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Rapid declaration of TB freedom in possums in the Hokonui Hills

TBfree New Zealand

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Rapid declaration of TB freedom in possums in Hokonui Hills

Graham Nugent, Jackie Whitford, Cecilia Latham, Grant Morriss

Landcare Research

Prepared for:

TBfree New Zealand

15 Willeston Street
PO Box 3412
Wellington 6140

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Landcare Research, Gerald Street, PO Box 69040, Lincoln 7640, New Zealand,
Ph +64 3 321 9999, Fax +64 3 321 9998, www.landcareresearch.co.nz

Reviewed by:	*Approved for release by:*
Andrew Gormley Ecologist/Researcher Landcare Research	Phil Cowan Team Leader - Wildlife Ecology and Management Landcare Research
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Summary

Project and Client

- A new surveillance concept combining probabilities of detection and eradication was trialled in the Hokonui Hills Vector Control Zone, Southland, and evaluated for its utility for confidently declaring areas free of bovine tuberculosis (TB) in possums. The project was commissioned by TBfree New Zealand, with partial funding from the Ministry of Business, Innovation and Employment, and was undertaken by Landcare Research between March and December 2014.

Objectives

Demonstrate and evaluate (via a pilot trial conducted in conjunction with aerial 1080 poisoning in the Hokonui Hills VCZ in winter 2014) a new 'Survey then Control' (StC) surveillance strategy for declaring TB freedom, by:

- Predicting (through simulation modelling) the amount of possum surveillance required to prove TB freedom using the StC approach
- Conducting pre- and post-control surveys to determine possum density, likely TB infection status, the percentage kill (%kill) achieved by the aerial 1080 operation and surveillance sensitivity
- Combining probabilities of TB detection, eradication, and future persistence to estimate the probability of TB freedom in possums, and comparing outcomes with those in surrounding 'fringe' VCZs subject to ground control.

Methods

- The StC approach was demonstrated by conducting (1) a formal pre-control Trap Catch Index (TCI) survey and simultaneously radio-collaring possums; (2) a pre-control necropsy survey to determine surveillance sensitivity; and (3) post-control surveys to estimate %kill and possum density.
- Epidemiological simulation modelling was used to predict the probability that TB had already been eradicated by historical control and then Bayesian updating was used to combine that estimate with the probabilities of detection, eradication, and future persistence calculated in 2014.

Results

The pre-control TCI survey captured 240 possums, of which 216 were ear-tagged and released (99 with radio-collars). The average 3-night TCI was 6.7% (range 0.0–10.0%). The subsequent main necropsy survey caught 265 additional possums, and 34 more were captured or found dead after control. None were infected. Overall, 539 different possums (~12.9% of the estimated population) were inspected for TB, giving an estimate of direct surveillance sensitivity (SS) of 0.108.

The TCI index declined by 98.9% after the poisoning operation. Of 88 radio-collared possums present in the poisoned area before toxic baiting only 4 survived the operation, indicating a 95.5% kill. All of the killed possums from which radio-collars were recovered had ceased moving before nightfall the day after 1080 bait was sown. Field staff encountered about four baits an hour. Distances between capture and kill locations averaged 161 m for non-dispersers (n = 81) and 1789 m for dispersers (n = 7). During a post-poisoning search (unaided by radio-telemetry) of 374 ha known subsequently to contain 56 radio-collared carcasses, 34 possum carcasses were found, 7 with radio-collars. This suggests an area-wide density of 0.64 possums/ha and a total population size of ~4200 possums.

Given a 2014 prior P_{Free} = 0.90, the 'Proof of Freedom' model predicted an *indirect* posterior P_{Free} = 0.920. In contrast, combining the probability of eradication and detection under the StC approach resulted in a *direct* posterior P_{Free} = 0.985. Assuming that the probability of TB persistence or re-establishment during the next 10 years is <0.5, there is a >0.99 probability that TB is now functionally extinct in the Hokonui Hills VCZ.

Between 2007 and 2014, 89 604 traps were set in the 10 373 ha of forest in the six fringe VCZs, with an average 43.2 trap nights/ha resulting in capture, necropsy, and culture of 3519 possums and a mean Operational Trap Catch Index (OTCI) of 0.79%. Most areas were surveyed 5–7 times, with predicted %kills of 26–63%. Simulation of a conservative control history predicted TB extinction by 2007 in 89% of runs. Bayesian updating of that belief using the predicted %kill as a proxy for SS provided *direct* posterior P_{Free} of 0.978- 0.996 by 2014 in the six VCZs. *Indirect* spatial SS estimates predicted from trapping effort were higher, resulting in even higher posterior P_{Free} values (>0.999) for all areas by 2014.

The total farmland area (10 240 ha) of these six fringe VCZs was similarly intensively surveyed during 2007-2014 (average of 28.7 trap nights per *trapped* ha), with 495 possums captured and necropsied and a mean OTCI of 0.29%. *Direct* posterior P_{Free} of 0.950-0.982 were calculated for 2014, while use of spatial SS estimates predicted near certain TB freedom (P_{free} > 0.99) in all six farmland strata.

Conclusions

- The Survey-then-Control approach can be translated into a practical approach for rapid declaration of TB freedom in a heavily forested area of difficult terrain. Although uncertainty has not yet been incorporated into our StC estimate of P_{Free}, a low estimate based on an unrealistically high possum density still exceeded the 0.95 target for declaring TB freedom.
- TB is likely to have been absent from the Hokonui fringe VCZs for some years, reflecting a long history of ground control. If similar effort has been applied in the outer VCZs of the Hokonui VRA, it is likely they are also free of TB in possums.
- There was strong evidence that the proof-of-freedom-calculated SS values were biased high, suggesting that this model may overestimate possum trappability.
- The cessation of movement by all of the killed radio-collared possums within ~30 hours of toxic baiting suggests that a 2-night window of fine weather could be sufficient for ensuring population-wide exposure to 1080.
- Pre-control assessment of possum abundance using the TCI method, coupled with radio-collaring of possums for %kill estimation, provided a credible and more

immediate estimate of control effectiveness than would conducting pre- and/or post-control TCI surveys. The distance between capture and kill locations of radio-collared possums suggested range sizes larger than usual for forest-dwelling possums.

- The high probability that TB has been eradicated from the Hokonui Hills and the surrounding fringe VCZs suggests VRA status could be revoked well before the targeted date of 2026.
- The StC approach is practical to implement and has the potential to speed up the declaration of TB freedom, in effect by eliminating the eradication phase. Further development is needed to incorporate uncertainty into the calculations, to account for non-random sampling of spatially clustered TB+ve possums, and to identify which of the techniques explored here for density and SS estimation would be most practical and suitable for standard operational use.
- Including consideration of post-control TB persistence and probability of re-establishment also has the potential to greatly reduce the time and cost required to enable declaration of TB freedom.

