

Witness Name: P.B. RONAN

Statement No: WITN3029001

Exhibits: WITN3029002 – WITN3029004

Dated: June 2019

INFECTION BLOOD INQUIRY

EXHIBIT WITN3029004

Sporadic Creutzfeldt-Jakob Disease in 2 Plasma Product Recipients, United Kingdom

Patrick Urwin, Kumar Thanigalkumar, James W. Ironside, Anna Molesworth, Richard S Knight,
Patricia E. Hewitt, Charlotte Llewelyn, Jan Mackenzie, Robert G. Will



JOINTLY ACCREDITED PROVIDER
OF CONTINUING MEDICAL EDUCATION
FOR PROFESSIONAL CONTINUING EDUCATION

Medscape **ACTIVITY** EDUCATION

In support of improving patient care, this activity has been planned and implemented by Medscape, LLC and Emerging Infectious Diseases. Medscape, LLC is jointly accredited by the Accreditation Council for Continuing Medical Education (ACCME), the Accreditation Council for Pharmacy Education (ACPE), and the American Nurses Credentialing Center (ANCC), to provide continuing education for the healthcare team.

Medscape, LLC designates this Journal-based CME activity for a maximum of 1.00 AMA PRA Category 1 Credit(s)[™]. Physicians should claim only the credit commensurate with the extent of their participation in the activity.

All other clinicians completing this activity will be issued a certificate of participation. To participate in this Journal CME activity: (1) review the learning objectives and author disclosures; (2) study the education content; (3) take the post-test with a 75% minimum passing score and complete the evaluation at <http://www.medscape.org/journal/eld>; and (4) view/print certificate. For CME questions, see page XXX.

Release date: May 12, 2017; Expiration date: May 12, 2018

Learning Objectives

Upon completion of this activity, participants will be able to:

- Recognize the clinical features of 2 cases of sporadic Creutzfeldt-Jakob disease (sCJD) reported in patients with clotting disorders treated with fractionated plasma products.
- Identify the laboratory and pathology findings of 2 cases of sCJD reported in patients with clotting disorders treated with fractionated plasma product.
- Determine the clinical implications of 2 cases of sCJD reported in patients with clotting disorders treated with fractionated plasma products.

CME Editor

Jude Rutledge, BA, Technical Writer/Editor, Emerging Infectious Diseases. *Disclosure: Jude Rutledge has disclosed no relevant financial relationships.*

CME Author

Laurie Barclay, MD, freelance writer and reviewer, Medscape, LLC. *Disclosure: Laurie Barclay, MD, has disclosed the following relevant financial relationships: owns stock, stock options, or bonds from Aplynn; Biogen; Pfizer.*

Authors

Disclosures: Patrick Urwin, MBBS, MA (CANTAB); Kumar Thanigalkumar, MBBS, MRCP, FRCPATH; James W. Ironside, MD; Anna Molesworth, PhD; Patricia E. Hewitt, MD, FRCPATH; Charlotte A. Llewelyn, PhD; and Jan Mackenzie, PG Cert Epidemiology, have disclosed no relevant financial relationships. Richard S. Knight, BMBCh, FRCP (E), has disclosed the following relevant financial relationships: served as a speaker or a member of a speakers bureau for Pfizer Inc. Robert G. Will, MD, has disclosed the following relevant financial relationships: served as an advisor or consultant for LFB (Paris); Ferring Pharmaceuticals.

Author affiliations: University of Edinburgh Western General Hospital, Edinburgh, Scotland, UK (P. Urwin, J.W. Ironside, A. Molesworth, R.S. Knight, J. Mackenzie, R.G. Will); University Hospital Lewisham, London, UK (K. Thanigalkumar); National Health Service Blood and Transplant, London (P.E. Hewitt); National Health Service Blood and Transplant/Public Health England Epidemiology Unit, Cambridge, UK (C. Llewelyn)

DOI: <https://dx.doi.org/10.3201/eid2306.161884>

Sporadic Creutzfeldt-Jakob disease (sCJD) has not been previously reported in patients with clotting disorders treated with fractionated plasma products. We report 2 cases of sCJD identified in the United Kingdom in patients with a history of extended treatment for clotting disorders; 1 patient had hemophilia B and the other von Willebrand disease. Both patients had been informed previously that they were at increased risk for variant CJD because of past treatment with fractionated plasma products sourced in the United

SYNOPSIS

Kingdom. However, both cases had clinical and investigative features suggestive of sCJD. This diagnosis was confirmed in both cases on neuropathologic and biochemical analysis of the brain. A causal link between the treatment with plasma products and the development of sCJD has not been established, and the occurrence of these cases may simply reflect a chance event in the context of systematic surveillance for CJD in large populations.

