This is an electronic appendix to the paper by Arnold & Wilesmith 2003 Modelling studies on bovine spongiform encephalopathy occurrence to assist in the review of the over 30 months rule in Great Britain. *Proc. R. Soc. Lond. B* [270](https://doi.org/10.1098/rspb.2003.2494), 2141–2145. (DOI 10.1098/rspb.2003.2494.)

Electronic appendices are refereed with the text. However, no attempt is made to impose a uniform editorial style on the electronic appendices.

**Electronic Appendix A**

**BSE Surveillance Data**

Data on the occurrence of BSE in the cattle population were derived from the three sources. First, the clinical incidence of BSE in GB was obtained from the ongoing passive surveillance system whereby the owners/keepers must report cattle with suspect clinical signs. Secondly, the results of the screening of all cattle OTM of age slaughtered as casualties or as fallen stocks since 1 July 2001, and over 24 months of age since January 2002, as required by the EU. Thirdly, results of the screening of all animals OTM of age which were born from 1 August 1996 to 31 July 1997 together with random samples of cattle OTM of age born after this period, and of cattle born before August 1996. Results from both the passive and active surveillance between 1 July 2001 and 30 June 2002 are given in Table 3. BSE was confirmed by histopathology, immunohistochemistry or electron microscopy for the presence of scrapie associated fibrils (OIE 2000)

**Back-calculation model**

The probability of being detected as a clinical case at age $a$ in cohort $c$, $P_c(c,a)$, given survival to age $a$, was given by

$$P_c(c,a) = r(c)\psi(0)O(a)$$

where $r(c)$ was the proportion of animals infected in the cohort (to be estimated), $O(a)$ was the probability of clinical onset at age $a$, and $\psi(t)$ was the probability that the diagnostic test detected infection at time $t$ before clinical onset (where we assume that $\psi(0) = 0.99$). This approach of using a single age of onset distribution over all the birth cohorts is similar in nature to (Donnelly *et al* 1999; Donnelly 2000).

The probability of being detected as an OTM survey case at age $a$ in cohort $c$, if culled at age $a$, was the product of the probability of being infected and the probability of being detected by the post-mortem test (which depends on the length of time before clinical onset). This was given by

$$P_s(c,a) = r(c)\int_a^\infty O(x)\psi(x-a)dx.$$ 

Since we were incorporating both the data on the number of clinical cases and the OTM survey data into the back-calculation model, the log-likelihood function consisted of two parts: $L_c$ and $L_s$, the log-likelihood functions of the data for the number of clinical animals and the number of OTM survey test positives respectively. The testing data for clinical cases and OTM survey test positives for animals of age $a$ in cohort $c$ each arose from a binomial distribution. Therefore, ignoring additive constants, the log-likelihood function for being a clinical case at age $a$ in cohort $c$ was given by
\[ L_c(c,a) = (N(c,a) + \tau_{FS}(c,a) + \tau_{CS}(c,a) - D_C(c,a)) \log(l - P_c(c,a)) + D_C(c,a) \log(P_c(c,a)) \]

where \( N(c,a) \) was the number of animals surviving beyond age \( a \) in cohort \( c \), \( \tau_{CS}, \tau_{FS} \) were the number of negatives from the testing of the casualty slaughter and fallen stock animals respectively, and \( D_C \) was the sum of the reported clinicals, and the fallen stock and casualty slaughter positives. The log-likelihood for the survey animals of age \( a \) in cohort \( c \) was given by

\[ L_S(c,a) = \tau_S(c,a) \log(l - P_S(c,a)) + D_S(c,a) \log(P_S(c,a)) \]

where \( \tau_S \) was the number of test negative samples and \( D_S \) the number of test positives from the OTM screening.

We estimated the prevalence for each annual birth cohort (1 July – 30 June) by dividing each annual birth cohort into quarterly groups (July-September, October-December, etc). The log-likelihood function was summed over the appropriate range of ages (from the age at 1 July 2001 to the age at 30 June 2002) for each of the four quarterly cohorts that made up the annual cohort.