Recommendations

TBfree New Zealand should consider:

- Funding further theoretical development and broadening of the StC concept, giving priority to incorporation of uncertainty.
- Inclusion of a probability of persistence and/or potential for re-establishment (based on Spatial Possum Model modelling) in the Proof of Freedom (PoF) framework, factors that are not fully accounted for in the current framework.
- Undertaking further operational field trials to (1) refine the StC approach in contexts similar to the Hokonui Hills, aiming towards a short single-year eradication phase; (2) extend the approach to conducting surveillance in conjunction with either the second, or even the first, of the three aerial operations usually planned for the pre-eradication phase; and (3) apply the StC approach to ground-based ‘performance’ contracting.
- Revisiting the default parameters currently used in the PoF calculator, specifically to account for (1) the suspected overestimation of SS from high-intensity input surveys in the fringe VCZ forest strata and (2) the indications of possible underestimation of SS from input surveys in the fringe VCZ farmland strata that we suspect reflects inclusion of substantial areas not used by possums.
- Beginning the process for declaring TB freedom in possums and livestock in the Hokonui Proof of Concept area.
- Whether a 2-day window of fine weather (rather than the current 3 days) is sufficient to allow for full exposure of the possum population to 1080.
- Switching to the use of radio-telemetry (coupled with pre-control TCI monitoring) as an alternative approach to post-control Residual TCI monitoring for assessing the effectiveness of control.

1 Introduction

A new surveillance concept combining probabilities of detection and eradication was trialled in the Hokonui Hills Vector Control Zone (VCZ), Southland, and evaluated for its utility for confidently declaring areas free of bovine tuberculosis (TB) in possums. The project was jointly funded by TBfree New Zealand and the Ministry of Business, Innovation and Employment and undertaken by Landcare Research between March and December 2014.

2 Background

This project directly addresses one of TBfree New Zealand's primary objectives – establishing the feasibility of eradicating bovine TB from vectors from two extensive bush areas by 2026 (specifically the Hokonui and Hauhungaroa ranges). It aimed primarily to determine whether a novel strategic approach could be used to accelerate the declaration of TB freedom in possums in the Hokonui Hills. Under the current strategic approach, TB freedom may not be declared until well after 2020, but we show here that, with the new approach, the area could be declared free of TB in possums as early as 2015. In reality, consideration of issues such as establishing the credibility (to stakeholders) of the new approach, and the continued presence in non-possum hosts such as deer will likely delay the actual declaration for some years.

The standard approach for rugged, heavily forested terrain such as the Hokonui Hills is based on a period of extended possum control (usually by aerial 1080 baiting) to eradicate TB from possums. That is then followed by wildlife surveillance (population sampling aimed at detecting TB in wildlife), which, provided no TB is found, enables managers to statistically determine the likelihood that TB is indeed absent from wildlife. The new approach partially inverts that strategy, such that surveillance is conducted in conjunction with, and immediately before, the final control operation. The core concept is that, *provided no TB is detected*, the percentage kill (%kill) achieved can be used to estimate the likelihood that any TB-infected possums (TB+ve possums) still present would *all* have been killed. The more-or-less simultaneous conduct of survey and then control, in that order in a single operation, makes it statistically valid to calculate the joint probability, for any plausible number of infected possums, that no TB+ve possums were detected *and* that all of them were eradicated during the particular operation. Put another way, we aim to estimate the chance that any TB+ve possum survived the particular operation undetected.

The approach is aimed at reducing the amount of surveillance required to declare TB freedom by taking into account the probability that the final control operation would have completely eradicated the small number of TB+ve possums still present (if any). At present that probability is taken into account via the epidemiological modelling used to predict the Bayesian prior probability of TB freedom, but because there are usually no historical prevalence data available to accurately parameterise the model, and the model assumptions themselves may not be accurate, a maximum prior is imposed (currently 0.9). Imposing that default maximum prior will often, effectively, discount the impact of the final control operation on the probability of freedom.

The logic of the approach is as follows:

1. A low-to-moderate intensity possum necropsy survey is undertaken at the same time (actually, for practical reasons, just before) as the final planned control operation. The power of the survey to detect TB is estimated and expressed as a per-individual detection probability (which is the same as the classical surveillance sensitivity estimate if just a single possum were present). The expectation is that no TB is found, which confirms that the area contains very few TB+ve possums, if any. If, against expectations, TB is found, managers will undoubtedly want to continue possum control beyond the current operation (i.e.; the 'Proof of Freedom' process will be postponed for several years.

2. Once the survey has been completed, control is applied immediately, and its efficacy (%kill) estimated. This equates to the per-individual probability of 'eradication'.

3. Once the detection probability has been measured, it is used to calculate the probability that no TB+ve possums would have been detected for each plausible number of infected possums potentially present (i.e.; 0, 1, 2, 3, 4,....N, where N is the population size). As the potential number of TB+ve possums increases, that probability rapidly becomes very small – i.e.; it becomes increasingly unlikely that many TB+ve possums are actually present.

4. Next, the eradication probability is likewise calculated for each possible number of TB+ve possum present - this is the probability (given the %kill) that all of the 0, 1, 2, 3, 4, …N TB+ve possums potentially present were killed. For low numbers of TB+ve possums, that probability is high and vice versa – the opposite of the detection probabilities.

The two probabilities are then combined to estimate the joint probability, and the worst case probability of a possum surviving undetected is identified. The complement of that probability is used as an approximate measure of the minimum probability that all residual TB (if any) was eradicated by the control operation.

That probability of eradication achieved by the operation is then weighted by the prior probability of TB freedom, akin to the way surveillance sensitivity estimates are in the Proof of Freedom process. This Survey-then-Control (StC) strategy was developed in late in 2013 and was, at that stage, an untested concept. This project was therefore initiated to determine *how* the idea could be implemented. It involved undertaking a large-scale demonstration trial in conjunction with a final TB-related aerial 1080 baiting operation planned for the Hokonui Ranges in winter 2014. As such, it was far more elaborate and complex than would be necessary in an operational context.

The two key data requirements for the StC approach are estimates of surveillance sensitivity (SS; effectively the proportion of the possum population necropsied adjusted as required for diagnostic sensitivity) and %kill (the percentage of the population killed by control). For this pilot trial, these parameters were measured *both directly and indirectly*, in an effort to provide some within-project corroboration of the estimates.

Direct assessment of SS was achieved by surveying possums before the operation and, in parallel, estimating possum density (and therefore population size) so that the percentage of the population sampled could be estimated. That percentage was converted to an SS estimate simply by multiplying it by the diagnostic sensitivity of the various inspection processes used for detecting TB in an infected possum. Direct assessment of % kill involved determining the percentage of a sample of radio-collared possums killed by the aerial 1080 operation.

An indirect assessment of %kill was obtained by comparing indices of possum abundance (Trap Catch Index; NPCA 2011) before and after control. An indirect estimate of SS was obtained using the Proof of Freedom (PoF) calculator (Anderson 2011) to estimate the probability of TB detection given the trapping effort expended to obtain the sample of possums inspected (Anderson et al. 2013).

The operational aim of the trial was to help TBfree New Zealand demonstrate to stakeholders by (or before) 2016 that TB can be eradicated from an area representing relatively difficult operational terrain containing vector infection. To support that, the project also aimed to summarise the history of TB presence in wildlife (and livestock) from the 1970s onward, and to evaluate the likely effectiveness of the possum control conducted in the area prior to the 2014 aerial operation. This component of the work focused on the operational areas surrounding the central core of the Hokonui Hills.

3 Objectives

Demonstrate and evaluate (via a pilot trial conducted in conjunction with aerial 1080 poisoning in the Hokonui Hills VCZ in winter 2014) a new ‘Survey then Control’ (StC) surveillance strategy for declaring TB freedom, by:

- Predicting (through simulation modelling) the amount of possum surveillance required to prove TB freedom using the StC approach
- Conducting pre- and post-control surveys to determine possum density, likely TB infection status, the %kill achieved by the aerial 1080 operation, and surveillance sensitivity
- Combining probabilities of TB detection, eradication, and future persistence to estimate the probability of TB freedom in possums, and comparing outcomes with those in surrounding ‘fringe’ VCZs subject to ground control.