Human prion diseases are a group of rare and fatal neurodegenerative diseases that include idiopathic (sporadic), genetic (inherited), and acquired (infectious) disorders (1). All are associated with the accumulation of an abnormal isoform of the prion protein (PrP^{Sc}) in the central nervous system (1). The most common human prion disease is the sporadic form of Creutzfeldt-Jakob disease (sCJD), which occurs worldwide with a relatively uniform incidence of 1–2 cases per million population per year, a peak incidence in the 7th decade of life, and a median duration of illness of 4 months. The relatively consistent mortality rates associated with sCJD, the overall random spatial and temporal distribution of cases, and the absence of any confirmed environmental risk factor have led to the hypothesis that sCJD occurs because of the spontaneous generation of PrP^{Sc} in the brain (1). In contrast, variant Creutzfeldt-Jakob disease (vCJD) is an acquired disorder that is most likely caused by the consumption of meat or meat products contaminated with the bovine spongiform encephalopathy agent. The median age at death in vCJD is 30 years, with a median duration of illness of 14 months. Most cases of vCJD have occurred in the United Kingdom, which has had the largest epizootic of bovine spongiform encephalopathy in the world. Of the 178 UK vCJD cases, 3 have been identified as cases of secondary transmission caused by the transfusion of nonleukodepleted red blood cell components from vCJD-infected blood donors.

Lookback studies have shown no evidence of transmission through blood transfusion in sCJD (2,3), despite the identification of PrP^{Sc} in some peripheral tissues (4) and experimental evidence, which demonstrated infectivity in blood (5), by using intracerebral inoculation of highly sensitive transgenic mice. The absence of clinical cases causally linked to past treatment with fractionated plasma products has been used as evidence of the safety of these products in relation to sCJD (6). These products are generally manufactured from the pooled plasma from several thousand donors; production using UK plasma was discontinued in 1999.

We describe 2 cases of sCJD in patients who had previously received treatment with UK plasma-sourced plasma products; both patients had been informed that they were at increased risk for vCJD because of that treatment. The clinical features and investigations in these cases were

typical of sCJD; the neuropathologic diagnosis in both cases was sCJD (subtype MM1).

The Investigation

The UK National CJD Research and Surveillance Unit has been carrying out systematic epidemiologic study of CJD since 1990. The methodology of this study has been published previously (7). In brief, patients with suspected CJD are referred by clinicians and visited by a research registrar, who obtains details of the clinical history and investigations, information on a range of possible risk factors, and past medical history. The Transfusion Medicine Epidemiology Review study investigates potential links between donors and recipients of labile blood components and, in cases of sCJD, investigates patients who have a history of blood donation or having received a blood transfusion.

Coordinated surveillance of CJD has been undertaken in the European Union since 1993 (8). National surveillance programs for CJD also are in place in several other countries, including Australia, Canada, Japan, and the United States.

Case 1

In 2014, a 64-year-old woman suffered a rapidly progressive dementia with deterioration in driving skills and balance disturbance, then limb coordination deficits with handwriting impairment. In the second month, her gait deteriorated, becoming shuffling and unsteady, she struggled to dress herself, and she had onset of daytime hypersomnolence. She became distractible, had visual misperceptions, emotional lability, and spatial memory problems. She was hospitalized at the beginning of the third month of her illness and had onset of cortical blindness, myoclonus, and akinetic mutism. She experienced rapid decline and died after a total illness duration of 3 months.

An electroencephalogram performed during the final stages of illness showed background slowing and runs of periodic complexes, and a magnetic resonance imaging (MRI) brain scan showed high signal in the caudate heads with posterior cortical ribboning. A cerebral spinal fluid (CSF) 14–3–3 assay and real-time quaking-induced conversion test for PrP^{Sc} both were positive. Prion protein gene (*PRNP*) sequencing showed no mutations with methionine homozygosity at codon 129.

Postmortem examination of the brain showed widespread spongiform encephalopathy of predominantly microvacuolar type. Immunocytochemistry for prion protein gave a widespread positive reaction in a granular/synaptic pattern (Figure). No plaques or plaque-like structures were identified. Results of immunocytochemistry for disease-associated prion protein were negative in peripheral nerve, liver, lymph node, appendix, and spleen. Western blot analysis of frontal cortex and cerebellum confirmed

