# The efficacy of badger population reduction by controlled shooting and cage trapping, and the change in badger activity following culling from 27/08/2013 to 28/11/2013

Report to Defra



6<sup>th</sup> February 2014

# **Executive summary**

- 1) Government policy on control of bovine tuberculosis requires those licensed to cull badgers to remove at least 70% of the population living within the licensed areas within a six-week period without causing a local extinction.
- 2) Defra required an appraisal of delivery of the policy in two pilot areas (West Gloucestershire and West Somerset) according to three criteria: efficacy, safety and humaneness.
- 3) The efficacy and humaneness of badger removal during the first six-week period has been reported previously, and those reports were externally audited and reviewed and approved by the Independent Expert Panel. At the end of the six week cull period both pilot areas were granted an extension in order to remove more badgers.
- 4) Here we present the results of monitoring work commissioned by Defra to evaluate the efficacy of industry-lead badger culling by controlled shooting and cage trapping, and to assess the change in badger activity following culling up to the end of the extension periods.
- During the entire period of industry-lead culling in 2013, a total of 924 badgers were reported culled under licence in the Gloucestershire pilot area, 610 of which were taken by controlled shooting. The total number reported culled in the Somerset pilot area was 955, 439 of which were taken by controlled shooting.
- 6) We used two methods to estimate the proportion of the pilot area populations that these culls represented. Both methods were reviewed, finalised and approved by the Independent Expert Panel, appointed by Defra, before commencing data collection. The primary method (cull sample matching) compared the genetic profiles of all ear tips from culled badgers with genetic profiles of hair samples collected from hair traps at random locations. The secondary method (capture-mark-recapture) used observations of the frequency with which each unique genetic profile was sampled by hair traps to estimate population size. The numbers of badgers culled were divided by the population estimates for the respective area to derive secondary estimates of the proportion of the populations removed.
- 7) We found the secondary method less reliable than the primary method, producing a confidence interval of the proportion of the population removed in Somerset that was not credible: The upper limit of the estimate was well above 100% of the population, in spite of abundant signs of badger activity following the cessation of culling. The secondary method was also potentially sensitive to interference (e.g. sabotage of hair traps or fraudulent submission of carcasses).
- 8) Hair traps were located around active badger setts and along runs that crossed the boundary of land that was not participating in the cull within approximately 50 1km by 1km squares. We collected 615 hair samples representing 233 individuals from 562 hair traps in Gloucestershire and 801 hair samples representing 216 individuals from 581 hair traps in Somerset.
- 9) From the primary method of efficacy estimation the proportion of the population removed by both controlled shooting and cage trapping during entire cull period in Gloucestershire was estimated to lie somewhere between 43.0 and 55.7% (95% confidence interval), with between 29.2 and 42.2% removed by controlled shooting alone.
- 10) From the primary method of efficacy estimation the proportion of the population removed by both controlled shooting and cage trapping during the entire cull period in Somerset was

estimated to lie somewhere between 37.0 and 50.9 (95% confidence interval), with between 14.2 and 25.3% removed by controlled shooting alone.

- 11) The number of active latrines at setts declined significantly in both pilot areas following culling (from 68 to 15 in Gloucestershire and from 38 to 10 in Somerset). However, the number of active entrance holes did not change significantly in Somerset (from 65 to 68), and increased significantly in Gloucestershire (from 73 to 145).
- 12) We conclude that in neither area was at least 70% of the population removed. Substantially less than 70% was removed in both areas by shooting and cage trapping. Badgers were highly likely to persist in both areas following culling.

# Contents

Exe	cutive summary	2
1.	Introduction	5
2.	Methods	6
3.	Results	7
4.	Discussion1	1
5. A	ppendix 1. Estimates of the population size in the Gloucestershire and Somerset pilot areas by	
hair	trapping and sett survey1	4
S	ummary1	4
Ν	1ethod1	4
R	esults1	5
	Active setts1	5
	Hair trap and sample numbers1	6
	Estimates of the number of badgers1	6
C	iscussion1	7
6.	Appendix 2. Quality assurance1	8
Ν	1ethods1	8
S	taff training and auditing1	8
Р	eer Review1	8
7	. Appendix 3. References1	9

