Table 1. Transmissibility of L-BSE in mice

Mice	Number of mice affected/inoculated	Incubation period (days)
Mouse	0/5	>700*
tga20	0/5	>668*
MHM2	0/5	>755*
MH2M	0/5	>853*
TgHaNSE	2/3	567†, 853†

The brain homogenate of L-BSE was intracerebrally inoculated into mice. *Mice showed no clinical signs, and tested negative for PrP^{Sc} in the brain. †PrP^{Sc} was present in the brain.

Table 2. Incubation period of hamsters inoculated with C-BSE and L-BSE

Inoculum	Passage numbers	Number of hamsters diseased/inoculated	Incubation period mean (SD) (days)
C-BSE	1st	0/5	>600*
Mouse passaged C-BSE†	1st	10/10	349.5 (6.6)
	2nd	10/10	267.3 (19.2)
	3rd	10/10	271.6 (16.2)
L-BSE	1st	3/4	576.8 (127.8)
	2nd	4/4	208 (15.5)

*Hamsters showed no clinical signs and were negative for PrP^{Sc} in the brain. †After one passage of BSE from cattle in mice (incubation period, 408.6 (28.2) days) and subsequent transmission to TgHaNSE mice (incubation period, 153.1 (1.1) days).[14] The brain of a diseased TgHaNSE mouse was inoculated in a hamster.

Table 3. Comparison of the PrP amino acid sequences in mouse and hamster

Species	Susceptibility		PrP amino acid*							
	C-BSE†	L-BSE	109	112	139	155	170	203	205	215
Mouse	Sus‡	Res§	L	V	I	Y	S	V	M	V
MHM2	Sus†	Res	M	M	-¶	-	-	-	-	-
MH2M	Res†	Res	M	M	M	N	N	-	-	-
Hamster	Res†	Sus	M	M	M	N	N	I	I	T
Cattle#	Sus	Sus	M	-	-	N	-	I	-	I

*Residue numbers correspond to those in hamster PrP. †Transmission results of C-BSE were reported previously.[14] ‡Susceptible. §Resistant. ¶Indicates the same amino acid sequence as mouse PrP. #Transmission results of cattle were reported previously.[8]

