Bovine tuberculosis in cattle and badgers in localised culling areas

Rosie Woodroffe[a,b], Christl A. Donnelly[b,c], D.R. Cox[b,d], Peter Gilks[c], Helen E. Jenkins[c], W. Thomas Johnston[c], Andrea M. Le Fevre[c*], F. John Bourne[b], C.L. Cheeseman[e], Richard S. Clifton-Hadley[f], George Gettinby[b,g], R. Glyn Hewinson[f], John P. McInerney[b], A.P. Mitchell[f], W. Ivan Morrison[b,h] and Gavin H. Watkins[i]

[a]Department of Wildlife, Fish & Conservation Biology, University of California, One Shields Avenue, Davis, California 95616, USA. Tel 530-754-9513; E-mail rwoodroffe@ucdavis.edu.

[b]Independent Scientific Group on Cattle TB, c/o Department for Environment, Food & Rural Affairs, 1A Page Street, London SW1P 4PQ, UK

[c]Department of Infectious Disease Epidemiology, Faculty of Medicine, Imperial College London, St. Mary's Campus, Norfolk Place, London W2 1PG, UK

[d]Nuffield College, Oxford OX1 1NF, UK.

[e]Central Science Laboratory, Sand Hutton, York YO41 1LZ, UK

[f]Veterinary Laboratories Agency, Woodham Lane, Surrey KT15 3NB, UK

[g]Department of Statistics and Modelling Science, University of Strathclyde, Richmond St, Glasgow G1 1XH, UK

[h]Centre for Tropical Veterinary Medicine, Royal (Dick) School of Veterinary Studies, University of Edinburgh, Easter Bush, Roslin, Midlothian EH25 9RG, UK

[i]Veterinary Laboratories Agency Carmarthen, Job's Well Road, Johnstown, Carmarthen SA31 3EZ, UK.

*Present address: Infectious Disease Epidemiology Unit, London School of Hygiene & Tropical Medicine, Keppel Street, London WC1E 7HT, UK.

Running head: Localised badger culling

Word count: 7,979 words including all text

Summary

Bovine tuberculosis (TB), caused by *Mycobacterium bovis*, has serious consequences for Britain's cattle industry. European badgers (*Meles meles*) can transmit infection to cattle, and for many years badgers were culled on and around TB-affected farms to remove animals which might have transmitted infection to cattle. Such localised "reactive" culling would be expected to reduce the incidence of cattle TB if it removed infectious badgers, and if this removal reduced the risk of *M. bovis* transmission to cattle. However, both experimental evidence and national trends indicate that localised badger culling has failed to control cattle TB.

We used data from a recent field trial to evaluate whether reactive culling, targetted at localised areas that had experienced recent cattle TB outbreaks, was effective at removing badgers which might have transmitted infection to cattle. Badgers taken on reactive culls showed a higher prevalence of *M. bovis* infection than did those from more widespread culls conducted in the same regions. However, *M. bovis* infected badgers from reactive culls were no more likely to have TB lesions – a potential indicator of infectiousness – than were those from widespread culls.

There was a very high (80.3%, 95% confidence interval 75.3-85.4%) probability of finding the same *M. bovis* strain type in infected badgers from a particular cull, and in infected cattle from the herds that prompted the cull. Although analyses indicate that random sampling of local infected badgers could have generated the similarity observed across culls, comparisons with cattle TB outbreaks after completion of culling suggest a temporal association.

Synthesis and applications: Since *M. bovis* infections in cattle and badgers were associated, reactive culling could have removed badgers causally linked to cattle TB outbreaks. Despite this association, reactive culling has been shown not to reduce subsequent TB risks for cattle, suggesting that this approach is unlikely to contribute positively to future TB control.

Keywords: badger; *Meles meles*; *Mycobacterium bovis*; perturbation; proactive culling; randomised badger culling trial; reactive culling; tuberculosis; wildlife disease; zoonosis

Introduction

Bovine tuberculosis (TB) is a disease with serious consequences for Britain's cattle industry. A nationwide eradication programme reduced incidence to very low levels by the 1970s, but infection rates among cattle have been rising since the mid-1980s (Krebs et al., 1997).

European badgers (*Meles meles*) are implicated in transmitting *Mycobacterium bovis* – the causative agent of bovine TB – to cattle. For this reason, cattle-based controls were for many years supplemented by badger culling on, and sometimes around, cattle farms that had experienced recent TB outbreaks (Krebs *et al.*, 1997).

National culling policies conducted during 1973-98 (Dunnet, Jones & McInerney, 1986; Zuckerman, 1980) were based on the assumption that infections in cattle and badgers were spatially associated, so that cattle could act as sentinels for *M. bovis* infection in badgers. Removing badgers spatially associated with infected cattle herds was intended to reduce the risks of *M. bovis* transmission from infected badgers to cattle in the same herd, and also to cattle in neighbouring herds likely to be in contact with the same badgers. Recent findings confirm that patterns of infection in badgers and cattle are associated on a scale of 1-2km, with particularly close associations among animals infected with the same strain of *M. bovis* (Woodroffe et al., 2005c). These results suggest that, by targetting badgers spatially associated with particular TB outbreaks in cattle, localised culling might indeed be expected to preferentially remove infected badgers that are potentially infectious to cattle.

Although such localised "reactive" badger culling might be expected to reduce TB risks to cattle, a large scale field trial completed recently in Britain (the Randomised Badger Culling trial, RBCT) found no evidence of such beneficial effects (Donnelly *et al.*, 2003; Le Fevre *et al.*, 2005). Nine 100km^2 areas subjected to a reactive culling treatment over periods of 1-5 years experienced higher cattle TB incidence than did matched areas where no culling was conducted (Donnelly *et al.*, 2003; Le Fevre *et al.*, 2005).

The failure of reactive culling to control cattle TB within RBCT areas mirrors the failure of past policies, likewise based on localised badger culling, to halt the increasing national trend in cattle TB incidence (Krebs et al., 1997). These failures are unfortunate, since localised culling is expected to be cheaper, to be more publicly acceptable, and to have a smaller environmental impact, than an alternative approach involving widespread badger culling (Defra, 2006; White & Whiting, 2000).

Designing better strategies for the control of cattle TB therefore demands an understanding of the reasons for the failure of reactive culling.

The anticipated utility of reactive culling depended on two assumptions: that culling would preferentially remove infectious badgers, and that this removal would reduce subsequent transmission of infection to cattle. Here, we evaluate the first assumption by determining whether reactively culled badgers were more likely to (i) be infected with *M. bovis*; (ii) show TB lesions which might indicate infectiousness; and (iii) share *M. bovis* strain types with associated cattle, than were badgers culled in similar landscapes, but in a less targetted manner.