# **1.** Introduction

- 1.1. As set out in Defra's policy statement published in December 2011<sup>1</sup>, the Government's policy is to allow controlled culling and vaccination of badgers in areas of high incidence of bovine TB in cattle in a carefully regulated way for the purpose of controlling the spread of the disease. The requirements of the policy are set out in Defra's Guidance to Natural England: "Licences to kill or take badgers for the purpose of preventing the spread of bovine TB under section 10(2)(a) of the Protection of Badgers Act 1992" (Defra publication PB13692).
- 1.2. The Guidance sets out that in the first year of culling, a minimum number of badgers must be removed during an intensive cull which must be carried out throughout the land to which there is access, over a period of not more than six consecutive weeks. This minimum number should be set at a level that in Natural England's judgement should reduce the estimated badger population of the application area by at least 70% (para 10(c)(i) and (ii) of the Guidance).
- 1.3. The guidance also sets out that Natural England should aim to ensure that culling will "not be detrimental to the survival of the population concerned" within the meaning of Article 9 of the Bern Convention on the Conservation of European Wildlife and Natural Habitats. For that purpose Natural England should determine appropriate area-specific licence conditions, and set a maximum number of badgers to be removed from the licence area (para 12 of the Guidance).
- 1.4. Two pilot areas in southwest England were selected to test whether industry-lead badger culling, licensed in accordance with this policy, could be undertaken effectively, humanely and safely. During the summer of 2013 licensed badger culling operations were implemented in these areas.
- 1.5. After an initial six-week culling period, licenses were extended to allow continued badger removal for a further two weeks in the West Somerset pilot area and a further eight weeks in the West Gloucestershire pilot area. The Somerset extension was completed but the Gloucestershire extension was halted two weeks before the license was due to expire.
- 1.6. Defra commissioned a programme of independent monitoring in order to estimate the proportion of the badger population from within the outer boundary of each pilot area that was removed by industry-led culling operations. A major challenge that this presented was identifying a monitoring method that was robust to interference, while offering estimates with a high degree of accuracy (see Donnelly and Woodroffe, 2012). High accuracy is required so that it can be confidently concluded whether industry-led culling removed 70% of the population, significantly more than 70% or significantly less.
- 1.7. Here we present the results of independent monitoring of the efficacy of badger removal during the pilots and including the extension periods. The underlying assumption of the primary analysis was that the proportion of badgers individually identified prior to culling that were subsequently observed in the cull returns provided an unbiased and relatively precise estimate of the proportion of the pilot area population that had been culled. This method of quantifying

<sup>&</sup>lt;sup>1</sup> https://www.gov.uk/government/uploads/system/uploads/attachment\_data/file/69463/pb13691bovinetb-policy-statement.pdf

cull efficacy was agreed with the Independent Expert Panel.

- 1.8. To be consistent with the report on the efficacy of badger culling during the first six weeks, a separate analysis requested for that report by the Independent Expert Panel was also used. This followed a more traditional capture-mark-recapture approach to provide an estimate of the population prior to the cull against which to compare the number of badgers reported culled. We analysed the frequency with which individual badgers were hair trapped in order to estimate the size of the population immediately before culling. This was compared with the number of badgers culled to estimate the proportion of the population removed by culling. It was expected *a priori* that this approach would produce estimates with greater confidence intervals.
- 1.9. To assess whether the requirement had been achieved to not be detrimental to the survival of the population concerned, we monitored activity at badger setts before and after culling in order to measure the change in activity caused by culling and to confirm whether badgers persisted in the pilot areas.

