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Epidemiological features of the bovine spongiform encephalopathy epidemic in Japan

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Summary
On 10 September 2001, the first case of bovine spongiform encephalopathy (BSE) in Japan was confirmed in a five-year-old dairy cow born in Hokkaido and raised in Chiba prefecture. Subsequently, BSE surveillance was enhanced. As a result, 35 additional cases were detected by the end of March 2009, with two to ten cases being detected each year up to the end of 2007. The epidemic appeared to peak in 2006. Cases are detected mostly in dairy cattle; in cattle born in 1996 and 2000; and in cattle born in Hokkaido. Two were atypical cases of BSE and the remainder classical cases. This paper describes and discusses the demographic structure of dairy and beef cattle, surveillance programmes in place and the epidemiological features of the BSE epidemic in Japan.

Keywords
Bovine spongiform encephalopathy – Epidemiology – Japan – Surveillance.

Introduction

Bovine spongiform encephalopathy (BSE) is a progressive and fatal disease affecting the central nervous system of cattle. The disease was first observed in the United Kingdom (UK) in 1986, and subsequently reported in other countries in the 1990s and early 2000s. Meat-and-bone meal (MBM) contaminated with a scrapie-like agent is considered to be the primary vehicle of infection (27).

The Japanese government has taken various measures to protect the cattle population from exposure to contaminated feed: since July 1990 importation of live cattle and MBM has been prohibited from the UK and other countries with cases of BSE (except for MBM heat-treated at 133°C/3 bar/30 min); since March 1996 importation of MBM from the UK has been totally prohibited; in April 1996 an administrative guidance was issued to prohibit the use of ruminant MBM for ruminant feed; and in January 2001 importation of MBM from Switzerland, Liechtenstein and Member States of the European Union (EU) was prohibited.

In spite of these measures, a case of BSE was detected in Japan on 10 September 2001. After the detection of the first case, the Japanese government introduced the following measures:

– the mandatory removal and incineration of specified risk material (SRM) from all cattle slaughtered for human consumption from 27 September 2001. Specified risk material was initially defined as the brain, the spinal cord, the eyes and the distal ileum and expanded on 16 February 2004 to include the vertebral column;

– a legal ban on the domestic use of ruminant protein for ruminant feed was implemented on 18 September 2001, followed by a ban on the domestic use and importation of all processed animal protein for the production of fertiliser or feed for ruminants, pigs, and chickens, effective from 4 October 2001;

– enhancement of BSE surveillance, involving mandatory reporting and investigation of all clinical BSE suspects (passive surveillance), testing of fallen stock and all cattle slaughtered for human consumption (active surveillance);

– an animal identification system, which enables trace-back to the farm of origin and access to other relevant information, including the date of birth, was introduced in December 2003.

As a result of the enhanced BSE surveillance, 35 additional cases were detected by the end of March 2009. Several epidemiological studies have been conducted to explore the possible source of introduction of the BSE agent, to identify the possible risk factors, to estimate the prevalence of infection in different birth cohorts, and to predict the future epidemic. This paper describes and discusses the demographic structure of dairy and beef cattle populations, surveillance programmes in place and epidemiological features of the BSE epidemic in Japan.

Demographic structure of the cattle population in Japan

Owing to the fact that BSE is a disease with a long incubation period and that the presently available diagnostic methods are capable of detecting infected animals only at the last three to 12 months of the incubation period (6), the demographic structure of the cattle population is an important factor that affects the incidence of BSE.

As of 1 February 2007, the total cattle population was 4,398,300, of which 2,806,300 were beef cattle and 1,592,000 dairy (17). The dairy cattle are dairy females of Holstein (99%), Jersey, and other breeds. They are culled at various ages, but on average at the age of six or seven after three or four parturitions. Highly concentrated rations are fed to dairy cattle to keep their average milk yield close to 8,000 kg per cow per one lactation period. Hokkaido, Japan's second largest island and largest prefecture, is the major dairy production area, representing 53% of the total dairy cattle population. Hokkaido also supplies dairy heifers to the other prefectures.

The Japanese beef cattle population comprises three types of cattle:

– Wagyu breeds (Japanese indigenous breeds including Japanese Black, Japanese Brown, Japanese Shorthorn and Japanese Polled)

– Holstein steers

– cross-breeds between Wagyu bulls and Holstein dairy cows.

Most Wagyu breed cattle are slaughtered at around the age of 30 months as steers or heifers, but more than 10% of females are used for breeding until they are five years of age and 5% until 12 years of age. Male Holstein calves, born from Holstein dairy cows, are castrated and slaughtered as steers at around the age of 20 months. Wagyu-Holstein-cross calves, also born from Holstein dairy cows by crossing with a Wagyu breed bull, are slaughtered as steers or heifers at around the age of 25 months. The calves are usually separated from their dams at two to three weeks of age and immediately transported to beef cattle rearing farms where they are initially fed with milk replacers followed by calf starter, forage and compound feed. Many of these calves are again transferred to beef fattening farms at six to seven months of age. The number of cattle by type and breed in the major cattle-rearing areas of Japan is shown in Table I. The Kyushu region, Miyazaki and Kagoshima prefectures in particular, supply Wagyu calves to beef cattle feeding and fattening areas in other prefectures.

Figure 1 shows the survival curves of dairy and different types of beef cattle in Japan obtained using the cattle traceability database (1).