7. Comoy EE, Casalone C, Lescoutra-Etchegaray N, Zanusso G, Freire S, Marce D, et al. Atypical BSE (BASE) transmitted from asymptomatic aging cattle to a primate. PLoS One 2008; 3:3017.
8. Fukuda S, Iwamaru Y, Imamura M, Masujin K, Shimizu Y, Matsuura Y, et al. Intraspecies transmission of L-type-like Bovine Spongiform Encephalopathy detected in Japan. Microbiol Immunol 2009; 53:704-7.
9. Kong Q, Zheng M, Casalone C, Qing L, Huang S, Chakraborty B, et al. Evaluation of the human transmission risk of an atypical bovine spongiform encephalopathy prion strain. J Virol 2008; 82:3697-701.
10. Lombardi G, Casalone C, A DA, Gelmetti D, Torcoli G, Barbieri I, et al. Intraspecies transmission of BASE induces clinical dullness and amyotrophic changes. PLoS Pathog 2008; 4:1000075.
11. Masujin K, Shu Y, Yamakawa Y, Hagiwara K, Sata T, Matsuura Y, et al. Biological and biochemical characterization of L-type-like bovine spongiform encephalopathy (BSE) detected in Japanese black beef cattle. Prion 2008; 2:123-8.
12. Capobianco R, Casalone C, Suardi S, Mangieri M, Miccolo C, Limido L, et al. Conversion of the BASE prion strain into the BSE strain: the origin of BSE? PLoS Pathog 2007; 3:31.
13. Yokoyama T, Itohara S, Yuasa N. Detection of species specific epitopes of mouse and hamster prion proteins (PrPs) by anti-peptide antibodies. Arch Virol 1996; 141:763-9.
14. Yokoyama T, Masujin K, Iwamaru Y, Imamura M, Mohri S. Alteration of the biological and biochemical characteristics of bovine spongiform encephalopathy prions during interspecies transmission in transgenic mice models. J Gen Virol 2009; 90:261-8.
15. Scott M, Groth D, Foster D, Torchia M, Yang SL, DeArmond SJ, et al. Propagation of prions with artificial properties in transgenic mice expressing chimeric PrP genes. Cell 1993; 73:979-88.
16. Fischer M, Rulicke T, Raeber A, Sailer A, Moser M, Oesch B, et al. Prion protein (PrP) with amino-proximal deletions restoring susceptibility of PrP knockout mice to scrapie. EMBO J 1996; 15:1255-64.
17. Race RE, Priola SA, Bessen RA, Ernst D, Dockter J, Rall GF, et al. Neuron-specific expression of a hamster prion protein minigene in transgenic mice induces susceptibility to hamster scrapie agent. Neuron 1995; 15:1183-91.
18. Westaway D, Goodman PA, Mirenda CA, McKinley MP, Carlson GA, Prusiner SB. Distinct prion proteins in short and long scrapie incubation period mice. Cell 1987; 51:651-62.
19. Oesch B, Westaway D, Walchli M, McKinley MP, Kent SB, Aebersold R, et al. A cellular gene encodes scrapie PrP 27–30 protein. Cell 1985; 40:735-46.
20. Goldmann W, Hunter N, Martin T, Dawson M, Hope J. Different forms of the bovine PrP gene have five or six copies of a short, G-C-rich element within the protein-coding exon. J Gen Virol 1991; 72:201-4.
21. Yokoyama T, Kimura KM, Ushiki Y, Yamada S, Morooka A, Nakashiba T, et al. In vivo conversion of cellular prion protein to pathogenic isoforms, as monitored by conformation-specific antibodies. J Biol Chem 2001; 276:11265-71.
22. Hayashi HK, Yokoyama T, Takata M, Iwamaru Y, Imamura M, Ushiki YK, et al. The N-terminal cleavage site of PrP^{Sc} from BSE differs from that of PrP^{Sc} from scrapie. Biochem Biophys Res Commun 2005; 328:1024-7.
23. Hagiwara K, Yamakawa Y, Sato Y, Nakamura Y, Tobiume M, Shinagawa M, et al. Accumulation of mono-glycosylated form-rich, plaque-forming PrP^{Sc} in the second atypical bovine spongiform encephalopathy case in Japan. Jpn J Infect Dis 2007; 60:305-8.
24. Fraser H, Dickinson AG. Distribution of experimentally induced scrapie lesions in the brain. Nature 1967; 216:1310-1.
25. Masujin K, Matthews D, Wells GA, Mohri S, Yokoyama T. Prions in the peripheral nerves of bovine spongiform encephalopathy-affected cattle. J Gen Virol 2007; 88:1850-8.
26. Shimizu Y, Kaku-Ushiki Y, Iwamaru Y, Muramoto T, Kitamoto T, Yokoyama T, et al. A novel anti-prion protein monoclonal antibody and its single-chain fragment variable derivative with ability to inhibit abnormal prion protein accumulation in cultured cells. Microbiol Immunol 2010; 54:112-21.
27. Thomzig A, Cardone F, Kruger D, Pocchiari M, Brown P, Beekes M. Pathological prion protein in muscles of hamsters and mice infected with rodent-adapted BSE or vCJD. J Gen Virol 2006; 87:251-4.
28. Thomzig A, Spassov S, Friedrich M, Naumann D, Beekes M. Discriminating scrapie and bovine spongiform encephalopathy isolates by infrared spectroscopy of pathological prion protein. J Biol Chem 2004; 279:33847-54.
29. Collinge J, Sidle KC, Meads J, Ironside J, Hill AF. Molecular analysis of prion strain variation and the aetiology of 'new variant' CJD. Nature 1996; 383:685-90.
30. Kim JI, Cali I, Surewicz K, Kong Q, Raymond GJ, Atarashi R, et al. Mammalian prions generated from bacterially expressed prion protein in the absence of any mammalian cofactors. J Biol Chem 2010; 285:14083-7.
31. Nicot S, Baron T. Strain-specific barriers against bovine prions in hamsters. J Virol 2011; 85:1906-8.
32. Beringue V, Herzog L, Reine F, Le Dur A, Casalone C, Vilotte JL, et al. Transmission of atypical bovine prions to mice transgenic for human prion protein. Emerg Infect Dis 2008; 14:1898-901.
33. Beringue V, Andreoletti O, Le Dur A, Essalmani R, Vilotte JL, Lacroux C, et al. A bovine prion acquires an epidemic bovine spongiform encephalopathy strain-like phenotype on interspecies transmission. J Neurosci 2007; 27:6965-71.

