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Spatial and Temporal Distribution of Bovine Tuberculosis in Badgers in Four Areas in Ireland: Preliminary Findings

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Introduction.

Bovine tuberculosis (BTB) is a continuing problem in cattle in several countries, but it is only in Great Britain and the Republic of Ireland that badgers are recognized as the principal wildlife reservoir. BTB was first recorded in badgers in Switzerland by Bouvier and co-authors in 1962, and they were first suspected as a reservoir in the early 1970's, in England (Krebs, 1997). Tuberculosis in badgers in Ireland was identified first in 1973 (Noonan *et al.*, 1975). Currently, *Mycobacterium bovis* infection is endemic in the badger populations in Great Britain (Cheeseman *et al.*, 1989; Clifton-Hadley *et al.*, 1993) and in Ireland. Evidence for the involvement of tuberculous badgers in the development of *Mycobacterium bovis* infection in cattle in Ireland is available elsewhere (O'Connor and O'Malley, 1989; Dolan 1993; Martin *et al.*, 1997). O'Connor & O'Malley (1989) reported that "in many areas of the country (Ireland) it may not be possible to control bovine tuberculosis without controlling the badger population". Meanwhile, identical strains of *M. bovis* have been found in local cattle and badger populations (Costello *et al.*, 1999) in Ireland.

In Ireland, if it appears that badgers are a likely source of an outbreak, badgers can be removed under licence, in areas around the affected farm. As well, an earlier study based on a badger removal project was conducted in East Offaly County, in order to assess the extent to which the prevailing high levels of BTB in that area might be influenced by badgers (Eves, 1993). More recently, a formal badger removal project in four different areas in Ireland (Cork North, Donegal, Kilkenny and Monaghan) began in November of 1996, with snaring commencing in September 1st 1997. This is referred to herein as the "Four-Area Project" (FAP; Griffin *et al.*, 1998).

The purpose of this study was to utilise the data currently available from these four areas in order to make a descriptive analysis of badger distribution, as well as to investigate whether or not tuberculous badgers had a clustered distribution either within a sett or within a geographic area. Such information could be an important tool in the further design and analysis of strategies for the control of tuberculosis in badger populations.

Materials and Methods

The badger data were obtained in two main files, one for the FAP (n=1702 badgers) and one for the same areas in the period prior to the FAP (n=712 badgers). Records in the first data set were complete, whereas in the second data set 122 badger records were deleted (by us) because they had either no sett identification, and/or no date. The files were merged, using SAS V6.12© (Statistical Analytical System Institute Inc., Cary, NC), giving a combined file of 2292 badgers from 897 setts over the period 1989-1999. The sett identifications used in this study were based on the surveys conducted as part of the FAP and included 3187 setts; 2290 of these had no badgers recorded against them and were deemed to be inactive setts. Prior to 1997, most of the badger examinations for tuberculosis

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were based on a gross *post-mortem* examination only. In the FAP gross *post-mortem* examination, histopathological examination and routine culture of specified organs was were also used (Griffin *et al.*, 1997). Descriptive data were obtained using SAS.

The spatial data were exported to SPLUS© (Insightful Corporation, 1700 Westlake Avenue N, Suite 500, Seattle, WA 98109-3044) and arranged in files for examination of nearest neighbour distances. Basically, for each badger, separately for negative and positive badgers, the distance to the nearest badger of the same health status was calculated. We assumed that if badger families are stable geographically and if *Mycobacterium bovis* is spread by aerosol or contact (direct and/or by vehicles), then tuberculous badgers would be located closer together than negative badgers. When determining distances, badgers from the same sett were assigned a distance of 0.1 metre apart.

To identify if there were areas of sett, or badger, clustering the data were prepared for analysis in SaTScanV 2.1.3 (available as freeware from National Cancer Institute, NIH in the USA, Kulldorff and Nagarwalla, 1995; Kulldorff, 1997). This technique uses the geographic coordinates of each sett as a focal point and then internally creates circles of increasing diameters.

Under the Bernoulli option, it compares the number of infected and non-infected badgers (or setts) within the circle to the numbers outside of the circle, and computes the probability (likelihood ratio) that the number of infected badgers (or setts) within the circle might have arisen by chance alone. The maximum circle size was limited to 10% of the geographic area. The method was extended also to examine for space-time clusters. Using the Poisson option, the number of badgers per sett was included as the denominator for the number of positive badgers.

Results

Descriptive data on setts, by area, are shown in Table 1. The percent of active setts differed by area ($X^2 = 22.9, p < 0.001$); the area in Cork North was the lowest, followed by the areas in Kilkenny, Donegal and Monaghan counties (the latter two do not differ significantly). However these data are somewhat biased in that more badgers were caught in Cork North and Monaghan prior to 1997, but were excluded from the working data set, because of missing sett or date information, than in the other two areas; these include 43, 1, 2 and 76 badgers with missing values for Cork North, Kilkenny, Donegal and Monaghan respectively. If each of these "excluded" badgers came from a different sett, the main conclusion would be that Monaghan had the highest percentage (39%) of active setts, whereas the other three areas had between 26% and 31% active setts.

