

# **Exposure of the human population to BSE infectivity over the course of the BSE epidemic in Great Britain and the impact of changes to the Over Thirty Month Rule.**

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## **Abstract**

The Over Thirty Month Rule (OTMR) was introduced in 1996 as a measure to reduce the exposure of the population to the BSE agent by limiting the age of animals that could be slaughtered for food. This study was carried out as part of the process of reviewing the OTMR that is currently being carried out by the UK Food Standards Agency (FSA).

We review data on the infectivity of bovine tissues, the development of infectivity through the incubation period and the cattle to human species barrier. All the possible routes by which infective material could be included in food for human consumption, including contamination with infected tissues in the abattoir, embolism following slaughter, dorsal root ganglia in meat, mechanically recovered meat and failure of SRM controls are evaluated over the period of the BSE epidemic.

It is estimated that the exposure from one fully infected animal slaughtered for food at the present time would be about 27 bovine oral ID<sub>50</sub> units, this compares to a peak value of about 1900 early in the epidemic. At present the exposure is primarily due to dorsal root ganglia, but over the course of the epidemic the main contributor to total infectivity is estimated to have been mechanically recovered meat (MRM). However, we also show that the potential exposure to infectivity due to MRM in any one meal would have been very small, casting doubt on whether this could have been an effective source of infection.

By combining these estimates with estimates of numbers of infected animals by incubation period and year it is estimated that a total of some 54 million bovine oral ID<sub>50</sub> units would have been consumed from 1980 to date, reaching a peak of about 11 million units in 1993, but falling rapidly following the introduction of SBO ban and the OTMR. In 2001 it is estimated that the exposure for the whole UK population is only 2.5 bovine oral ID<sub>50</sub> units. The study has helped demonstrate that current exposure to BSE infective material in the UK is very low, and would remain very low if the OTMR was amended or abolished.

## **1. Introduction**

Bovine Spongiform Encephalopathy (BSE) was first diagnosed in Great Britain in 1986 (Wells *et al*, 1987). From then to the end of 2002 there have been more than 178,000 confirmed clinical cases; these peaked in 1992 with 36,680 confirmed cases, and then fell rapidly due to the effect of the ruminant feed ban of 1988. The number of clinical cases continues to fall year on year, to less than 500 in 2002 (DEFRA, 2003). In March 1996 the Spongiform Encephalopathy Advisory Committee (SEAC) announced that a new variant of the fatal neurological disease in humans, variant Creutzfeldt-Jakob Disease (vCJD), had been

identified and that the most likely explanation was exposure to the infective agent of BSE through consumption of beef prior to the specified bovine offal ban in 1989. 122 cases of variant CJD have been confirmed up to February 2003, with an additional 8 probable cases still alive.

The Over Thirty Month Rule (OTMR) was introduced in 1996 as a measure to protect human health. It was designed to reduce the exposure of the population to the BSE agent. With two exceptions, meat from cattle aged over thirty months at slaughter is banned from sale for human consumption in the UK. The exceptions are firstly a few cattle with very low BSE risk in the beef assurance scheme, and secondly imports from some defined countries with low BSE risk. The OTMR does not affect prime beef cattle as most of these are finished and slaughtered at about 24 months. When other cattle (e.g. dairy cows or beef suckler cows) reach the end of their useful lives and are older than 30 months but would otherwise be fit for human consumption they are sold into the Over Thirty Month Scheme (OTMS). The animals are slaughtered in an abattoir approved for OTM slaughter, the complete carcass, including all the SRM and other offal, is sent for rendering and the resulting meat and bone meal is disposed of by incineration.

With the continuing decline of the BSE epidemic, the Food Standards Agency (FSA) initiated a review of the OTMR in 2002. Two groups were set up to advise on any changes to the rule, a Risk Assessment Group to advise on changes to the risk of exposure to the UK population and a Core Stakeholder Group to advise on whether or not the OTMR may be varied without unacceptable risk to consumers and if so to make recommendations on appropriate measures. If cattle older than 30 months were allowed to be used for food they would be subject to rapid BSE testing in line with EC rules. The work of the Risk Assessment group included two studies to assess the expected number of infected animals that could be slaughtered for food under alternative strategies for the OTMR (Ferguson & Donnelly, 2003; Arnold & Wilesmith, 2003) and a study to assess the likely exposure of people to infective material if an infected animal is slaughtered for food. This paper presents the results of this third study which have been used by Ferguson & Donnelly (2003) in their overall assessment of the risk impact of changes to the OTMR.

## **2. Data and Assumptions**

### **Infectivity of Bovine Tissues**

A key input to the risk assessment is the amount of infectivity present in the material to which people may be exposed. There are four factors that need to be assessed in order to determine the potential infectivity of any bovine tissue to humans:

1. The infectivity of central nervous system (CNS) tissues from an animal with clinical BSE to another bovine;
2. The relative infectivity of non-CNS tissues in an animal with BSE;
3. The development of infectivity through the incubation period of the disease; and
4. The cattle-human species barrier.

These have recently been reported in detail in Comer and Huntly (2003), and a summary is given below.

### ***Infectivity of CNS tissues from an animal with BSE***

Data from the “attack rate” experiment carried out by the Veterinary Laboratory Agency (VLA) in the UK, and its interpretation by the Scientific Steering Committee (SSC) of the European Commission (SSC, 2000) have been used to define a distribution for use in risk assessment studies. For this study it was assumed that the bovine oral infective dose is represented by a log normal distribution with a mean of 90 bovine oral ID<sub>50</sub>/g and a standard deviation of 150; in simulations, truncation was introduced by discarding values less than 10 or greater than 10<sup>3</sup>. The median value of the resultant distribution is about 50 bovine oral ID<sub>50</sub>/g. It will be necessary to update this distribution formally as more data become available from a second attack rate study using lower oral doses of infection that is still in progress. Initial results indicate that the bovine oral ID<sub>50</sub> estimated from the combined attack rate experiments is likely to be in the range from 2 - 10 ID<sub>50</sub>/g so that the proposed distribution will tend to overestimate exposures.

### ***Infectivity of non-CNS tissues from an animal with BSE***

The VLA’s pathogenesis study (Wells *et al.* 1998) was designed to test the infectivity of a range of tissues from experimentally infected cattle at various times through the incubation period of BSE. Calves from farms with no history of BSE were each dosed orally with 100g of pooled brain stems from 75 cases of BSE and then killed sequentially at approximately four month intervals from two to 40 months after inoculation. A wide range of tissues was collected for bioassay in mice and for detection of PrP-res. The pathogenesis study found presence of infectivity in the distal ileum of cattle killed from 6 months to 18 months post inoculation (p.i.) and from 36 to 40 months p.i., and in the brain, spinal cord, dorsal root ganglia and trigeminal ganglia from 32 to 40 months p.i. The earliest onset of clinical signs was noted in an animal 35 months p.i. (Wells *et al.* 1998).