SHORT COMMUNICATION

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Comparative analysis of Japanese and foreign L-type BSE prions

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Key words: prion, atypical BSE, L-type BSE

Abbreviations: BSE, bovine spongiform encephalopathy; BSE/JP8, the 8th BSE case in Japan; BSE/JP24, the 24th BSE case in Japan; BASE, bovine amyloid spongiform encephalopathy; CNS, central nervous system; C-BSE, classical BSE; H-type BSE, high-type BSE; L-type BSE, low-type BSE; mAb, monoclonal antibody; PET blot, paraffin-embedded tissue blot; PK, proteinase K; PNGaseF, N-glycosidase F; PrP, prion protein; PrPcore, proteinase K-digested PrP^{Sc}; PrP^{Sc}, abnormal prion protein; TSEs, transmissible spongiform encephalopathies

L-type bovine spongiform encephalopathy (BSE) is an atypical form of BSE. To characterize the Japanese L-type BSE prion, we conducted a comparative study of the Japanese and foreign L-type BSE isolates. The L-type BSE isolates of Japan, Germany, France and Canada were intracerebrally inoculated into bovinized prion protein-overexpressing transgenic mice (TgBoPrP). All the examined L-type BSE isolates were transmitted to TgBoPrP mice, and no clear differences were observed in their biological and biochemical properties. Here, we present evidence that the Japanese and Canadian L-type BSE prions are identical to those from the European cases.

Bovine spongiform encephalopathy (BSE) is one of the transmissible spongiform encephalopathies (TSEs), or prion diseases, in cattle. TSE is characterized by spongiform changes in the central nervous system (CNS) and the accumulation of an abnormal prion protein (PrP^{Sc}) in the CNS.[1] PrP^{Sc} has been regarded as the major component of TSE pathogens.[2]

BSE was detected in the UK in 1986,[3] and subsequently spread to the other European countries, Japan and North America.[4-6] BSE is thought to be caused by a single prion strain, based on the analyses of its biological and biochemical characteristics.[7] From 2003, however, several atypical neuropathological and molecular phenotypes of BSE (atypical BSE) have been detected in Japan, several European countries and North America.[6,8-17] Currently, based on the molecular size of the proteinase-digested non-glycosylated form of PrP^{Sc}, atypical BSE is classified into two groups (L-type and H-type).[14]

L-type BSE cases have been identified in the European countries, including Italy, France, Germany, Netherland, Poland and in Canada and Japan.[8-15] Two L-type BSE cases have been identified in Japan. One case was detected in a healthy 23-mo-old Holstein steer (BSE/JP8),[8] and the other was detected in a 14-y-old black Japanese beef cattle (BSE/JP24).[9] The latter case was successfully transmitted to bovinized transgenic mice and cattle, and the biological and biochemical properties differed from that of classical BSE (C-BSE).[18,19] However, it is unclear whether Japanese L-type BSE prion is identical to that of L-type BSE isolates from other countries. To characterize the Japanese L-type BSE isolate, we performed a comparative study of the Japanese and foreign L-type BSE isolates.

A transmission study using experimental animals is a useful approach for prion characterization. Therefore, we performed a transmission study of the L-type BSE isolates in bovinized prion protein (PrP)-overexpressing transgenic mice (TgBoPrP).[20] Brain samples of L-type BSE-affected cattle from Japan (BSE/JP24),[9] France,[10] Germany[11] and Canada[12] were used in this study. The brain homogenates were intracerebrally inoculated into TgBoPrP using previously described methods in reference 18. All animal experiments were reviewed by the Committee of the Ethics on Animal Experiment of the National Institute of Animal Health.

All the examined L-type BSE isolates were transmitted to TgBoPrP, and the affected mice developed progressive neurological diseases. Japanese L-type BSE isolate-affected TgBoPrP exhibited a unique clinical sign, the circling behavior. The same phenotype was observed when TgBoPrP were inoculated with German, French and Canadian L-type BSE isolates. On the other hand, in the first passage the incubation period for the Japanese L-type BSE isolate was significantly different from that of the other L-type BSE isolates (**Table 1**). We then performed serial passages of these isolates for further characterization. The incubation periods in the second passage were shorter than those in the first passage. In the third passage, the incubation periods for all the L-type BSE isolates converged at about 145 d. These results suggest that the L-type BSE isolates in the primary passage were not fully adapted to the TgBoPrP mice. Furthermore,

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Table 1. Transmission of L-type BSE isolates in TgBoPrP mice