3.1 Glossary

OTCI – Operational Trap Catch Index

P_D – probability of detection

P_E – probability of eradication

P_{Free} – probability of TB freedom

PoF – proof of freedom

%kill – percentage of population killed

+ve/−ve – positive/negative

SPM – Spatial Possum Model

SS – surveillance sensitivity

StC – survey then control

TB+ve possum – possum infected with bovine TB

VCZ – Vector Control Zone

4 Methods

4.1 Study area and TB control history

4.1.1 Study area

The 6567-ha Hokonui Hills VCZ lies within the centre of a larger tract of more-or-less continuous forest and shrub land, mostly native. It is surrounded by six VCZs each of which includes some forest contiguous with Hokonui Hills and some farmland (Figure 1). Those are in turn surrounded by a further 18 VCZs, with the 25 VCZs together comprising the Hokonui Vector Risk Area (Figure 2). The six VCZs adjoining the Hokonui Hills VCZ (total area ~ 20 600 ha) have been intensively surveyed and controlled since mid-2007, with a ground-based trapping effort of >450 000 trap nights (~22 trap nights/ha) resulting in >4000 possums being caught. No TB was found in any of those possums, nor in any other wild animal (115 ferrets, 13 pigs) inspected during that time. By early 2014, these data provided a high level of qualitative confidence that TB was unlikely to remain in those six VCZs (a belief that is more formally assessed in this report; Section 5.6).

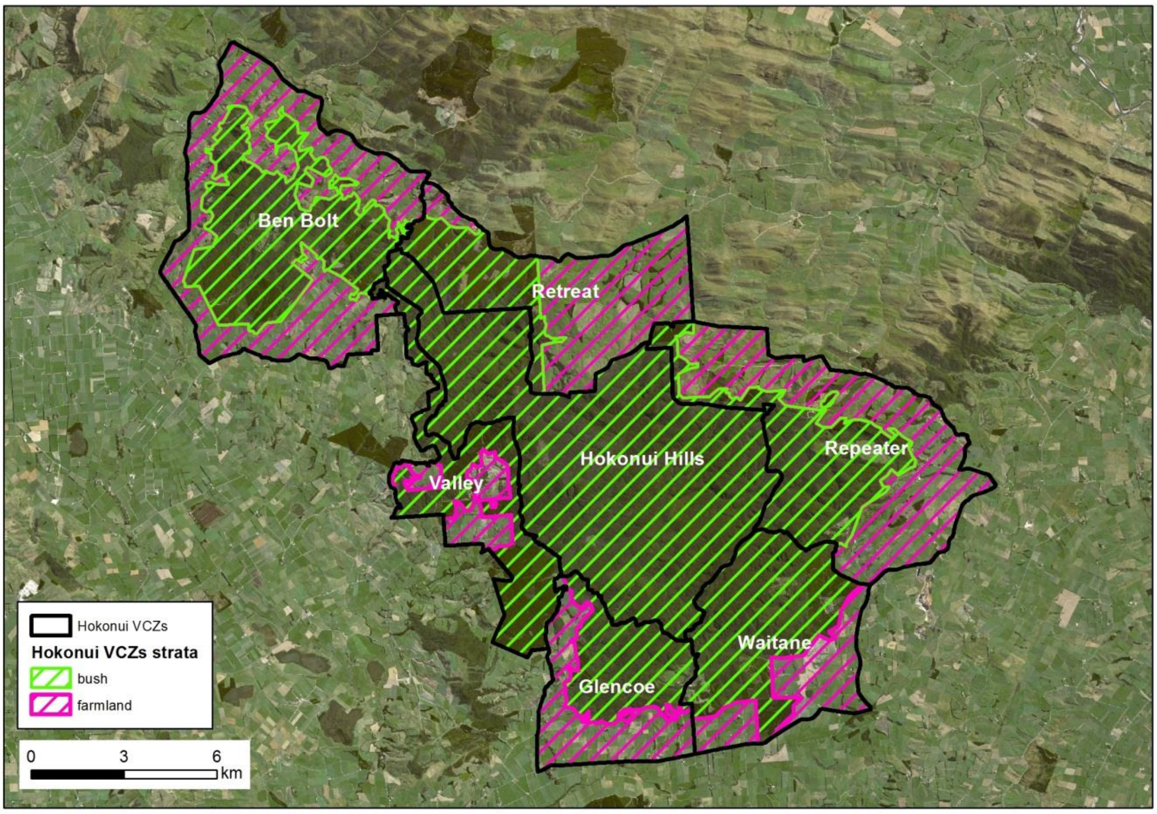

Figure 1 Map of the Hokonui Hills Vector Control Zone (VCZ) and the six surrounding VCZs, with the two major habitat types or 'strata' (bush and farmland) in the latter VCZs shown separately.

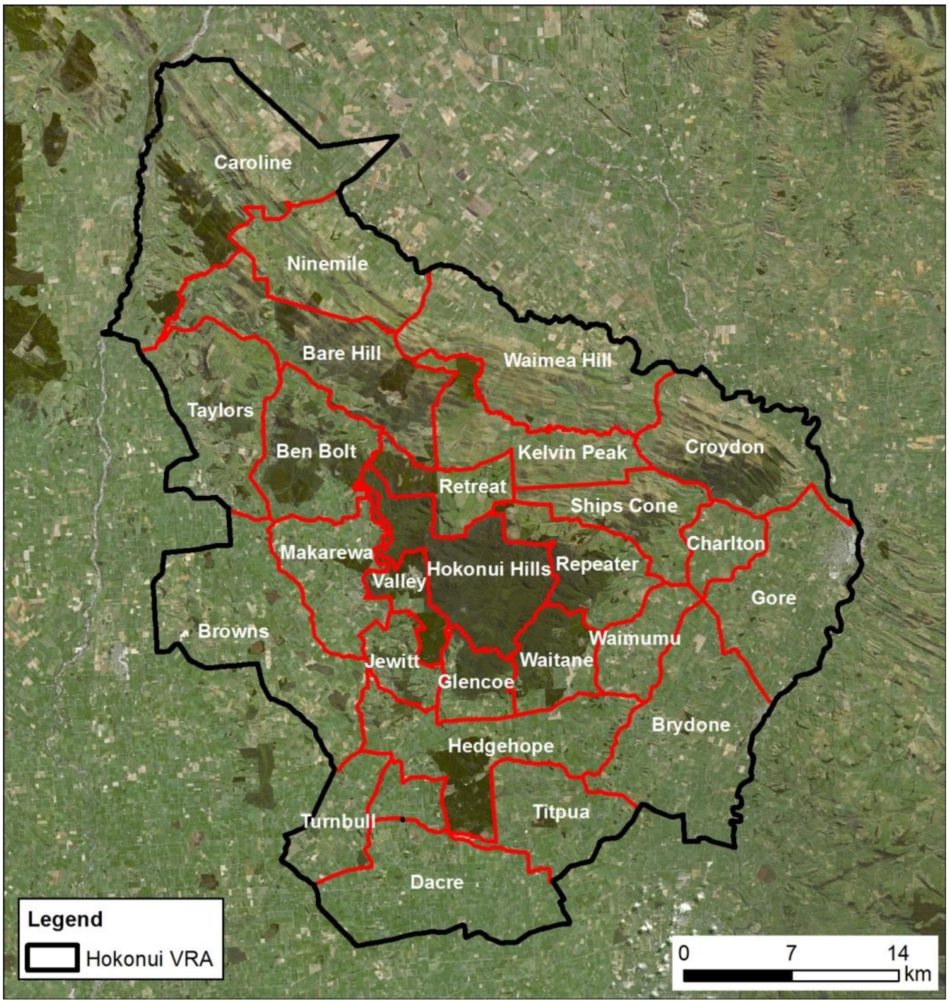

Figure 2 Hokonui Vector Risk Area.