Methods

Badger culling

All data presented here were collected in the course of the RBCT, a large-scale field trial of the effectiveness of badger culling as a control measure for cattle TB (Donnelly *et al.*, 2006; Donnelly *et al.*, 2003). Thirty 100km^2 trial areas were situated in areas of high cattle TB risk and recruited sequentially as 10 'triplets' (designated A-J) between 1998 and 2002; trial area locations are shown in Supplementary Material. Within each triplet, trial areas were surveyed for signs of badger activity and then allocated to receive either proactive (widespread), reactive (localised), or no badger culling. Treatments were allocated at random.

The proactive treatment involved a single initial cull across all accessible land, with follow-up culls repeated approximately annually thereafter. The reactive treatment involved a series of localised culls in response to specific outbreaks of TB in cattle herds ("breakdowns"). When TB was confirmed in a cattle herd within a reactive trial area, field staff mapped the land used by the affected herd. Survey data were then used to estimate the likely home ranges of badgers using this land (Woodroffe, Frost & Clifton-Hadley, 1999), and to identify setts (dens) used by these badgers (sometimes on neighbouring properties). Areas targetted for culling in this way often coalesced where multiple cattle herds in the same vicinity were affected by TB; hence the number of breakdowns that prompted reactive culling operations exceeded the number of operations. The average reactive culling operation targetted an area of 8.8km^2 and involved eight nights of trapping.

Badgers were captured in cage traps and killed by shooting. The majority of badgers received no injuries from confinement in the trap (Woodroffe et al., 2005b)

and independent audit deemed dispatch methods "humane" (Kirkwood, 2000). No culling occurred during February-April each year to avoid killing females with dependent cubs confined to the sett (Woodroffe et al., 2005a). Culling was also suspended in May 2001-January 2002 due to a nationwide epidemic of foot and mouth disease (FMD). This delayed a number of reactive culling operations, and several operations scheduled for 2001 were cancelled as more recent breakdowns were prioritised once culling resumed. Reactive culling was discontinued in November 2003 when its detrimental effects became apparent (Donnelly et al., 2003); hence no reactive culls were conducted in Triplet J. Proactive culling continued until October 2005 (Donnelly et al., 2006).

Diagnosis and severity of M. bovis infection in badgers

Each badger carcass was chilled after death and necropsied (at one of nine laboratories), usually within 72 hours of dispatch. A proportion of carcasses (9.2% of the total) were stored (almost always frozen) for >7 days before necropsy. Veterinarians conducting necropsies first recorded gender, tooth wear (a measure of age, Neal & Cheeseman, 1996), and any fresh bite wounds. Eighteen pre-specified tissue sites, in five body compartments (head, lungs, chest, abdomen, peripheral), were then incised and examined for lesions suggestive of TB. If a lesion was detected at any of these sites, the badger was considered "lesioned". Each site was scored for lesion severity as: 1 = a single lesion; 2 = 2-3 lesions; 3 = multiple (>3) lesions affecting parts of tissue; 4 = diffuse lesions throughout the tissue. A sample was collected from every lesion, along with one half of each retropharyngeal, both bronchial, and the mediastinal lymph nodes. Badgers were considered infected if *M. bovis* was detected from any sample by bacteriological culture (at one of three laboratories), or if acid-fast bacteria were detected in lesions by Ziehl Neelsen staining (Gallagher & Clifton-Hadley, 2000).

Isolates of *M. bovis* were genotyped by spacer oligonucleotide typing ("spoligotyping", Kamerbeek et al., 1997). This allowed allocation of each isolate to one of the small number of readily identifiable *M. bovis* clones which occur in Britain (Smith *et al.*, 2003); exploratory analysis revealed that an alternative typing method (using variable number tandem repeats) provided no additional information.

Cattle TB data

Data on TB in cattle were taken from routine surveillance. In trial areas, surveillance involved annual tuberculin skin testing as well as ongoing surveillance in slaughterhouses. If any herd showed evidence of *M. bovis* infection ("disclosure"), all skin test positive animals were compulsorily slaughtered and subjected to necropsy. Within trial areas, policy was to culture tissue samples from all compulsorily slaughtered cattle. A breakdown was considered "confirmed" (and hence prompted badger culling in reactive areas) only if lesions suggestive of TB were recorded at necropsy, or if *M. bovis* was isolated following bacteriological culture. The median period between disclosure and slaughter was 21 days; confirmation through detection of TB lesions at necropsy would be immediate, whereas confirmation by culture would take at least another 42 days. Slaughter date is therefore used as a conservative estimate of confirmation date. All *M. bovis* isolates were spoligotyped as for badgers.

Statistical analyses

Primary analyses compared badgers taken in reactive and proactive culling operations. We hypothesised that reactively culled badgers would have (i) higher *M. bovis* prevalence; (ii) greater evidence of TB lesions; and (iii) a higher probability of sharing *M. bovis* strain types with associated cattle because, while reactive culling selectively removed badgers from the vicinity of TB-affected herds, proactive culling was conducted across the landscape without regard to specific breakdowns. Matching of trial areas within triplets suggested that reactive and proactive data would come from badger populations experiencing similar environmental conditions as well as similar overall patterns of *M. bovis* infection. The sensitivity of diagnostic tests was likewise expected to be similar in the two treatments, since the same laboratories were used, with similar proportions of badgers going to each laboratory. Between four and eight laboratories conducted the necropsies on badgers taken from each triplet.

The prevalence of *M. bovis* infection among reactively and proactively culled badgers was compared using logistic regression models adapted from Woodroffe *et al.* (2006b). As in previous analyses, adults and cubs were analysed separately because they showed very different patterns of *M. bovis* infection (Jenkins *et al.*, in review; Woodroffe *et al.*, 2006b; Woodroffe *et al.*, 2005c). These models included several covariates known to influence the probability of infection: triplet, gender, age (measured as tooth wear for adults and days since 1st February for cubs), carcass storage, necropsy laboratory, culture laboratory and date (2002 vs other years; the

suspension of cattle TB testing during the 2001 FMD epidemic was associated with elevated *M. bovis* prevalence in badgers in 2002, and this binary formulation was shown to describe interannual variation in prevalence as effectively as a multi-level categorical variable considering each year separately; Woodroffe *et al.*, 2006b). Model results were corrected for overdispersion (details in Supplementary Material). Since prevalence was known to increase on successive proactive culls (Woodroffe et al., 2006b), a multi-level categorical variable "cull type" was developed to compare baseline prevalence in reactively and proactively culled badgers. This used initial proactive culls as the comparison group for subsequent proactive culling operations, as well as for reactively culled badgers taken on the initial, and all subsequent, culls conducted in a particular land parcel. Comparisons of prevalence under reactive and proactive culling excluded data from 2004-5 when reactive culling had been discontinued (Donnelly et al., 2003).