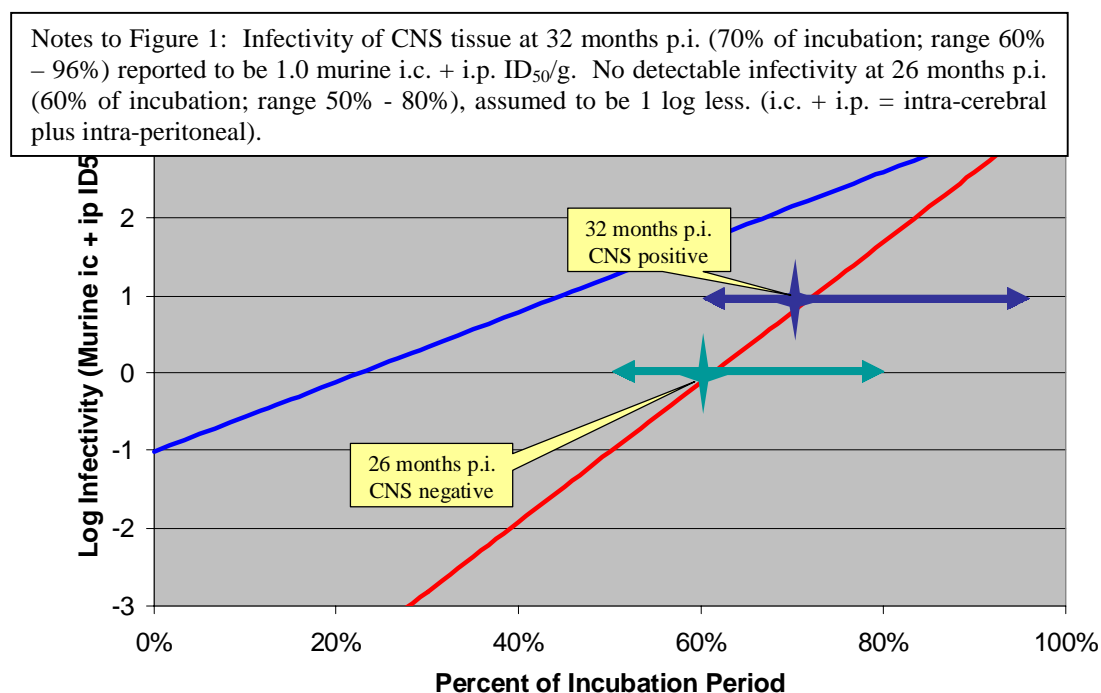
Subsequently, some tissues from the pathogenesis study have been tested by bioassay in cattle to increase the sensitivity of detection by avoiding the species barrier. It was shown that there is a 500 fold underestimation of the infectivity when titrated across the species barrier from cattle to mice (SSC, 2002a). Initial findings reported in SSC (2002a) confirmed the results from the mouse bioassay, and found no other positive tissues, despite the increased sensitivity. Tissues found to have infectivity were distal ileum at 6, 10 and 18 months p.i., and brain stem/spinal cord at 32 months p.i. (Tissues from the later kills at 36, 38 and 40 months p.i. were not tested in the cattle bioassay). More recently a positive result has also been found in the tonsil from the kill at 10 months p.i., but with an incubation period double that of the earlier results, indicating a very low titre (SEAC, 2002).

### ***Development of infectivity through incubation period***

Exponential growth of infectivity in CNS tissues has been demonstrated in other species (e.g. Beekes, *et al.* 1996) and is suggested as an appropriate model for the development of BSE infectivity in cattle in Cummins *et al.* (2001). Data from the pathogenesis study, as reported in SSC (2002b), are shown to be compatible with exponential growth of infectivity with a 2 month doubling time (see Figure 1). However, the sample sizes in the pathogenesis study were small, and there are insufficient data to provide any real confidence in this assumption. Cohen *et al.* (2001), in the BSE risk assessment for the US, adopted an exponential growth model with a one month doubling time, but provided no basis for the assumption.

For a 2 month doubling time the model indicates an infectivity 3 logs less than the clinical value at 70% of the incubation and 4.5 logs less than the clinical value at 50% of the incubation period (see Figure 1).

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### ***The cattle-human species barrier***

The infectivity of BSE for humans is believed to be lower than in cattle due to the species barrier: if the cattle–human species barrier was 100, it would mean that 100 times more infective material would be required in order to have a similar probability of infecting a human as a bovine.

The size of the species barrier between BSE in ruminants and BSE in humans (vCJD) is not known. The SSC (2000) suggested that a range from 10,000 to 10 be considered and that the worst case scenario of no species barrier be included (i.e. species barrier = 1), although available evidence (Raymond *et al*, 1997) indicates that values greater than 1 are more realistic. In DNV risk assessments, the cattle-human species barrier had been represented as 10, 100, 1000 or 10,000 with equal probabilities, and as 1 with 1% probability (DNV, 1997a). The results of this work suggest that the species barrier is more likely to be at the top end of this range.

### ***Total Infectivity in a Clinical Case***

The total infectivity in a clinical case of BSE is summarised in Table 1. The weights of the various tissues are taken from the LFRA (1997) report and the infectivity values are as discussed above, with the infectivity for whole brain taken to be 50 cattle oral ID<sub>50</sub>/g. It can be seen that 90% of the infectivity is associated with central and peripheral nervous system tissues, with about 10% associated with the distal ileum.

**Table 1: Infectivity in a Clinical Case of BSE (Bovine oral ID<sub>50</sub>)**

Tissue	Weight g/animal	Infectivity		%
		ID50/g	ID50/animal	
Brain	500	50	25000	60.2%
Spinal cord	200	50	10000	24.1%
Dorsal root ganglia	30	50	1500	3.6%
Trigeminal ganglia	20	50	1000	2.4%
Tonsil	50	0.005	0.25	0.0%
Distal ileum	800	5	4000	9.6%
<b>TOTAL</b>	<b>1600</b>		<b>41500</b>	

It could be argued that infectivity may be present in other tissues at some level below the limit of detection in the mouse or bovine bioassay. If it is assumed that this is the case, and that infectivity were present in all other tissues at a level an order of magnitude below that found for tonsil, which is 5 logs less than the infectivity in brain and spinal cord, then there would be an additional 270 bovine oral ID<sub>50</sub> units (less than 1% of the total), of which 90 would be from the edible meat. This theoretical possibility is not considered further in this study.

### **Routes of Exposure**

The possible routes of exposure through food to any infectivity present in the tissues identified above are considered below. Both the current exposure and exposures over the course of the epidemic have been considered. There have been many changes to legislative controls over the BSE epidemic; those that are thought to have had the main impact on exposure to infectivity in food are summarised in Table 2.

#### ***Brain***

There are three possible ways by which people could have been exposed to infectivity in brain: direct consumption or incorporation into meat products before this was banned; contamination of head meat and tongue; and embolism.

#### ***Contamination of Head Meat***

Meat cut from the head of the animal is of interest as there is a greater risk that this could have been contaminated with brain material from the slaughter wound at the time of slaughter or during transport of the head to specialist head boning facilities. If it was planned to remove the brain as well as take the head meat, the head meat would normally be removed before the head was split. However there was a period in 1989/90 when the brain may have been removed first as a result of commercial pressures due to SBO legislation that made the whole head SBO if it included the brain.