However, there had been no similar control or survey effort in the central forested Hokonui Hills VCZ since 2005/06, so the primary goal of this project was to determine whether adopting a StC approach could cheaply and quickly attain the same or higher level of confidence in TB absence. The history (since the 1970s) of TB presence in livestock and wildlife in the Hokonui area is summarised in Appendix 1, along with a summary of historical possum control efforts up until 2007.

4.2 Post-2006 control and 2014 aerial control operation

4.2.1 2007–2014 ground control

Ground-based control was implemented in all or most of the bush strata in the six 'fringe' VCZs adjoining the central forested area of the Hokonui Hills over the period 2007–2014. This control was undertaken on an input-funded basis whereby leg-hold traps were deployed at a specified spacing for a specified number of nights (usually five, but four in 2013/14), and the possums captured were recovered and necropsied to determine TB status. The data for these surveys were extracted from Vector Net (the TBfree New Zealand operational database for such surveys) and included trap location, trap type, the dates on which traps were set (at least in recent years), and (with some omissions, especially in 2007/08) whether a possum was captured. These data were provided by K Crews, TBfree New Zealand National Disease Manager, or obtained directly from Vector Net by Landcare Research staff. They were used to determine the total number of possums captured, the total number of trap nights, and an operational trap-catch index (OTCI; number of possums captured/number of trap nights, expressed as a percentage) in each area in each year (Appendix 2).

4.2.2 2014 aerial control

The aerial 1080 operation primarily involved dual pre-fed helicopter-sown broadcast baiting. A flight path spacing of 220 m was used both for prefeeding and for toxic baiting, with no alignment of the prefeed and toxic bait flight-paths. The two prefeeds of non-toxic cereal bait (cinnamon-lured 6-g RS5 baits; Animal Control Products, Whanganui; all baits coated with Epro deer repellent) were broadcast at 0.75 kg/ha 15 days apart on 30 April and 15 May 2014, and were followed 16 days later by 0.15% 1080 cereal bait (cinnamon-lured 12-g RS5 baits) broadcast at 1.5 kg/ha on 31 May 2014. No rain fell in the area until 5 June when a combined total of 4.1 mm was recorded on that and the following two days. On 8 June, a further 18 mm of rain fell.

The operation extended beyond our study area to cover 9166 ha in total (Figure 3). A small area in the central southern part of our study area was excluded from aerial control. Input-based ground control was applied in this area (and over all of the remaining area in the six VCZs surrounding the aerial zone) before or soon after the aerial operation.

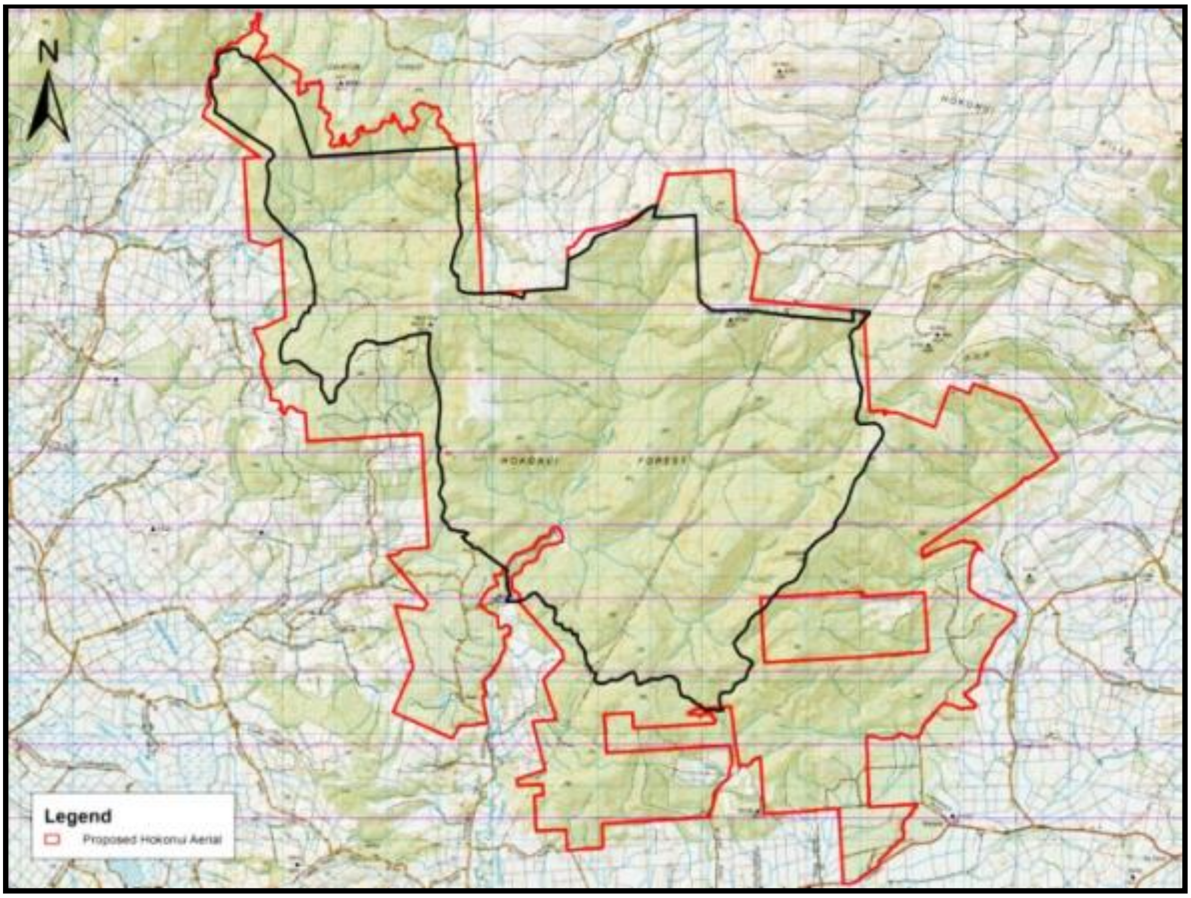

Figure 3 Boundaries of the Hokonui aerial 1080 consent area approved for winter 2014 (red line) and the study area for this project (Hokonui Hills Vector Control Zone, black line). The red-lined area inside the black line shows where a small part of the Hokonui Hills was excluded from the aerial poisoning operation.

4.3 Measurement of possum abundance, possum density, and percent kill

4.3.1 Modelling Survey-then-Control

To assess what level of pre-control TB+ve possum surveillance was likely to be required for successful implementation of the StC approach, a generic conceptual model was re-parameterised specifically for the Hokonui Hills VCZ. The simple spreadsheet model was used to calculate, for all plausible (given non-detection of TB) numbers of infected possums (0–200), the probability of detection (P_D), the probability of eradication (P_E), and, from their joint probability, the probability that no infected possums survived undetected (which equates to an empirical point estimate of the converse of probability of TB freedom (P_{Free})). This P_{Free} estimate was then combined, following Bayesian theory, with a predicted or assumed prior probability of TB freedom (prior P_{Free}) to produce a posterior probability of TB freedom (posterior P_{Free}).