**GB Cattle Demography**

The survival rates of all cattle born on or after 1 July 1996 were obtained from data held in the Cattle Tracing System (CTS). This provided information on cattle surviving up to 31 January 2002, beyond which the data were incomplete. We used the CTS data to provide an age-distribution for each annual birth cohort for cattle up to 54 months of age. For cattle older than 54 months, survival rates were estimated for suckler and dairy animals separately and a weighted-average of the two used to create an overall survival profile of the entire cattle population. The dairy survival rates were obtained from information on herds in the BSE database between 1988 and 1996, which held information on the number of animals alive in each year by lactation (Wilesmith et al 1992). To translate this into age in years we assumed a first lactation of 28 months (Kossaibati & Esslemont 1997), and an average lactation interval of 12.7 months (Esslemont & Kossaibati 2002). The age-structure of the suckler herd was obtained from a Meat and Livestock Commission survey (of 170 herds). In each case, the proportion of animals surviving to 3 monthly age intervals was estimated using geometric interpolation between the known age points. The survival rates by cohort were assumed to remain constant through time. Foot-and-Mouth disease may have increased the survival probability of the pre-July 1996 born animals during 2001, although we do not expect this would affect the model conclusions. We estimated that the total cattle population over 24 months of age at July 2001 was 5.0 million. The estimated survivorship distribution for cattle is given in Fig. 3A.

Estimates of the number of animals born in each cohort post 1 July 1996 were obtained from the CTS. For pre-July 1996 birth cohorts, estimates of the total born were obtained using June agricultural census data between 1986 and 1996 (Ministry of Agriculture Fisheries and Food 1987-1997; Department of Agriculture and Fisheries for Scotland 1987-1990; Scottish Office 1991-1997) on the number of animals alive under 1 year of age. The estimated number born into each quarterly cohort were then estimated using survivorship estimates along with calving
seasonality estimates. The estimates of calving seasonality were obtained from CTS data, and were assumed to remain constant through time.

**Age of onset distribution**

For the age of onset probability distribution function, $O(a)$, we fitted a truncated lognormal distribution to the clinical case data, adjusted for cattle survivorship, for annual birth cohorts 1986-1995. The truncation point for each birth cohort was the maximum age of the youngest animal in the cohort at 30 June 02. The same age of onset distribution was used for each of the annual birth cohorts included in the model.

There was uncertainty in the age of onset distribution of animals born after August 1996 (BARB), as it is possible that these animals will have experienced lower doses resulting in a lengthened age of onset distribution. This would result in a higher prevalence than that estimated using the age of onset distribution from the 1986-1995 case data as fewer of the infected animals would have been detected by surveillance. Therefore we investigated the effect of a lengthening age of onset distribution for the BARB animals on the number of infected animals entering the food chain. We fitted a lognormal distribution to the BARB age of onset data, assuming that casualty slaughter and fallen stock animals were clinical at the time of death/testing ($n=25$ for combined reported clinical animals, fallen stock and casualty slaughter). To obtain a pessimistic estimate, we used the upper 99.9% confidence interval for $\mu$ (obtained using profile likelihoods) when obtaining our estimates of infecteds entering the food chain.

**Sensitivity and specificity of diagnostic test**

There are only limited data available on the sensitivity of currently available diagnostic tests throughout the incubation period of infected cattle. Application of one particular test to cattle orally dosed with 100g of brain homogenate showed that the test could detect infection 32 months post exposure (Grassi et al 2001). This suggests that the diagnostic tests used will typically detect infection in the last few months of the incubation period, but the length of time before detection occurs and its precise performance throughout the incubation period are not known. We assumed that the sensitivity of the diagnostic test decreased as a logistic function as the time from disease onset increased. We ran the model with two different logistic curves for the test sensitivity. For both curves we assumed a maximum sensitivity of 99% at disease onset, but with one we assumed 50% sensitivity at 6 months before onset, and 0% more than 12 months before onset, and for the second we assumed 50% and 0% test sensitivity at 3 and 6 months before onset respectively (Fig. 4).

Given the low proportion of tests which have resulted in false positive results from active surveillance in the EU and UK, and previous work examining the performance of diagnostic tests (Moynagh et al 1999) test specificity is likely to be very close to 100%. Therefore we assumed 100% test specificity.

**Risk of infection since August 1996**

The long incubation period, combined with the lack of a diagnostic test that could detect infection before the last few months of the incubation period, meant that there was very little information on infection prevalence in annual birth cohorts post July 1997. We therefore produced results with two different assumptions regarding the change in risk since August 1996: constant risk of infection from August 1996...
onwards, and a constant risk up to the end of the year 2000, and a risk which halved each year from January 2001.