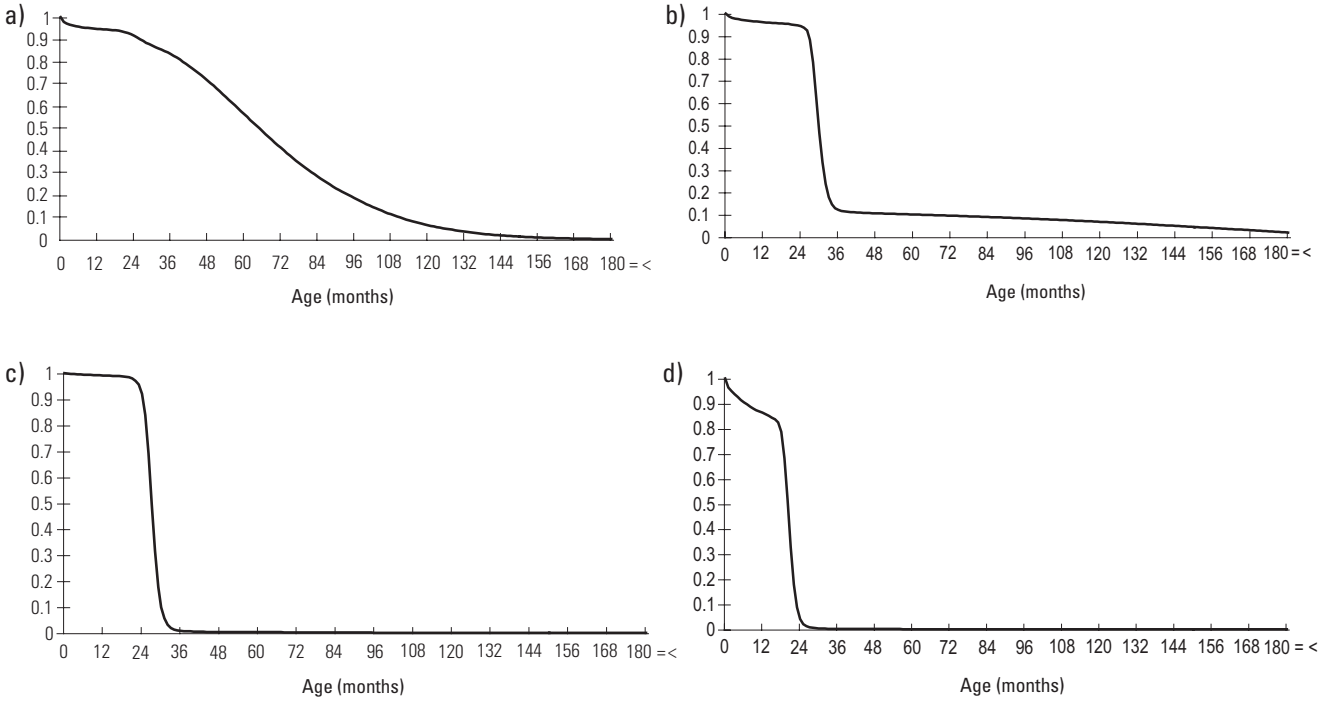
Bovine spongiform encephalopathy surveillance in place in Japan

Prior to October 2001, only passive surveillance was practised, targeting clinical suspects (animals examined on

Table I
Number of cattle by type and breed in the major cattle-rearing areas of Japan as at 1 Feb 2007

Type of cattle	Location	Number of animals	Percentage
Dairy	Japan	1,592,000	100.0%
	Hokkaido prefecture	836,000	52.5%
Beef	Japan	1,742,000	100.0%
(Wagyu)	Kyushu region*	841,300	48.3%
Beef	Japan	460,300	100.0%
(Holstein	Hokkaido prefecture	222,600	48.4%
steers)			
Beef	Japan	604,000	100.0%
(Wagyu	Hokkaido prefecture	115,700	19.2%
x Holstein)	Kanto region**	142,800	23.6%
	Kyushu region*	125,200	20.7%
All breeds	Japan	4,398,300	

Source: Ministry of Agriculture, Forestry and Fisheries, Japan
* Kyushu region consists of Fukuoka, Saga, Nagasaki, Kumamoto, Oita, Miyazaki and Kagoshima prefectures
** Kanto region consists of Ibaraki, Tochigi, Gunma, Saitama, Chiba, Tokyo, Kanagawa, Yamanshi and Nagago prefectures



a) dairy females
b) Wagyu-beef cattle (mostly steers and heifers)
c) Wagyu-Holstein-cross steers and heifers
d) Holstein steers

Fig. 1
Survival curves of cattle in Japan, obtained from the cattle traceability database (1)

farms and showing neurological signs consistent with a clinical diagnosis of BSE). Since October 2001, systematic screening of cattle of all ages slaughtered for human consumption has been introduced. Since 1 April 2004, all fallen stock ≥ 24 months old have been tested.

In December 2003, a National Cattle Identification Scheme was introduced which records the date and farm of birth of every bovine animal. As a result, no cattle, except fallen stock < 24 months old can leave the population without being tested (slaughter cattle < 21 months old have been exempted from testing since 1 August 2005).

BSE testing is first done with one of the three validated rapid screening tests: Platelia enzyme-linked immunosorbent assay (ELISA) kit (Bio-Rad Laboratories), Enfer BSE test (Enfer) and FRELISA BSE kit (Fujirebio). If the result of the screening test is not negative, chilled obex samples are sent to the National Institute of Animal Health, National Institute of Infectious Diseases, Obihiro University of Agriculture and Veterinary Medicine or Hokkaido University for confirmatory diagnosis. Diagnosis is confirmed either by detection of infectious prion protein (PrP^{Sc}) by Western blot or immunohistochemistry. The decision on the final diagnosis is made by the Expert Committee for BSE Diagnosis (an expert panel of the Ministry of Health, Labour and Welfare [MHLW]) or by the Subcommittee on Prion Diseases of the Advisory Committee on Food, Agriculture and Rural Policy (an expert panel of the Ministry of Agriculture, Forestry and Fisheries [MAFF]) (16).