Before the use of head meat for human food was banned when the whole head (other than the tongue) was made SBM in March 1996, head meat would have been taken from most carcasses (Cooper & Bird, 2002-b). Head meat is a very lean meat and it was a valuable product. It was often used in burgers and mince as well as other meat products (DNV, 2002).

<b>Table 2: Legislative changes affecting human exposure</b>	
November 1989	The Bovine Offal (Prohibition ) Regulations 1989 (SI 1989 No 2061) came into force in England and Wales (January 1990 in Scotland). These banned the use of certain specified bovine offals (SBO) for human consumption. SBO was defined as the brain, spinal cord, spleen, thymus, tonsils and intestines of bovine animals slaughtered in the UK.
March 1992	The Bovine Offal Prohibition (Amendment) Regulations 1992 (SI 1992 No 306) implemented recommendations made by Advisory Committees to prohibit the use of the head after the skull is opened (effectively minimising risks of contamination of head meat by the process of brain removal) and the removal of the brain except in areas which are free at all times from any food intended for human consumption.
December 1995	The Specified Bovine Offal (Amendment) Order 1995 and the Export of Goods (Control) (Amendment Number 2) Order 1995 prohibited the use of the bovine vertebral column in the manufacture of all mechanically recovered meat and also in the production of some other products for human consumption.
March 1996	The Specified Bovine Material Order 1996 (SI 1996 No 963) extended the definition of SBO (subsequently known as Specified Bovine Material - SBM) to include the head (including the brain but excluding the tongue) of a bovine aged over six months old.
March 1996	The Beef (Emergency Control) Order 1996 prohibited the sale for human consumption of meat from any bovine animals aged over thirty months at the time of slaughter.
December 1997	The Beef Bones Regulations 1997 required the boning of all beef derived from cattle aged over 6 months at slaughter.
December 1999	The Beef Bones (Amendment) (England) Regulations 1999 lifted the ban on the sale of beef on the bone. Similar regulations came into force in other parts of the UK.

Cooper and Bird (2002-b) derived estimates for the amount of brain material removed with head meat from each bovine head for each of four legislative periods and these have been used here. They estimate that the mean amount of brain material removed with head meat increased from 1.65 g prior to 1989 to 1.83 g in 1990 as some brains were being removed before the head meat. This then reduced to 1.3 g as this practice and the use of pithing declined. The detailed values from Cooper & Bird are given in Table 3.

In March 1996 the complete bovine head, but excluding the tongue, was included in the definition of SRM (see Table 2). It is not known if the tongue could be contaminated with any CNS tissue during slaughter, but for the purposes of this assessment it will be assumed that between 1 and 10% of the infectivity removed with head meat could contaminate the tongue. Prior to 1996 this is assumed to be included in the infectivity in head meat.

**Table 3: Estimates of quantity of brain material removed with head meat**

Legislative period	Quantity of brain material removed with head meat from each bovine head (g)		
	Mean	Median	95% credible range
Period 1 Before November 1989	1.65	1.46	0.38 – 3.99
Period 2 November 1989 to May 1990	1.83	1.50	0.38 – 5.15
Period 3 June 1990 to February 1992	1.32	1.16	0.35 – 3.10
Period 4 March 1992 to March 1996	1.28	1.15	0.35 – 2.93

Source: Cooper and Bird (2002-b)

### ***Brain Embolism***

Most cattle in the UK are stunned with a penetrating captive bolt gun (CBG) prior to slaughter. The heart continues pumping for some minutes following stunning, during which time the animal is killed by bleeding out by severing the main blood vessels to the head. Any brain tissues (emboli) dislodged by the action of the CBG could be within the blood so released and/or trapped within the heart and the lung. Particles smaller than the diameter of pulmonary capillaries could theoretically be disseminated throughout the body.

A number of studies have been undertaken to try and determine the extent of contamination that could occur during stunning and slaughter, some of which are still on-going (Anil *et al*, 1999 & 2002; Love *et al*, 2000; Daly *et al*, 2002; Lücker *et al*, 2002). Anil (personal communication) has estimated that some degree of embolism occurs in 4% of slaughters using a CBG without pithing. Anil *et al* (2002) estimate that “an average of 10g of brain tissue can be dislodged” and that “This figure represents, approximately, the maximum potential load of prion contaminated tissue that might be disseminated haematogenously as a result of using a CBG on an animal with BSE.” They also note that “showers of embolic brain tissue include many fragments of sufficiently small size to be capable, in principle, of passing through the pulmonary capillary bed.” In the past pithing was used in some abattoirs and may have resulted in a greater risk of embolism due to additional disruption of brain tissue. This possible greater historical exposure has not been taken into account in this study as the risks from embolism have been found to be very low.

At present no quantification of the size range of the brain particles in the blood has been carried out. However, it is understood that about 50% of the particles are small (< 5micron) and may be able to pass through the capillary bed, but that these would make up less than 10% of the total mass of material (personal communication, Professor S. Love).

The interim results from these studies do not yet allow for reliable quantification that can be used in a risk assessment and we are not aware of other data to support these findings. For the purpose of this risk assessment we therefore propose to make a set of assumptions regarding embolism that can be tested against further results when these become available.

1. Embolism occurs in 4% of slaughters using a penetrating CBG.
2. Between 1 to 10g of brain material may be dislodged during stunning and may be available to pass into the blood.
3. Between 1 and 10% by mass of the emboli released into the blood pass through the capillary bed and remain in the meat.
4. Any possible contamination of the small amount of bovine blood used for food products and the use of heart for human food is assumed to be included within assumption 3.

### ***Brain Consumption***

The Ruminant Products Audit (LFRA, 1997) found that about 9% of independent butchers sold bovine brains directly for human consumption. From this LFRA estimated that some 200,000 brains would have been consumed in 1980, reducing to about 100,000 in 1989 when the SBO legislation came into effect. None of the respondents to the survey reported the use of brains in meat products. These numbers would represent about 5% of the total cattle slaughtered for food in 1980, reducing to about 3% in 1989. These estimates indicate a potentially high exposure to infectivity that is confounded by the fact that none of the victims of vCJD have reported eating bovine brain (CJD Surveillance Unit, 2001). Cohen *et al* (2001) assume that 1% of brains are consumed in the US, but provide no basis for this assumption.

The report of the investigation into the North Leicestershire cluster of variant CJD (Bryant and Monk, 2001) found that four of the five vCJD cases in this area bought meat from one of two butchers, both of whom removed the brain as a regular activity. In a case control study 22 butchers were identified, of which four (18%) split the heads and removed the brain. The two butchers linked to the cases each processed between 3 and 5 carcasses a week. If they removed brains from 50% of their carcasses this would be 200 brains per year, a significant fraction of the national total for one small area. The report gives no details as to how the brains were used, but there was some anecdotal evidence of use in meat products such as cheap burgers (Bryant, personal communication).