	Incubation period (days)			
	JPN	CAN	GER	FRA
First passage	197.7 (3.4)†	172.8 (4.0)*	173.3 (3.3)*	175.7 (5.6)*
	(10/10·)	(12/12)	(12/12)	(10/10)
Second passage	152.0 (1.7)	145.7 (1.8)	143.1 (5.7)	143.1 (3.9)
	(24/24)	(23/23)	(18/18)	(18/18)
Third passage	145.1 (3.6)	143.7 (4.6)	145.3 (8.6)	141.6 (4.7)
	(21/21)	(25/25)	(12/12)	(20/20)

†Mean (standard deviation); ·, Number of affected mice/number of inoculated mice; *$p < 0.05$ for Japanese L-type BSE isolate vs. other L-type BSE isolates in the first passage (Student's t-test).

the different incubation periods in the first passage may be caused by the lower titer of the Japanese L-type BSE prion.

Neuropathological examination of the L-type BSE isolate-affected TgBoPrP were performed using previously described methods.[18] Lesion profile analysis revealed that the degree of brain vacuolation due to the Japanese L-type BSE isolate was similar to that caused by the other L-type BSE isolates (**Fig. 1A**). All the L-type BSE isolates caused severe spongiform changes in the hippocampus, septal nuclei of the paraterminal body and cerebral cortex. We next examined the PrP^{Sc} deposition pattern in the brain using paraffin-embedded tissue (PET) blot, as described previously in reference 18. The distributions of PrP^{Sc} deposits in Japanese L-type BSE isolate-inoculated mice were similar to that of mice inoculated with the other L-type BSE isolates; fine punctate and fine granular PrP^{Sc} were predominantly and uniformly distributed in the pons, cerebellar medulla, midbrain, thalamus and corpus callosum (**Fig. 1B**). Furthermore, similar PrP^{Sc} deposits and distribution patterns were observed in the brain in the first and subsequent passages of all the L-type BSE isolates (data not shown).

We further examined the biochemical properties of PrP^{Sc}, such as the glycoform ratio and molecular mass of proteinase K (PK)-digested PrP^{Sc} (PrPcore). PrP^{Sc} were extracted from the brain of L-type BSE isolate-affected TgBoPrP using previously described methods in reference 18. Western blotting analysis revealed that the glycoform patterns and molecular mass of the PrPcore of the Japanese L-type BSE isolate resembled that of the other L-type BSE isolates. In contrast, clear differences were observed between C-BSE and L-type BSE isolates (**Fig. 2A and B**). Next, we examined the relative PK resistance of PrP^{Sc} from L-type BSE isolate-affected TgBoPrP, as described previously in reference 18. The PrP concentration of the sample was adjusted using the signal intensity of western blot. The PK resistance of PrP^{Sc} from the Japanese L-type BSE was similar to that of the foreign L-type BSE isolates. The PrP^{Sc} of C-BSE-affected TgBoPrP was resistant to digestion with 1,000 μg/ml of PK. In contrast to C-BSE, the PrP^{Sc} signal from the L-type BSE isolates decreased when digested with 500 μg/ml of PK (**Fig. 2C**).

The analyses of L-type BSE cases have been performed using different bovinized PrP-overexpressing transgenic mice, such as TgBoPrP,[18] Tgbov XV[11,21] and Tg540.[22] Thus, it has been impossible to compare the properties of L-type BSE isolates in detail. In this study, therefore, we performed a transmission study of the L-type BSE isolates using identical bovinized PrP-overexpressing transgenic mice to further characterize the Japanese L-type BSE prion. All the L-type BSE isolates transmitted to TgBoPrP, and their incubation periods converged at approximately 145 d following serial passages (**Table 1**). Similar degrees of vacuolation and PrP^{Sc} deposition patterns in the brain were observed among the L-type BSE isolates (**Fig. 1A and B**). Besides the biological characteristics, no differences were observed in the biochemical characteristics of PrP^{Sc} from the L-type BSE isolates (**Fig. 2A–C**). These findings suggest that the examined L-type BSE cases were caused by prions with identical characteristics.