Passive surveillance

Bovine spongiform encephalopathy was designated a notifiable disease under the Domestic Animal Infectious Diseases Control Law in April 1996. However, only between 20 and 36 clinical suspects were officially reported annually for histopathological examination of brains. In April 2001 passive surveillance was enhanced by increasing the number of brain samples to be tested, in accordance with the international standard set by the World Organisation for Animal Health (OIE). In September 2001, the first case of BSE was detected under this enhanced passive surveillance. In October 2001, immediately after the detection of the first case of BSE, surveillance was enhanced again so that all on-farm cattle showing neurological signs consistent with BSE would be reported, examined and slaughtered for laboratory diagnosis.

To encourage reporting of clinical suspects, farmers are paid compensation of 80% of the market value for a suspected cow culled under the Domestic Animal

Infectious Diseases Control Law. In addition to legal compensation, the rest of the value is paid by mutual assistance for confirmed cases and a premium is paid for fallen stock, taking into account how much the animals were insured for.

The clinical suspects are reported to prefecture governors; they are then investigated by a veterinary inspector from the prefecture Livestock Hygiene Service Center and samples are taken for laboratory diagnosis.

Active surveillance

Fallen stock collection and brain sampling

Between 1 April 1996 and 31 March 2001, between 194 and 237 fallen stock ≥ 24 months old were subjected to histopathological examination of the brain annually. Under the law concerning Special Measures for the Prevention of Bovine Spongiform Encephalopathy enacted in July 2002, all fallen stock must be reported to the prefecture governor. Brain samples are taken from the dead animals by a prefecture veterinary inspector for BSE testing, in accordance with the Domestic Animal Infectious Diseases Control Law. If the results are positive, the entire carcass is incinerated. If the results are negative, the carcass is rendered and transformed into MBM for incineration. Since 1 April 2004, all fallen stock ≥ 24 months old have been subjected to BSE testing. In recent years, no distinction is made between clinical suspects and fallen stock: the clinical suspects are euthanased and tested for BSE in the same way as fallen stock.

Testing for bovine spongiform encephalopathy in cattle slaughtered for human consumption

As of 18 October 2001, all cattle slaughtered in abattoirs for human consumption have been subjected to BSE testing under the Abattoir Law. All cattle for slaughter are subjected to ante-mortem inspection before they are slaughtered in abattoirs. If cattle are found to be showing neurological signs or signs compatible with BSE, they are rejected for slaughter. Meat inspectors, who are veterinarians, collect brain-stem samples from all cattle slaughtered, which are sent to prefecture meat-inspection laboratories for the BSE screening test. The carcass of any animal testing positive for BSE is incinerated.

In September 2004, based on the result of risk assessment by the Food Safety Commission (FSC), the MHLW decided to exempt cattle < 21 months old from BSE testing. The MHLW amended the regulations so that all cattle ≥ 21 months old were tested as of 1 August 2005. However, reflecting the concern of consumers, all cattle slaughtered for human consumption continued to be tested voluntarily by the prefecture governments with financial support from the MHLW (the financial support ceased on 1 August 2008 but testing continues).

The number of clinical suspects, fallen stock animals and slaughtered cattle subjected to BSE testing is shown in Table II. The details of the 36 cases detected between 10 September 2001 and the end of March 2009 are shown in Table III.

Epidemiological features of the bovine spongiform encephalopathy epidemic in Japan

Higher incidence in the dairy population

Of the 36 cases detected by the end of March 2009, 31 cases were in dairy cattle, two were Holstein steers and three were Wagyu beef cattle (Fig. 2).

The demographic structure of the dairy population contributes to the higher incidence in this population. As shown by comparing Fig. 1a with Fig. 1b, Fig. 1c or Fig. 1d, dairy cattle are usually older than beef cattle when they are culled and consequently a large proportion of infected dairy cattle are at the end or last stage of the incubation period when they are culled. In contrast, the majority of beef cattle, in particular Holstein steers and Wagyu-Holstein-cross cattle, leave the population early in the incubation period when the presence of BSE cannot be detected.

Table II
Evolution of the number of cattle subjected to bovine spongiform encephalopathy testing recorded during fiscal years 1996 to 2007 in different risk subpopulations

Risk subpopulation	Fiscal years											
	1996	1997	1998	1999	2000	2001*	2002	2003	2004	2005	2006	2007**
Clinical suspects	23	20	36	36	24	132	420	4,505	6,241	6,957	1,777	90,829
Fallen stock	194	203	210	237	227	727	3,756	43,645	92,333	88,144	92,832	
Slaughter cattle	0	0	0	0	0	523,591	1,253,811	1,252,630	1,265,620	1,232,252	1,218,285	1,228,256

Source: Ministry of Agriculture, Forestry and Fisheries and Ministry of Health, Labour and Welfare, Japan
* The first case of bovine spongiform encephalopathy in Japan was detected on 10 September 2001
** No distinction was made between clinical suspects and fallen stock in this year

Table III
Date of diagnosis, date of birth, place of birth, clinical signs and results of laboratory tests of cases of bovine spongiform encephalopathy detected by the end of March 2009 in Japan