## 2. Methods

- 2.1. We aimed to sample as many badgers as possible in 50 1km x 1km cells selected at random from a grid covering approximately 300km<sup>2</sup> in each of the two pilot areas. This aimed to provide a representative sample, covering 17% of cells and 12% of the population, assuming an even distribution and 70% trapping/sampling efficiency.
- 2.2. We deployed hair traps (short lengths of barbed wire suspended between two short stakes or trees) at all active setts and along badger runs associated with setts and adjacent to non-participating land, following the procedures described by Frantz *et al.* (2004). Hair samples were removed from all barbs daily for 18 days and each sample was stored in a uniquely coded plastic ziplock bag containing a sachet of desiccant. Following sample collection, hair traps were de-contaminated by flaming with a cigarette lighter. DNA from the bulb of a randomly-selected single guard hair from each hair trap was amplified and sequenced to provide a unique genetic finger print for each badger. DNA was extracted from hairs using a suspension of chelex resin (Frantz et al. 2004). The purified DNA was genotyped using 10 loci (Dawnay et al. 2008) on an Applied Biosystems *3130xl* Genetic Analyzer. Repeat samples from individuals were identified on the basis of all 10 loci matching (probability of identity approximately 1 in 100,000 based on observed allele frequencies).
- 2.3. If a profile including all 10 loci was not isolated from a hair, a further guard hair from the same sample was sequenced. In circumstances where no guard hairs were present within a sample all under-fur was pooled from the sample and these were amplified and sequenced. This allowed assessment of whether the sample constituted a single individual or multiple individuals. Only samples from single individuals were used in the estimates of efficacy. By sampling in this way across the 50 1km x 1km squares in each pilot we built a list of individual profiles to constitute the 'marked' population.

- 2.4. Tissue samples (ear tips) removed from every culled and retrieved badger were submitted by industry contractors in sealed tubes containing 5ml of ethanol for DNA profiling.
- 2.5. Profiles of culled badgers were matched to profiles of the 'marked' population to estimate culling efficacy as the proportion of the marked population that was identified in the cull in each pilot area (cull sample matching see Appendix 1 in AHVLA 2014 for details of the analysis).
- 2.6. Profiles were also used to identify the frequency with which each hair-trapped badger was sampled in order to estimate pre-cull population size using a capture-mark-recapture approach (see Appendix 1 in AHVLA 2014 for details on how the impact of incomplete genetic profiles was addressed and Appendix 1 of the current report for details of the main analysis and results). The number of badgers culled, identified from the unique profiles submitted for genetic analysis, was compared with this population estimate in order to estimate the proportion of the population culled.
- 2.7. Badger activity was monitored by placing small sticks over unobstructed (potentially active) sett entrance holes of all setts that had been hair trapped, and at as many setts as possible, given time constraints, that were not hair trapped. Setts were re-surveyed three days after sticks had been placed, and the number of entrance holes at which sticks were disturbed and the number that were undisturbed were recorded. The number of active latrines from the centre to within 5m of the outermost entrance holes of each sett was also recorded. This exercise was undertaken approximately two weeks before badger control started and approximately two weeks after it ended. The statistical significance of the change in the number of active entrance holes and active latrines per sett from before culling started to after it had ceased was estimated using a Wilcoxon test for matched pairs.

# 3. Results

- 3.1. All ranges quoted in this section are 95% confidence intervals.
- 3.2. From cull sample matching (the primary method) we estimate that the cull in Gloucestershire removed between 43.0 and 55.7% of the population by both cage trapping and shooting (Table 3.1). This is not consistent with removal of at least 70% of the population.
- 3.3. The number reported removed by shooting was 610 and was 314 by cage trapping, giving an estimated shooting efficacy of 29.2 to 42.2% from cull sample matching and 18.5 to 43.1% from capture-mark-recapture.
- 3.4. Dividing the number of badgers culled in Gloucestershire by the proportion estimated to be culled by cull sample matching (Table 3.1) gives a pre-cull population estimate for Gloucestershire of 1658 to 2151.
- 3.5. The Gloucestershire population estimate based on the capture-mark-recapture analysis and a survey of setts in 2012 was between 1416 and 3304 badgers immediately before the cull (Appendix 1). Removal of 924 badgers (Table 3.1) from this population estimate results in an

estimated removal of 28.0% to 65.3%. This is not consistent with removal of at least 70% of the population.

