blood and frozen cerebral tissue were available from case 4 for molecular genetic analysis. All four specimens were homozygous for methionine at codon 129 of the PrP gene.

Discussion

AETIOLOGY AND PATHOGENESIS OF CJD
Spongiform encephalopathies are a group of human or animal diseases, transmissible in the laboratory, and caused by unconventional agents, with the common histopathological finding of spongiform degeneration of the CNS.¹ CJD and kuru in humans, and scrapie, bovine spongiform encephalopathy, and mink encephalopathy in animals, are the most well known examples of these diseases.¹ Initially the agent responsible for CJD was thought to be a "slow virus". Virus like particles have also been described, and recently a "prion" (protein with no identifiable nucleic acid) has been suggested as the causative agent.² Although several epidemiological studies of CJD exist, the cause of spontaneous cases is unknown.^{1-3 7-9}

There are two recognised means of transmission of CJD: experimental and iatrogenic. CJD has been described as the result of hormone treatment with extracts of pooled cadaveric pituitary glands (growth hormone and gonadotropins).¹⁰⁻¹⁶ In these instances the incubation period ranged from four to 22

Iatrogenic CJD (Lyodura): clinical features

No	Age, sex	Operation, date	Incubation period (months)	Initial symptoms	EEG	Biopsy	Outcome (months after onset)
1	17 M	Cerebellar astrocytoma, 22 Nov 1983	16	Unsteadiness blurred vision	+ / + +	Yes	Dead (21)
2	57 F	Arnold-Chiari post fossa decompression 1 Jan 1984	43	Unsteadiness, dysarthria	+ / + +	Yes	Dead (25)
3	10 M	Cerebellar astrocytoma, 18 Apr 1983	79	Unsteadiness dysarthria	+ + +	Yes	Dead (3)
4	25 M	Arnold-Chiari, post fossa decompression, 5 Dec 1983	105	Unsteadiness dysarthria	+ / + +	Yes	In hospital (15)

EEG: + = slowing; ++ = slowing plus periodic waves; +++ = triphasic waves.

years. In hormone related CJD cerebellar signs are prominent, and dementia and myoclonus are either absent or present late during the course of the disease.¹⁶ EEG rarely shows periodic wave activity. Iatrogenic CJD has also been amply documented after surgical procedures, especially neurosurgical interventions.^{7 8 17-22} In these instances the incubation period has ranged from 15 months to 10 years (average 32 months).

IATROGENIC CASES OF CJD AFTER CADAVERIC DURA MATER GRAFTS

Recently, Thadani *et al* described a patient who acquired CJD from a dura mater graft.⁵ Six additional cases of CJD after dural grafts of cadaveric origin have also been documented, five after Lyodura implants,²³⁻²⁶ and one after a dural graft from a non-commercial tissue bank.²⁷ In these reports the incubation period averaged 47 months. Although these seem to be the last cases to be reported after the use of cadaveric dura mater grafts, the recent occurrence of CJD in our third and fourth patients, eight and nine years after surgery respectively, has convinced us of the possibility of other instances evolving with a more protracted course. The unusually long incubation period of our last two cases could be related either to a less virulent strain of the agent, or to less infectivity in the implanted grafts.

Disease transmission by contaminated instruments or by other means remains a possibility in our cases. The hypothesis that they became infected by the dural graft is sustained, however, by the following facts: (1) CJD has been described in young patients almost exclusively as the result of iatrogenic transmission; (2) no other instances of CJD have been noted among the 1052 patients subjected to neurosurgical operations at our hospital during the period 1983-4 other than the four out of 36 who received dural grafts; (3) similar reports exist in the current literature of CJD associated with the use of cadaveric dural grafts; and (4) grafts of cadaveric origin were incompletely sterilised (The CJD agent is resistant to the methods of sterilisation used until 1985 by the Lyodura manufacturers.^{28 29})

DIAGNOSIS OF IATROGENIC CJD

The triad of dementia, myoclonus, and triphasic waves in the EEG seems to constitute sufficient criteria for the clinical diagnosis of CJD.¹ Iatrogenic cases of CJD usually follow a different clinical course, however, in which cerebellar manifestations are more evident, and the EEG may not present the classical triphasic pattern of periodic activity.¹⁶ CT and MRI are unremarkable in early stages of CJD, showing atrophy only in the latest phases of its evolution. At present, the only way of establishing the diagnosis is by histological verification; however, the practicality of conducting a cerebral biopsy may be questioned, in view of the reluctance of medical and paramedical personnel to perform biopsies or necropsies on patients with CJD.^{1 28}

Also, some authors have noted a worsening of the patients' condition after brain biopsy, although this did not occur in our patients.