Italian L-type BSE cases are called bovine amyloid spongiform encephalopathy (BASE). We could not compare the characteristics of the Japanese L-type BSE with those of the Italian isolates. In a transmission study using transgenic mice, the French L-type BSE isolate and BASE exhibit similar biological characteristics.[22] Our data indicated that the properties of the Japanese L-type BSE prion are identical to those of the French L-type BSE isolate. It has also been reported that the characteristics of Japanese L-type BSE isolate closely resemble those of BASE in an experimental transmission study in cattle.[19]

The origin of L-type BSE prion is unknown. The present study showed that the Japanese and Canadian L-type BSE prions are identical to those from the European cases. The fact that identical L-type BSE prions exhibit a worldwide distribution is important insight for devising atypical BSE control measures.

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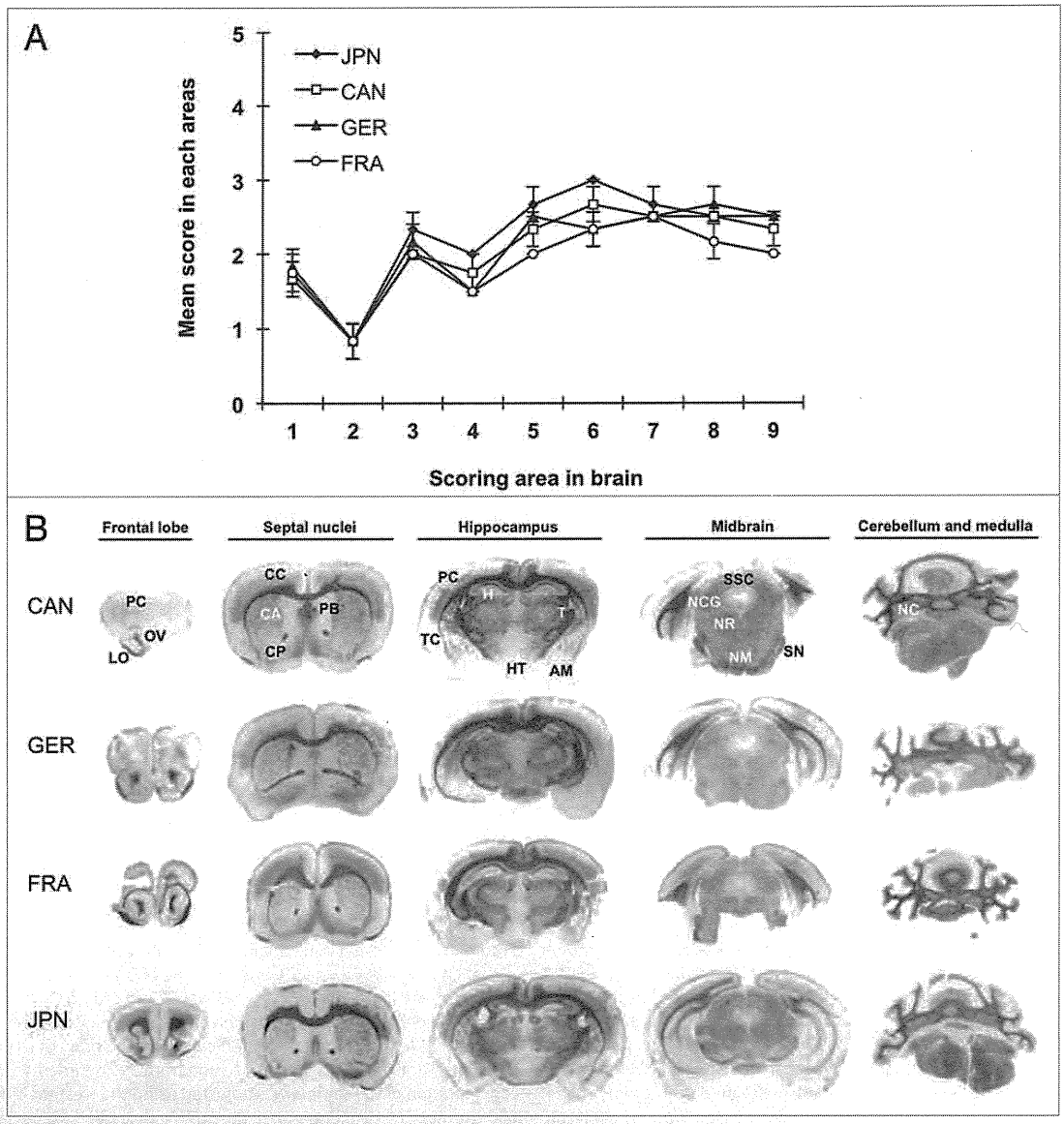