**Estimation of number of infected animals which would enter the human food chain**

The number of infected animals slaughtered for human consumption at age \(a\) in cohort \(c\), \(H(c,a)\), which would not be identified by the diagnostic test (and therefore enter the food chain) was given by

\[
H(c,a) = N(c,0)\mu(a) r(c) \int_{x=a}^{\infty} O(x)(1 - \psi(x-a))dx
\]

where \(N(c,0)\mu(a)\) was the number of animals slaughtered at age \(a\), \(r(c)\) was the overall proportion of animals ever infected in the cohort, and the integral term represented the proportion of infected animals which survive to age \(a\) and evade detection by the post-mortem diagnostic test. We assumed that all animals over 30 months would be tested. For animals under 30 months, the number of infected animals was calculated as above but with the \(\psi(t)\) term removed since none of the infected animals would be detected. The slaughter rates for human consumption, \(\mu(a)\), were assumed to be equal to the culling rates derived from the survivorship curve, adjusted to take into account the proportion of fallen stock animals by age, none of which enter the food chain (Fig. 3B). We estimated that, with these culling rates, 2.2 million animals currently enter the food chain. This would increase to 3.2 million if all animals over 30 months were allowed into the food chain. We assumed that all animals become infected at 3 months of age. However, epidemiological evidence (Wilesmith et al 1988) suggests that most animals are infected relatively early in life. Furthermore, due to low culling rates of animals between 2 and 12 months of age, there would be little effect on the outputs if this assumption were changed to a distribution of infection age in the first 12 months of life. The total number of infected animals entering the human food chain was calculated for each calendar year by summing the \(H(c,a)\) over the range of cohorts and ages determined by the scenario considered.

**Supplementary results**

Fig. 5 shows the number of animals in the last year of the incubation period which enter the food chain under the more pessimistic assumption of 50% test sensitivity at 3 months before onset. The number of screen test positives predicted each year was approximately half that of that assuming the more sensitive test (approximately 2.5 instead of 5). As mentioned in the letter, the results are qualitatively the same as for the more sensitive test, but with relatively larger numbers of infecteds getting through to the food chain.

**Using BARB age of onset distribution**

The maximum likelihood estimates of the parameters of the BARB age of onset distribution were \(\mu = 1.76, \sigma = 0.238\), with the upper one-sided 99.9% confidence interval estimate for \(\mu = 2.0\) (\(\mu = 1.74, \sigma = 0.25\) for the 1986-1995 case data). The upper estimate of \(\mu\) resulted in a BARB infection prevalence of 160 per million (from approximately 60 per million using the age of onset distribution from the 1986-1995 case data). The estimate of the number of animals in the last 12 months of the
incubation period entering the food chain, assuming 3 month test sensitivity and constant risk of infection from August 1996, and using the upper value of $\mu$ in the age of onset distribution, was approximately 16 per year by 2010 in the birth date cut-off scenarios. The figure using the age of onset distribution from the 1986-1995 case data was approximately 7 (Fig. 2B). The equivalent age-based values were 1.1, 1.3, and 1.8 for the age limits of 30, 36, and 42 months respectively, compared with 0.45, 0.55, 0.74 (Table 2). The relative increase in numbers of animals entering the food chain is slightly less than the relative increase in prevalence of infection, since for a given prevalence a smaller proportion of the culled animals are in the last year of the incubation period under a longer age of onset distribution. The number of clinicals and the number of screen test positives fell to an ultimate level of approximately 80 per year and 10 per year respectively assuming the lengthened age of onset distribution.

References
Department of Agriculture and fisheries for Scotland. 1987-1990 Agricultural statistics, Scotland. HMSO.
Figure 3. A- The probability of survival with age for each annual birth cohort. B) The proportion of each cohort culled by age.
Table 3
Active and passive surveillance data collected between 1 July 2001 and 30 June 2002 for post 1985 annual birth cohorts (1 July – 30 June).

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<th>OTM animals total tested</th>
<th>Fallen stock &amp; Casualty slaughter positive</th>
<th>Fallen stock &amp; Casualty slaughter total tested</th>
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Figure 4. The two distributions of how the sensitivity of the diagnostic test varies with the length of time before clinical onset, the less sensitive detecting 50% of infections with 3 months before clinical onset, and the more sensitive detecting 50% of infections 6 months before onset.
Figure 5. Total number of animals in the last year of the incubation period which are predicted to enter the human food chain in each of the calendar years 2003-2010, with A - different age limits, and B - different birth date cut-offs. The results are produced assuming 50% test sensitivity at 3 months prior to onset, and constant risk of infection post August 1996.