MOLECULAR GENETIC ANALYSIS

All four specimens studied by molecular genetic analysis were homozygous for methionine at codon 129 of the PrP gene, as were all five previously tested cases from other countries. All of the patients received Lyodura grafts between 1983 and 1985, and the uniform occurrence of methionine homozygosity in these cases raises the interesting possibility that the contaminating donor (or donors) may have also been homozygous for methionine at codon 129, in which event a genetic "match" may have increased susceptibility to iatrogenic infection. This speculation unfortunately cannot be verified, as donor grafts from the 1983-5 period are no longer available for analysis.

PREVENTIVE MEASURES

Precautions for handling cases of CJD, and sterilisation measures for instruments and objects in contact with these patients, have been amply publicised.^{28 29} Although it has been suggested that surgical instruments may be decontaminated by autoclaving at 134°C for one hour, or by immersion in 5% sodium hypochlorite for one hour, current evidence suggests that this may be inadequate. In the United Kingdom present policy requires destruction of all neurosurgical instruments used in patients with suspected CJD.

Many neurosurgical conditions, including head trauma, cranial and spinal tumours, and congenital malformations (cephalocele repair, Arnold-Chiari decompression, etc.), may require dural grafting. Dura mater grafts are also used in general and paediatric surgery for large defects of the abdominal wall, and in maxillofacial procedures. At present, the preferred meningeal substitute is the dura mater homograft from cadaveric origin. Diringer³⁰ investigated chemical disinfection of dural tissue, and found a decrease in infectivity of this tissue after treatment with 1N NaOH, but even this method does not guarantee its complete sterilisation. Cases of CJD associated with the use of cadaveric dural implants show the necessity of a careful selection of organs and tissue donors, although the most rigorous screening cannot ensure the exclusion of donors with preclinical CJD.^{4 5 31} Neurosurgeons must consider the alternative use of autologous tissues, such as temporalis fascia or fascia lata, or of synthetic dural substitutes. In the event that cadaveric dural grafts are used, the lot number should be recorded in the operation protocols, and if feasible, a piece of tissue should be kept for the investigation of possible future cases of iatrogenic CJD (or other transmissible diseases).

Conclusions

We have reported four patients who most probably acquired CJD as a consequence of the use of cadaveric dural grafts.

Unfortunately when these operations were performed, the possibility of CJD transmission by dural grafting was unknown. The potential risks of using tissues and extracts from cadaveric origin should be strongly weighed against their beneficial effects, and disposable medical and surgical instruments should be used in cases of suspected CJD.

The publication of this article was supported by Grant No 93/9028 from the Fondo de Investigaciones Sanitarias of the Spanish Social Security. We acknowledge the collaboration of Drs V Navarro and M G Villanueva (Ramón y Cajal Hospital) and Dr A Cabello (Doce de Octubre Hospital) for sharing tissue specimens from patients 1 and 2. We also appreciate the secretarial assistance of Mrs C G Navarro and Mr A L Alarcón.