From TBfree New Zealand's February 2013 trend monitoring results (RTCI = 2.6%; B. Rohloff, pers. comm.), we assumed (1) a current Trap-Catch Index (TCI) of 4%, (2) that a 4% TCI equates to ~0.4 possums/ha (Sweetapple & Nugent 2014), and (3) that the dual-pre-fed aerial 1080 operation would achieve a >90% kill. The model was used to predict the minimum surveillance sensitivity (SS) required to achieve a posterior P_{Free} = 0.95 for each value of a range of prior P_{Free} probabilities.

A wide range of sampling intensities was simulated in a Global Information System, emulating systematic survey of possums with leg-hold traps set 50 m apart along parallel transects spaced a range of distances (0.5–2.0 km) apart. The PoF calculator was then used to predict surveillance sensitivity estimates for each of the simulated trapping designs. The results were used to identify the transect spacing needed to deliver the desired SS.

4.3.2 Assessment of pre-control possum abundance

To estimate possum abundance and, at the same time, radio-collar a large sample of resident possums for direct assessment of %kill, a survey was conducted in March 2014 using a variant of the standard Residual Trap Catch method (RTCI; NPCA 2011). Ten 1.8-km-long transects were established systematically across the ~6500-ha operational area (Figure 4). Five standard TCI traplines were nested within each transect, separated from each other by a ‘gap’ of 200 m to provide some degree of independence. However, to maximise the number of possums captured and inspected for TB per field day, traps were also set at ‘best sign’ in those ‘gaps’, resulting in a total of 90 traps per 1.8-km transect. Traps were set for 5 days, to facilitate calculation of a removal estimate of total number of trappable possums in the vicinity of the traplines. This entailed use of a least-squares minimisation analysis to fit an exponential ‘capture decay’ curve (with an ad hoc first-night ‘familiarisation’ adjustment) to the observed data, and then determining the number of possum captures predicted if trapping had been extended until the catch rate fell to close to zero.

Other than the few injured during capture, the captured possums were radio-collared and/or ear-tagged and then released at their capture sites. All possums were visually inspected and palpated for TB lesions, a process we assumed had 50% diagnostic sensitivity ($DS_{palpate}$). Those animals injured during capture were killed and necropsied for TB by Landcare Research staff, and a pooled sample of nodes was submitted for culture (assumed $DS_{culture}$ = 0.95).

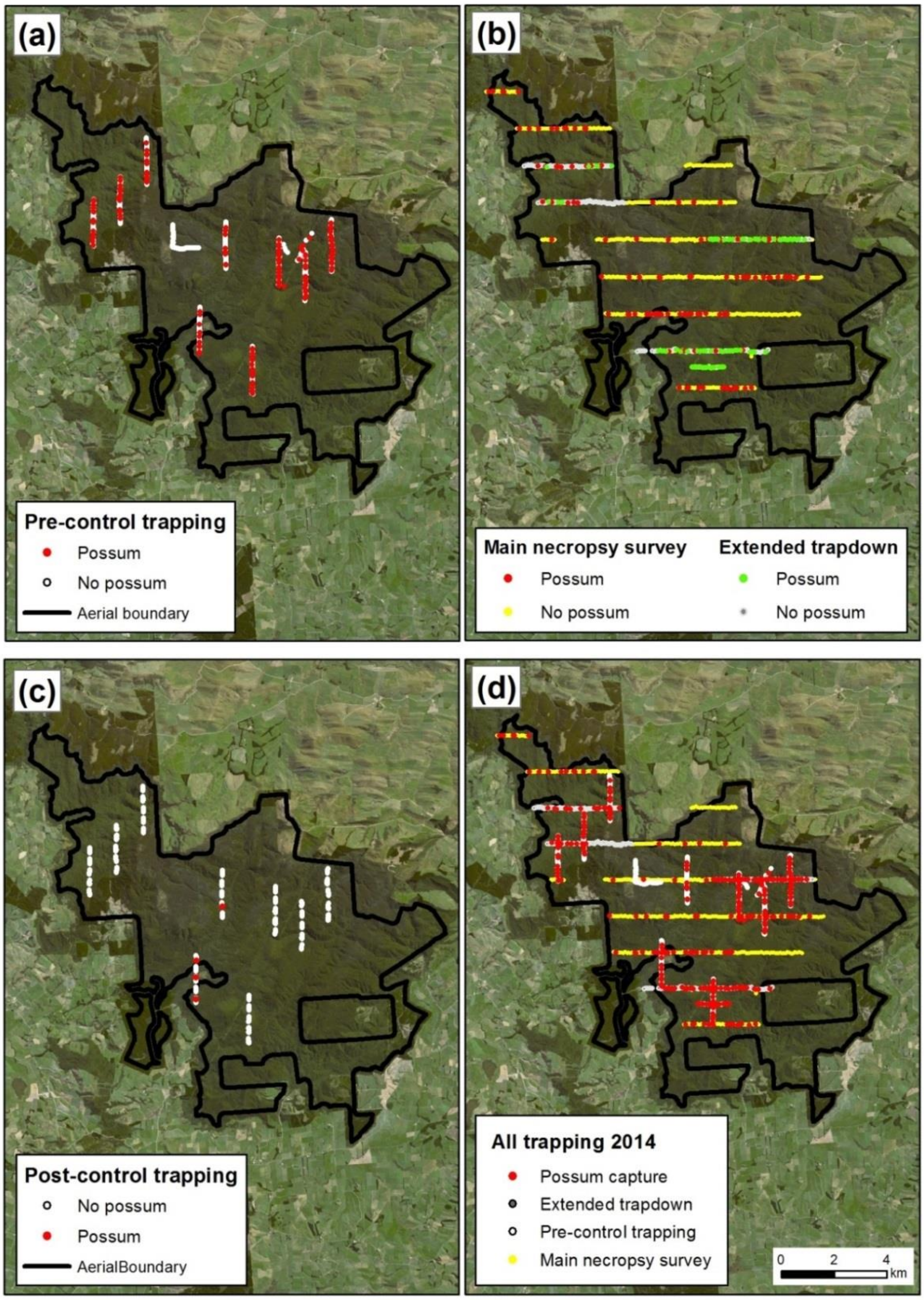

Figure 4 Trapping efforts in the Hokonui Hills in winter 2014, showing the location of traps and possums captured in (a) 10 1.8-km-long transects along which traps were set every 20 m and at 'best sign' and surveyed before the control operation; (b) east–west transects along which traps were set every 50 m and surveyed for 4 nights (main necropsy survey) but some of which were surveyed for an additional 6 nights (extended trap-down); (c) nine 1.8 km-long transects from the pre-control survey which were resurveyed after the control operation; and (d) all of the trapping effort conducted in 2014 combined.

4.3.3 Main necropsy survey of TB prevalence in possums

A necropsy survey was conducted by Contract Wild Animal Control, contracted (and paid) directly by TBfree New Zealand. Leg-hold trapping was conducted along variable-length transects spaced 1400 m apart, with a 50-m spacing between traps and with traps set for 4 nights. The east–west transects spanned the entire operational area, and were placed at right angles to the TCI transects described in Section 4.3.2 (Figure 4).

In an effort to increase the number of possums obtained for necropsy, a subset of the trap sites were trapped for a further six nights (a total of 10 nights) along four different sections of the east–west traplines and a fifth 10-night trapline was established in an accessible area (Figure 4).