The severity and distribution of lesions in *M. bovis* infected badgers were assessed using an index developed by Jenkins *et al.* (in review). This index was calculated as:

Lesion index = *(average score of lesioned sites) x (number of sites per affected body compartment)^2 x (number of affected body compartments)*

This index was based on the distributions, particularly the variances, of the three lesion variables included within it (Jenkins *et al.*, in review). For a badger with one lesioned site, the index was equal to the score at that site. The index was higher if more body compartments were lesioned, and if lesions were present in multiple sites in one body compartment. In case freezing influenced the detection of lesions, indices were not calculated for badgers which had been stored >7 days before necropsy.

The *M. bovis* spoligotypes found in badgers on each reactive culling operation were compared with those detected in the cattle herd breakdown(s) which prompted the operation. For each operation, we calculated the average probability that a randomly chosen badger (from those culled on that operation) would share the same spoligotype as a randomly chosen bovine from the associated breakdown(s). This probability (detailed in Supplementary Material) provided a measure of the agreement between spoligotypes from badgers and cattle.

To determine whether operations preferentially removed badgers with spoligotypes matching those in associated cattle, we calculated similar agreement measures for two comparison groups of badgers. For each reactive operation, we

determined the probability that a randomly chosen bovine would share the same spoligotype as a randomly chosen badger from (i) the proactive culling area in the same triplet and year (except that in triplet A proactive data from 1999 were compared with reactive data from 2000 since triplet A received no proactive culling in 2000); and (ii) all other reactive operations conducted in the same triplet (across all years). These measures of spoligotype agreement were then compared on the basis of the weighted average within-operation difference (see Supplementary Material for further details). These analyses were performed for all breakdowns, and also for the subset of breakdowns in which there was no evidence that any tuberculin-positive cattle had moved in from another herd in the previous 12 months.

We also calculated the agreement between spoligotypes from reactively culled badgers, and those from cattle slaughtered in the course of subsequent breakdowns, which occurred in culling-associated herds, but after culling operations had been conducted. This measure was compared with the spoligotype agreement between the same badgers and the cattle which prompted culling. A similar approach was used to compare agreement values for operations conducted at different times after confirmation of infection in cattle, and in response to infection in single or multiple herds.

Results

In total, there were 169 confirmed cattle herd breakdowns which prompted reactive culling, leading to 76 culling operations. The average breakdown involved the slaughter of 12.2 cattle (19.2 SD, range 1-134), of which 4.4 (7.5 SD, range 1-68) were confirmed to be infected (Table 1). The average reactive culling operation captured 27.2 badgers (22.6 SD, range 2-87), including 4.0 (4.5 SD, range 0-25) found to be infected with *M. bovis*. The median time lag between the first cattle slaughter date on a breakdown, and the date the first badger was culled on the associated reactive operation was 211 days (inter-quartile range 146-323 days). Time lags that did not span the FMD epidemic were shorter (median 186 days, inter-quartile range 139-285 days, n=147 breakdowns) than those which did (median 646 days, inter-quartile range 562-718 days, n=22 breakdowns). When breakdowns are divided into clusters (with each cluster prompting a single culling operation), the median time lag between the earliest cattle slaughter date in the cluster and the first badger cull date was 254 days (inter-quartile range 166-453 days).

Prevalence of M. bovis infection in badgers

Of 2,064 badgers taken on reactive culling operations for which culture data were available, 307 (14.9%) showed evidence of *M. bovis* infection. The prevalence recorded amongst adults (15.8%, n=1,654) was higher than that in cubs (10.7%, n=410).

After adjusting for other known predictors of *M. bovis* infection, adult badgers culled under the reactive strategy were more likely to show evidence of infection than were those taken on initial proactive culls (Table 2; overall). Prevalence in badger cubs showed a comparable, albeit non-significant, trend in the same direction (details in Supplementary Material). As shown in Table 2, prevalence tended to be higher among reactively culled adults taken from land parcels where one or more (maximum four) operations had already occurred (n=337 animals) than on land receiving reactive culling for the first time (n=1,317).

Pathology of M. bovis infection in badgers

Of 247 *M. bovis* infected reactively culled adult badgers for which pathology data were available, 103 (41.7%) had lesions suggestive of TB and 19 (7.7%) were considered to have severe or widely distributed lesions (lesion indices ≥8; Table 3). Equivalent figures for reactively culled cubs were 40.5% and 14.3% respectively (n=42). Detailed descriptions of lesion distribution and severity are provided in Supplementary Material.

Table 3 shows that *M. bovis* infected badgers taken on proactive and reactive culls had similar probabilities of being lesioned. Overall, the pattern of lesion severity was similar among proactively and reactively culled badgers (Figure 1). Adult badgers taken on both types of cull were more likely to be infected with *M. bovis* if they had fresh bite wounds (Table 3). However, while proactively culled adults had higher lesion prevalence if they were bite wounded (Jenkins et al., in review), this pattern was not observed among reactively culled badgers (Table 3).

Comparison of infections in cattle and badgers

Reactive culling was conducted in response to confirmed breakdowns in cattle; therefore all herds considered here contained at least one confirmed infected bovine. Of 76 reactive operations, 60 (79%) captured one or more infected badgers. A

logistic regression showed that the overall probability of capturing at least one infected badger increased with the (log transformed) total number caught (odds ratio associated with doubling the number captured 1.76; χ^2=6.36, p=0.012). However, some operations which caught no infected animals captured large numbers of badgers (range 2-62). There was no evidence that the probability of capturing an infected badger was lower where infection involved bought-in cattle. Seventy-eight tuberculin-positive cattle, associated with 24 breakdowns and 21 culling operations, had moved in from other herds within the previous 12 months. However, these operations had a probability of catching one or more infected badgers (17/21 operations) very similar to that recorded on breakdowns not involving bought-in cattle (43/55 operations, χ^2=0.07, d.f.=1, p=0.79).

Of 169 breakdowns associated with reactive culling, 155 produced isolates of *M. bovis* which were successfully spoligotyped. Of these, 139 involved a single spoligotype, 14 involved two spoligotypes, and two involved three spoligotypes. Spoligotype frequencies recorded among cattle in breakdowns associated with reactive culling were broadly similar to those found in badgers in the same trial area (Table 4). While some spoligotypes were recorded in one species but not the other within a trial area, these never accounted for more than 16% of infections within a species (Table 4).