- 3.6. From cull sample matching we estimate that the cull in Somerset removed between 37.0 to 50.9% of the population by both cage trapping and shooting (Table 3.1). This is not consistent with removal of at least 70% of the population.
- 3.7. The number reported to be removed by shooting was 439 and was 516 by cage trapping, giving an estimated shooting efficacy of 14.2 to 25.3% from cull sample matching and 23.2 to 52.2% from capture-mark-recapture.
- 3.8. Dividing the number of badgers culled in Somerset by the estimated proportion culled (Table 3.1) gives a pre-cull population estimate for Somerset of 1876 to 2584 with 95% confidence.
- 3.9. The Somerset population estimate based on the capture-mark-recapture analysis and a survey of setts in 2012 was between 841 and 1898 immediately before the cull (Appendix 1). Removal of 955 badgers (Table 3.1) from this population estimate results in an estimated removal of 50.4% to 113.6%. From the capture-mark-recapture method, the possibility that 70% or more of the population was removed cannot be excluded, but neither can we conclude that at least 70% was removed. However, from the more reliable cull sample matching approach we can conclude removal of less than 70% because we estimate that between 37.0 and 50.9% was removed.
- 3.10. The robustness of the efficacy estimates can be assessed from whether the assumptions on which they were based were met or violated (Table 3.2).
- 3.11. In Gloucestershire 92 setts were monitored for entrance hole activity before and after culling. Of these 36 experienced an increase in the number of active entrance holes following culling, 41 experienced no change (although 38 of these were inactive before and after culling) and 15 experienced a decline. The number of active entrance holes summed across all 92 setts was 73 before culling and was 145 after culling. This represents a statistically significant increase in sett activity (P = 0.0012).
- 3.12. In Gloucestershire latrines at 94 setts were monitored before and after culling. Of these six experienced an increase in the number of active latrines following culling, 55 experienced no change (53 of these had no active latrines before or after culling) and 33 experienced a decline, of which only two retained active latrines. The number of active latrines summed across all 94 setts was 68 before culling and was 15 after culling. This represents a statistically significant decline in latrine activity at setts (p = 0.00016).
- 3.13. In Somerset 105 setts were monitored for entrance hole activity before and after culling. Of these 22 experienced an increase in the number of active entrance holes following culling, 64 experienced no change (57 of these were inactive before and after culling) and 19 experienced a decline. The number of active entrance holes summed across all 105 setts was 65 before culling and was 68 after culling. This represents no significant change in sett activity (p = 0.6034).

3.14. In Somerset latrines at 109 setts were monitored before and after culling. Of these five experienced an increase in the number of active latrines following culling, 89 experienced no change (87 of these had no active latrines before or after culling) and 15 experienced a decline of which only two retained active latrines. The number of active latrines summed across all 109 setts was 38 before culling and was 10 after culling. This represents a statistically significant decline in latrine activity at setts (p = 0.007).

	West	West
	Gloucestershire	Somerset
Number of 1km by 1km grid squares with hair traps (number of original, randomly-selected survey squares in parenthesis)	71 (40)	78 (46)
Number of setts hair trapped	93	130
Number of hair traps at setts	408	444
Number of hair traps away from setts	114	137
Total number of hair samples (counted as one per hair trap per night)	615	801
Number of unique hair profiles	233	216
Number of badgers reported culled	924	955
Number reported shot	610#	439 <sup>#</sup>
Number reported cage trapped	314	516
Number of ear tip samples received	926*	965*
Number of unique ear tag-genetic profile combinations from ear tips	924	955
Number of matches among ear tips (one match = a pair of matching ear tips)	3	2
Number of badgers identified from ear tips	921	953
Number of ear tips matching hair profiles	113	87
Number of matches shot with rifles and shotguns	77	41
Number of matches cage trapped	36	46
Estimated Cull efficacy (proportion of population removed by cull using both methods; 95% confidence interval)	0.430 to 0.557	0.370 to 0.50

**Table 3.1.** Summary of survey effort and measurements contributing to the cull sample matchingestimates of efficacy for each pilot area.

<sup>#</sup>One badger in Gloucestershire and 11 in Somerset were shot with shotguns.

\*Two samples were sent in duplicate from Gloucestershire and five from Somerset. Two additional samples were from road-killed badgers. Duplicates and road-killed badgers were subsequently removed from all analyses.

Note: 1) Cull efficacy cannot be estimated by dividing the number of ear tips matching hair profiles by the number of unique hair profiles because these figures are not corrected for false negatives or population churn (Appendix 1 in AHVLA 2014).