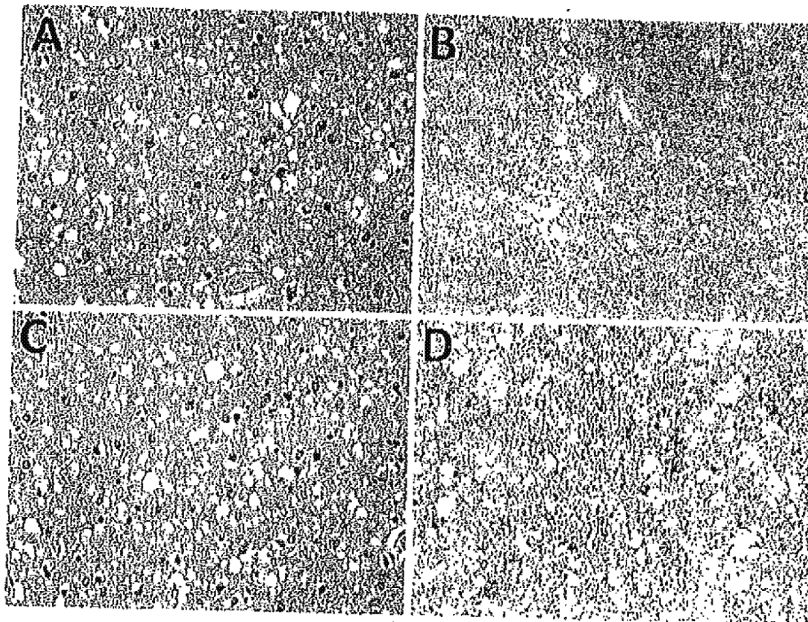


Figure. Results of neuropathologic examinations of the brains of the 2 patients with sporadic Creutzfeldt-Jakob disease, United Kingdom, 2014. A) Microvacuolar spongiform change in the frontal cortex (case 1). Hematoxylin and eosin stain; original magnification $\times 400$. B) Fine granular/synaptic accumulation of abnormal prion protein in the cerebral cortex (case 1). 12F10 anti-prion protein antibody; original magnification $\times 400$. C) Microvacuolar spongiform change with neuronal loss and gliosis in the frontal cortex (case 2). Hematoxylin and eosin stain; original magnification $\times 400$. D) Focally intense granular/synaptic accumulation of abnormal prion protein in the cerebral cortex (case 2). 12F10 anti-prion protein antibody; original magnification $\times 400$.

the presence of protease-resistant prion protein with a type 1A isoform.

The patient had been diagnosed with von Willebrand disease in childhood. Her early therapies include numerous transfusions of red blood cells and platelets; in more recent years, she received plasma-derived and recombinant factor VIII and additional blood component transfusions at times of hemorrhage. Factor VIII was administered on 4 occasions in the 1990s and during 2000–2004 and von Willebrand factor/factor VIII (Haemate-P) during 2001–2013. Because of her history of exposure to UK-sourced plasma products, for public health purposes she had been informed that she was at risk for vCJD, although she was not known to have been exposed to factor VIII derived from a batch including a vCJD donation. She had no history of potential iatrogenic exposure to CJD and no family history of CJD.

Donors for all blood or platelet transfusions since 2001 have been identified. Of the 107 donors, 106 are still alive, with a median age of 55 years (range 27–80 years). (Table 1). One donor of leukodepleted platelets, which were transfused

12 years before clinical onset in the recipient, died in 2013 at 76 years of age, and the diagnoses on the death certificate were vascular dementia and bladder cancer. Identification of donors for transfusions before 2001 has not been possible.

Case 2

In 2014, a 64-year-old woman reported day/night reversal of sleep patterns and, 3 months later, excessive tearfulness, for which she was started on antidepressants. She then had onset of writing problems, followed during the next few days by increasing language problems that led to expressive dysphasia. She deteriorated rapidly thereafter, requiring assistance with her activities of daily living and having coordination and memory problems, jerking movements suggestive of myoclonus, and itching in both arms. She was admitted to the hospital and experienced a probable focal seizure with secondary generalization. She had onset of a homonymous hemianopia and limb rigidity and then became bedbound and mute, dying 7 months after the onset of symptoms.

Table 1. Selected characteristics of blood donors to the patient with sporadic Creutzfeldt-Jakob disease described in case 1, United Kingdom, 2014*

Interval from transfusion to onset, y	Component	No. donors	No. donors alive	No. donors dead
3	RBC LD	4	4	0
6	RBC LD	6	6	0
7	RBC LD	19	19	0
9	RBC LD	3	3	0
10	RBC LD	4	4	0
12	Whole blood LD; RBC LD; platelets LD	2; 27; 42	2; 27; 41	0; 0; 1

*LD, leukodepleted; RBC, red blood cells. Median age of donors, 56 years (range 27–80 years).

SYNOPSIS

An electroencephalogram performed during the final stages of illness showed widespread slowing, more evident on the left. An MRI brain scan showed left-sided caudate head and anterior putaminal high signal. Diffusion weighted imaging showed areas of cortical high signal. Results of a CSF 14-3-3 assay and real-time quaking-induced conversion tests were positive. Consent for full sequencing of the *PRNP* was not obtained; methionine homozygosity at codon 129 was identified.

Postmortem neuropathologic examination of the brain showed a widespread spongiform encephalopathy with microvacuolar spongiform change, neuronal loss, and gliosis. Immunostaining for prion protein showed widespread positivity with a granular/synaptic pattern (Figure). No amyloid plaques were identified. Western blot analysis confirmed the presence of protease resistant prion protein with a type 1A isoform. There was no evidence of abnormal prion protein accumulation in spleen and appendix either on immunocytochemistry or high sensitivity Western blot analysis.

The patient was known to have hemophilia B since 1964 and had received plasma-derived and recombinant factor IX during 1984–2012. For public health purposes, she had been informed that she was at risk for vCJD and in 1991 had received factor IX derived from a pool containing plasma from a donor who subsequently had vCJD. She had no history of potential iatrogenic exposure to CJD and no family history of CJD.

In 1985, the patient received 6 units of fresh frozen plasma (FFP). Tracing of donors has not been possible.

Discussion

This report describes 2 cases of sCJD in patients with a history of treatment with UK-sourced plasma products, 1 with a history of hemophilia B and 1 with von Willebrand's disease. To our knowledge, no previous case of sCJD in a person with a history of extended exposure to plasma products has been reported. It is clearly of concern that there have been 2 such cases in a relatively short period in the UK, where many plasma product recipients have

been informed that they are at increased risk for vCJD. However, a causal link between the treatment with plasma products and the onset of sCJD has not been established, and the occurrence of these cases may simply reflect a chance event in the context of systematic surveillance of CJD in large populations.