Figure 1. Neuropathological analysis of L-type BSE isolate-affected TgBoPrP. (A) Lesion profile in the first passage. The vacuolation in the following brain regions was scored on a scale of 0–5 (mean values): 1, dorsal medulla; 2, cerebellar cortex; 3, superior cortex; 4, hypothalamus; 5, thalamus; 6, hippocampus; 7, septal nuclei of the paraterminal body; 8, cerebral cortex at the levels of the hypothalamus and thalamus; and 9, cerebral cortex at the level of the septal nuclei of the paraterminal body. The data are presented as mean ± standard deviation (n = 5). ◆, Japanese L-type BSE (JPN); □, Canadian L-type BSE (CAN); ▲, German L-type BSE (GER); ○, French L-type BSE (FRA). (B) The neuroanatomical distribution of PrP^{Sc} in the brain of TgBoPrP mice infected with Canadian (CAN), German (GER), French (FRA) and Japanese (JPN) L-type BSE isolate by PET-blot analysis. The PET-blot analysis reveals preferential and intense PrP^{Sc} immunolabeling along with periventricular areas, corpus callosum and cerebellar gray matter. Widespread PrP^{Sc} immunolabeling is also detected in the thalamic and brainstem nuclei, while PrP^{Sc} immunostaining in the cerebral and cerebellar cortices and basal ganglia is less conspicuous. Dewaxed membranes were treated with PK (80 μg/mL), followed by denaturation with 3 M guanidine thiocyanate. The monoclonal antibody (mAb) SAF84 was used. Blots corresponding to the brain areas at the level of frontal lob, septal nuclei, hippocampus, midbrain and medulla and cerebellum. FC, frontal cortex; OV, olfactory ventricle; LO, lateral orbital cortex; CC, cingulated cortex; CP, caudate putamen; PB, paraterminal body; CP, caudate putamen; PC, parietal cortex; TC, temporal cortex; H, hippocampus; T, thalamus; HT, hypothalamus; AM, amygdala; SSC, stratum moleculare of the cerebellum; NCG, nucleus corporis geniculati; NR, nucleus rubber; SN, substantia nigra; NM, nucleus mammillaris; NC, deep nuclei of the cerebellum.