The location of all trap sites, and the location, sex, and age class of all radio-collared possums caught were recorded, and possums that were killed were carried out of the forest and frozen until they could be necropsied by TBfree New Zealand's necropsy contractors (Asure Quality) as per TBfree New Zealand's standard practice or by Landcare Research staff. For most, the peripheral and lung lymph nodes were removed and submitted for culture in pools of up to 10 possums.

4.3.4 Assessment of percentage killed and carcass searches

To determine the percentage of possums killed during the aerial 1080 poisoning operation, the radio-collared possums were tracked using a fixed-wing aircraft on 9 April (approximately 3 weeks after possums were radio-collared), and on 17 May (2 weeks prior to the 1080 operation on 31 May). Ground-based radio-tracking commenced 3 days after the poison operation and continued for approximately 10 days, followed up by an additional aerial radio-tracking on 7 July.

An effort was subsequently made to recover all collars on killed possums using ground-based searches, and, at the same time, use the known number and location of dead radio-collared possums to derive an estimate of possum density. Starting 3 days after the aerial poisoning operation, 2–3 observers walking in parallel searched either 100- or 140-m-wide strips on both sides of eight of the north–south RTCI traplines described in Section 4.3.2, and recorded the location, sex, and age class of any possum found, along with the details of ear-tag or radio-collar IDs for previously captured possums. These searches did not use radio signals to find the locations of killed possums. Once the searches were completed, as many as possible of the unfound radio-collared possums were then radio-tracked to confirm their fate, and (where possible) recover the radio-collars.

About 9 weeks later, the five 'RTCI' sections in nine of the 1.8-km transects described in Section 4.3.2 were resurveyed, with 50 traps per transect set for three fine nights. A further effort was made during this survey to recover any remaining radio-collars not previously accounted for, both on killed possums and on any survivors.

All possums found or captured during this phase of the project were necropsied as above.

4.4 Assessment of TB freedom

4.4.1 Prior probability of TB Freedom and future persistence (prior P_{Free})

The Spatial Possum Model (SPM; Ramsey & Efford 2010) was used to estimate the probability (prior P_{Free}) that TB had already been eradicated from the Hokonui Hills VCZ, assuming a conservative (and simplified) version of the control history described in Appendix 1. The standard process prescribed by TBfree New Zealand's protocol was followed. The same modelling exercise was used to estimate the likelihood that TB would persist for 10 years after the 2014 control operation.

4.4.2 Post-control probability of TB freedom (posterior P_{Free})

A direct surveillance sensitivity estimate (SSe) was derived by expressing the number of possums captured as a proportion of the estimated total population. Population size was estimated using the density estimate we considered likely to be the most accurate, and an alternative density estimate derived from a modelled calibration of the relationship between TCI and possum density (Ramsey et al. 2005). Recent empirical evidence indicates that modelled calibration predicts possum densities that are too high relative to other estimators when possum densities are low (Sweetapple & Nugent 2014), so use of this calibration arguably results in a conservative estimate of SS and therefore of the posterior P_{Free} that we have used as a de facto margin of error.

An indirect estimate of SS was derived from the PoF calculator (Anderson 2011), using the number of traps and trap nights separately for each of the three surveys (initial TCI survey, main necropsy survey, and extended trap-down) and combining the resultant SS estimates into an overall one.

As in the modelling above, the estimates of SS were used to calculate P_D, and the %kill and density estimates to calculate P_E, then the posterior P_{Free}. The latter was then adjusted for the estimated probability of future TB persistence.

5 Results

5.1 Modelling Surveillance-then-Control in the Hokonui Hills

The probability of at least one TB+ve possum surviving undetected was predicted for each of a wide range of combinations of SS and %kill under a conservative prior P_{Free} of 0.5. Assuming that the dual-pre-fed 1080 operation would deliver a >95% kill, the model predicted that a pre-control SS of 0.15 was required to achieve the target posterior P_{Free} of 0.95.

From the GIS-simulated placement of traps, the PoF calculator indicated that an SS of 0.15 would be achieved by 3 nights' trapping with traps spaced 50 m apart along transects spaced 1.4 km apart. These specifications were therefore adopted for the main necropsy survey, but with the belated addition of an extra night of trapping.

5.2 Assessment of possum abundance

5.2.1 Initial TCI survey

A total of 274 possum captures was recorded (including 34 escapes) from 4376 trap nights, with 232 possums captured on the 10 1.8-km-long transects and eight in the off-transect traps. Of the total possums handled (240), one was a recapture, 11 were killed, 99 were radio-collared, 117 were released with ear-tags, and 12 were released unmarked.

Along the designated RTCI segments of each transect (five traps per transect, 50 in total) an average TCI (including possum escapes) of 6.7 ± 2.2% (range 0.0–10.0%) was recorded over the first three fine nights. This was higher than expected given the TCI of 2.6% recorded by TBfree New Zealand trend monitoring in early 2013.

The 3-night TCI was almost identical (6.8%) for the segments in between the designated formal RTCI lines where trap were set at ‘best sign’. This suggested that, within the Hokonui forest area, trap location had little impact on catch rate. Over 5 nights, the overall TCI was 5.97%.

5.2.2 Main necropsy survey

Nine east–west transects between 1.3 and 8.4 km long were surveyed, with 26–169 traps on each. A total of 164 possums were captured (4-night TCI (including escapes) = 4.4%, 3-night TCI = 4.5%; range 2.1–9.0%). The lower catch rate than the one described in Section 5.2.1 is presumed to reflect the impact of poor weather, with >10 mm of rainfall recorded nearby (at Invercargill) on three dates during the trapping period, at times when between 232 and 591 traps were set. Only three of the 216 radio-collared or ear-tagged possums present were recaptured during this survey, too few for precise mark–recapture density estimation. The reasons for the low recapture rate are not known, but all three possums were radio-collared, suggesting the possibility that contractors may not have noticed ear-tagged animals (although it seems unlikely that the necropsy staff would also not have noticed them).

The extension of trapping at four locations along the east–west transects (and at one additional location) resulted in the capture of a further 110 possums. These five traplines spanned 1.35–4.35 km, with 10-night OTCIs varying widely among lines (1.4% to 13.1%), but averaging 5.7%. There was only a slight decline in nightly TCI over the 10 nights (Figure 5), probably reflecting suppression of trap-catch rates by heavy rainfall early during some of the trapping periods. As a result, estimation of the size of the trappable population in the vicinity of these transects from the decline in catch rate was considered not to be worthwhile.

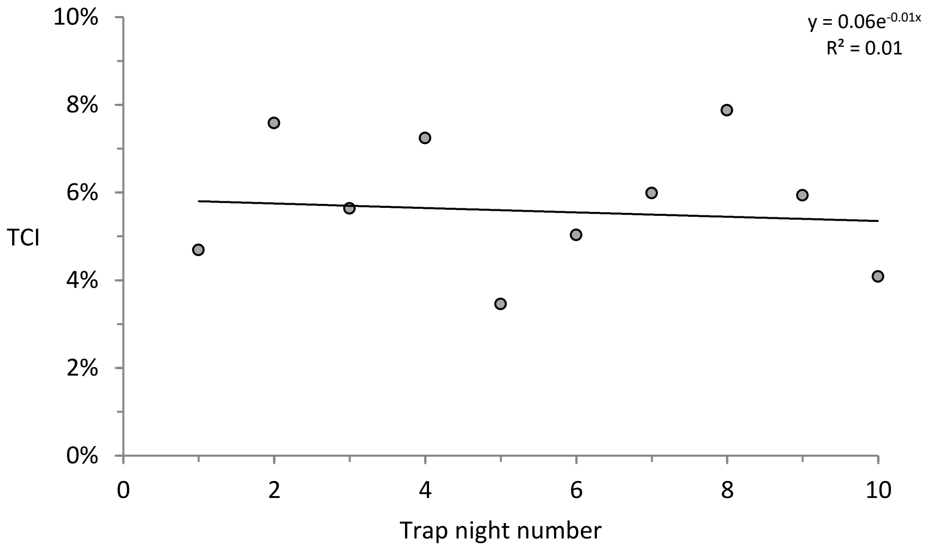

Figure 5 Nightly trap-catch index (TCI) recorded over 10 consecutive nights' trapping on each of five transects, with 27–87 traps set on each transect.