Both patients had been informed that they were at increased risk for vCJD, and considering the evidence for the type of CJD in the 2 cases is important. Both patients had a clinical phenotype suggestive of sCJD, including a short duration of illness, typical early symptoms, a suggestive MRI scan, and, in 1 patient, a typical EEG. Notably, both patients had a positive real-time quaking-induced conversion test result for PrP^{Sc} in CSF; previously this test had not been positive in any case of vCJD evaluated in our laboratory (Table 2) (9). However, neuropathological examination was critical; it showed appearances typical of sCJD in both patients and no evidence of peripheral pathogenesis on immunostaining of lymphoreticular tissues, a feature that is observed in all tested specimens of vCJD patients to date (10). Furthermore, both patients had a type 1A isoform PrP^{Sc} on Western blot consistent with a diagnosis of sCJD subtype MM1 (11). Neither patient had a history of potential iatrogenic exposure or a family history of CJD, and for the case for which sequencing of the *PRNP* was performed, no mutations were detected. In both cases, an MM genotype occurred at codon 129 of *PRNP*, which does not distinguish between sCJD and vCJD. Laboratory transmission studies to provide evidence of agent strain in the cases have not been possible.

One patient had received multiple transfusions of blood components over an extended period, and the other had received 6 units of FFP 19 years before clinical onset, raising the possibility that these cases could have resulted from secondary transmission through blood components. In the case of the patient with von Willebrand disease, 107 donors have been traced, and none appear in the register of cases of CJD kept at the National CJD Research and Surveillance Unit. However, it has not been possible to obtain information

Table 2. Selected characteristics and clinical features of the 2 patients with sporadic Creutzfeldt-Jakob disease described in cases 1 and 2, United Kingdom, 2014*

Characteristic/clinical feature	Case 1 64/F	Case 2 64/F
Age, y/sex of patient		
Symptoms/signs	Ataxia, cognitive impairment, visual impairment, myoclonus	Somnolence/depression, dysphasia, cognitive impairment, myoclonus/ataxia
MRI	+	+
EEG	+	Slow activity
CSF 14-3-3 assay	+	+
RT-QuIC	+	+
Genotype	MM	MM
Diagnosis	Definite sCJD	Definite sCJD
Duration	3 mo	7 mo

*CSF, cerebrospinal fluid; EEG, electroencephalogram; MRI, magnetic resonance imaging; RT-QuIC, real-time quaking-induced conversion; sCJD, sporadic Creutzfeldt-Jakob disease.

on blood transfusions for this patient before 2001 nor on the FFP transfusions for the patient with hemophilia B. Lookback studies in the United States and United Kingdom have provided no evidence of transfusion-transmission of sCJD (2,3), and although 1 study suggested an increase in risk after a lag period of 10 years (12), this finding was not confirmed in another study (13). The balance of evidence indicates that, if sCJD is transmitted by blood transfusion, it must be a rare event, if it happens at all, and transfusion transmission is probably not the explanation for the 2 cases we describe.

Systematic surveillance for CJD, including a coordinated study in Europe (14), has been carried out in many countries over the past 25 years and is continuing. Many of these studies obtain information on potential risk factors, including details of past medical history. To date, no case of sCJD has been reported in a person who has received treatment for a clotting disorder. In fact, the absence of such a case has been used to argue against the possibility that plasma-derived products pose a risk for sCJD transmission (6). CJD surveillance centers are aware of the relevance of this issue, and sCJD patients with a history of treatment with plasma products probably would have been identified and reported if they occurred. Although it is surprising that 2 cases of sCJD have been identified among a population of 4,000–5,000 patients in the UK who have been treated for clotting disorders with fractionated plasma products, the total population under surveillance for CJD in Europe and internationally exceeds 500 million. Assuming an annual incidence rate of sCJD of 1.5–2.0 per million population (15), the occurrence of 2 cases of sCJD in this total population may not imply a causal link between the treatment and the occurrence of the disease. The 2 cases were identified over a period of months, and no further cases have been found since 2014; however, continuing to search for such cases through CJD surveillance programs is essential.

Acknowledgment

We thank Mark W. Head for the Western blot data.

The National CJD Research and Surveillance Unit is supported by the Policy Research Program of the Department of Health and the Government of Scotland (grant no. PR-ST-0614-00008). This report is independent research in part funded by the Department of Health Policy Research Programme and the Government of Scotland. The views expressed in this publication are those of the authors and not necessarily those of the Department of Health or the Government of Scotland.

Dr. Urwin worked as a research registrar at the National CJD Research and Surveillance Unit and is currently training in neurology. His primary research interests include human prion diseases.