Of 76 reactive culling operations, 55 had spoligotype data from both badgers and associated cattle. Badgers and cattle were found to share at least one spoligotype on 51 (94%) of these operations. Overall, there was an estimated 80.3% probability (95% confidence interval (CI): 75.3-85.4%) that a (spoligotyped) badger chosen at random from a particular reactive operation would share the same *M. bovis* spoligotype as a (spoligotyped) bovine chosen at random from the associated breakdown(s).

The extent of agreement between spoligotypes recorded in associated cattle and badgers appeared unrelated to the time lag between breakdowns and the culling operations that they prompted. Time lags were arbitrarily considered "short" if ≤270 days elapsed between the median date of first cattle slaughter in a cluster of breakdowns, and the subsequent associated badger cull. The probability of associated cattle and badgers sharing the same *M. bovis* spoligotype was similar for operations subject to "short" and "long" time lags (78.8% and 82.8% respectively, difference

3.9% less for short time lags, 95% CI: 13.4% less to 5.6% greater, p=0.42). Similarly, within clusters of TB-affected herds, particular breakdowns were considered "early" if they occurred on or before the median date for the cluster, and "late" if they occurred after the median. The spoligotype agreement between cattle and badgers was similar for "early" and "late" breakdowns (84.9% and 86.0% respectively, difference 1.1% less for early breakdowns, 95% CI: 4.1% less to 1.9% greater, p=0.46). Agreement between badger and cattle spoligotypes was also similar for operations associated with single and multiple herd breakdowns (73.9% and 82.1% respectively, difference 8.2% less for single breakdowns, 95% CI: 22.9% less to 6.4% more, p=0.27).

The spoligotype agreement between cattle from breakdowns prompting reactive operations, and associated reactively-culled badgers (80.3%), was greater than that between the same cattle and badgers taken on proactive culls in the same year and triplet (75.6%, difference 4.7% greater for associated reactive badgers, 95% CI: 1.4-8.0% greater, p=0.005). However, this agreement between associated cattle and badgers was not significantly different from that between the same cattle and all other reactively culled badgers from the same triplet (79.8%, difference 0.6% greater for associated reactive badgers, 95% CI: 1.7% less to 2.8% greater, p=0.62). Results were very similar if analyses excluded breakdowns involving tuberculin-positive cattle which had been bought in during the previous 12 months (associated reactive compared with proactive: 81.9% *vs* 76.5%, difference 5.4% greater, 95% CI: 2.1-8.8% greater, p=0.001; associated reactive compared with all other reactive: 81.9% *vs* 80.8%, difference 1.1% greater, 95% CI: 0.9% less to 3.0% greater, p=0.28).

By the end of 2005, 79 further confirmed breakdowns had been recorded in the herds originally associated with reactive culling. These herds had been targetted by 42 culling operations, of which 31 provided spoligotype data for both host species (Table 5). The agreement between spoligotypes from these repeat breakdowns, and the badgers culled previously (82.5%) was significantly lower than that between the same badgers and the breakdowns that originally prompted culling (86.7%, difference 4.2% less, 95% CI: 2.0-6.4% less, p<0.001).

Discussion

The findings presented here suggest that, as in proactive areas (Woodroffe et al., 2005c), *M. bovis* infections in cattle and badgers were associated in reactive culling areas. Badgers taken on reactive culling operations showed a higher

prevalence of *M. bovis* infection than did those from proactive culls, as would be expected if badger infections were spatially associated with those in cattle. Moreover, the agreement between *M. bovis* spoligotypes found in cattle and badgers was greater for breakdowns that prompted reactive culls than for subsequent breakdowns in the same herds. It should be noted, however, that these associations provide no information on the relative importance of badger-to-cattle and cattle-to-badger transmission.

Although the prevalence of *M. bovis* infection differed between badgers taken from reactive and proactive areas, no such differences were apparent for lesion prevalence or severity. This mirrors the situation recorded within proactive areas, where associations with infected cattle were no closer for lesioned infected badgers than for infected badgers without TB lesions (Woodroffe et al., 2005c). It appears likely that badgers without detected lesions might nevertheless have been able to transmit infection (Jenkins et al., in review), as has been recorded in cattle (McCorry et al., 2005). It is also possible that transmission of infection from cattle to badgers may have contributed to the spatial association of infection in the two host species; evidence suggests that such transmission was widespread when testing and removal of infected cattle was temporarily suspended (Woodroffe et al., 2006b), showing that cattle can be a source of infection for badgers.

The majority of *M. bovis* spoligotypes were shared between badgers and associated cattle. Although the spoligotype agreement observed on reactive culls was higher than that expected based on spoligotype frequencies from proactively culled badgers, this difference was driven by two triplets where spoligotype frequencies were markedly different in proactive and reactive areas (Table 5). In triplet F, spoligotypes SB0140 and SB0145 accounted for, respectively, 56% and 44% of badger spoligotypes in the proactive area in 2002-3, but 0% and 9% of those in reactively culled badgers in the same years. Likewise, in Triplet I, spoligotypes SB0263 and SB0272 accounted for 73% and 7% of badger spoligotypes in the proactive area in 2003, but 21% and 79% of those in reactively culled badgers. When cattle spoligotypes from particular reactive operations were compared with badger spoligotypes from non-associated operations conducted in the same trial area, the level of agreement was not significantly different from that with badgers taken on the associated operation. This indicates that, on the basis of spoligotype data, reactive

culling appeared equivalent to random – rather than targetted – sampling of infected badgers in the vicinity of infected cattle.

There are several possible explanations for this pattern. One possibility is that the amalgamation of multiple breakdowns into a smaller number of culling operations led to sampling of badgers on a spatial scale larger than that on which cattle and badger infections are associated. However, the similar level of agreement between badger and cattle spoligotypes in operations associated with single and multiple breakdowns provides no support for this hypothesis.

An alternative explanation is that the comparatively small number of spoligotypes in each trial area (Table 4) provided insufficient precision to link infections in cattle and badgers. While this is consistent with the localised geographic distribution of most *M. bovis* clones in Britain (Smith *et al.*, 2003), we were able to detect differences in spoligotype agreement with badgers between breakdowns which prompted reactive culls, and subsequent breakdowns in the same herds. This suggests that spoligotyping offered sufficient precision to detect temporal associations between infections in the two host species, and should therefore have been able to detect spatial associations.