Table 3.2. Assumptions underpinning estimates of efficacy

Assumption	Evidence
Up to 10% of the population (including marked animals) within 3km of the outer boundary of the pilot area move out of the area and are replaced by badgers moving in.	Rogers <i>et al.</i> (1998) found that up to 10% of badgers moved from their social group in Woodchester Park, and moved up to 3km away. Therefore, simulating up to 10% replacement is very conservative (10% is worst case) and may have resulted in slightly wider confidence intervals.
Equality in the probability of being culled between individual hair-trapped and non-hair trapped badgers.	The proportions of the marked population that was taken by cage trapping and by shooting were much the same as for the un-marked population. This equivalence between marked and un-marked badgers for two different techniques is consistent with the assumption. Failure to meet this assumption would lead to bias.
Receipt of a genetic sample from every badger culled.	All badgers in the cull data base were represented by at least one sample received by the laboratory. Badgers culled prior to hair trapping or from elsewhere would have no effect on the efficacy estimates. Failure to submit genetically marked badgers would bias the efficacy estimates low.
The hair trapped sample population was representative of the wider population within each area.	Hair trapped badgers were a random sample from a random sample of selected locations. A non-random sample could lead to biased estimates.
Culling operations were not spatially correlated (positively or negatively) with hair traps.	Cull returns were not associated with hair traps (Appendix 5, AHVLA 2014).
The additional effect of spatial variation in culling efficacy was small relative to other sources of variation.	The mean of proportions of the marked population returned per sample square and the total proportion returned are approximately equivalent. Differences may mean that our confidence intervals were too tight.

## 4. Discussion

4.1. The culling of badgers is a highly sensitive issue, which tends to polarize opinions. We anticipated that activities employed to provide evidence to evaluate Government policy on badger population control would experience interference. Consequently we developed a rapid, robust approach to estimate the proportion of the badger population that was removed by industry-lead culling operations in each of the two pilot areas. We aimed to sample badgers in 50 1km x 1km cells selected from a grid covering approximately 300km<sup>2</sup> in each of the two pilot

areas. This aimed to provide a representative sample, covering 17% of cells and 12% of the population, assuming an even distribution and 70% trapping/sampling efficiency.

- 4.2. The primary method of efficacy estimation (cull sample matching) required far fewer assumptions and data manipulations to produce estimates of cull efficacy and population size than did the secondary method (capture-mark-recapture). Nevertheless, both estimates relied on reliable reporting of the number of animals culled (including provision of samples from all culled badgers) and several other assumptions.
- 4.3. Sampling of badgers prior to the start of culling was from within and around 1km squares that were randomly selected from all those lying wholly within the outer boundary of each pilot area. From the number of unique genetic profiles collected by hair sampling, we might have sampled somewhere between 8 and 13% of the population in pilot areas at least once. It is reasonable to assume that the samples constitute a representative sample of the population.
- 4.4. The key assumption affecting the accuracy of the primary efficacy estimates is that there was no spatial association between culling operations and our hair traps. If contractors had invested more effort at hair trapped sites, our primary efficacy estimates would be biased high. If contractors invested less effort at hair trapped sites, our estimates would be biased low. We investigated whether the density of badgers culled was different in areas with hair traps compared with areas without hair traps and found no significant differences (Appendix 5 in AHVLA 2014). This does not preclude the possibility of an effect, but if there was an effect it was unlikely to be large. Therefore we conclude that the efficacy estimates were not significantly biased by any factors that were more prevalent at hair trapping sites.
- 4.5. Analysis of the proportions of marked badgers returned from each survey square indicated that variation in returns per square contributed very little additional uncertainty to the efficacy estimates over-and-above that associated with other sources of uncertainty, such as sample sizes, genetic sample mis-match rates, and replacement of genetically marked badgers with unmarked badgers. Therefore, it is likely that the confidence limits of our estimates are appropriate (*i.e.* have good coverage; Appendix 2 in AHVLA 2014).
- 4.6. In the Gloucestershire pilot area a total of 924 badgers were reported removed during the entire culling period, with an estimated cull efficacy of somewhere between 43.0 and 55.7% from cull sample matching. The number reported removed by shooting was 610 and by cage trapping was 314, giving an estimated shooting efficacy of 29.2 to 42.2%.
- 4.7. From the capture-mark-recapture analysis the estimated total cull efficacy in Gloucestershire was somewhere between 28.0 and 65.3% and was 18.5 to 43.1% for shooting alone. The minimum cull target for Gloucestershire was 2856 from an estimated population of 2657 to 4079 badgers (Anon. 2013a).
- 4.8. In the Somerset pilot area a total of 955 badgers were reported removed during the entire culling period, with an estimated cull efficacy of 37.0 to 50.9% from cull sample matching. The number reported to be removed by shooting was 439, and by cage trapping was 516, giving an estimated shooting efficacy of 14.2 to 25.3%.