References

1. Prusiner SB. Molecular biology of prion diseases. *Science*. 1991;252:1515–22. <http://dx.doi.org/10.1126/science.1675487>
2. Dorsey K, Zou S, Schonberger LB, Sullivan M, Kessler D, Notari E IV, et al. Lack of evidence of transfusion transmission of Creutzfeldt-Jakob disease in a US surveillance study. *Transfusion*. 2009;49:977–84. <http://dx.doi.org/10.1111/j.1537-2995.2008.02056.x>
3. Urwin PJ, Mackenzie JM, McEvoy CA, Will RG, Hewitt PE. Creutzfeldt-Jakob disease and blood transfusion: updated results of the UK Transfusion Medicine Epidemiology Review Study. *Vox Sang*. 2016;110:310–6. <http://dx.doi.org/10.1111/vox.12371>
4. Olatzel M, Abela E, Maissen M, Aguzzi A. Extraneural pathologic prion protein in sporadic Creutzfeldt-Jakob disease. *N Engl J Med*. 2003;349:1812–20. <http://dx.doi.org/10.1056/NEJMoa030351>
5. Douet JY, Zafar S, Parrot-Llaudet A, Lacroix C, Lagan S, Aron N, et al. Detection of infectivity in blood of persons with variant and sporadic Creutzfeldt-Jakob disease. *Emerg Infect Dis*. 2014;20:114–7. <http://dx.doi.org/10.3201/eid2001.130353>
6. European Medicines Agency. CHMP position statement on Creutzfeldt-Jakob disease and plasma-derived and urine-derived medicinal products. London, 23 June 2011 [cited 2015 May 1]. http://www.ema.europa.eu/docs/en_GB/document_library/Position_statement/2011/06/WC500108071.pdf
7. Cousens SN, Zeldner M, Esmonde TF, De Silva R, Wifesmith JW, Smith PG, et al. Sporadic Creutzfeldt-Jakob disease in the United Kingdom: analysis of epidemiological surveillance data for 1970–96. *BMJ*. 1997;315:389–95. <http://dx.doi.org/10.1136/bmj.315.7105.389>
8. Wientjens DPWM, Will RG, Hoffman A. Creutzfeldt-Jakob disease: a collaborative study in Europe. *J Neurol Neurosurg Psychiatry*. 1994;57:1285–99.
9. McGuire LI, Peden AH, Orr CD, William JM, Appleford NE, Mallinson G, et al. Real time quaking-induced conversion analysis of cerebrospinal fluid in sporadic Creutzfeldt-Jakob disease. *Ann Neurol*. 2012;72:278–85. <http://dx.doi.org/10.1002/ana.23589>
10. Head MW, Ritchie D, Smith N, McLoughlin V, Wallon W, Samuel S, et al. Peripheral tissue involvement in sporadic, iatrogenic, and variant Creutzfeldt-Jakob disease: an immunohistochemical, quantitative, and biochemical study. *Am J Pathol*. 2004;164:143–53. [http://dx.doi.org/10.1016/S0002-9440\(10\)63105-7](http://dx.doi.org/10.1016/S0002-9440(10)63105-7)
11. Head MW, Bunn TJR, Bishop MT, McLoughlin V, Lowrie S, McKinnon CS, et al. Prion protein heterogeneity in sporadic but not variant Creutzfeldt-Jakob disease: UK cases 1991–2002. *Ann Neurol*. 2004;55:851–9. <http://dx.doi.org/10.1002/ana.20127>
12. Puopolo M, Ladogana A, Vetrugno V, Pocchini M. Transmission of sporadic Creutzfeldt-Jakob disease by blood transfusion: risk factor or possible biases. *Transfusion*. 2011;51:1556–66. <http://dx.doi.org/10.1111/j.1537-2995.2010.03004.x>
13. Molesworth AM, Mackenzie J, Everington D, Knight RSG, Will RG. Sporadic Creutzfeldt-Jakob disease and risk of blood transfusion in the United Kingdom. *Transfusion*. 2011;51:1872–3. <https://doi.org/10.1111/j.1537-2995.2011.03198.x>
14. Ladogana A, Puopolo M, Croes EA, Budka H, Jurlin C, Collins S, et al. Mortality from Creutzfeldt-Jakob disease and related disorders in Europe, Australia, and Canada. *Neurology*. 2005;64:1586–91. <http://dx.doi.org/10.1212/01.WNL.0000160117.56690.B2>
15. Minikel EV, Vallabh SM, Lok M, Estrada K, Samchoke KB, Sathirapongsasati JP, et al.; Exonome Aggregation Consortium (ExAC). Quantifying prion disease penetrance using large population control cohorts. *Sci Transl Med*. 2016;8:322ra9. <http://dx.doi.org/10.1126/scitranslmed.aad5169>

Address for correspondence: R.G. Will, National CJD Research and Surveillance Unit, Western General Hospital, Edinburgh, Scotland EH4 2XU, UK; email: r.g.will@gro-c

Sporadic Creutzfeldt-Jakob disease in two plasma product recipients in the United Kingdom

Patrick Urwin¹, James Ironside¹, Anna Molesworth¹, Richard Knight¹, Patricia Hewitt², Charlotte Llewelyn³, Jan Mackenzie¹, Robert Will¹

¹National CJD Research & Surveillance Unit, Western General Hospital, Edinburgh, EH4 2XU

²NHS Blood and Transplant, Colindale Centre, Charcot Road, London, NW9 5BG

³NHSBT/PHE Epidemiology Unit, Long Road, Cambridge, CB2 0PT

Corresponding author: Professor R G Will
National CJD Research & Surveillance Unit
Western General Hospital
Edinburgh EH4 2XU
Tel: GRO-C
Fax: GRO-C
Email: r.g.will@ GRO-C

Reprints are not required.

Funding Body: Policy Research Program of the Department of Health and the Scottish Government
Grant No. PR-ST-0614-00008

No conflicts of interest

Word count: 1921

Short running head: sCJD in two plasma product recipients

ABSTRACT

Background

No cases of sporadic Creutzfeldt-Jakob disease (sCJD) have been reported in patients with clotting disorders treated with fractionated plasma products.