A final explanation is that not all infections in cattle and badgers may have been causally linked. Past policies conducted localised culling only in response to breakdowns considered to have been "badger related", excluding breakdowns thought to have been caused by factors such as bought-in cattle (Krebs et al., 1997). In contrast, RBCT reactive culling was conducted in response to confirmed breakdowns with no attempt to identify possible causes. It is therefore likely, on the basis of national patterns, that some breakdowns which prompted reactive culling were caused by cattle-to-cattle transmission (Cox *et al.*, 2005; Gilbert *et al.*, 2005). Infections might or might not be detected among badgers culled in association with such breakdowns, but would not necessarily be causally linked to the cattle infection. We could find no support for this hypothesis using this dataset: excluding herds with evidence of infection in recently bought-in cattle altered neither the probability of catching an infected badger, nor the patterns of spoligotype agreement. However, these analyses cannot rule out the possibility that herd breakdowns caused by cattle-to-cattle transmission reduced agreement between infections in cattle and badgers, because such infections may not always be detected in the bought-in cattle themselves, and because transmission could also occur between herds through other

means. The possible role of cattle-to-cattle transmission in explaining the patterns presented here could be investigated in future by tracing cattle on a case-by-case basis, although the imperfect sensitivity of the tuberculin test (Morrison *et al.*, 2000) means that some breakdowns could have been caused by infected cattle that remained undetected. It is important to note that re-stocking of herds immediately after the 2001 FMD epidemic is unlikely to have had a large impact on our findings. Only 7% of herds in RBCT areas were affected by FMD (Bourne *et al.*, 2005), and still fewer of these would have subsequently experienced TB. Moreover, of 76 reactive operations, 60 commenced after June 2002, and 45 commenced more than 12 months after the end of the FMD epidemic on 28th November 2001.

The time lag between confirmation of infection in cattle and culling of badgers was comparable – when not interrupted by FMD – with that operating under the previous "interim" culling strategy (approximately 6 months in both cases, Krebs *et al.*, 1997). It should be noted that, given annual testing, cattle could become infected many months before infection is confirmed. This time lag was considered a weakness of the interim strategy, since it allowed opportunities for badgers associated with particular breakdowns to infect additional cattle (Krebs *et al.*, 1997). Delays were an inevitable component of the reactive strategy since (i) reactive operations were often postponed until herds contiguous with the original breakdown herd had been tested, to ensure inclusion of all land associated with a breakdown cluster; (ii) additional surveying was needed to prepare for culling; (iii) reactive and proactive operations were conducted by the same teams, necessitating that the two strategies follow complementary timetables; and (iv) no culling could be conducted during the closed season. Despite concerns about the delays intrinsic to reactive culling, we found no difference in spoligotype agreement between operations associated with "long" and "short" time lags.

Overall, our findings suggest that *M. bovis* infections in cattle may be used as a sentinel for infections in badgers, though probably an imperfect one. This provides some support for the first assumption on which the reactive culling strategy was based, namely that localised culling would preferentially remove infectious badgers. The second assumption – that such removal would reduce transmission of infection to cattle – therefore warrants further scrutiny. Both reactive and proactive culling disrupted badger territoriality and prompted expanded ranging behaviour (Woodroffe *et al.*, 2006a); this would have increased the number of cattle herds encountered by

each badger, and hence the number of herds to which an infected badger could transmit *M. bovis*. The same behavioural changes appear to have increased badger-to-badger transmission: repeated culling in the same areas was associated with increased *M. bovis* prevalence among proactively culled badgers (Woodroffe *et al.*, 2006b), and data presented here suggest a similar pattern for reactively culled badgers. These changes in badger behaviour and infection prevalence would both increase the cattle TB risk associated with each badger, potentially undermining – or even overcoming – beneficial effects caused by reduced badger density. This may explain why reactive culling did not reduce the risks of transmission to cattle, even though it successfully targetted infected badgers.

The problems encountered with the reactive treatment are likely to apply to other strategies involving localised culling. Other ways of targetting culling at "the right badgers" entail their own problems. A serological test developed for badgers lacked sufficient sensitivity to identify infected animals or social groups (Woodroffe et al., 1999). More recently, molecular methods have been used to detect mycobacteria in the environment (Courtenay et al., 2006), but positive sample rates are extremely high and specificity – as well as relevance to transmission – are unknown.

An additional concern is that any form of localised culling is likely to cause behavioural change in badgers and, hence, increased transmission. Although *M. bovis* infections are clustered in badgers, the edges of these clusters are not sharply defined (Delahay et al., 2000; Woodroffe et al., 2005c) so, even if every infected animal, or every member of an infected social group, could be identified and removed, it is likely that some animals immigrating into the cleared area would be infected. Imperfect detection of infection in badgers, and imperfect badger removal, elevate the chances of increased contact rates leading to increased transmission, constraining the ability of localised culling to reduce TB risks to cattle.

These findings suggest that localised badger culling, using currently available methods, is unlikely to contribute to future strategies for cattle TB control in Britain. The study also highlights the critical need for good ecological data in designing control strategies for wildlife diseases.

Acknowledgements

This study was funded and implemented by the Department of Environment, Food and Rural Affairs (Defra). We gratefully acknowledge the work of the Defra Wildlife Unit which conducted all RBCT fieldwork, and the Veterinary Laboratories Agency and Central Science Laboratory which performed all diagnostic work. We also wish to thank the many landholders who allowed data collection on their land.

References

Bourne, J., Donnelly, C.A., Cox, D.R., Gettinby, G., McInerney, J.P., Morrison, W.I., & Woodroffe, R. (2005) *An epidemiological investigation into bovine tuberculosis - towards a science-based control strategy. Fourth Report of the Independent Scientific Group on Cattle TB.* Defra, London.

Courtenay, O., Reilly, L.A., Sweeney, F.P., Hibberd, V., Bryan, S., Ul-Hassan, A., Newman, C., Macdonald, D.W., Delahay, R.J., Wilson, G.J., & Wellington, E.M.H. (2006) Is *Mycobacterium bovis* in the environment important for the persistence of bovine tuberculosis? *Biology Letters*, **2**, 460-462.

Cox, D.R., Donnelly, C.A., Bourne, F.J., Gettinby, G., McInerney, J.P., Morrison, W.I., & Woodroffe, R. (2005) Simple model for tuberculosis in cattle and badgers. *Proceedings of the National Academy of Sciences of the United States of America*, **102**, 17588-17593.

Defra (2006) *Public consultation on controlling the spread of bovine tuberculosis in cattle in high incidence areas in England: badger culling – Summary of responses* Department for Environment, Food and Rural Affairs, London.

Delahay, R.J., Langton, S., Smith, G.C., Clifton-Hadley, R.S., & Cheeseman, C.L. (2000) The spatio-temporal distribution of *Mycobacterium bovis* (bovine tuberculosis) infection in a high-density badger population. *Journal of Animal Ecology*, **69**, 428-441.

Donnelly, C.A., Woodroffe, R., Cox, D.R., Bourne, F.J., Cheeseman, C.L., Wei, G., Gettinby, G., Gilks, P., Jenkins, H., Johnston, W.T., Le Fevre, A.M., McInerney, J.P., & Morrison, W.I. (2006) Positive and negative effects of widespread badger culling on cattle tuberculosis. *Nature*, **439**, 843-846.