5.2.3 Post-control RTCI

Between 16 and 28 August, 10–13 weeks after the poisoning operation, an RTCI of 0.3% was recorded on 45 200-m-long traplines located along nine transects. Of the four possums captured, one was trapped within the small exclusion zone included in our study area (Figure 3), and two more were within a few hundred metres of it. Excluding the transect (#12) on which those three possum were captured, and presuming that the RTCI for transect #10 would have been zero (because no possums were captured on it before the poisoning operation), the overall RTCI for the area away from the exclusion zone was 0.07 ± 0.01%.

5.3 Density and %kill

5.3.1 Density

Assuming the OTCIs recorded in the initial TCI survey and the main necropsy survey are equivalent to standard TCI measures, the TCI: possum density calibration predicted from modelling by Ramsey et al. (2005) suggests possum densities of 1.36 and 0.96 possums/ha, respectively. At the other extreme, an empirical calibration measured in the Hauhungaroa Ranges, central North Island, for very low (<2% TCI) possum densities (Sweetapple & Nugent 2014) suggest 0.45 and 0.32 possum/ha, respectively. These 'ballpark' estimates suggest a total population size of 2000–10 000 possums.

During the pre-control TCI survey (Section 5.2.1), catch rate declined only slightly over the 5 nights (Figure 6). From the capture rates predicted for nights 6–30 (Figure 6), we estimate a total 'trappable population' of ~400 possums, but it is difficult to assess what the size of 'trapping catchment' might be for that 'trappable population'. The distribution of capture–recapture distances recorded in this study suggests a median capture radius of ~160 m (see details in Section 5.4), which in turn suggests a possum density estimate of 0.59 possum/ha.

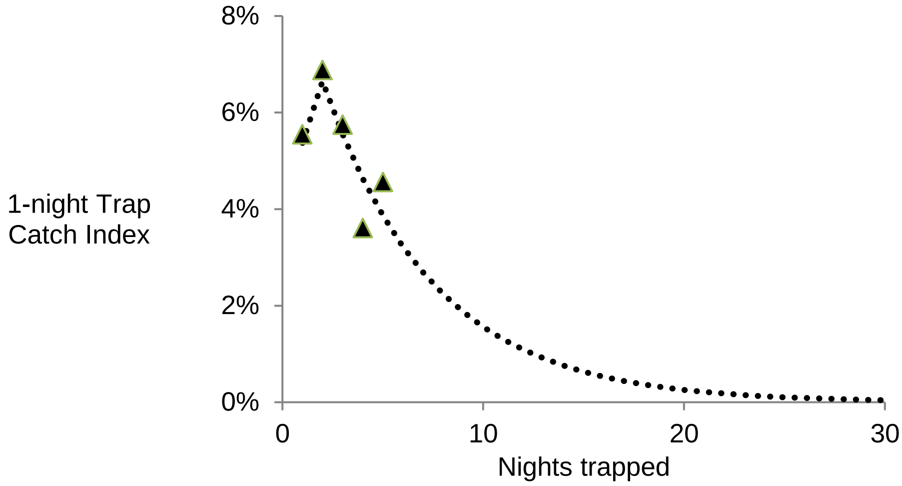

Figure 6 Measured (triangles) and predicted (dashed line) decline in trap-catch rate on 10 90-trap transects trapped for five consecutive fine nights. The predicted decline is based on an equation developed using least-squares regression to fit an exponential decay curve (with an ad hoc first-night adjustment) to the observed data.

We did not calculate similar removal estimates for either the main necropsy survey or the extended trap-down of parts of some transects because the substantial decline in trap catch that is required to obtain robust estimates using this removal method did not occur. This was because there only marginally enough trap nights (four) in the main survey, and because rain reduced trapping success greatly on some nights (creating high variability between nights; Figure 5).

However, the carcass searches produced a possum density estimate that appears to be free of the major uncertainties of the TCI calibration and trap-down mark–recapture methods. Within the 374-ha searched (unaided by radio-telemetry) after poisoning, 34 carcasses were found, of which seven had radio-collars. Subsequent radio-tracking identified that 56 radio-collared carcasses were present in the search area, with a Lincoln Index indicating a total population of carcasses of 264 possums. Assuming (from Section 5.3.2) that 4.5% survived, and adjusting for the higher average TCI for the lines searched (7.8%) than overall (6.7%), this suggests an area-wide density of 0.64 possum/ha and therefore a total population size of about 4200 possums.

5.3.2 % kill

Radio telemetry

Only 88 radio-collared possums were confirmed as both being alive and present in the poisoned area immediately before toxic baiting and definitely re-located inside the poisoned area soon after poisoning. The other 11 had either slipped their collars, died, moved out of the poisoned area, or were not re-located either before or after poisoning. Of these 88 animals, only four survived, indicated a 95.5% (95% CI: 88.9–98.2%) kill. Another possum, not included amongst the 88 above, also survived, but was not counted as it was not detected until long after the operation, when it was found inside the poisoned area but a long way from where it had been collared, so its location during the poison operation is unclear.

Reduction in TCI

Adjusting for the assumed 0% TCI on the non-surveyed transect #10, the overall RTCI (i.e. after control) was 0.27%, indicating a 96% reduction. Excluding the transect that spanned the exclusion zone (transect #12), the reduction was 98.9%.

5.4 Incidental findings

Of the radio-collared possums that were killed, time-since-death data were obtained for 81. Of these, one ceased moving before dark on the day the toxin was sown, and three others before 2200 hours, with almost 80% of possums ceasing movement before daybreak the following day (Figure 7). All 81 animals had ceased moving before nightfall the following day.

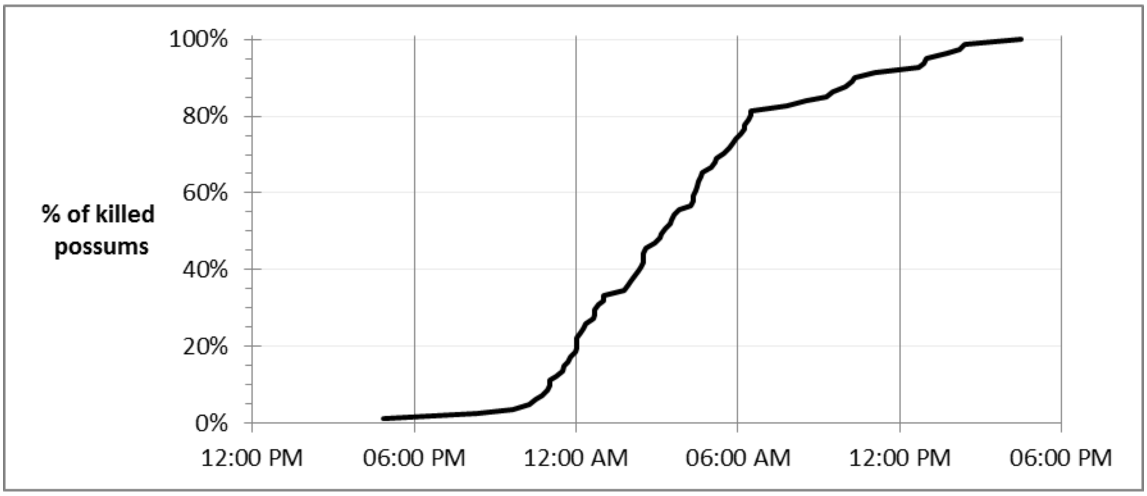

Figure 7 Cumulative times to cessation of movement during the day 1080 bait was sown and the following day. Sowing was completed for most of the area before midday on 31 May 2104.