Case reports

Two cases of sCJD have been identified in the United Kingdom (UK) with a history of extended treatment for clotting disorders, one with Haemophilia B and the other with von Willebrand's disease. Both individuals had previously been informed that they were at an increased risk of variant CJD (vCJD) because of treatment with UK sourced fractionated plasma products.

Results

Both cases had clinical and investigative features suggestive of sCJD. This diagnosis was confirmed in both cases on neuropathological and biochemical analysis of the brain. One case had also received multiple blood component transfusions, but a partial look-back study has not identified a donor with either sCJD or vCJD.

Conclusions

A causal link between the treatment with plasma products and the development of sCJD has not been established and the occurrence of these cases may simply reflect a chance event in the context of systematic surveillance of CJD in large populations.

Keywords: sporadic Creutzfeldt-Jakob disease; variant Creutzfeldt-Jakob disease; clotting disorders; plasma products; blood transfusion

INTRODUCTION

Sporadic Creutzfeldt-Jakob disease (sCJD) is a human prion disease of unknown aetiology. The relatively consistent mortality rates, an overall random spatial and temporal distribution of cases and the absence of any confirmed environmental risk factor has led to the hypothesis that this disease is due to the spontaneous generation of disease-associated prion protein (PrP^{Sc}) in the brain.¹ Look-back studies have shown no evidence of transmission through blood transfusion in sCJD,^{2,3} despite the identification of PrP^{Sc} in peripheral tissues⁴ and experimental evidence demonstrating infectivity in blood, using intracerebral inoculation of highly sensitive transgenic mice.⁵ The absence of cases of CJD causally linked to prior treatment with fractionated plasma products has been used as evidence of the safety of these products in relation to prion diseases.⁶

This report describes two cases of sCJD with a history of prior treatment with UK plasma sourced plasma products, both of whom had been informed that they were at increased risk of developing variant CJD (vCJD) because of this treatment. The clinical features and investigations in these cases were typical of sCJD; the neuropathological diagnosis in both cases was sCJD (MM1 subtype).

METHODS

The UK National CJD Research and Surveillance Unit (NCJDRSU) has been carrying out systematic epidemiological study of CJD since 1990. The methodology of this study has been published previously,⁷ but, in brief, suspect cases are referred by clinicians and visited by a research registrar who obtains details of the clinical history and investigations together with information on a range of putative risk factors, including past medical history.

The Transfusion Medicine Epidemiology Review (TMER) study investigates potential links between donors and recipients of labile blood components and in sCJD investigates cases with a history of blood donation or having received a blood transfusion.

Coordinated surveillance of CJD has been undertaken in the European Union since 1993⁸ and national surveillance programmes for CJD are in place in a number of other countries, including Australia, Canada, Japan and the USA.

RESULTS

Case 1:

In 2014 a 64 year old woman suffered a rapidly progressive dementia with deterioration in driving skills and balance disturbance, then limb coordination deficits with handwriting impairment. In the second month, her gait deteriorated, becoming shuffling and unsteady, she struggled to dress herself, and she developed daytime hypersomnolence. She became distractible, developed visual misperceptions, emotional lability, and spatial memory problems. She was admitted to hospital at the beginning of the third month of her illness and developed cortical blindness, myoclonus and akinetic mutism. There was a rapid decline and she died with a total illness duration of 3 months.

The EEG showed background slowing and runs of periodic complexes, the MRI brain scan showed high signal in the caudate heads with posterior cortical ribboning. The CSF 14-3-3 assay and RT-QuIC for PrP^{Sc} were both positive. Prion protein gene (*PRNP*) sequencing showed no mutations with methionine homozygosity at codon 129.

Post mortem examination of the brain showed widespread spongiform encephalopathy of predominantly microvacuolar type. Immunocytochemistry for prion protein gave a widespread positive reaction in a granular/synaptic pattern (Figure 1). No plaques or plaque like structures were identified. Immunocytochemistry for disease-associated prion protein was negative in peripheral nerve, liver, lymph node, appendix and spleen. Western blot analysis of frontal cortex and cerebellum confirmed the presence of protease-resistant prion protein with a type 1A isoform.

She had been diagnosed with Von Willebrand's Disease in childhood. Her early therapies include numerous transfusions of red cells and platelets; in more recent years she received both plasma derived and recombinant factor VIII, with further blood component transfusion at times of haemorrhage. In light of her history of exposure to UK sourced plasma products, she had been informed that she was "at risk of vCJD for public health purposes", although she was not known to have been exposed to Factor VIII derived from a batch including a vCJD donation. There was no history of potential iatrogenic exposure to CJD and no family history of CJD.

Donors for all blood or platelet transfusions since 2001 have been identified. Of the 107 donors, 106 are still alive with an average age of 55 years (Table 1). One donor of leucodepleted platelets, which were transfused 12 years before clinical onset in the recipient, died in 2013, aged 76 years, of vascular dementia and bladder cancer. It has not been possible to identify the donors for transfusions prior to 2001.

Case 2:

In 2014 a 64 year old woman described day/night reversal of sleep patterns and three months later excessive tearfulness, for which she was started on antidepressants. She then developed writing problems, followed over the next few days by increasing language problems, leading to an expressive dysphasia. She deteriorated rapidly thereafter, requiring assistance with her activities of daily living, developing coordination and memory problems, jerking movements suggestive of myoclonus and itching in both arms. She was admitted to hospital and experienced a probable focal seizure with secondary generalisation. She developed a homonymous hemianopia and limb rigidity and then became bedbound and mute, dying 7 months after the onset of symptoms.