No.	Date of diagnosis	Date of birth	Type of cattle	Prefecture of birth	Clinical signs	Type of surveillance	Laboratory diagnosis*	Typical/ atypical
1	10 Sep 2001	26 March 1996	Dairy cow	Hokkaido	Downer	CS	WB, IHC, HP	Typical
2	21 Nov 2001	4 April 1996	Dairy cow	Hokkaido	None	SH	WB, IHC	Typical
3	2 Dec 2001	26 March 1996	Dairy cow	Gunma	None	SH	WB, IHC, HP	Typical
4	13 May 2002	23 March 1996	Dairy cow	Hokkaido	Muscular split of forelimb	SH	WB, IHC, HP	Typical
5	23 Aug 2002	5 Dec 1995	Dairy cow	Kanagawa	Hip joint dislocation	SH	WB, IHC	Typical
6	20 Jan 2003	10 Feb 1996	Dairy cow	Hokkaido	Downer	SH	WB, IHC, HP	Typical
7	23 Jan 2003	28 March 1996	Dairy cow	Hokkaido	None	SH	WB, IHC	Typical
8	6 Oct 2003	13 Oct 2001	Holstein steer	Tochigi	None	SH	WB	Atypical
9	4 Nov 2003	13 Jan 2002	Holstein steer	Hyogo	None	SH	WB	Typical
10	22 Feb 2004	17 March 1996	Dairy cow	Kanagawa	Hip joint dislocation	SH	WB, IHC, HP	Typical
11	9 March 2004	8 April 1996	Dairy cow	Hokkaido	Hip joint dislocation	FS	WB, IHC, HP	Typical
12	13 Sep 2004	3 July 1999	Dairy cow	Kumamoto	None	SH	WB, IHC, HP	Typical
13	23 Sep 2004	18 Feb 1996	Dairy cow	Hokkaido	Hip joint dislocation	SH	WB, IHC, HP	Typical
14	14 Oct 2004	8 Oct 2000	Dairy cow	Hokkaido	Suffocation	FS	WB, IHC, HP	Typical
15	26 Feb 2005	5 Aug 1996	Dairy cow	Hokkaido	Arthritis	FS	WB, IHC, HP	Typical
16	27 March 2005	23 March 1996	Dairy cow	Hokkaido	None	SH	WB, IHC, HP	Typical
17	8 April 2005	11 Sep 2000	Dairy cow	Hokkaido	Downer	FS	WB, IHC, HP	Typical
18	12 May 2005	31 Aug 1999	Dairy cow	Hokkaido	Hip joint dislocation	SH	WB, IHC, HP	Typical
19	2 June 2005	16 April 1996	Dairy cow	Hokkaido	None	SH	WB, IHC	Typical
20	6 June 2005	12 Aug 2000	Dairy cow	Hokkaido	None	SH	WB, IHC	Typical
21	10 Dec 2005	13 Feb 2000	Dairy cow	Hokkaido	Heart failure	FS	WB, IHC	Typical
22	23 Jan 2006	1 Sep 2000	Dairy cow	Hokkaido	Abomasum displacement	FS	WB, IHC	Typical
23	15 March 2006	8 July 2000	Dairy cow	Hokkaido	None	SH	WB, IHC, HP	Typical
24	17 March 2006	10 Feb 1992	Beef cow	Nagasaki	Downer	SH	WB, IHC, HP	Atypical
25	19 April 2006	18 April 2000	Dairy cow	Hokkaido	None	SH	WB, IHC	Typical
26	13 May 2006	11 Aug 2000	Dairy cow	Hokkaido	Arthritis	FS	WB, IHC	Typical
27	19 May 2006	20 Aug 2000	Dairy cow	Hokkaido	Mastitis	FS	WB, IHC, HP	Typical
28	11 Aug 2006	21 Nov 1999	Dairy cow	Hokkaido	Hip joint dislocation	FS	WB, IHC	Typical
29	28 Sep 2006	24 June 2000	Dairy cow	Hokkaido	Ketosis	FS	WB, IHC, HP	Typical
30	13 Nov 2006	28 June 2001	Dairy cow	Hokkaido	Heart failure	FS	WB, IHC	Typical
31	8 Dec 2006	12 Nov 1999	Dairy cow	Hokkaido	Staggering	FS	WB, IHC	Typical
32	5 Feb 2007	26 Aug 2001	Dairy cow	Hokkaido	Hip swelling	SH	WB, IHC, HP	Typical
33	2 July 2007	21 June 2000	Dairy cow	Hokkaido	Fatty liver	FS	WB, IHC	Typical
34	21 Dec 2007	1 July 1992	Beef cow	Hokkaido	None	SH	WB, IHC	Typical
35	24 March 2008	12 Oct 2000	Beef cow	Hokkaido	Heart failure	FS	WB, IHC, HP	Typical
36	30 Jan 2009	5 Aug 2000	Dairy cow	Hokkaido	Downer	FS	WB, IHC	Typical

Source: Ministry of Agriculture, Forestry and Fisheries and Ministry of Health, Labour and Welfare, Japan
CS: clinical suspect (passive surveillance targeting animals on farms with clinical signs compatible with bovine spongiform encephalopathy)
FS: fallen stock (active surveillance targeting all FS animals of 24 months of age or older, dead on farms or during transportation)
SH: slaughterhouse (active surveillance targeting all animals slaughtered for human consumption)
WB: Western blot
IHC: immunohistochemistry
HP: histopathology
* All three tests were carried out in each case, but only those that were positive have been listed

The authors estimated the prevalence of infected animals in birth cohorts from 1995 to 2001 of different subpopulations using the surveillance data obtained up to the end of 2008 and the model described by Sugiura (24, 25). The results showed that the prevalence of infected animals in dairy birth cohorts was not significantly higher than that of the other subpopulation cohorts (Fig. 3). On the basis of the data available, it is not known whether the