Donnelly, C.A., Woodroffe, R., Cox, D.R., Bourne, J., Gettinby, G., Le Fevre, A.M., McInerney, J.P., & Morrison, W.I. (2003) Impact of localized badger culling on TB incidence in British cattle. *Nature*, **426**, 834-837.

Dunnet, G.M., Jones, D.M., & McInerney, J.P. (1986) *Badgers and bovine tuberculosis: review of policy* H.M.S.O., London.

Gallagher, J. & Clifton-Hadley, R.S. (2000) Tuberculosis in badgers: a review of the disease and its significance for other animals. *Research in Veterinary Science*, **69**, 203-217.

Gilbert, M., Mitchell, A., Bourn, D., Mawdsley, J., Clifton-Hadley, R.S., & Wint, W. (2005) Cattle movement and bovine tuberculosis in Great Britain. *Nature*, **435**, 491-496.

Jenkins, H.E., Morrison, W.I., Cox, D.R., Donnelly, C.A., Johnston, W.T., Bourne, F.J., Clifton-Hadley, R.S., Gettinby, G., McInerney, J.P., Watkins, G.H., & Woodroffe, R. (in review) The prevalence, distribution and severity of pathological lesions in badgers naturally infected with *Mycobacterium bovis*. *Epidemiology and Infection*.

Kamerbeek, J., Schouls, L., Kolk, A., van Agterveld, M., van Soolingen, D., Kuijper, S., Bunschoten, A.E., Molhuizen, H., Shaw, R., Goyal, M., & van Embden, J. (1997) Simultaneous detection and strain differentiation of *Mycobacterium tuberculosis* for diagnosis and epidemiology. *Journal of Clinical Microbiology*, **35**, 907-914.

Kirkwood, J.K. (2000). Humaneness of MAFF's badger dispatch procedures. Ministry of Agriculture, Fisheries and Food, London.

Krebs, J.R., Anderson, R., Clutton-Brock, T., Morrison, I., Young, D., Donnelly, C., Frost, S., & Woodroffe, R. (1997) *Bovine tuberculosis in cattle and badgers* H.M.S.O., London.

Le Fevre, A.M., Donnelly, C.A., Cox, D.R., Bourne, F.J., Clifton-Hadley, R.S., Gettinby, G., Johnston, W.T., McInerney, J.P., Morrison, W.I., & Woodroffe, R. (2005) The impact of localised reactive badger culling versus no culling on TB incidence in British cattle: a randomised trial, pp. http://www.defra.gov.uk/animalh/tb/isg/pdf/lefevre1005.pdf.

McCorry, T., Whelan, A.O., Welsh, M.D., McNair, J., Walton, E., Bryson, D.G., Hewinson, R.G., Vordermeier, H.M., & Pollock, J.M. (2005) Shedding of *Mycobacterium bovis* in the nasal mucus of cattle infected experimentally with tuberculosis by the intranasal and intratracheal routes. *Veterinary Record*, **157**, 613-618.

Morrison, W.I., Bourne, F.J., Cox, D.R., Donnelly, C.A., Gettinby, G., McInerney, J.P., & Woodroffe, R. (2000) Pathogenesis and diagnosis of infections with *Mycobacterium bovis* in cattle. *Veterinary Record*, **146**, 236-242.

Neal, E. & Cheeseman, C. (1996) *Badgers* Poyser, London.

Smith, N.H., Dale, D., Inwald, J., Palmer, S., Gordon, S.V., Hewinson, R.G., & Smith, J.M. (2003) The population structure of *Mycobacterium bovis* in Great Britain: Clonal expansion. *Proceedings of the National Academy of Sciences, USA*, **100**, 15271-15275.

White, P.C.L. & Whiting, S.J. (2000) Public attitudes towards badger culling to control bovine tuberculosis in cattle. *Veterinary Record*, **147**, 179-184.

Woodroffe, R., Bourne, F.J., Cheeseman, C.L., Cox, D.R., Donnelly, C.A., Gettinby, G., McInerney, J.P., & Morrison, W.I. (2005a) Welfare of badgers (*Meles meles*) subjected to culling: development and evaluation of a closed season. *Animal Welfare*, **14**, 19-25.

Woodroffe, R., Bourne, F.J., Cheeseman, C.L., Cox, D.R., Donnelly, C.A., Gettinby, G., McInerney, J.P., & Morrison, W.I. (2005b) Welfare of badgers (*Meles meles*) subjected to culling: patterns of trap-related injury. *Animal Welfare*, **14**, 11-17.

Woodroffe, R., Donnelly, C.A., Cox, D.R., Bourne, F.J., Cheeseman, C.L., Delahay, R.J., Gettinby, G., McInerney, J.P., & Morrison, W.I. (2006a) Effects of culling on badger (*Meles meles*) spatial organization: implications for the control of bovine tuberculosis. *Journal of Applied Ecology*, **43**, 1-10.

Woodroffe, R., Donnelly, C.A., Jenkins, H.E., Johnston, W.T., Cox, D.R., Bourne, F.J., Cheeseman, C.L., Delahay, R.J., Clifton-Hadley, R.S., Gettinby, G., Gilks, P., Hewinson, R.G., McInerney, J.P., & Morrison, W.I. (2006b) Culling and cattle controls influence tuberculosis risk for badgers. *Proceedings of the National Academy of Sciences of the United States of America*, **103**, 14713-14717.

Woodroffe, R., Donnelly, C.A., Johnston, W.T., Bourne, F.J., Cheeseman, C.L., Clifton-Hadley, R.S., Cox, D.R., Gettinby, G., Hewinson, R.G., Le Fevre, A.M., McInerney, J.P., & Morrison, W.I. (2005c) Spatial association of *Mycobacterium bovis* infection in cattle and badgers. *Journal of Applied Ecology*, **42**, 852-862.

Woodroffe, R., Frost, S.D.W., & Clifton-Hadley, R.S. (1999) Attempts to control tuberculosis in cattle by culling infected badgers: constraints imposed by live test sensitivity. *Journal of Applied Ecology*, **36**, 494-501.

Zuckerman, S. (1980) *Badgers, cattle and tuberculosis* H.M.S.O., London.

Table 1 – Descriptive data on reactive culling operations within nine RBCT triplets; no reactive culling was conducted in Triplet J.