- Matthews WB. Slow viruses and the central nervous system. In: Lambert HP, ed: *Infections of the central nervous system*. Philadelphia: Decker, 1991:329-42.
- Prusiner SB. Genetic and infectious prion diseases. *Arch Neurol* 1993;50:1129-53.
- Brown P, Cathala F, Raubertas RF, Gajdusek DC, Castaigne P. The epidemiology of Creutzfeldt-Jakob disease: conclusion of a 15-year investigation in France and review of the world literature. *Neurology* 1987;37:895-904.
- Janssen RS, Schonberger LB. Creutzfeldt-Jakob disease from allogeneic dura: a review of risks and safety. Discussion. *J Oral Maxillofac Surg* 1991;49:274-5.
- Thadani V, Penar PL, Partington J, et al. Creutzfeldt-Jakob disease probably acquired from a cadaveric dura mater graft. Case report. *J Neurosurg* 1988;69:766-9.
- Martínez-Lage JF, Sola J, Poza M, Esteban JA. Pediatric Creutzfeldt-Jakob disease: probable transmission by a dural graft. *Childs Nerv Syst* 1993;9:239-42.
- Matthews WB. Epidemiology of Creutzfeldt-Jakob disease in England and Wales. *J Neurol Neurosurg Psychiatry* 1975;38:210-3.
- Will RG, Matthews WB. Evidence for case-to-case transmission of Creutzfeldt-Jakob disease. *J Neurol Neurosurg Psychiatry* 1982;45:235-8.
- Harries-Jones R, Knight R, Will RG, Cousens S, Smith PG, Matthews WB. Creutzfeldt-Jakob disease in England and Wales, 1980-1984: a case-control study of potential risk factors. *J Neurol Neurosurg Psychiatry* 1988;51:1113-9.
- Brown P, Gajdusek DC, Gibbs CJ Jr, Asher DM. Potential epidemic of Creutzfeldt-Jakob disease from human growth hormone therapy. *N Engl J Med* 1985;313:728-31.
- Brown P. Human growth hormone therapy and Creutzfeldt-Jakob disease: a drama in three acts. *Pediatrics* 1988;81:85-92.
- Cochius JJ, Burns RJ, Blumbergs PC, Mack K, Alderman CP. Creutzfeldt-Jakob disease in a recipient of human pituitary-derived gonadotropin. *Aust N Z J Med* 1990;20:592-3.
- Fradkin JE, Schonberger LB, Mills JL, et al. Creutzfeldt-Jakob disease in pituitary growth hormone recipients in the United States. *JAMA* 1991;265:880-4.
- Collinge J, Palmer MS, Dryden AJ. Genetic predisposition to iatrogenic Creutzfeldt-Jakob disease. *Lancet* 1991;337:1441-2.
- Brown P, Gólfarb LG, Cervenáková L, et al. Iatrogenic Creutzfeldt-Jakob disease as a case study for the interplay of genes and modern medicine. *Neurology* 1994;44:291-3.
- Brown P, Preece MA, Will RG. "Friendly fire" in medicine: hormones, homografts and Creutzfeldt-Jakob disease. *Lancet* 1992;340:24-7.
- Duffy P, Wolf J, Collins G, DeVoe AG, Streeten B, Cowen D. Possible person-to-person transmission of Creutzfeldt-Jakob disease. *N Engl J Med* 1974;290:692-3.
- Nevin S, McMenemey WH, Behrman S, Jones DP. Subacute spongiform encephalopathy—a subacute form of encephalopathy attributable to vascular dysfunction (spongiform cerebral atrophy). *Brain* 1960;83:519-64.
- Gaches J, Supino-Viterbo V, Oughourlian JM. Unusual electroencephalographic and clinical evolution of a case of meningioma of the left temporo-occipital convexity. *Europ Neurol* 1971;5:155-64.
- Bernoulli C, Siegfried J, Baumgartner G, et al. Danger of accidental person-to-person transmission of Creutzfeldt-Jakob disease by surgery. *Lancet* 1977;i:478-9.
- Foncin J, Gaches J, Cathala F, El Sherif E, LeBeau J. Transmission iatrogène interhumaine possible de maladie de Creutzfeldt-Jakob avec atteinte des grains du cervelet. *Rev Neurol (Paris)* 1980;136:280.
- Tange RA, Troost D, Limburg M. Progressive fatal dementia (Creutzfeldt-Jakob disease) in a patient who received homograft tissue for tympanic membrane closure. *Eur Arch Otorhinolaryngol* 1990;247:199-201.
- Nisbet TJ, MacDonaldson I, Bishara SN. Creutzfeldt-Jakob disease in a second patient who received a cadaveric dura mater graft. *JAMA* 1989;261:1118.
- Massullo C, Pocchiari M, Mascchi G, Alema G, Piazza G, Panzera MA. Transmission of Creutzfeldt-Jakob disease by dural cadaveric graft. *J Neurosurg* 1989;71:954-5.
- Miyashita K, Inuzuka T, Kondo H, et al. Creutzfeldt-Jakob disease in a patient with a cadaveric dural graft. *Neurology* 1991;41:940-1.
- Willison HJ, Gale AN, McLaughlin JE. Creutzfeldt-Jakob disease following cadaveric dura mater graft. *J Neurol Neurosurg Psychiatry* 1991;54:940.
- Pocchiari M, Massullo C, Salvatore M, Genuardi M, Galgani S. Creutzfeldt-Jakob disease after non-commercial dura mater graft. *Lancet* 1992;340:614-5.
- Gajdusek DC, Gibbs CJ Jr, Asher DM, Brown P, Diwan A, Hoffman P, Nemo G, Rohwer R, White L. Precautions in medical care of, and in handling materials from, patients with transmissible virus dementia (Creutzfeldt-Jakob disease). *N Engl J Med* 1977;297:1253-8.
- Brown P, Gibbs CJ Jr, Amyx HL, Kingsbury DT, Rohwer RG, Sulima MP, Gajdusek DC. Chemical disinfection of Creutzfeldt-Jakob disease virus. *N Engl J Med* 1982;306:1279-82.
- Düringer H, Braig HR. Infectivity of unconventional viruses in dura mater. *Lancet* 1989;i:439-40.
- Gottesdiener KM. Transplanted infections: donor-to-host transmission with the allograft. *Ann Int Med* 1989;110:1001-16.