- 4.9. From the capture-mark-recapture analysis the estimated total cull efficacy was somewhere between 50.4 to 113.6% and was 23.2 to 52.2% for shooting alone. The minimum cull target for Somerset was 2081 from an estimated population of 1972 to 2973 badgers (Anon. 2013a).
- 4.10. Confidence intervals produced using cull sample matching were tighter than those commonly produced when estimating the size of wildlife populations at similar scales to the pilot areas (see for example Parrott *et al.* 2012). This is in part due to the large sample sizes provided by the cull (approximately 29 to 51% of the population was in the final 'sample'). Confidence intervals produced using capture-mark-recapture were consistent with the width of those commonly produced when estimating the size of wildlife populations at similar scales to the pilot areas (see for example Parrott *et al.* 2012). Cull sample matching was less prone to interference, required fewer assumptions and offered greater estimate precision than did capture-mark-recapture. Therefore, our conclusions are based on the estimates derived from cull sample matching.
- 4.11. Concomitant with the removal of badgers from the pilot areas was an increase in the number of active entrance holes in Gloucestershire and no change in Somerset and a decrease in the number of active latrines at setts in both areas. Since it is clear that large number of badgers were removed from both populations by culling, the number of active sett entrance holes may not provide a reliable indicator of population change immediately following culling. Moreover, the substantial decline in the number of active latrines was inconsistent with the estimated proportion of the populations removed, so may equally provide an unreliable indicator of change immediately following culling. Nevertheless, the persistence of signs of badger activity provides evidence for the persistence of badger populations following culling within both pilot areas.
- 4.12. From the results presented above we conclude that industry-lead controlled shooting of badgers during the entire culling period (including the initial six week period and the extensions) did not remove at least 70% of the population inside either pilot area. In both areas significantly fewer than 70% were removed by controlled shooting. The combined approach of controlled shooting and cage trapping also did not remove at least 70% of the population inside either pilot area; substantially fewer than 70% were removed in both areas. Populations of badgers were highly likely to persist within both pilot areas following culling.

# **5. Appendix 1.** Estimates of the population size in the Gloucestershire and Somerset pilot areas by hair trapping and sett survey

#### **Summary**

- 5.1 Here we describe how sett surveying, hair trapping and genetic profiling were used to estimate the badger population sizes within the pilot areas during 2013.
- 5.2 Estimates of the average number of badgers per active sett for each area during each year were multiplied by the estimates of the number of active setts within the pilot areas to produce estimates of population size with a measure of uncertainty.
- 5.3 Surveys to estimate the number of active setts were undertaken in 2012. These 2012 results were also used in 2013 under the assumption that the number of active setts was the same in 2013 as in 2012. Estimates of the number of badgers per active sett were obtained in 2013 using hair trapping data collected to measure the efficacy of badger culling.
- 5.4 Population size was estimated between 841 to 1898 in Somerset and 1416 to 3304 in Gloucestershire, with 95% confidence.

#### **Method**

- 5.5 Each study area was overlaid with a grid of 1km squares, and a number of these were randomly selected for hair trapping. Not all land within each randomly-selected square was accessible so accessible land in adjacent squares was used when this was the case. Hair trapping was undertaken from 6 to 30 August 2013 for two purposes:
  - a) To genetically identify as many badgers as possible to provide a cull efficacy estimate from the number of known badgers returned by the cull.
  - b) To provide an estimate of the pre-cull population size by an analysis of the mark-recapture profile of badger identities.
- 5.6 The primary objective required the identification of as many individual badgers as possible, whereas the secondary objective required the identification and re-sampling of badgers within defined sampling area associated with setts. Sett location and activity status was assigned during a survey of setts within the survey areas undertaken from 17 August to 21 September 2012 in Gloucestershire and 5 August to 1 October 2012 in Somerset. The purpose of the survey was to provide an estimate of the number of active setts in each of the pilot areas. The mean number of active setts per nominal 1km square was used to estimate the number of active setts in the pilot areas and the observed between-square variation in the number of setts was used to estimate the uncertainty of the estimate. The survey included an expert review of photographs of putatively active holes at setts assessed to be active by surveyors, in order to exclude from further analysis any setts that appeared to be mis-identified, and to apply a correction to the estimated number of active setts in the pilot areas.
- 5.7 Hair traps consisted of a loop of barbed wire fixed to wooden stakes, on fence-lines or natural objects such as logs and suspended approximately 20cm above the ground. They were placed at active setts and areas of obvious badger activity such as runs crossing land boundaries and at latrines. Each hair trap was given a unique alphanumeric identifier and its location taken with a GPS unit. Samples from hair traps were collected on each day for up to 18 days. During the first three days hair traps were moved or additional hair traps were placed to maximise sample