Approximately 31%, 36% and 41% of all badgers were caught prior to the FAP in Donegal, Kilkenny and Monaghan respectively. Only a small percentage of all badgers (11%) were caught in Cork North prior to the FAP. Cork North had a significantly ($X^2 = 108, p < 0.001$) lower percent of tuberculous badgers caught prior to the formal study (0.6%), then Donegal (16%) and Monaghan (19.6%), whereas Kilkenny had a significantly higher percent of tuberculous badgers caught prior to the formal FAP (51%).

The data in Table 2 show the number of badgers caught from the same sett over a period of years. The diagonal cells indicate the numbers of badgers caught in a calendar year; the off-diagonal row numbers indicate the number of badgers caught in subsequent years from the same setts; for example, 11 badgers were caught at setts in 1990; 1 badger was caught at one of the same setts in 1991. The number of badgers caught at each sett decreased rapidly after the initial snaring activity (e.g. 610 badgers caught in the initial snaring in 1997, 226 badgers at these setts in 1998 and 159 at these same setts in 1999).

The percentages of all badgers caught by year are shown at the column base; the FAP activity had a large impact on these numbers.

The data in Table 3 show the number of tuberculous badgers caught from the same sett over a period of years. The prevalence of tuberculous badgers was 19%. Similar to the data for all badgers, after the FAP began, the number of tuberculous badgers at a given sett decreased annually after the year of initial snaring.

The overall distribution of badgers per sett is shown in Table 4. Over all setts there were 0.7 badgers per sett (S^2=2.5), whereas in active setts the average number was 2.5 badgers (S^2 =4.3). An expected number of badgers per sett was determined assuming a Poisson distribution; there was evidence of significant over-dispersion in the observed data consistent with the variance being greater than the mean. The average number of badgers caught per sett varied from a low of 0.6 in Cork North to a high of 0.9 in Monaghan. In active setts the average number of badgers caught per sett varied from 2.2 in Donegal to 2.9 in Kilkenny. For tuberculous badgers, the mean per sett was 0.13 (S^2 = 0.24) and 1.4 (S^2 = 0.64) for those with positive badgers. A chi-square test confirmed that there was no trend towards an increased percentage positive with number of badgers per sett. The percentage of positive badgers differed among DVOs (X^2= 44.6) from a low of 12.2% in Donegal to a high of 21.2% in Cork North; Kilkenny and Monaghan had a 17% risk.

There were very few setts with more than 2 positive badgers, and the difference between the numbers observed and expected (based on Poisson or binomial distributions) was not large. The data for 1997-1999 were recast in order to identify setts that had badgers caught after the initial snaring success, in contrast to those setts that did not have badgers caught after the initial year of snaring. On inspection the two types of setts were very similar in the first year of snaring, having 19% of tuberculous badgers in setts without a badger being caught after the initial year of snaring, and 21% in those setts that had badgers caught in the second year. Furthermore, the risk of finding a positive badger in the second year (18%) of snaring was very similar to that in the first year (21%; p= 0.25).

The results of the nearest neighbour analysis are shown in Table 5. For each of the four areas the average of the nearest neighbour distances were shorter for the negative than for the tuberculosis positive badgers. For Kilkenny, we examined the data prior to 1997 and the same result was obtained; the negative badgers were, on average, located closer together than the positive badgers. The same interpretation, or one of no difference, was made when the median distances were used.

Using the Bernoulli model and a maximum 10% scanning window size, we found clusters of 18, 4 and 16 setts for Cork North, Donegal and Kilkenny respectively. Monaghan had no cluster of positive setts. At a scanning window of 5% diameter of each study area, 8, 4 and 10 setts were found in the same three areas respectively. Again, Monaghan did not have significant clusters.

Using the Poisson model, Cork North was the only study area that showed a clustered distribution for the number of tuberculous positive badgers among setts after adjusting for the number of badgers per sett. At a scanning window of 10% of diameter of the geographical area, we found one primary cluster composed with 24 setts. A secondary cluster was found with 16 setts (p=0.113). At a scanning window of 5%, 14 setts formed a cluster (p=0.06).

Discussion
The major aim of this study was to provide a descriptive analysis of the distribution of setts, badgers and tuberculous badgers in each area of the FAP. Furthermore, understanding the dynamics and behaviour of an animal population structure, as well the way in which certain events of interest are occurring within that population, in this case *M. bovis* tuberculosis in badgers, can provide important information to help control the disease both in badgers and cattle.