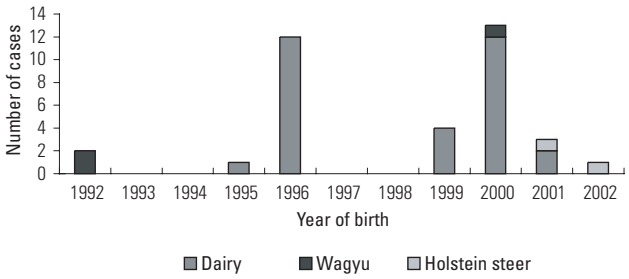
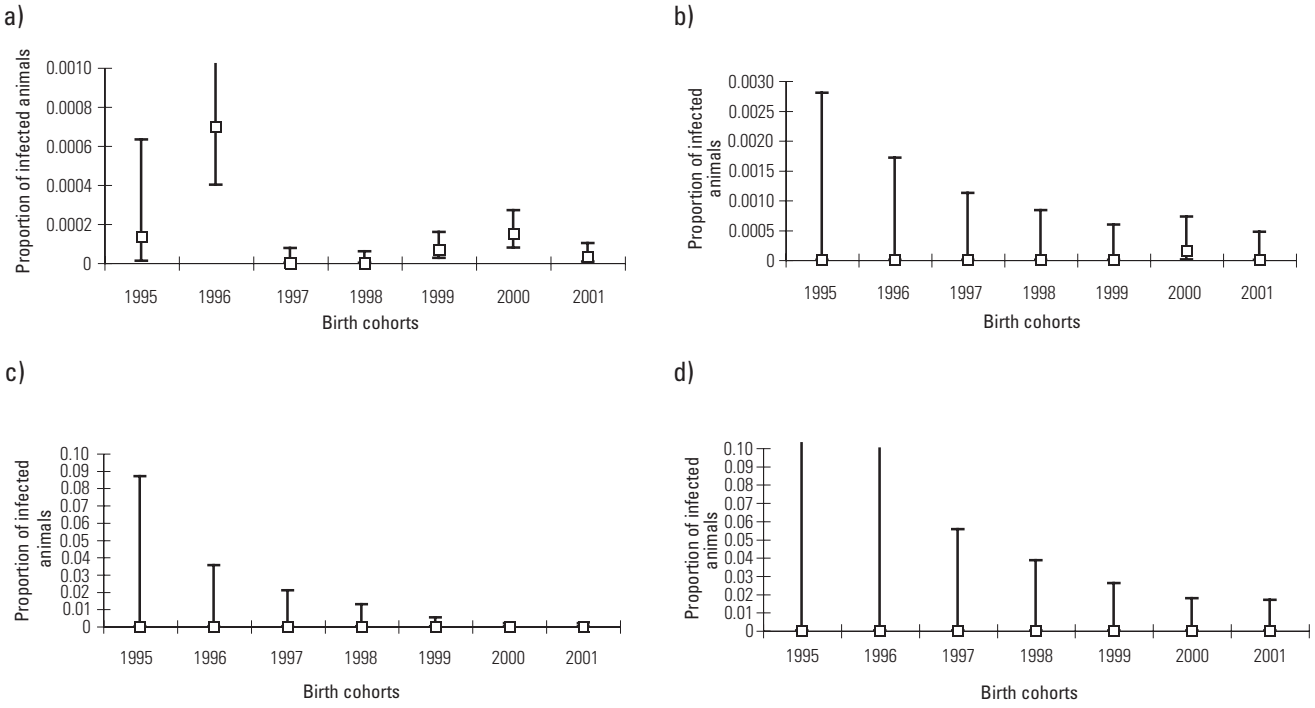


Fig. 2
The 36 bovine spongiform encephalopathy cases detected in Japan between September 2001 and the end of March 2009: year of birth and type of cattle

higher incidence in the dairy population is due in part to the higher prevalence of infection in this subpopulation or if it is due only to the demographic structure of this subpopulation.

Higher prevalence in 1996 and 2000 birth cohorts

Of the 36 cases detected by the end of March 2009, 12 were born in 1996, 13 in 2000, four in 1999, three in 2001, two in 1992 and one each in 1995 and 2002 (Fig. 2). Because no BSE cases were born in 1997 and 1998, those born in 1995 and 1996 and those born in 1999, 2000 and 2001 form two independent temporal clusters, assuming that the culling rate and type of cattle kept each year do not change over time. In particular, of the 13 BSE cases born between 1995 and 1996, 11 were born within a two-month period from 10 February 1996 to 8 April 1996. Considering that cattle usually get infected within one year of birth (2), it is likely that most animal feed consumed in these years was contaminated with the



a) dairy cattle
b) Wagyu beef cattle
c) Wagyu-Holstein cross beef steers and heifers
d) Holstein steers

Fig. 3
Proportion of infected animals in birth cohorts from 1995 to 2001, estimated using surveillance data up to the end of 2008 and the model described by Sugiura *et al.* (22, 26)

Open squares indicate the most likely estimates and error bars 95% confidence intervals. Most likely estimates of the proportion of infected animals in all cohorts of Wagyu beef, Wagyu-Holstein-crosses and Holstein steers (except for the 2000 Wagyu beef cohort) are zero, because no cases have been detected in these cohorts. However, the confidence intervals of these subpopulations are larger than those of dairy subpopulations, because most

of the animals in these subpopulations are culled at a young age before the disease becomes detectable by a rapid test, and therefore do not provide information for estimating the proportion of infected animals. In estimating the proportion of infected animals in the 2001 Holstein steer birth cohort, the atypical case born in 2001 and detected in 2003 (number 8 in Table III) was not considered

BSE agent. Using Bayesian theory, Sugiura *et al.* (24) estimated that the number of BSE-infected dairy cattle born in 1996 and 2000 was 461 (95% confidence interval: 262-802) and 82 (42-148), respectively. Yamamoto *et al.* (30), using a simulation-based model, estimated that BSE-infected cattle born in 1996 numbered 155 (90-275). Hamasaki and Yamamoto (9), using BSurvE method, estimated that BSE-infected cattle born in 1996 numbered 288.