It is probable that brains for consumption would mainly have been extracted from prime beef animals, which would be less likely to have significant levels of infectivity. (Prime beef animals will normally have been slaughtered from 24 to 30 months of age). This likelihood is reinforced by the fact that the brains were reported as being sold by the independent butchers who would be less likely to use adult animals. There is no report of brains being harvested at abattoirs handling cull cows.

For this assessment, the estimates of brain consumption from LFRA (1997) have been used, but with an additional assumption that 99% of these brains would have been from prime beef animals. This assumption will reduce the average exposure, but there would be a relatively high chance (6%) that an infected brain would be used for human consumption in the event that an animal reaches clinical stage within about 2 years. However, there were relatively few BSE cases in animals less than 2 years old, 0.5% of cases in 1989 reducing to less than 0.1% by 1992 and to zero by 1997. Overall 0.1 % of BSE cases were less than 2 years old (DEFRA, BSE Progress Report, December 2001, Table 2).

### ***Spinal Cord***

Infectivity from the spinal cord may be transferred to food in a number of ways; contamination of the carcass when it is split following slaughter; failure of SRM controls;

and, in the past, possible direct use in food products and mechanically recovered meat (MRM). MRM is considered under dorsal root ganglia.

### ***Contamination of Carcass***

During dressing, beef carcasses are normally split down the vertebral column using a band or reciprocating saw or rarely by a circular saw. Band saws have a fine blade and teeth and the spinal cord may remain visually more or less intact. Reciprocating saws are thicker with larger teeth and may mutilate the spinal cord more than band saws due to the wider cut. The spinal cord is removed as SRM. Inspections at abattoirs in Great Britain have shown that the removal of spinal cord since 1996 is virtually 100% (see below). However, there will be some contamination of the cut surfaces of the carcass with macerated spinal cord as a result of the action of the saw (Helps *et al*, 2002). The degree of contamination with spinal cord is being investigated in two FSA/EC funded projects.

The studies, which have been carried out both in a controlled research abattoir and at commercial abattoirs throughout the EC, have confirmed that spinal cord is deposited on carcasses and that normal washing procedures do not remove the contamination. Knight (2002) reported that typically 25-50mg of CNS material was found on each of the medial carcass surfaces, particularly along the cut vertebral surface with less in the body cavity. It was also reported that considerable variation was found in carcass contamination levels between abattoirs, both in the same country and between countries. A variation of about a factor of 5 between the lowest and highest values was found over the 7 UK abattoirs tested (personal communication H. Anil).

One of the objectives of the study had been to assess contamination in the boning hall and in retail packs as well as on the carcass. At present there are only a few preliminary results available from one abattoir (in France). This indicated a significant reduction in contamination in the boning hall and only occasional positive results in retail packs and at a much lower level (personal communication H. Anil).

The following assumptions have been made on carcass contamination for the risk assessment:

1. A total of 100mg of CNS tissue present on the cut surfaces of each carcass following splitting and washing.
2. 1% of the contamination on the carcass will be transferred to meat during boning and packing operations.

### ***Failure of SRM Controls***

Slaughterhouses handling red meats are inspected regularly by the State Veterinary Service to ensure full compliance with the SRM controls. Over the three years 1998 to 2000 there were a total of more than 12,000 inspections, representing about 10 inspections per plant per year. Assuming that an inspection would cover one day's production, about 4% of the total number of cattle slaughtered would be inspected. Over the three years there were a total of 7 unsatisfactory reports relating to inadequate separation from materials fit for human consumption, of which only one involved a section of spinal cord left in place. This would represent a failure frequency of 1 in 240,000.

In addition to the SVS inspections, carcasses are routinely inspected and checked both by abattoir staff and by the MHS inspectors at the plant. There is therefore good confidence that there is a very high degree of compliance with the SRM controls. If some spinal cord were to be left on a carcass this would increase the risk of contamination, but it is unlikely that it

would remain un-noticed and be included in a food product. It is assumed that 10% of any infectivity left on the carcass would be consumed.

Failure of SRM controls after 1996 are represented by 5% of the spinal cord being left on the carcass with a frequency of 1 in 240,000, with 10% of this being consumed. Before 1996, inclusion of spinal cord due to failures of SRM controls is included in the estimates for MRM (see below).

### ***Dorsal Root Ganglia***

Dorsal root ganglia (DRG) are peripheral nervous system tissues located within the vertebral column and shown in the pathogenesis experiment to have similar levels of infectivity as CNS tissues (Wells *et al*, 1998). The potential exposure to infectivity associated with DRG has been assessed both for the UK and for the Republic of Ireland (DNV, 1997b & 2001).

There are two DRG associated with each of the 31 vertebrae along the vertebral column, one on each side. Each DRG weighs about 0.5 g, so that there is a total weight of about 31 g of DRG in one carcass. They would not generally be removed with the spinal cord but remain attached to the vertebral column. If an animal with late stage infection were to be slaughtered for food there is the potential for exposure to infectivity if either the meat is boned and some DRG is cut away with the meat, or when a cut of meat including part of the vertebral column is cooked and eaten. In the past there was also the possibility the bones would have been used to produce mechanically recovered meat (MRM).

### ***Exposure to DRG in Meat***

The potential for exposure to any infectivity present in DRG will depend on how the cuts of meat along the vertebral column are used. There are only two cuts of meat that are routinely sold with parts of the vertebral column, and so which may contain DRG; rib of beef and T-bone steaks. The section of the carcass used for rib roasts has four vertebrae. This may be sold as rib on the bone, but is also prepared as a boneless rolled joint or as steaks. When sold bone-in, it is common for the bone of the vertebral column to be removed, and to leave only the rib bone. This would remove the DRG. T-bones are cut from the sirloin which includes 9 of the vertebrae. The sirloin section is used to produce both fillet steak and sirloin steak, both boneless, and also T-bone steaks.

Information from the Meat and Livestock Commission indicates that sales of bone-in beef have gradually increased since the beef on the bone ban was lifted in 1999, but are still well below the pre 1997 level. Total sales of bone-in beef that could include the vertebral column (essentially rib of beef and T-bone steaks) are estimated to be 10,000 tonnes in 2001 as compared with 28,000 tonnes in 1997. The data also indicate that few of the multiple restaurant chains list T-bones, so that the proportion of bone-in sales from T-bones would be less than in 1997.

Of the 10,000 tonnes of bone-in sales we assume that 500 tonnes is T-bones, which would be equivalent to about 50,000 carcasses. 6,650 tonnes of retail ribs are sold at about 7kg each, representing about 475,000 carcasses, plus 2,850 tonnes of catering ribs at about 8kg each, representing about 178,000 carcasses. The total number of prime beef animals slaughtered for food in 2001 was 2.073 million and some 38% of the meat supply was imported. Thus about 1.5% of the UK production is used for T-bones, 14% for retail fore-ribs and 5% for catering ribs. As indicated above, it is common for the vertebral column to be removed from the rib joint. No new data are available so it has been assumed that the data from DNV (1997) would

still apply (30% of retail ribs and 10% of catering ribs sold with vertebral column), although it is likely that a higher proportion would now have the vertebral column removed.