An EEG showed widespread slowing, more evident on the left. MRI brain showed left sided caudate head and anterior putaminal high signal, together with areas of cortical high signal, on DWI images. CSF 14-3-3 and Rt-QuIC analyses were positive. Consent for full sequencing of the *PRNP* was not available; methionine homozygosity at codon 129 was identified.

Neuropathological examination of the brain showed a widespread spongiform encephalopathy with microvacuolar spongiform change, neuronal loss and gliosis. Immunostaining for prion protein showed widespread positivity with a granular/synaptic pattern (Figure 1). No amyloid plaques were identified. Western blot analysis confirmed the presence of protease resistant prion protein with a

type 1A isoform. There was no evidence of abnormal prion protein accumulation in spleen and appendix either on immunocytochemistry or high sensitivity Western blotting.

This individual was known to have haemophilia B since 1964 and had received plasma derived and recombinant factor IX. She was informed that she was 'at risk of vCJD for public health purposes' and had received factor IX derived from a pool containing plasma from a donor who subsequently developed variant CJD. There was no history of potential iatrogenic exposure to CJD and no family history of CJD.

In 1985 she received 6 units of FFP, but it has not been possible to trace the donors.

DISCUSSION

This report describes two cases of sCJD with a history of treatment with UK sourced plasma products, one with a history of Haemophilia B and the other von Willebrand's disease. No previous case of sCJD with a history of extended exposure to plasma products has been reported and it is clearly of concern that there have been two such cases in a relatively short period in the UK, where many plasma product recipients have been informed that they are at increased risk of developing vCJD. However, a causal link between the treatment with plasma products and the development of sCJD has not been established and the occurrence of these cases may simply reflect a chance event in the context of systematic surveillance of CJD in large populations.

Both cases had been informed that they were at increased risk of developing vCJD and it is important to consider the evidence for the type of CJD in the two cases. Both had a clinical phenotype suggestive of sCJD, including a short duration of illness, typical early symptoms, suggestive MRI scan and, in one case a 'typical' EEG. Notably both had a positive CSF RT-QuIC, an investigation that has never been positive in vCJD in our laboratory⁹ (Table 2). However, the critical investigation was the neuropathological examination, which showed appearances typical of sporadic CJD in both cases and no evidence of peripheral pathogenesis on immunostaining of lymphoreticular tissues, a feature that is present in all tested cases of vCJD to date.¹⁰ Furthermore, both had a type 1A isoform PrP^{Sc} on Western Blot consistent with a diagnosis of sCJDMM1.¹¹ There was no history of potential iatrogenic exposure nor a family history of CJD in either case and one had sequencing of the *PRNP*, which showed no mutations. Both cases had an MM genotype at codon 129 of *PRNP*, which does not distinguish between sCJD and vCJD. Laboratory transmission studies to provide evidence of agent strain in the cases have not been possible.

One case had received multiple transfusions of blood components over an extended period and the other 6 units of FFP 19 years before clinical onset, raising the possibility that these cases could result from secondary transmission via blood components. In the von Willebrand's case 107 donors have been traced and none appear in the register of cases of CJD held at the NCJDRSU. However, it has not been possible to obtain information on blood transfusions in this case prior to 2001 nor on the FFP transfusions in the Haemophilia B case. Look-back studies in the USA and the UK have provided no evidence of transfusion transmission of sCJD^{7,3} and, although one study suggested an increase in risk after a lag period of 10 years,¹² this was not confirmed in another study.¹³ The balance of evidence indicates that should sCJD be transmitted by blood transfusion this must be a rare event, if

It happens at all, and it is unlikely that transfusion transmission can be the explanation for the two cases in this report.

Systematic surveillance for CJD has been carried out in many countries over the past 25 years, including a coordinated study in Europe.¹⁴ Many of these studies obtain information on potential risk factors, including details of past medical history and no case of CJD in an individual with treatment for a clotting disorder has been reported. It is of note that the absence of such a case has been used to argue against the possibility that plasma derived products pose a risk of transmission of human prion diseases.⁶ CJD surveillance centres are aware of the relevance of this issue and it is likely that cases of CJD with a prior history of treatment with plasma products would have been identified and reported, should these have occurred. The population of plasma product recipients in the UK is relatively small and, even with a period of observation of more than 25 years, it is surprising that two cases of sCJD have been identified in this population if this was a chance occurrence. However, the total population under surveillance for CJD in Europe and internationally exceeds 500 million and extrapolating from the size of the plasma product recipient population in the UK to other countries, the duration of surveillance programmes for CJD and assuming an annual incidence rate of sCJD of 1.5–2/ million,¹⁵ the occurrence of 2 cases of sCJD in this total population may reflect the size of the population under observation rather than implying a causal link between the treatment and the occurrence of the disease. The two cases were identified over a period of months and no further cases have been found since 2014, but it is essential to continue to search for such cases through CJD surveillance programmes.