Higher incidence in cattle born in Hokkaido

Of the 36 cases detected by the end of March 2009, 28 were born in Hokkaido prefecture, two in Kanagawa, one each in Gunma, Tochigi, Hyogo, Shimane, Nagasaki and Kumamoto prefectures. Considering that the number of cases born in Hokkaido represents 78% of the total number of cases and that the dairy population in Hokkaido represents 53% of the national population (Table I), the incidence in Hokkaido standardised for cattle population is still higher than that in mainland Japan, assuming that the culling rate does not differ between the dairy and mainland dairy subpopulations. This indicates that the Hokkaido-born cattle were more exposed to the BSE agent through contaminated feed than cattle born in other prefectures. Many BSE cases are reported in the densely populated areas in Hokkaido. Most of the BSE cases detected in Hokkaido were detected in the eastern districts of Tokachi, Abashiri, Kushiro and Nemuro, where approximately 75% of dairy cattle in Hokkaido are reared. Although none of the farms with BSE cases had a record of using MBM for cattle, their feed might have been cross-contaminated with MBM for feed for non-ruminant species (31). Nonaka *et al.* conducted a geographical analysis using a geographical information system to examine the possible spatial relationship between the suspected feed and BSE cases in Hokkaido, with no significant relationship found (18).

The map in Figure 4 shows the birth prefectures of BSE cases detected by the end of March 2009.

Relatively high incidence risk in slaughter cattle

BSE cases were detected more in slaughter cattle than in fallen stock up until 2004, after which there were more cases in the fallen stock than in slaughter cattle. This is attributed to the enhanced surveillance of fallen stock for BSE as of 1 April 2004 (Fig. 5). Sugiura, using an 'adjusted incidence risk', which is comparable with incidence risks

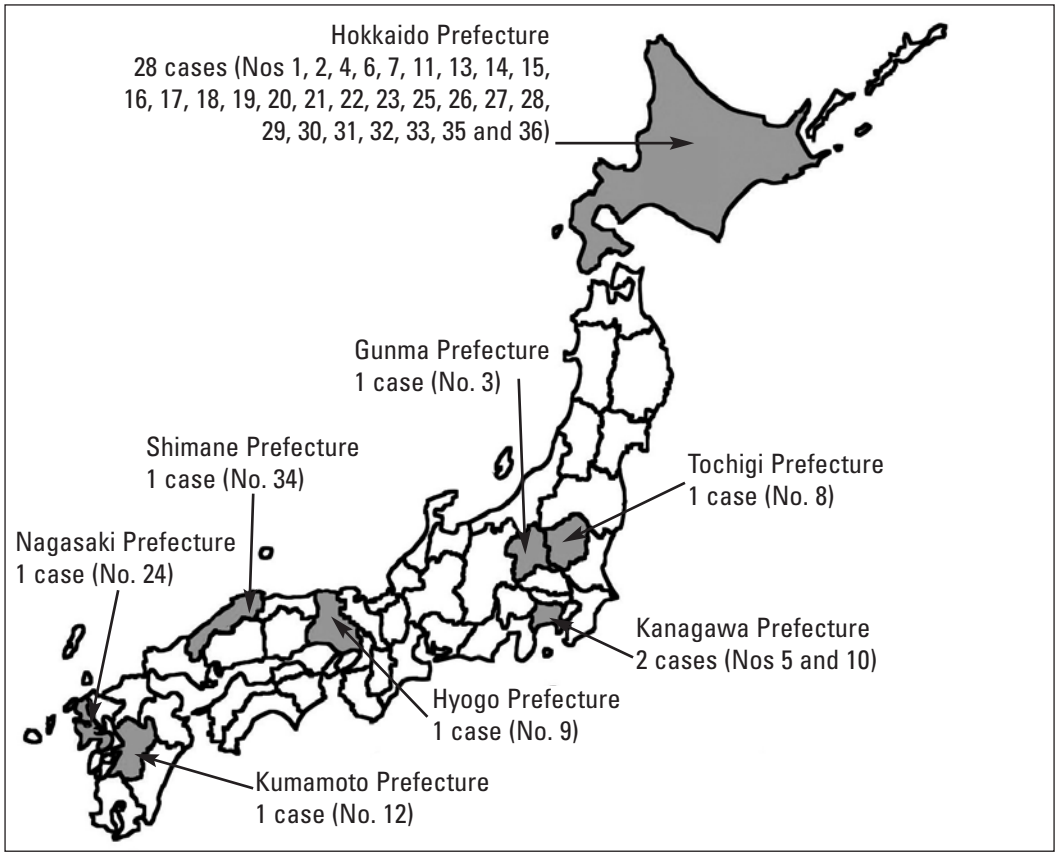


Fig. 4

Prefectures of birth of bovine spongiform encephalopathy cases detected in Japan between September 2001 and March 2009

Numbers in brackets correspond to the numbers in Table III

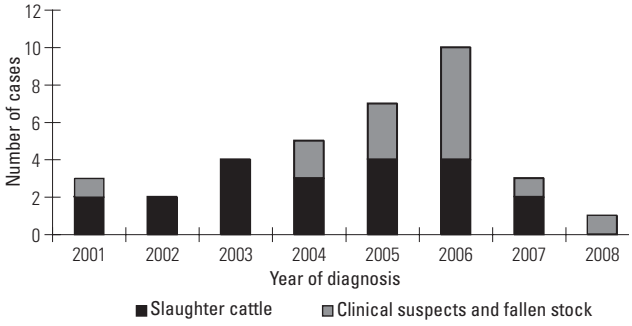


Fig. 5
Evolution of the number of bovine spongiform encephalopathy cases detected in Japan by the risk subpopulation

in Europe, noted that the ratio of incidence risk in slaughter cattle to that in fallen stock is higher in Japan than in the EU and Switzerland (23). He suggested that the relatively high BSE incidence in slaughter cattle in Japan might have been because sick or moribund cattle in Japan are more likely than in Europe to be sent to abattoirs for human consumption, without being investigated for BSE either on farm or in rendering plants (23).