**Meat sold off the bone:** Some limited trials have been carried out to examine whether the DRG is likely to be removed when meat is cut from the bone by the butcher or whether it would remain with the back bone (Dore, 2001). In this study, the vertebral columns removed from four beef carcasses were examined for the presence or absence of DRGs. They reported that the only nerve tissues remaining were spinal nerve roots at each intervertebral foramen, associated with which were the DRGs, easily discernible as small brown masses. In all the segments examined DRGs remained on the vertebral column and were not cut away with the meat. The only exception to this was the DRG of the first cervical spinal nerve. This nerve emerges through the lateral vertebral foramen of the atlas on each side. Its DRG lies towards the outside of this foramen. In one instance a portion of this ganglion was removed.

These limited data suggest that about 1 in 248 (0.4%) of DRG may be cut away with the meat when the bone is removed. Whilst this is useful to confirm the likely range of values, it was only a limited trial and does not provide robust evidence. It is therefore proposed to assume that 1% of DRG would be cut away with the meat when the bone is removed. This is in line with the assumption made in DNV (1997), and should err on the side of caution.

In commercial cutting plant, which operate at high throughput, DRG may be less likely to be removed than in small butcher shops, where the butcher may have more time to cut residual tissue from the bone for inclusion in mince etc. This effect has not been included.

**Meat sold on the bone:** For meat sold on the bone, it is necessary to estimate how much of the DRG contained within the vertebral column of a cut such as a T-bone steak would be consumed. There are no data for this and it would be difficult to determine with any certainty. In DNV (1997) it was considered that the DRG would not normally be eaten and, following discussions with the meat industry and SEAC, it was assumed that any DRG present would be consumed 5% of the time. A worst case of the DRG being consumed 100% of the time was also considered.

This was reconsidered in the study for Ireland (DNV, 2001) where some members of the project steering group considered that 5% was low, as once the meat was cooked the DRG would come away from the bone easily. In addition, there is also the possibility that the bones would be used to make stock. In this situation it is more likely that all tissues would be removed from the bone.

For this assessment it is proposed to use the same distribution of values as that used in DNV (2001), with a normal distribution having a 95 percentile range from 5% to 95%.

### ***Mechanically Recovered Meat***

MRM is produced by subjecting the bones from which the meat has been cut to high pressure to extract most of the remaining edible tissue. Beef MRM was produced throughout the main period of the BSE epidemic up to December 1995 and the bovine vertebral column would normally have been included when this had been boned. The use of the vertebral column raises the likelihood that MRM would be contaminated with any spinal cord not removed and also with dorsal root ganglia (DNV, 2002; Cooper & Bird, 2002a; LFRA, 1997).

As indicated above, when the vertebral column is boned most (assumed to be 99%) of the DRG would remain with the bone. It is likely that the MRM process would remove most of

this tissue so that it would be included in the MRM product; Cooper & Bird (2002a) assume 90% and this value is adopted here.

LFRA (1997) estimated the amount of spinal cord likely to be included in MRM, and this was reviewed and updated by Cooper & Bird. They developed an event tree to estimate the amount of spinal cord recovered per carcass as a mean of 3.3 g (median 2.3 g; 95% credible interval 0.2 – 12.0 g) before the SBO legislation and mean 1.5 g (median 0.6 g; 95% credible interval 0.02 – 8.3 g) after it.

Cooper & Bird (2002a) estimated the production of MRM as increasing from about 4000 tonnes per year in 1980 to about 5000 tonnes per year in 1987, and then falling rapidly in 1988 and 89 with the initial concerns about BSE. Production then started to increase again slowly, but only reaching about 2000 tonnes per year in 1995 before MRM production was effectively halted by legislation banning the use of bovine vertebral column in the manufacture of MRM. These estimates have been used here, although DNV (2002) were not able to verify this variation in production. The MRM production figures indicate that bones from about 12% of animals slaughtered would have been used for MRM production in 1980 increasing to 20% in 1987. Cooper and Bird (2002a) also report that vertebral columns for use in MRM were preferentially sourced from prime beef animals. DNV (2002) found no evidence for this and in this assessment we assume that there is no age selection bias for use in MRM.

MRM was widely used in economy burgers and frozen mince (DNV, 2002).

### *Tonsil*

Recent results from the pathogenesis experiment using the bovine bioassay have shown evidence of infectivity in palatine tonsil. One of five cattle that received pooled palatine tonsil, taken 10 months after experimental inoculation with BSE, has shown clinical evidence of the onset of BSE at 45 months post-inoculation (SEAC, 2002). At the time of reporting the 4 remaining animals in the group are still alive without evidence of clinical signs of BSE and there have been as yet no other positive results from tonsil sampled at 6, 18 or 26 months after inoculation.

The long incubation period in the positive test animal suggests that the infectivity titre would be very low, estimated to be about  $10^1$  bovine i/c ID<sub>50</sub>/g (personal communication, Dr D. Matthews, VLA). In considering the significance of the result SEAC noted that tonsil is SRM from 6 months of age in the UK and that although the quantity of tonsil tissue that may be attached to tongues is not known, it is likely, at most, to be small. The FSA have initiated a study to assess the potential risk of exposure to BSE infectivity in tonsil but this is still on going.

For this assessment, the maximum likely exposure to infectivity in tonsil has been assessed by assuming:

- The infectivity in tonsil is  $10^1$  bovine i/c ID<sub>50</sub>/g;
- 1 in 20 tongues may be contaminated with tonsil tissue, in which
- 10% of the infectivity in the tonsil would remain with the tongue and be consumed.

### ***Distal Ileum and Other SRM***

The only other bovine tissue that is known to contain infectivity is the distal ileum. This is SRM together with the rest of the intestine and mesentery. It is not difficult to remove and most unlikely now to be used in any food products, though historically, before the SBO ban in 1989, bovine intestine from UK cattle was used for the manufacture of natural sausage casings, surgical catgut and strings for musical instruments and racquets. The potential risk of exposure from other SRMs is thought to be minimal, taking account of both the low level of infectivity and the high compliance with the SRM controls, and have not been considered further.

## **3. Results**

The potential exposure from one undiagnosed infected bovine slaughtered for food has been assessed by combining the various routes of exposure for each year from 1980 to 2001. The median exposure, in terms of bovine oral ID<sub>50</sub> units is shown in Figure 2. From 1980 to 1990 the results are shown separately for adult (over 30 months old) animals and prime beef. The difference between these is that it has been assumed that most (99%) of brains consumed would be from prime stock. In Figure 3 the same results for the prime animals are shown on a logarithmic scale together with the 95 percentile range. The contributions from the main sources of exposure are then shown in Figures 4 & 5, for adult and prime animals respectively.