collection. Only samples from the last 15 days were used to estimate population sizes. Each sample consisted of hairs from a single tuft. Where a hair trap had tufts on multiple barbs all tufts were taken and stored in separate plastic ziplock bags.

- 5.8 Hair samples from a hair trap were placed into a uniquely coded bag which was matched with the time of collection and trap location. DNA was extracted from hairs using a suspension of chelex resin (Frantz et al. 2004). The purified DNA was genotyped using 10 loci (Frantz et al. 2004; Dawnay et al. 2008) on an Applied Biosystems 3130xl Genetic Analyzer. Repeat samples from individuals were identified on the basis of sufficient loci matching to provide a probability of identity approximately 1 in 100,000 based on observed allele frequencies).
- 5.9 The profile of captures expressed as the number of individuals hair-trapped once, twice, three times etc was analysed with the mark-recapture CAPWIRE package in R. (Miller et al. 2005) using its 'two intrinsic rates model' (TIRM). This model is designed to estimate population size from sparse datasets where the majority of individuals are only captured once. It is particularly suitable for estimating population size of badgers, which vary in capture probability according to age/sex class (Valiere 2002).
- 5.10 A second order Monte Carlo model was used to estimate the effect of missed matches (as described in Appendix 1 of AHVLA 2014) and the uncertainty associated with TIRM estimates via a parametric bootstrap.
- 5.11 The estimates are based on the assumption that the populations were closed both geographically and demographically (*i.e.* no dispersal or mortality) over the trapping periods, with two sub-sets of individuals that have higher and lower trap probabilities due to some individuals being more easy to sample than others. We assumed closure was met by the short sampling period and by locating hair traps close to setts because badgers tend to defend territories within which their setts are located. The subsequent trapping probability of an individual is assumed not to be affected by being previously hair trapped. The model is a two-probability mixture model version of Chessel's equation (Valiere 2002; Frantz and Roper 2006).
- 5.12 The sett at which a hair trap was deployed was recorded or else it was recorded as not associated with a sett. Population estimates required estimation of the number of badgers per active sett from those individuals closely associated with active setts at which hair traps were deployed. Consequently, data from hair traps deployed away from setts (deployed as part of the effort to estimate the proportion of the population removed by culling) were excluded from this analysis.
- 5.13 Estimates of the average number of badgers per active sett were converted into estimates of the number of badgers within each of the pilot areas by multiplying by the estimated number of active setts in each pilot area as described in Anon. (2013b).

#### **Results**

#### **Active setts**

- 5.14 All sett surveying was undertaken during 2012. In Gloucestershire 183 setts were identified as active by observers, but 10.3% of the setts assessed during the photograph review were judged as misidentified, so were excluded from further analysis.
- 5.15 In Somerset 275 setts were surveyed as active, but 27.3% of the setts assessed during the photograph review were judged as misidentified as active badger setts, so were excluded from further analysis.

#### Hair trap and sample numbers

- 5.16 A total of 852 hair traps associated with setts yielded 758 putative samples, of which 535 produced viable genetic profiles, representing 274 individuals (Table 5.3). The average number of alleles per locus was 9.24 (range 4 10). Genetic profiles were obtained from 57 of the 93 active setts trapped in Gloucestershire and 60 of the 130 active setts trapped in Somerset. Despite failure to collect hair samples from many setts, the inability to sample every individual from every sett means it is not possible to conclude that the identification of these setts as active was incorrect.
- 5.17 Data from five setts in Gloucestershire and three setts in Somerset were excluded from analysis because their hair traps were operative for fewer than six days before being disturbed by third parties.