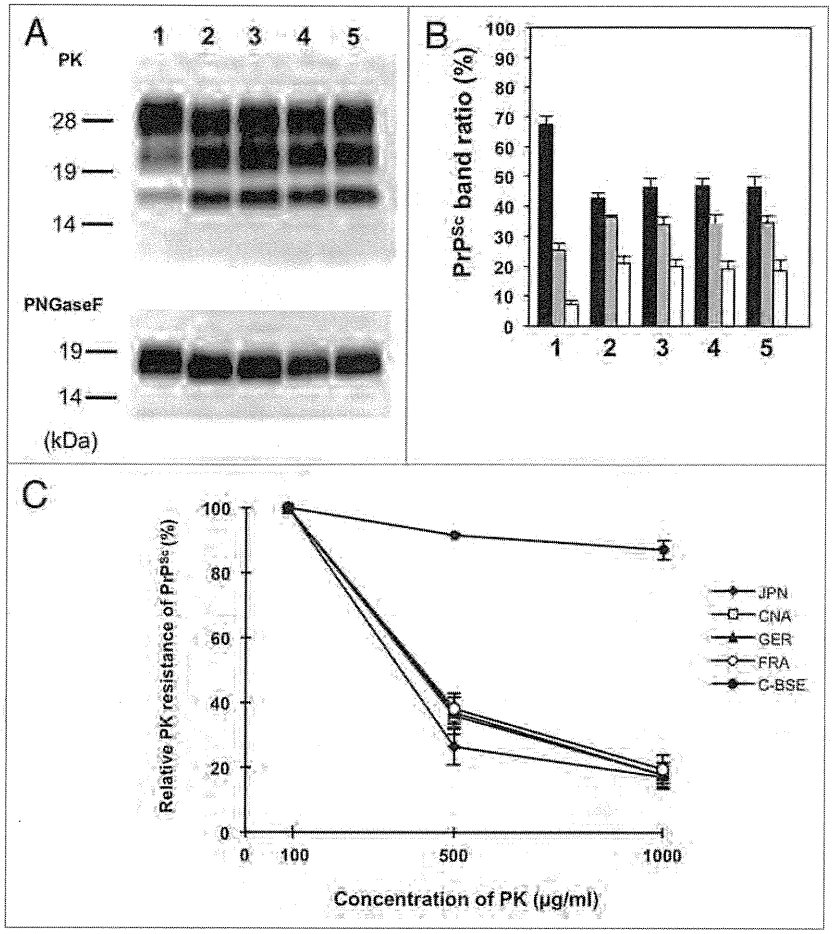

Figure 2. Western blot analysis of proteinase K (PK)-digested prion protein (PrPcore) from the brain of L-type BSE isolate-affected TgBoPrP. (A) Lane 1, Classical-BSE; Lane 2, Japanese L-type BSE; Lane 3, Canadian L-type BSE; Lane 4, French L-type BSE; Lane 5, German L-type BSE. All the samples were digested with 50 μg/ml PK at 37°C for 1 h (upper part), and digested aliquots were treated with N-glycosidase F (PNGaseF), according to the manufacturer's instructions (bottom part). PrPcore was detected with mAb 6H4. Molecular markers are shown on the left (kDa). (B) The relative amounts of the diglycosylated (solid black bar), monoglycosylated (gray bar), and unglycosylated (clear bar) forms in the PrPcore from the brain of L-type BSE isolate-affected TgBoPrP. The lane numbers are as listed in (A). The results are presented as mean ± standard deviation from 5 experiments. (C) Relative PK resistance of PrP^{Sc} from L-type BSE isolate-affected TgBoPrP. The PrP^{Sc} concentration of the sample was adjusted using the western blot signal intensity. The samples were treated with various concentrations of PK (100–1,000 μg/mL). The results are presented as mean ± standard deviation from 3 experiments. PrP^{Sc} was detected with mAb 6H4. ◆, Japanese L-type BSE (JPN); □, Canadian L-type BSE (CAN); ▲, German L-type BSE (GER); ○, French L-type BSE (FRA); ●, Classical-BSE (C-BSE).

References

1. Prusiner SB. Molecular biology of prion diseases. Science 1991; 252:1515-22; PMID:1675487; http://dx.doi.org/10.1126/science.1675487.
2. Bolton DC, McKinley MP, Prusiner SB. Identification of a protein that purifies with the scrapie prion. Science 1982; 218:1309-11; PMID:6815801; http://dx.doi.org/10.1126/science.6815801.
3. Wells GA, Scott AC, Johnson CT, Gunning RF, Hancock RD, Jeffrey M, et al. A novel progressive spongiform encephalopathy in cattle. Vet Rec 1987; 121:419-20; PMID:3424605; http://dx.doi.org/10.1136/vr.121.18.419.
4. Ducrot C, Arnold M, de Koeijer A, Heim D, Calavas D. Review on the epidemiology and dynamics of BSE epidemics. Vet Res 2008; 39:15; PMID:18187031; http://dx.doi.org/10.1051/vetres:2007053.
5. Giles J. Mad cow disease comes to Japan. Nature 2001; 413:240; PMID:11564996; http://dx.doi.org/10.1038/35095180.
6. Richt JA, Kunkle RA, Alt D, Nicholson EM, Hamir AN, Czub S, et al. Identification and characterization of two bovine spongiform encephalopathy cases diagnosed in the United States. J Vet Diagn Invest 2007; 19:142-54; PMID:17402608; http://dx.doi.org/10.1177/104063870701900202.
7. Bruce M, Chree A, McConnell I, Foster J, Pearson G, Fraser H. Transmission of bovine spongiform encephalopathy and scrapie to mice: strain variation and the species barrier. Philos Trans R Soc Lond B Biol Sci 1994; 343:405-11; PMID:7913758; http://dx.doi.org/10.1098/rstb.1994.0036.
8. Yamakawa Y, Hagiwara K, Nohtomi K, Nakamura Y, Nishijima M, Higuchi Y, et al. Expert Committee for BSE Diagnosis, Ministry of Health, Labour and Welfare of Japan. Atypical proteinase K-resistant prion protein (PrP^{res}) observed in an apparently healthy 23-month-old Holstein steer. Jpn J Infect Dis 2003; 56:221-2; PMID:14695437.
9. Hagiwara K, Yamakawa Y, Sato Y, Nakamura Y, Tobiume M, Shinagawa M, et al. Accumulation of mono-glycosylated form-rich, plaque-forming PrP^{Sc} in the second atypical bovine spongiform encephalopathy case in Japan. Jpn J Infect Dis 2007; 60:305-8; PMID:17881874.
10. Biacabe AG, Laplanche JL, Ryder S, Baron T. Distinct molecular phenotypes in bovine prion diseases. EMBO Rep 2004; 5:110-5; PMID:14710195; http://dx.doi.org/10.1038/sj.embor.7400054.
11. Buschmann A, Gretzschel A, Biacabe AG, Schiebel K, Corona C, Hoffmann C, et al. Atypical BSE in Germany—proof of transmissibility and biochemical characterization. Vet Microbiol 2006; 117:103-16; PMID:16916588; http://dx.doi.org/10.1016/j.vetmic.2006.06.016.
12. Dudas S, Yang J, Graham C, Czub M, McAllister TA, Coulthart MB, et al. Molecular, biochemical and genetic characteristics of BSE in Canada. PLoS One 2010; 5:10638; PMID:20498835; http://dx.doi.org/10.1371/journal.pone.0010638.