	Herd breakdowns associated with culling				Years§ of reactive culling			Reactive culling operations		Number of badgers captured on reactive culling operations		
Triplet	*n*	*cattle slaughtered**	*cattle with M. bovis**	*cattle with spoligotypes**	*n*	*first*	*last*	*culls*	*culls with M. bovis*	*badgers caught†*	*badgers with M. bovis†*	*badgers with spoligotypes†*
A	21	278 (13.2)	64 (3.0)	55 (2.6)	3	2000	2003	10	8	117 (11.7)	30 (3.0)	23 (2.3)
B	27	307 (11.4)	60 (2.2)	49 (1.8)	4	1999	2003	9	7	301 (33.4)	27 (3.0)	25 (2.8)
C	42	468 (10.4)	199 (4.7)	161 (3.8)	3	2000	2003	19	15	394 (20.7)	55 (2.9)	49 (2.6)
D	7	71 (10.1)	26 (3.7)	22 (3.1)	1	2003	2003	4	4	122 (30.5)	31 (7.8)	31 (7.8)
E	24	295 (12.3)	117 (4.9)	98 (4.1)	2	2002	2003	10	6	188 (18.8)	23 (2.3)	20 (2.0)
F	23	250 (10.9)	77 (3.3)	71 (3.1)	2	2002	2003	10	10	436 (43.6)	52 (5.2)	43 (4.3)
G	10	186 (18.6)	60 (6.0)	57 (5.7)	2	2002	2003	7	5	255 (36.4)	31 (4.4)	31 (4.4)
H	7	125 (17.9)	92 (13.1)	86 (12.3)	2	2002	2003	4	3	160 (40.0)	29 (7.3)	26 (6.5)
I	8	86 (10.8)	43 (5.4)	42 (5.3)	1	2003	2003	3	2	94 (31.3)	29 (9.7)	28 (9.3)
Total	169	2,066 (12.2)	738 (4.4)	641 (3.8)				76	60	2067‡ (27.2)	307 (4.0)	276 (3.6)

§"badger years", defined as running 1st February-31st January; *total across breakdowns (mean per breakdown); †total across culling operations (mean per operation);
‡culture data were lacking for 3 of these animals

Table 2 – Logistic regression model of *M. bovis* infection prevalence in adult badgers during 1998-2003. Results are adjusted for overdispersion (see Supplementary for overdispersion.

Predictor	**odds ratio (95% CI)**	χ^2	**d.f.**	**P**
Triplet		186.16	9	<0.001
Gender		20.59	1	<0.001
male *vs* female	1.45 (1.23-1.71)			
Tooth wear		7.35	5	0.196
2 *vs* 1	0.95 (0.47-1.91)			
3 *vs* 1	1.11 (0.55-2.21)			
4 *vs* 1	1.09 (0.54-2.20)			
5 *vs* 1	1.24 (0.60-2.57)			
not recorded *vs* 1	0.23 (0.03-1.63)			
Carcass storage		8.90	1	0.003
>7 days *vs* ≤7 days	0.59 (0.40-0.88)			
Necropsy laboratory		18.49	9	0.030
Culture laboratory		2.45	2	0.293
FMD		10.13	1	0.001
2002 *vs* other years	1.49 (1.18-1.88)			
Cull type		25.78	5	<0.001
first reactive *vs* first proactive	1.82 (1.27-2.61)			
subsequent reactive *vs* first proactive	3.20 (1.82-5.63)			
second *vs* first proactive	1.02 (0.76-1.35)			
third *vs* first proactive	1.46 (0.93-2.29)			
fourth *vs* first proactive	3.04 (1.75-5.29)			
fifth *vs* first proactive	2.23 (0.91-5.47)			

Material).

Predictor	**odds ratio (95% CI)**	χ^2	**df**	**P**
Triplet		186.16	9	<0.001
B *vs* A	0.19 (0.12-0.31)			
C *vs* A	0.22 (0.14-0.33)			
D *vs* A	1.14 (0.75-1.74)			
E *vs* A	0.24 (0.16-0.36)			
F *vs* A	0.18 (0.12-0.37)			
G *vs* A	0.22 (0.14-0.34)			
H *vs* A	0.30 (0.19-0.47)			
I *vs* A	1.03 (0.65-1.64)			
J *vs* A	0.31 (0.19-0.52)			
Gender		20.59	1	<0.001

male *vs* female	1.45 (1.23-1.71)			
Tooth wear		7.35	5	0.196
2 *vs* 1	0.95 (0.47-1.91)			
3 *vs* 1	1.11 (0.55-2.21)			
4 *vs* 1	1.09 (0.54-2.20)			
5 *vs* 1	1.24 (0.60-2.57)			
not recorded *vs* 1	0.23 (0.03-1.63)			
Carcass storage		8.90	1	0.003
>7 days *vs* ≤7 days	0.59 (0.40-0.88)			
Necropsy laboratory		18.49	9	0.030
Culture laboratory		2.45	2	0.293
FMD		10.13	1	0.001
2002 *vs* other years	1.49 (1.18-1.88)			
Cull type		25.78	5	<0.001
first reactive *vs* first proactive	1.82 (1.27-2.61)			
subsequent reactive *vs* first proactive	3.20 (1.82-5.63)			
second *vs* first proactive	1.02 (0.76-1.35)			
third *vs* first proactive	1.46 (0.93-2.29)			
fourth *vs* first proactive	3.04 (1.75-5.29)			
fifth *vs* first proactive	2.23 (0.91-5.47)			

Table 3 – Comparison of lesion prevalence and lesion indices among badgers taken on reactive (1999-2003) and proactive (1998-2005) culls. Numbers in parentheses are exact binomial 95% confidence intervals; n gives the sample size used to estimate each proportion. Details on infection patterns in cubs with and without bite wounds are omitted due to very small sample sizes.

	adults		**cubs**	
	proactive	*reactive*	*proactive*	*reactive*
Proportions lesioned				
% *M. bovis* infected animals with visible lesions	38.5% (35.5-41.6%) n=1020	41.7% (35.5-48.1%) n=247	55.5% (47.0-63.7%) n=146	40.5% (25.6-56.7%) n=42
% *M. bovis* infected animals with >1 body compartment lesioned	14.7% (12.6-17.0%) n=1020	12.6% (8.7-17.3%) n=247	28.1% (21.0-36.1%) n=146	26.2% (13.9-42.0%) n=42
% *M. bovis* infected animals with lesion indices ≥8	10.5% (8.7-12.5%) n=1020	7.7% (4.7-11.8%) n=247	23.3% (16.7-31.0%) n=146	14.3% (5.4-28.5%) n=42
Bite wounds				
% animals with fresh bite wounds	5.3% (4.7-6.0%) n=5117	4.9% (3.8-6.2%) n=1402	1.3% (0.8-2.0%) n=1385	0.9% (0.2-2.6%) n=333
% bite wounded animals with *M. bovis* infection	44.5% (38.5-50.6%) n=272	44.9% (32.9-57.4%) n=69	–	–
% non-bite wounded animals with *M. bovis* infection	16.8% (15.8-17.9%) n=4845	14.6% (12.7-16.6%) n=1333	–	–
% bite wounded, *M. bovis* infected animals with lesions	53.7% (44.4-62.8%) n=121	35.5% (19.2-54.6%) n=31	–	–
% non-bite wounded, *M. bovis* infected animals with lesions	37.0% (33.7-40.4%) n=816	42.8% (35.7-50.0%) n=194	–	–

Table 4 – Frequency of spoligotypes in reactively culled badgers and associated cattle herds. Data indicate the proportions of infected badgers and cattle (from breakdowns which prompted culling) in each triplet found to have each spoligotype of *M. bovis*. The last line gives the numbers of *M. bovis* isolates available from each species in each reactive trial area.