ACKNOWLEDGEMENTS

We thank Dr Mark W Head for the Western blot data. The National CJD Research & Surveillance Unit is supported by the Policy Research Program of the Department of Health and the Scottish Government (PR-ST-0614-00008). This report is independent research in part funded by the Department of Health Policy Research Programme and the Scottish Government. The views expressed in this publication are those of the author(s) and not necessarily those of the Department of Health or the Scottish Government.

REFERENCES

- 1 Prusiner SB. Molecular biology of prion diseases. *Science* 1991;252:1515-1522.
- 2 Dorsey K, Zou S, Schonberger LB, et al. Lack of evidence of transfusion transmission of Creutzfeldt-Jakob disease in a US surveillance study. *Transfusion* 2009;49:977-984.
- 3 Urwin PJ, Mackenzie JM, Llewelyn CA, et al. Creutzfeldt-Jakob disease and blood transfusion: updated results of the UK Transfusion Medicine Epidemiology Review Study. *Vox Sang* 2016;110:310-316.
- 4 Glatzel M, Abela E, Maissen M, et al. Extraneural pathologic prion protein in sporadic Creutzfeldt-Jakob disease. *New Engl J Med* 2003;349:1812-1820.
- 5 Douet JY, Zafar S, Perret-Liaudet A, et al. Detection of infectivity in blood of persons with variant and sporadic Creutzfeldt-Jakob disease. *Emerg Infect Dis* 2014;20:114-117.
- 6 European Medicines Agency: CHMP position statement on Creutzfeldt-Jakob disease and plasma-derived and urine-derived medicinal products. London, 23 June 2011.
http://www.ema.europa.eu/docs/en_GB/document_library/Position_statement/2011/06/WC500108071.pdf
- 7 Cousens SN, Zeidler M, Esmonde TF, et al. Sporadic Creutzfeldt-Jakob disease in the United Kingdom: analysis of epidemiological surveillance data for 1970-96. *BMJ* 1997;315:389-395.
- 8 Wientjens DPWM, Will RG, Hofman A. Creutzfeldt-Jakob disease: a collaborative study in Europe. *JNNP* 1994;57:1285-1299.
- 9 McGulre LI, Peden AH, Orrú CD, et al. Real time quaking-induced conversion analysis of cerebrospinal fluid in sporadic Creutzfeldt-Jakob disease. *Ann Neurol* 2012;72:278-285.
- 10 Head MW, Ritchie D, Smith N, et al. Peripheral tissue involvement in sporadic, iatrogenic, and variant Creutzfeldt-Jakob disease: an immunohistological, quantitative and biochemical study. *Am J Pathol* 2004;164:143-153.
- 11 Head MW, Bunn TJR, Bishop MT, et al. Prion protein heterogeneity in sporadic but not variant Creutzfeldt-Jakob disease: UK cases 1991-2002. *Ann Neurol* 2004;55:851-859.
- 12 Puopolo M, Ladogana A, Vetrugno V, et al. Transmission of sporadic Creutzfeldt-Jakob disease by blood transfusion: risk factor or possible biases. *Transfusion* 2011;51:1556-1566.
- 13 Molesworth AM, Mackenzie J, Everington D, et al. Letters to the editor: Sporadic Creutzfeldt-Jakob disease and risk of blood transfusion in the United Kingdom. *Transfusion* 2011;51:1872-1873.
- 14 Ladogana A, Puopolo M, Croes EA, et al. Mortality from Creutzfeldt-Jakob disease and related disorders in Europe, Australia, and Canada. *Neurology* 2005;64:1586-1591.
- 15 Minikel EV, Vallabh SM, Lek M, et al. Quantifying prion disease penetrance using large population control cohorts. *Sci Transl Med* 2016;8(322):1-12.

Table 1 Blood donors to Case 1

Interval from transfusion to onset in case	Component	Number of donors	Donors Alive	Donors Dead
3 years	RBC LD	4	4	0
6 years	RBC LD	6	6	0
7 years	RBC LD	19	19	0
9 years	RBC LD	3	3	0
10 years	RBC LD	4	4	0
12 years	WB LD	2	2	0
	RBC LD	27	27	0
	Platelets LD	42	41	1

RBC: red blood cells

WB: whole blood

LD: leucodepleted

Average age of donors = 55

Median age of donors = 56 (range 27-80)

Table 2 Clinical and Investigative features of the two cases

	Case 1	Case 2
Age/gender	64 F	64F
Symptoms/signs	Ataxia Cognitive Impairment Visual Impairment Myoclonus	Somnolence/depression Dysphasia Cognitive Impairment Myoclonus/ataxia
MRI	+	+
EEG	+	Slow activity
14-3-3	+	+
RT-QuIC	+	+
Genotype	MM	MM
Diagnosis	Definite sporadic CJD	Definite sporadic CJD
Duration	3 months	7 months

Legend for Figure 1

- (a) Microvacuolar spongiform change in the frontal cortex in Case 1 (Haematoxylin and eosin stain, x 400); (b) Fine granular/synaptic accumulation of abnormal prion protein in the cerebral cortex in Case 1 (12F10 anti-prion protein antibody, x 400); (c) Microvacuolar spongiform change with neuronal loss and gliosis in the frontal cortex in Case 2 (Haematoxylin and eosin stain, x 400); (d) Focally intense granular/synaptic accumulation of abnormal prion protein in the cerebral cortex in Case 2 (12F10 anti-prion protein antibody, x 400).