Initially it is important to note that the observed results in this study may depend on the badger capture methods. Most published studies have tended to use the badger capture methods described by Cheeseman *et al.* 1985. In this method badgers are caught using cage traps. Traps are deployed 21 days before trapping and pre-baited with peanuts for the final 14 days to increase trapping efficiency. Longitudinal studies using this method in which animals are captured, marked and released without unduly disturbing the badger population are also reported (Cheeseman, 1988).

The method of badger capture used in Ireland is based on snaring. This method is humane when applied in accordance with the rules outlined by the Department of Agriculture, Food and Rural Development (DAFRD), historically referred to as the Department of Agriculture, Food and Forestry, or DAFF. All setts must be surveyed, the presence of badgers established and snares laid. Legal requirements must also be met (DAFF, 1996). In the FAP, 30-40 snares were located at large active setts. The number of snares used was recorded as well their position. Snares were checked at each visit, and missing snares were reported. Snares were usually placed on a Monday, left for 10 nights, and checked each morning. Occasionally, if the field operator was of the opinion that all of the badgers associated with a sett had not been removed at the end of this period, the snares were left in place for a further 11 nights (Griffin, 1998). From 1998 to the end of 1999, each sett was examined three times per year, twice in the period January to June and once in the period October to December (Griffin, pers. comm.). Based on previous experience, with this method at least 90% of the badgers in an area are removed in a period of 2 years (Eves, 1993).
The four study areas are located in very divergent areas of Ireland and have different agricultural environments. However, badgers appear to have a similar density across these areas. As well, the mean badger density recorded for the four areas (1.99/ km^2) is similar to the density recorded in the midlands (East Offaly) of Ireland with a density of 2.1 badgers / km^2 (Eves, 1993). The mean number of badgers per active sett in the four areas varied from 2.2 in Donegal to 2.9 in Kilkenny. These findings suggest that badgers have a similar distribution throughout Ireland. This differs considerably from the picture in England, where several reports (Anderson and Trewhella, 1985; Cheeseman *et al.*, 1981; Cheeseman *et al.*, 1985; Cheeseman et *al.*, 1989; Krebs, 1997) show that the density of badgers is much higher in the South West of England, in contrast to the near total absence of badgers in more northern areas of England. In addition, the mean density recorded in our study differs from that recorded in Gloucestershire at 23.1 badgers/km^2 during a five-year period between 1981 to 1985.

In contrast to other studies which often present findings on a social group basis, given the snaring methods used in Ireland our focus was on badgers per sett. However, even within Ireland there may be differences in the sett structure. For example, the results obtained by Hammond *et al.*, 2001, from 1989 to 1994, in East county Offaly where the mean number per active sett was 4.5 badgers differs from our result of 2.5 badgers per active sett.

In terms of identifying tuberculosis in the badger, prior to the year 1997 the diagnosis was based solely on gross *post-mortem* examination. In the FAP, culture of all cadavers was used as the basis for diagnosis and this likely accounts for the difference in prevalence over time. Prior to the FAP, 13.7% of the badgers were reported as tuberculous, whereas since then 20.7% of the badgers were found to be positive for tuberculosis. Over the entire study period, 19% of the animals were positive. Among the four areas, Donegal had the lower risk (12.2%) of tuberculous badgers relative to the other 3 areas, whereas Cork North had a significantly higher risk (21.2%) of having tuberculous badgers.

Based on the assumption that badger-to-badger spread is by aerosol, direct contact, or indirect contact with contaminated materials, we expected to observe an increase in the number of infected badgers within the same sett. However, our analyses found no significant association between the number of infected badgers and the total number of badgers in a sett. An increased mortality rate in infected badgers could account for some of these findings. However, the pathogenesis of tuberculosis in badgers appears to be highly variable. Wilesmith, 1991 (cited in Nolan and Wilesmith, 1994) reported that 33% of naturally infected badgers survive at least one year. Thus, given that in the FAP, snaring took place 3 times per year the "loss" of tuberculous badgers would not be expected to have a large impact the results. Thus we have no clear explanation for the near "random" distribution of tuberculosis in badgers within a sett. Moreover, we did not find significant differences in tuberculosis prevalence in badgers caught in the first year of removal and those removed from the same sett in the following year.

As well as within sett distribution, we assessed if the tuberculous animals showed a clustered geographical distribution. Data from Monaghan did not show the presence of any clusters. The other 3 areas each had one significant cluster of tuberculous setts, based on defining infected and non-infected setts while ignoring the actual number of badgers present (Bernoulli model).