	Triplet																	
	A		**B**		**C**		**D**		**E**		**F**		**G**		**H**		**I**	
Spoligotype*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*	*badger*	*cattle*
SB0129	–	–	–	–	0.02	–	–	0.05	–	0.01	–	–	0.94	1.00	–	0.01	–	–
SB0134	–	–	–	–	–	–	0.10	–	–	–	–	–	–	–	–	–	–	–
SB0140	0.04	0.04	0.96	0.88	0.92	0.98	–	–	0.25	0.13	–	0.03	–	–	–	–	–	–
SB0145	–	–	–	–	–	–	–	–	–	–	0.09	0.20	–	–	–	0.01	–	–
SB0263	0.91	0.95	–	0.04	0.06	0.01	0.84	0.95	0.75	0.86	0.02	–	–	–	–	–	0.21	0.71
SB0271	–	–	–	–	–	0.01	–	–	–	–	0.86	0.77	–	–	–	–	–	–
SB0272	–	0.02	–	–	–	–	0.06	–	–	–	0.02	–	0.06	–	0.12	–	0.79	0.21
SB0274	0.04	–	–	0.08	–	0.01	–	–	–	–	–	–	–	–	0.88	0.97	–	0.07
SB0275	–	–	0.04	–	–	0.01	–	–	–	–	–	–	–	–	–	–	–	–
SB1073	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	0.01	–	–
N=	23	55	25	49	49	161	31	22	20	98	43	71	31	57	26	86	28	42

*International spoligotype identities; for equivalent VLA identiies see http://www mbovis.org/spoligodatabase/GBmetadata/frequency%20spoligo%20GB html

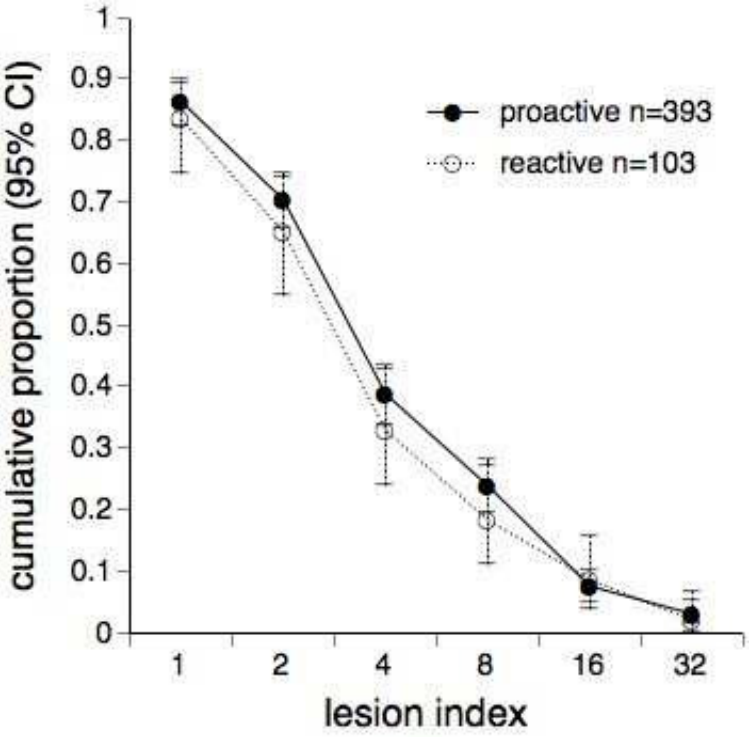
Table 5 – Matching of *M. bovis* spoligotypes from badgers taken on reactive culling operations and with those from the original herd breakdowns that prompted culling, and with subsequent breakdowns involving the same herds. Data are restricted to operations with spoligotype data from both species, hence numbers of animals reported as having spoligotype data differ from those given in Table 1.

	A	B	C	D	E	F	G	H	I	Total
Original breakdowns										
Operations with spoligotypes from both species	7	6	14	4	6	9	4	3	2	55
Operations with one or more spoligotypes found in both species	7	6	12	4	6	7	4	3	2	51
Badgers with spoligotypes	23	25	49	31	20	41	28	26	28	271
Cattle with spoligotypes	37	40	121	22	82	71	54	81	20	528
Probability associated cattle and badgers share same spoligotype	89.3%	81.7%	78.6%	80.3%	69.0%	67.6%	99.6%	85.2%	91.7%	80.3%
Probability cattle share spoligotype with proactively culled badgers in same triplet and year	83.8%	92.5%	96.4%	93.0%	53.3%	13.8%	99.8%	96.3%	43.3%	75.6%
Probability cattle share spoligotype with badgers from other reactive operations in same triplet	86.6%	89.6%	89.6%	79.4%	71.8%	67.6%	91.5%	85.6%	9.2%	79.8%
Subsequent breakdowns										
Operations with spoligotypes from both species	4	6	8	1	4	4	1	1	2	31
Operations with one or more spoligotypes found in both species	4	6	8	1	4	4	1	1	2	31
Badgers with spoligotypes	14	25	31	4	17	15	4	17	28	155
Cattle with spoligotypes	32	81	45	4	51	5	4	7	5	234
Probability associated cattle and badgers share same spoligotype	92.9%	92.7%	83.7%	75.0%	66.4%	93.2%	100%	84.6%	91.7%	86.7%
Probability repeat cattle share spoligotype with badgers culled on previous reactive operation	92.0%	96.0%	77.4%	75.0%	54.2%	93.3%	100%	75.6%	85.7%	82.5%

Figure Legends

Figure 1 – Variation in lesion indices on reactive and proactive culls. Points indicate the cumulative proportion of *M. bovis* infected badgers (adults and cubs combined) showing different levels of lesion severity. Error bars are exact binomial 95% confidence intervals.

Figure 1


cumulative proportion (95% CI)
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
proactive n=393
reactive n=103
1
2
4
8
16
32
lesion index