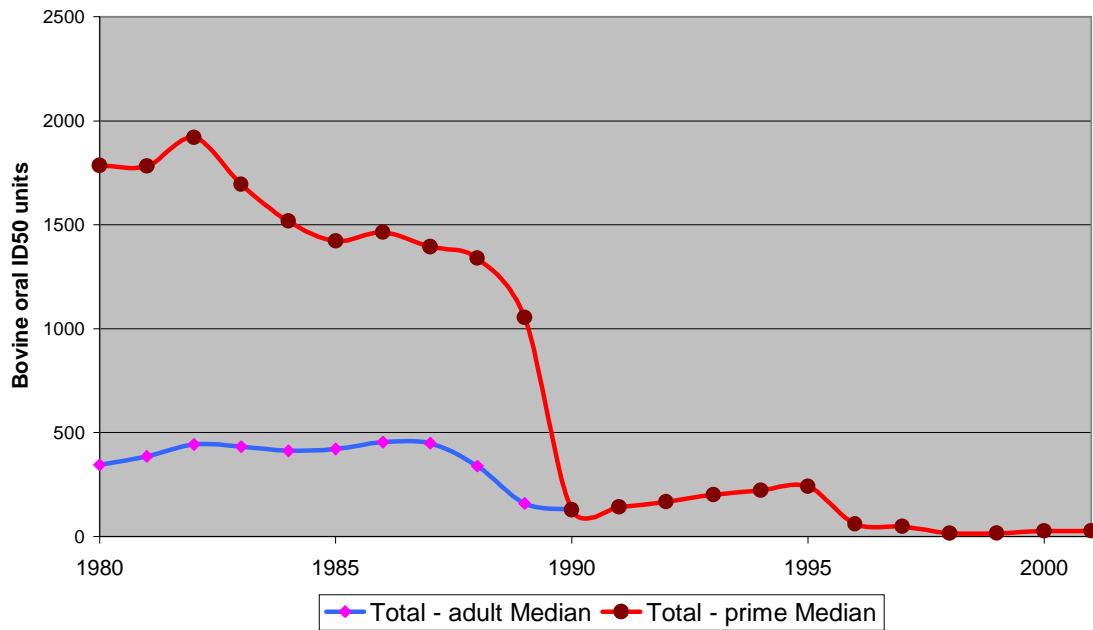
The results of the exposure assessment have been combined with the results from the back calculation model of the BSE Epidemic in Ferguson and Donnelly (2003). This model has been extended *inter alia* to make use of data from the Cattle Tracing Scheme and the results of proactive BSE testing in the UK. Ferguson and Donnelly (2003) present a detailed sensitivity assessment for different parameters and a range of results for different OTMR options. Here, we present only results from the base case which assumes 100% test sensitivity in the last 3 months of the incubation period and that all differential slaughter also occurs in the last 3 months of the incubation period. The results are shown in Figure 6, which shows the estimate of the total number of bovine oral ID<sub>50</sub> units consumed each year by the population of Great Britain from 1980 to 2009 for this scenario (Northern Ireland is being assessed separately). This is presented on a log scale in order to include the wide range of values predicted. Finally, the overall contributions from the different exposure routes considered are shown in Figure 7.

## **4. Discussion**

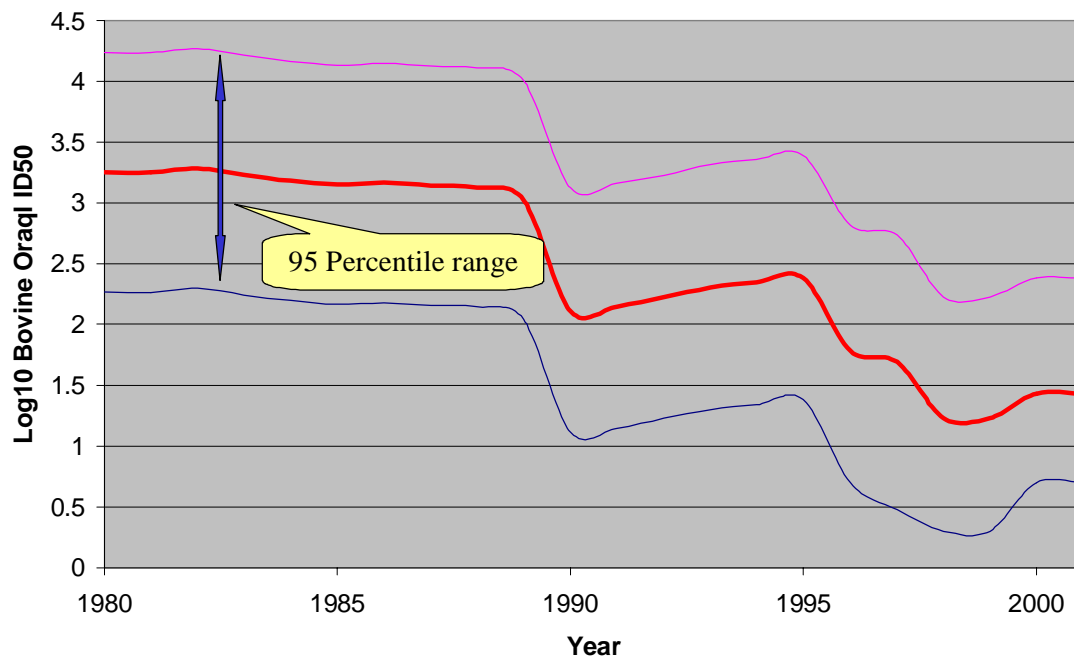
The results show that the maximum exposure from one infected animal would have been in 1982 with a median value of 1900 bovine oral ID<sub>50</sub> units (95 percentile range 200 – 18,500) for prime beef animals (440 bovine oral ID<sub>50</sub> units for adult animals). By the peak of the epidemic in 1992 the exposure from one infected animal is predicted to have reduced to 170 bovine oral ID<sub>50</sub> units (95 percentile range 17 – 1700), due mainly to the effect of the SBO regulations in 1989 banning the use of brain for human consumption and the reduction in MRM production. By 2001, the exposure is estimated to have reduced further to 27 bovine oral ID<sub>50</sub> units (95 percentile range 5 – 240). If all the meat was taken from the bone (as would happen for any animals older than 30 months if the OTM rule was changed) the exposure is estimated to be slightly less at 19 bovine oral ID<sub>50</sub> units. The reduction in

infectivity from an infected animal in 1998/99 (see Figure 3) was due to the implementation of the beef on the bone ban in December 1997 that was lifted in December 1999.