Our results indicate that there is insufficient evidence to conclude that tuberculosis in badgers has a clustered geographical distribution. This was not an anticipated finding for us, but could have occurred due to extensive badger movement away from the home (main) sett after removal began. The detailed individual surveying of all setts on a regular basis by trained personnel to identify active setts for snaring may have contributed to this disturbance. However, based on the data from Kilkenny before widespread snaring had taken place, there was no evidence of clustering. This might suggest that the badger populations in Ireland are not yet stable and the idea that most badgers stay within the same sett area may be incorrect. As the availability of high quality pasture land in an area increases, so too does the badger density (Kruuk and Parrish, 1982; Hammond *et al.*, 2001); thus, with ongoing improvement of agricultural lands, the badger populations may be increasing and very mobile filling the new niche areas.

Conclusion

Bovine tuberculosis is widely distributed among badgers in diverse areas in Ireland. However, the disease among badgers does not have a defined geographically clustered distribution. Future studies should continue to investigate these counter-intuitive findings.

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Table 1. The number of setts with and without badgers by area.

DVO	Active setts	Inactive setts	Total	% Active setts
Cork North	260	848	1108	23.5
Donegal	189	419	608	31.1
Kilkenny	204	526	730	27.9
Monaghan	244	497	741	32.9
Total	**897**	**2290**	**3187**	**28.4**

Table 2. Number of badgers caught by year, and in subsequent years from the same sett.

Calendar year	**1989**	**1990**	**1991**	**1992**	**1993**	**1994**	**1995**	**1996**	**1997**	**1998**	**1999**
Index year											
1989	7	0	0	0	0	0	0	1	0	0	0
1990		11	1	0	0	0	0	0	0	0	0
1991			81	0	0	0	0	3	0	4	0
1992				31	0	0	0	1	1	0	0
1993					17	17	0	0	2	7	1
1994						44	12	9	4	4	1
1995							129	57	5	4	5
1996								229	32	22	11
1997									610	226	159
1998										717	226
1999											416
										Total	**2292**
Percent by year	0.3	0.5	3.5	1.4	0.7	1.9	5.6	10.0	26.6	31.3	18.2
Cumulative percent	0.3	0.8	4.3	5.7	6.4	8.3	14.0	24.0	50.6	81.8	100.0

Table 3. Number of tuberculous badgers caught by year, and in subsequent years from the same sett.

Calendar year	**1989**	**1990**	**1991**	**1992**	**1993**	**1994**	**1995**	**1996**	**1997**	**1998**	**1999**
Index year											
1989	0	0	0	0	0	0	0	0	0	0	0
1990		0	0	0	0	0	0	0	0	0	0
1991			3	0	0	0	0	0	0	0	0
1992				5	0	0	0	0	0	0	0
1993					2	0	0	0	0	0	0
1994						2	0	0	0	0	0
1995							16	1	0	0	0
1996								48	0	1	1
1997									110	16	7
1998										169	20
1999											80
										Total	**435**
Percent by year	0.0	0.0	0.7	1.1	0.5	0.5	3.7	11.0	25.3	38.9	18.4
Cumulative percent	0.0	0.0	0.7	1.8	2.3	2.8	6.4	17.5	42.8	81.6	100.0

Table 4. Overall distribution of badgers in the four-area project locations.

No. of badgers per sett	**No. of setts**	**No. of badgers observed**	**Number expected**
0	2290	0	1552.6
1	386	386	1116.6
2	175	350	401.5
3	131	393	96.2
4	75	300	17.3
5	48	240	2.5
6	34	204	0.3
7	19	133	0.0
8	10	80	0.0
9	5	45	0.0
10	4	40	0.0
11	6	66	0.0
12	0	0	0.0
13	1	13	0.0
14	3	42	0.0
Total	**3187**	**2292**	

Table 5. Nearest neighbour badger-badger distances, by tuberculosis status, in the 4 areas, 1989-98.

	CK[1]_NEG	CK_POS	DL[2]_POS	DL_POS	KK[3]_NEG	KK_POS	KK96_NEG	KK96_POS	MN[4]_NEG	MN_POS
Minimum	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
1st Quarile	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Mean	122.8	346.9	165.8	902.8	603.3	603.3	146.7	286.2	159.8	764.5
Median	0.1	0.1	0.1	842.5	0.1	0.1	0.1	0.1	0.1	465.4
3rd Quarile	0.1	616.4	113.2	1522.3	1265.9	1265.9	0.1	0.1	0.1	1171.3
Maximum	1740.3	2138.3	1815.8	2585.7	2886.3	2886.3	3573.6	2393.5	2268.2	4282.8
Total No.	477.0	178.0	235.0	41.0	100	100	120	46	362.0	86.0
Std. Deviation	302.9	516.6	369.8	853.3	808.8	808.8	469.0	638.6	392.6	967.6

1 County Cork North_Neg indicates no tuberculosis; _Pos indicates tuberculosis present

2 County Donegal

3 County Kilkenny Overall and also just for year 1996

4 County Monaghan