Table 5.3. Hair trapping effort, numbers of samples, genetic profiles and individuals contributing to the estimates of the number of badgers per sett

Location	Hair traps	Average trap/sett	Putative samples	Genetic profiles	Individuals
Gloucestershire 2013	408	4.4	359	265	150
Somerset 2013	444	3.4	399	270	124

#### Estimates of the number of badgers

5.18 In both Gloucestershire the ratio of individuals caught once to those caught on multiple occasions was 1.7:1 and in Somerset it was 1.6:1 (Table 5.4). Table 5.5 presents the estimates of the number of badgers per active sett. Table 5.6 presents estimates of the population of badgers in the pilot areas during 2013.

Area	Number of individuals by number of times sampled															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Glos	97	29	9	8	3	1	3	1	0	0	0	0	0	0	0	0
Som	73	18	7	8	3	4	0	2	2	0	0	0	1	0	0	1

Note: Data in this table represent one possible iteration of the Monte Carlo sample, and should not be used to estimate population size in isolation.

The rate of missed matches was estimated to be 7 per 186 observed matches in Somerset. In Gloucester the rate of missed matches was estimated to be 10 per 111 observed matches.

Table 5.5. TIRM estimates of the average number of badgers per sett at setts trapped for at least 6 days.

Location Setts		Badgers per sett	(95 CI)		RSE (%)		
Gloucester	87	3.381	3.138	4.530	0.1621		
Somerset	127	1.584	1.496	2.101	0.1726		

Table 5.6. Population estimates for the two pilot areas

	Gloucestershire	Somerset
Survey year	2013	2013
Total area (km <sup>2</sup> )	311.00	256.05
Squares (nominal km <sup>2</sup> ) surveyed	80	67
Area surveyed (km <sup>2</sup> )	77.96	62.82
Active setts found	183	275
RSE between location(%)	14.51	11.39
Photo reliability	89.7	72.7
RSE photo reliability (%)	2.3	3.5
Mean badgers per sett	3.381	1.584
RSE badgers per sett (%)	0.1621	0.1726
Average density (80% one-tailed interval) (number per km <sup>2</sup> )	7.12 (5.80 to 8.34)	5.04 (4.14 to 5.88)
Population range as 95% confidence interval	1416 to 3304	841 to 1898

RSE = Relative Standard Error

#### **Discussion**

- 5.19 Two badgers in Somerset were sampled a surprisingly high number of times (13 and 16 times). The effect of these badgers on the estimate of numbers of badgers per sett has been quantified before (AHVLA 2014), and was found to be small. Therefore, these badgers were not excluded from the analysis.
- 5.20 Similarly, the effect of the missed match rate on estimates of the number of badgers per sett has also been quantified before (AHVLA 2014) and found to be small. Therefore, the missed match rate was retained in the analysis.
- 5.21 Using the same CMR approach AHVLA (2014) estimated that the pre-cull population in Gloucestershire was between 1394 and 3242 badgers, and for Somerset the pre-cull population was between 850 and 1905. The new estimates are sufficiently similar as to constitute the same estimates. The very small differences were due to the slightly larger database of genetic profiles (from the addition of ear tips taken during the extensions), which altered the allele frequencies, hence influencing the assessment of the frequency with which hair samples matched each other.

# 6. Appendix 2. Quality assurance

#### **Methods**

6.1 All survey methods, data to be collected and data analysis methods were reviewed, finalised and approved by the Independent Expert Panel before data collection started.

#### Staff training and auditing

6.2 Most staff engaged in hair trapping activities were experienced hair trappers who received one day of training in the use of the relevant standard operating procedures. Less experienced staff were paired with experienced staff during field data collection. In addition, staff received two days of training and 5 days of practical experience in sett surveying and hair trapping before starting hair trapping in the field. An independent assessment of surveying, data recording and manipulation, sample handling and laboratory processes was undertaken during the initial sixweek cull period by an experienced external auditor. Errors identified by the auditor were corrected before final estimates were calculated. One member of the Independent Expert Panel reproduced all of our calculations for the initial six-week cull period and the extension periods from our raw data. Discrepancies were explained or corrected as appropriate before production of final results.

#### **Peer Review**

6.3 Numerically competent staff and members of the Independent Expert Panel selected by Defra reviewed the assumptions used to underpin analyses, calculations and interpretation. Senior members of staff and internal auditors reviewed this report. Recommendations to improve data collection, analysis and reporting were followed. The reviewers accepted the final version of this report.

# 7. Appendix 3. References

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