13. Casalone C, Zanusso G, Acutis P, Ferrari S, Capucci L, Tagliavini F, et al. Identification of a second bovine amyloidotic spongiform encephalopathy: molecular similarities with sporadic Creutzfeldt-Jakob disease. Proc Natl Acad Sci USA 2004; 101:3065-70; PMID:14970340; http://dx.doi.org/10.1073/pnas.0305777101.
14. Jacobs JG, Langeveld JP, Biacabe AG, Acutis PL, Polak MP, Gavier-Widen D, et al. Molecular discrimination of atypical bovine spongiform encephalopathy strains from a geographical region spanning a wide area in Europe. J Clin Microbiol 2007; 45:1821-9; PMID:17442800; http://dx.doi.org/10.1128/JCM.00160-07.
15. Polak MP, Zmudzinski JF, Jacobs JG, Langeveld JP. Atypical status of bovine spongiform encephalopathy in Poland: a molecular typing study. Arch Virol 2008; 153:69-79; PMID:17896076; http://dx.doi.org/10.1007/s00705-007-1062-6.
16. Tester S, Juillerat V, Doherr MG, Haase B, Polak M, Ehrensperger F, et al. Biochemical typing of pathological prion protein in aging cattle with BSE. Virol J 2009; 6:64; PMID:19470160; http://dx.doi.org/10.1186/1743-422X-6-64.
17. Terry LA, Jenkins R, Thorne L, Everest SJ, Chaplin MJ, Davis LA, et al. First case of H-type bovine spongiform encephalopathy identified in Great Britain. Vet Rec 2007; 160:873-4; PMID:17586792; http://dx.doi.org/10.1136/vr.160.25.873.
18. Masujin K, Shu Y, Yamakawa Y, Hagiwara K, Sata T, Matsuura Y, et al. Biological and biochemical characterization of L-type-like bovine spongiform encephalopathy (BSE) detected in Japanese black beef cattle. Prion 2008; 2:123-8; PMID:19158500; http://dx.doi.org/10.4161/pri.2.3.7437.
19. Fukuda S, Iwamaru Y, Imamura M, Masujin K, Shimizu Y, Matsuura Y, et al. Intraspecies transmission of L-type-like Bovine Spongiform Encephalopathy detected in Japan. Microbiol Immunol 2009; 53:704-7; PMID:19954458; http://dx.doi.org/10.1111/j.1348-0421.2009.00169.x.
20. Scott MR, Safar J, Telling G, Nguyen O, Groth D, Torchia M, et al. Identification of a prion protein epitope modulating transmission of bovine spongiform encephalopathy prions to transgenic mice. Proc Natl Acad Sci USA 1997; 94:14279-84; PMID:9405603; http://dx.doi.org/10.1073/pnas.94.26.14279.
21. Capobianco R, Casalone C, Suardi S, Mangieri M, Miccolo C, Limido L, et al. Conversion of the BASE prion strain into the BSE strain: the origin of BSE? PLoS Pathog 2007; 3:31; PMID:17352534; http://dx.doi.org/10.1371/journal.ppat.0030031.
22. Béringue V, Andréoletti O, Le Dur A, Essalmani R, Vilotte JL, Lacroux C, et al. A bovine prion acquires an epidemic bovine spongiform encephalopathy strain-like phenotype on interspecies transmission. J Neurosci 2007; 27:6965-71; PMID:17596445; http://dx.doi.org/10.1523/JNEUROSCI.0693-07.2007.