Introduction of bovine spongiform encephalopathy into Japan, possibly in 1992 or earlier

The earliest born indigenous case was the 34th case; the animal was born in July 1992 (the 24th case also occurred in an animal born in 1992, but was atypical). Considering that cattle usually get infected within one year of birth (2), the BSE agent must have entered the animal feed chain in Japan around this year or earlier. Sugiura estimated that the BSE agent entered the animal feed chain most probably in 1992 or 1993 (with the probability of 17%), as a result of live cattle imports from the UK in the 1980s (22).

Two reports have been published by the MAFF BSE epidemiological study team. The first report, published in September 2003 (29), concluded that BSE most probably entered Japan through the importation of live cattle from the UK in the 1980s or MBM originating from Europe in the mid-1990s and denied the possibility of animal fat being the source of infection. The second report, published in December 2007 (33) before the 34th case was detected, concluded that the possible sources of infection were: animal fat and powdered animal fat imported from the Netherlands in 1995 and later; and rendered materials from a dairy cow imported from the UK and slaughtered in 1995.

Tsutsui *et al.* conducted a case-control study to test the association between milk replacers produced from a specific feed plant and a cluster of 13 BSE cases in cattle born between December 1995 and August 1996 (26). Their results indicated that the use of the milk replacers produced by a specific plant in Gunma prefecture, using tallow domestically produced and imported from the Netherlands, was associated with BSE infection. However, they did not rule out the possibility that the association of the calf concentrates with the BSE infection might have been masked by the use of specific milk replacers.

Two young cases

Of the 36 cases, two were rather young, being 21 and 23 months old. The animals, both Holstein steers, were born in October 2001 and January 2002, respectively, just a few months after the feed ban. Samples from these two young healthy Holstein steers yielded weakly positive primary ELISA (Platelia BSE) results. The ELISA positive sample was sent to the National Institute for Infectious Diseases for confirmation. The confirmatory diagnosis was made by Western blot only, which detected an accumulation of unusually small amounts of core fragments of PrP^{Sc} (PrP^{core}) (as low as 1/1,000 of PrP^{core} in a classical BSE case). Neither spongiform changes nor PrP deposition was found after histopathological and immunohistochemical examination (29). Attempts were made to transmit disease from these two animals by inoculating bovine PrP-overexpressing transgenic (TgBoPrP) mice with brain material. The transmission study was terminated in 2007 with none of these mice developing evidence of BSE infection, even with subsequent blind passages. The researchers involved in this study concluded that the amount of prion that the mice received would have been under the limit of sensitivity of the bioassay (32). They believed that these two infected animals, born immediately after the feed ban was enforced, were a result of cross-contamination of the remaining feed. Ozawa questions the results of the confirmatory diagnosis used in diagnosing these two cases, using the logic that if the animals had indeed been infected with BSE, they must have been exposed to large amounts of the BSE agent, and that if that were the case, there should have been other BSE cases detected amongst their cohorts (19).

Two atypical cases

Classical BSE is caused by a single major strain of the transmissible spongiform encephalopathy agent – BSE agent (7, 21). However, in recent years, two distinct forms of the disease have been described. These forms are phenotypically different from the previously identified classical BSE (3, 5). Western blot studies of the protease-resistant prion protein (PrPres) showed higher and lower molecular masses of unglycosylated PrPres in these two types (subsequently named H- and L-types, respectively) compared with classical type BSE (4). In L-type BSE, the most discriminant molecular feature is the lower level of diglycosylated PrPres (4, 5). Such atypical cases have now been identified in a number of countries, including Japan (8, 10, 20, 29). The origins and possible routes of transmission of H-type and L-type BSE are not known.

Of the 36 BSE cases detected by the end of March 2009 in Japan, two were atypical: both L-type cases. The results of the Western blot tests for these cases showed a lower concentration of the diglycosylated band, and a slightly lower molecular mass of the unglycosylated band of PrP^{Sc}, than would usually be seen with classical BSE. One of the animals was a Holstein steer aged 23 months (one of the young cases mentioned above), detected during active surveillance of healthy slaughtered animals (29). Considering the fact that most atypical BSE cases have been found in animals more than ten years old, the age of this Japanese atypical case was extremely young compared with other atypical cases (youngest atypical case in the world).

The other was a Wagyu cow born in 1992, aged 169 months, detected in an abattoir in 2006 during active surveillance. In this cow, plaques of PrP^{Sc} were detected in the brain by immunohistochemical analysis. At the level of the obex, the medulla oblongata was devoid of plaques of PrP^{Sc}, and a pathological phenotype similar to that of typical BSE specimens, with spongiform change and coarse granular/linear deposition of PrP^{Sc} was observed (8). Noting that the case is similar to an atypical case reported in Italy, Yoshikawa *et al.* did not rule out the possibility of the MBM imported from Italy between 1987 and 1993 being the source of infection (34), despite the conclusion by an OIE expert ad hoc group in 2003, based on the data available at that time, that there was no link identified between the Japanese and the Italian atypical cases (28).

Neither of these atypical cases showed clinical signs of BSE when detected, as is reported for most atypical cases in the rest of the world.

Prediction of the future bovine spongiform encephalopathy epidemic

The Prion Expert Committee of the FSC estimated that the annual number of cattle testing positive in Japan using a rapid BSE test would be fewer than 43 up to 1996; between 6 and 24 from 1996 to 2000; and between 3 and 14 from 2001 to 2005. (From this they estimated that the annual occurrence of BSE infection in animals < 21 months old would be 0.4 to 1.7 from July 2003 onward) (32). No further prediction has been made by the FSC since this report was published.