**Figure 2: Exposure from one Fully Infected Bovine Slaughtered for Food**



**Figure 3: Exposure from one Fully Infected Bovine (Prime Beef) Slaughtered for Food: Log plot showing 95 percentile range**



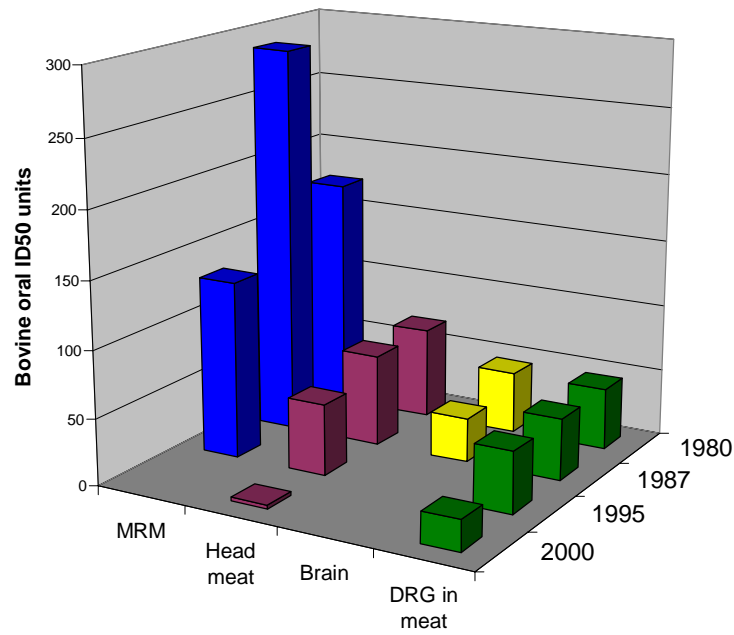
From Figure 2 it may seem that prime cattle pose a larger risk. In fact, as is shown below, there will be very few prime animals that would enter the food chain late in the incubation period. If that did happen prior to the SBO ban in 1989 then the exposure potential would have been high. This is well illustrated in Figure 5. From Figure 4 it can be seen that for the older animals, those more likely to be slaughtered for food with significant infectivity, over the main period of the BSE epidemic the exposure is dominated by infectivity in MRM. MRM represents more than 50% of the exposure until the use of bovine vertebral column in MRM production was stopped in 1995. The remainder of the exposure was due to head meat, DRG in meat and brain.

At the present time more than 90% of the exposure is due to DRG, whether in beef on the bone or from residual amounts left on boned meat. The only other significant contributor is estimated to be possible contamination of the tongue with CNS tissue following slaughter. There is little data available on this, but the assumptions made are thought to be highly pessimistic. The results show negligible exposure to infectivity from possible brain emboli, carcass contamination, failure of SRM controls or infectivity in tonsil.

The total quantity of bovine oral ID<sub>50</sub> units estimated to have been consumed by the population of Great Britain by year is shown in Figure 6. The exposure increases rapidly from the early 1980s, exceeding 1 million bovine oral ID<sub>50</sub> units in 1986 and reaching a peak of about 11 million in 1993. The reduction in infectivity per carcass due to the SBO ban in 1989 as seen in Figure 3 is masked here by the increase in the numbers of infected animals slaughtered. The exposure reduces from 1993, falling rapidly by 3 logs from 1996 to 1997 following the introduction of the OTMR, reducing to 200 bovine oral ID<sub>50</sub> units in 1997 and then to 19 in 1998. Exposure in 2001 is estimated to be only 2.5 bovine oral ID<sub>50</sub> units for the whole GB population and is predicted to fall to little over 1 bovine oral ID<sub>50</sub> units by 2004. For this set of assumptions, it is estimated that a total of 54 million bovine oral ID<sub>50</sub> units entered the human food chain from 1980 to 2001, 99.4% of which were from animals aged older than 30 months.

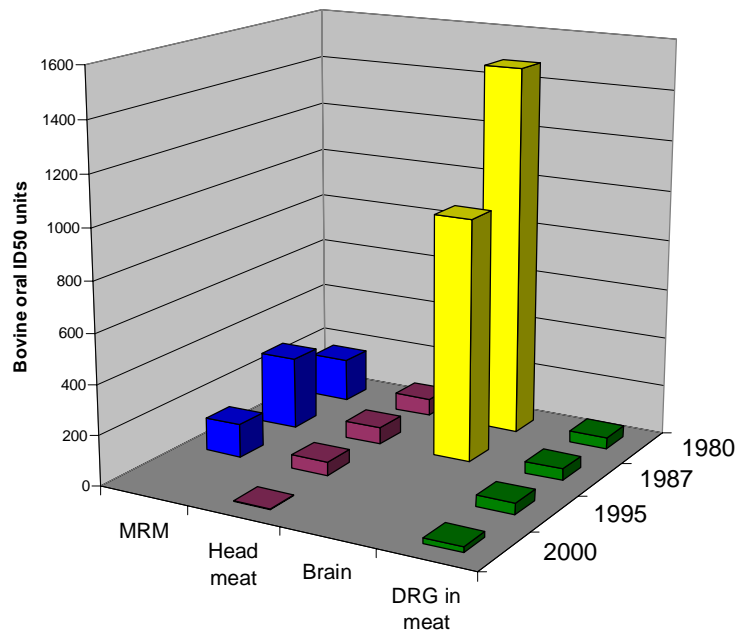
**Figure 4: Contributions to Total Exposure – Adult Beef**

**One fully infected bovine slaughtered for food**

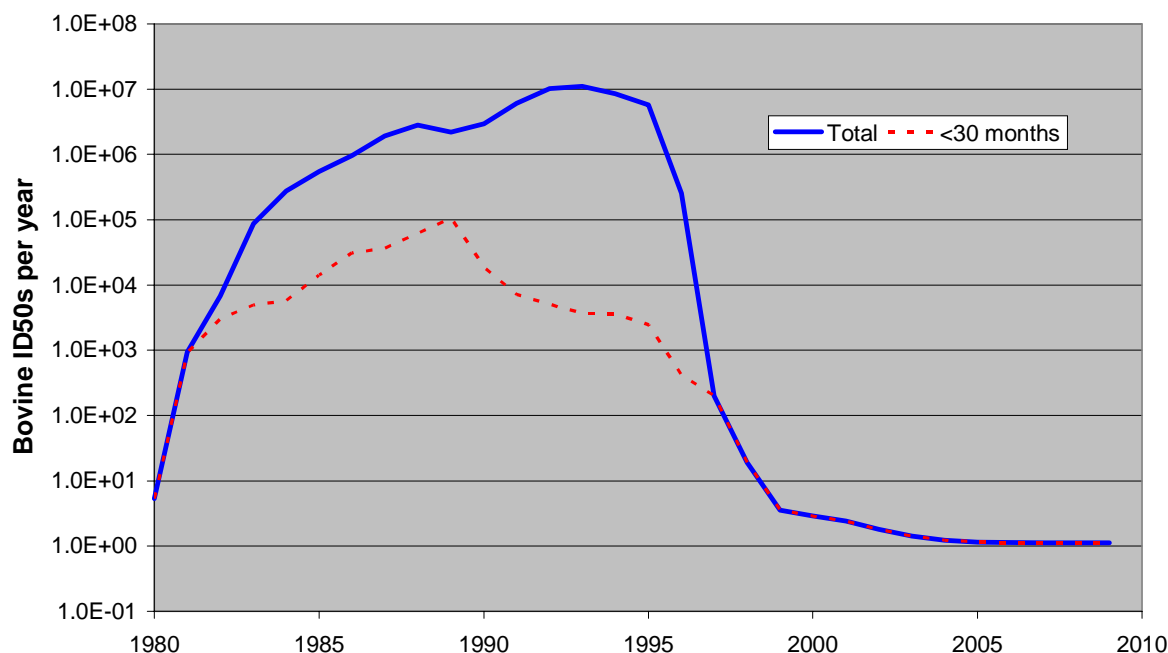


**Figure 5: Contributions to Total Exposure – Prime Beef**

**One fully infected bovine slaughtered for food**



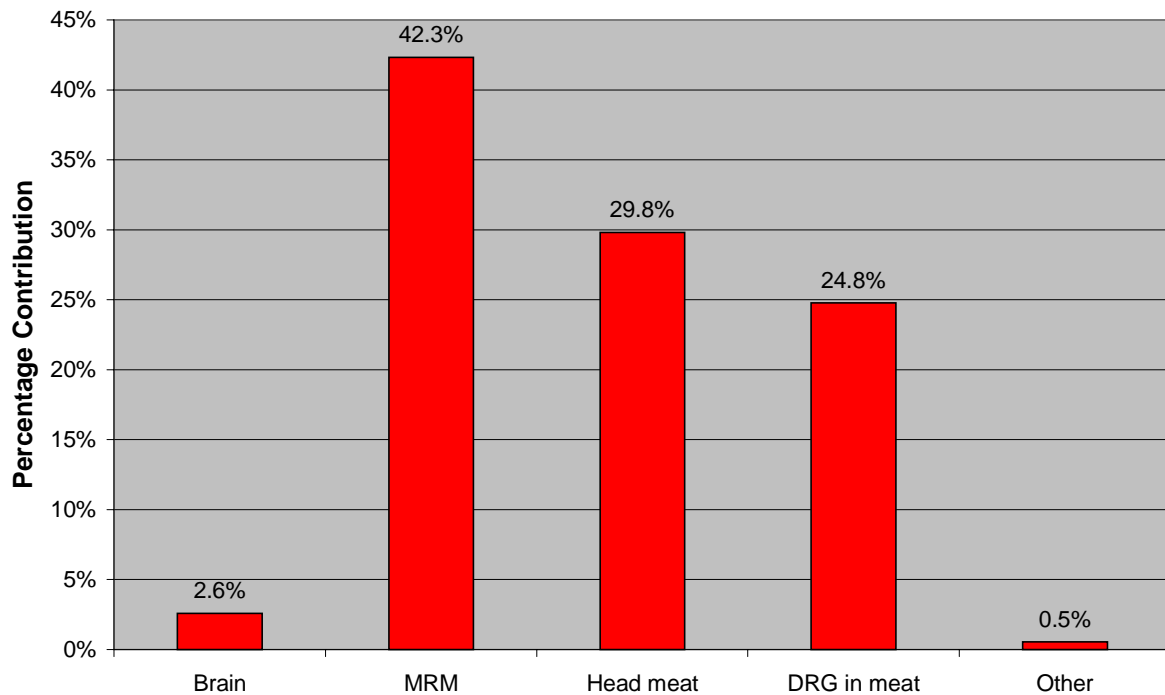
**Figure 6: Bovine Infectious units entering Food Supply by Year (GB)**



122 cases of variant CJD have been confirmed in the UK up to February 2003 with recent data indicating that there has been a slow down in the numbers of cases (Andrews *et al*, 2003). However, at the present time all cases in which the *PrP* genotype has been examined (the majority) encode methionine/methionine at codon 129. The frequency of this polymorphism in the general population is just under 40% and it is as yet unknown if any cases will arise in the future, or with what frequency, in the other genotypes. The maximum size of the vCJD epidemic in the UK is now being put at no more than 7,000 deaths (Ghani *et al*, 2003) with this upper estimate further reduced to 5000 in Ferguson and Donnelly (2003). The exposure estimates presented here allow an estimate to be made of the cattle to human species barrier. The results do not indicate the exposure for any one person, only the exposure for the total population. Using this gives an indication that the species barrier for methionine homozygous individuals may be of the order of 4,000, towards the top end of the range that has been assumed in risk assessment studies.

From Figure 7 it can be seen that MRM contributed 42% of the infectivity consumed over the whole epidemic, with head meat and DRG in meat contributing 30% and 25% respectively. MRM was used primarily in the cheaper range of burgers and economy mince, including use in institution cooking (DNV, 2002). Head meat was used in a wider range of products including quality mince and exposure to DRG would have been through the more expensive meat cuts. This result differs from that of Cooper and Bird (2002, b) who found that head meat accounted for 60% of the combined exposure of MRM and head meat. This difference may be because Cooper and Bird assume that MRM was produced mainly from prime beef, and thus lower risk, animals, whereas we have found no evidence for this (DNV, 2002).

**Figure 7: Contribution from Main Sources of Infectivity into Food, 1980 - 2001**



Direct consumption of brain is estimated to have contributed only 2.5% of the total. However, this is dependent on the assumption that 99% of brains consumed would have been from prime beef animals. If 5% of consumption was from older cattle the exposure from brains would increase from 2.5% to 10%; and if 10% of consumption was from older cattle the exposure from brains would increase further to 18%.

This overall distribution of the sources of infectivity may be misleading as the infectivity present in both MRM and head meat is likely to be dispersed, whereas consumption of brain would lead to high individual exposures. Some examples of exposures for typical “meals” are shown in Table 4, assuming that the tissues were from a) an animal close to clinical onset, or b) an animal 6 months prior to onset. From this it can be seen that the amount of infectivity consumed per meal from burgers or mince containing MRM or head meat would have been low, especially if the cattle human species barrier is greater than 1000. In addition, the exposure to infectivity in meals containing MRM would have been significantly less than this as, even at the height of the epidemic, only a small proportion of the animals used to make a batch of MRM would have had significant levels of infectivity. In order to consume high individual doses it would be necessary to consume a quantity of brain, either directly or included in a meat product. This indicates that it is unlikely that MRM would have been an effective source of infection, despite being the main route of infectivity into the food chain.

**Table 4: Typical exposures from individual meals  
(Bovine oral ID<sub>50</sub> units)**

	<b>Meal</b>	<b>Animal close to onset</b>	<b>6 months before onset</b>
1	250g burger with 10% inclusion of MRM (DNV, 2002), made from an infected animal (no additional dilution). Infectivity in MRM 0.25 bovine oral ID <sub>50</sub> /g	6	0.8
2	250g burger with MRM, including a single DRG	25	3
3	250g burger with 50% inclusion of head meat (DNV, 2002) with contamination from infected animal. Infectivity in head meat 0.04 bovine oral ID <sub>50</sub> /g	5	0.6
4	250g of bovine brain	12,500	1,600
5	250g burger or mince with 10% inclusion of bovine brain	1,250	160

The results presented here clearly show the effectiveness of the OTMR as a measure to protect human health when it was first introduced. Exposure of the UK population fell from 250,000 bovine oral ID<sub>50</sub> units in 1996 to 19 in 1998. With the ongoing reduction in BSE cases the current exposure is very low. Ferguson and Donnelly (2003) show that replacing the OTMR with BSE testing will result in a marginal increase in risk, but the level of exposure remains very low.

The low level of predicted exposure if an animal with significant infectivity were to be slaughtered for food at the present time is a testament to the effectiveness of the current SRM controls.

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