Using Bayes' theorem and Monte Carlo simulation, and assuming that dairy cattle born in and after 2002 are not infected, Sugiura *et al.* predicted that there would be zero to two BSE cases in the dairy cattle population in and after 2009 and that there would be no BSE-infected animals after 2013 (24, 25). Since the total feed ban was introduced in October 2001, audit inspections of feed mills and distributors have been conducted, and samples of domestic and imported feeds and animal by-products were analysed for contamination by ruminant protein, using microscopy, ELISA and polymerase chain reaction for species-specific proteins, with no samples of domestic and imported feed samples found contaminated with ruminant animal protein (11, 12, 13, 14, 15). These results, together with the fact that no BSE case has been detected in animals born after January 2002, indicate that the controls on feed implemented since October 2001 have been effective and that no animals are likely to have been exposed to contaminated feed except those born in early 2002, which might have been infected as a result of cross-contamination with the remaining contaminated feed. Consequently, the average age of BSE-infected animals has been increasing, as shown in Figure 6.

The detection in March 2008 of the 35th case, which was a Wagyu cow born in 2000, raises the possibility that some additional cases may be detected in Wagyu cattle in the future, as many Wagyu cattle born in that year remain alive.

Conclusion

The BSE epidemic in Japan has features both similar to and different from the BSE epidemic in the EU. Finding two young and two atypical cases among the total of 36 cases is unusual compared with the situation in the EU.

The higher incidence in dairy cattle than in beef cattle is similar to that in the EU. There is still controversy over whether or not the two young cases are indeed BSE cases, but this might never be solved because there is no sample left for further assay.

It is unknown how the most recent BSE case in a Wagyu beef cow born in 1992 was exposed to the BSE agent in Shimane prefecture in that year.

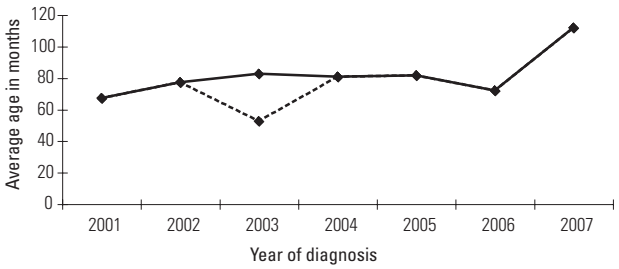


Fig. 6
Evolution of the mean age of bovine spongiform encephalopathy cases in Japan
Dotted line indicates the mean ages, including the two young cases detected in 2003, and the solid line indicates the mean ages without taking account of the two young cases

This is still one of the most sensitive food safety and animal health issues in Japan. The epidemiological features of the BSE epidemic in Japan, in particular the downward trend in the number of BSE cases detected in recent years (including fewer infected animals in the birth cohorts born in recent years) and the upward trend of the mean age of BSE cases, suggest that a review of BSE measures would be appropriate. Any review of BSE measures should follow a risk assessment (including a risk communication with all stakeholders, including consumers), taking account of these epidemiological features and ensuring that the guidelines provided by the World Health Organization and the OIE are followed.

Although the available evidence on atypical BSE cases in Japan and in the rest of the world in recent years does not seem to suggest that any changes should be made to the BSE measures currently in place, further research might be required to obtain more data on the aetiology and oral transmissibility of the disease to fully assess the animal and human health implications. ■

Caractéristiques épidémiologiques de l'épidémie d'encéphalopathie spongiforme bovine au Japon

K. Sugiura, T. Onodera & R. Bradley

Résumé
Le 10 septembre 2001, un premier cas d'encéphalopathie spongiforme bovine (ESB) a été confirmé au Japon chez une vache laitière âgée de cinq ans, née à Hokkaido et élevée dans la préfecture de Chiba. Suite à cet évènement, le Japon a relevé le niveau de surveillance de l'ESB. La surveillance exercée jusqu'à la fin du mois de mars 2009 a permis de détecter 35 autres cas, répartis en un nombre de cas nouveaux par an compris entre deux et dix, et ce jusqu'à la fin de l'année 2007. Le pic de l'épidémie a probablement eu lieu en 2006. La plupart des cas appartiennent à l'une au moins des catégories suivantes : bovins de race laitière ; bovins nés en 1996 et 2000 ; bovins nés à Hokkaido. A l'exception de deux cas catégorisés comme « atypiques », tous les autres étaient des cas classiques d'ESB. Les auteurs décrivent et analysent la structure démographique du cheptel bovin (races laitières et bovins de boucherie), les programmes de surveillance mis en place et les caractéristiques épidémiologiques de l'épidémie d'ESB au Japon.

Mots-clés
Encéphalopathie spongiforme bovine – Épidémiologie – Japon – Surveillance.
■

Características epidemiológicas de la encefalopatía espongiforme bovina en Japón

K. Sugiura, T. Onodera & R. Bradley

Resumen
El 10 de septiembre de 2001 se confirmó el primer caso de encefalopatía espongiforme bovina (EEB) en Japón, que afectó a una vaca lechera de cinco años nacida en Hokkaido y criada en la prefectura de Chiba. A partir de entonces se intensificó la vigilancia contra la enfermedad, con lo que a finales de marzo de 2009 se habían detectado otros 35 casos, a razón de entre dos y diez al año

hasta finales de 2007. La epidemia pareció llegar a su punto álgido en 2006. Los casos detectados afectan sobre todo a vacas lecheras, al ganado nacido en 1996 y 2000 y al nacido en Hokkaido. Dos de los casos resultaron corresponder a una EEB atípica, mientras que en todos los demás se observó la forma clásica de la enfermedad. Los autores describen y analizan la estructura demográfica del ganado lechero y cárnico, los programas de vigilancia en vigor y las características epidemiológicas de la EEB en Japón.

Palabras clave
Encefalopatía espongiforme bovina – Epidemiología – Japón – Vigilancia.

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