Given that all possums killed found bait within one day of sowing, the four survivors are likely to also have encountered baits. Supporting that, field staff had no difficulty in finding baits, encountering them at the rate of about four baits an hour (9–51 baits encountered on every carcass search day; 409 baits found in 106 carcass search hours, and 413 in 106 radio-tracking hours).

Distance between capture and kill location was calculated for the 88 possums. Seven animals (~8%) had moved >890 m from their capture locations, and were classed as dispersers. Most (74%) of non-dispersers were killed between 40 and 240 m from where they were captured (mean = 161 m; range 4–590 m), whereas the dispersers were killed up to 3959 m away (mean = 1789 m; Figure 8). Using the mean distance for non-dispersers as a measure of the average effective trapping radius around the traplines suggests a trapping area of 68 ha per trapline (680 ha in total). The trappable population of 400 possums estimated from Figure 6 suggests a density of 0.59 possum/ha.

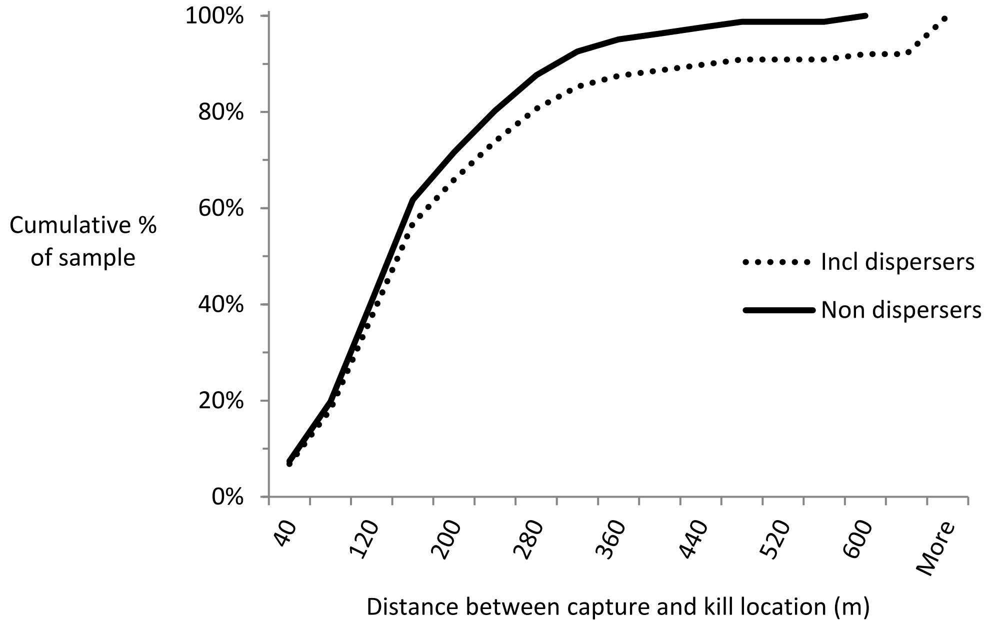

Figure 8 Cumulative distribution of distances between the locations where possum were captured and radio-collared and where they were subsequently killed or re-located, shown for the whole sample (including dispersers) and for non-dispersers with a capture-kill distance of <600 m.

In addition to the 118 possums found dead after poisoning, field staff also found carcasses of 20 rats, 1 hedgehog, 1 stoat, 1 hare, 2 deer, 1 sheep, and 4 pigs. The two deer, the hare and one of the pigs were tested for 1080 residues, as was a live hare captured at that time, with 1080 being found in the tested pig and one of the deer, but being below detection limits for the other animals. In addition, seven bird carcasses were found, none of them native (3 chaffinches, 2 dunnocks, a thrush and a blackbird). All seven were tested and 1080 residues were confirmed in six at 0.3–13.2 mg/kg.

5.5 Surveillance sensitivity and TB freedom

5.5.1 Direct estimation of surveillance sensitivity

A total of 539 different possums were captured. Of the 161 new possums obtained by contract trapping during the main necropsy survey, 51 were subject to ‘cut and chuck’ (i.e. no culture) necropsies by Asure Quality necropsy contractors. From the remainder, 14 pools of tissue from multiple animals and one pool from a single animal were cultured after necropsy, as were the 34 new animals found during carcass searches and all but two of the 101 new possums captured during the extended trap-down. Full necropsy and culture was also undertaken for 90 possums from the initial TCI survey that were subsequently recovered (mostly by radio-tracking or carcass searches), with the remaining 130 from the initial survey that were not recaptured or found dead after poisoning being subject to only visual inspection and palpation.

In summary, 150 possums were palpated (assumed 50% diagnostic sensitivity, DS), 53 were necropsied but not cultured (75% DS), and 336 were necropsied and cultured (95% DS). That is equivalent to surveying 433 possums with perfect DS. No TB was detected in any of them.

Assuming a population of 4200 possums (as estimated in Section 5.3.1), the 539 inspected possums equate to about 12.9% of the population. Adjusting for the less-than-perfect diagnostic sensitivity (DS = 0.804), this equates to a direct SS of 0.103 for a design prevalence of one TB+ve possum. If a single TB+ve possum had been present within the 4200 possums (i.e. at a prevalence of 0.025%), it would have been detected in only one of 10 surveys of this intensity.

From another perspective, the survey provided a direct SS of 0.95 for a design prevalence of 0.67% (i.e. 28/4200 TB+ve possums) – in other words, the survey provided 95% confidence that if 28 or more TB+ve possums were present, at least one TB+ve would have been detected.

There is a case for arguing that DS is irrelevant when animals are removed, at least when DS exceeds 50% (Gormley & Nugent 2012). That is because, in line with the overall StC logic, the killing and removal of possums that were potentially false negatives nonetheless removes those animals from the remaining population. If so, the direct *unadjusted* SS equates directly to the %capture of 0.129. For the main necropsy survey alone, the %kill (and therefore the *unadjusted* SS) estimates would be 3.9% (164/4200).

5.5.2 Indirect estimation of surveillance sensitivity

The indirect SS predicted by the PoF calculator for our combined 2014 surveys using a design prevalence of one TB+ve cell was 0.290 assuming imperfect diagnosis, and 0.317 assuming removal of possums equates to a perfect diagnosis (Table 1). That is 2.7–2.9 times higher than the direct estimates above.

Table 1 Estimates of P_{Free} for the 6567-ha Hokonui Hills VCZ obtained using the standard TBfree New Zealand protocol for combining a prior P_{Free} (predicted to be >0.90 by the Spatial Possum Model) with the Surveillance Sensitivity estimate (SSe) predicted by a data model of possum trapping effort for a cell design prevalence of 1. The SSe was based on 1875 traps set across the initial, main, and extended trap-down survey. It was assumed they were all set for 5 nights, and that there was no chance of reinvasion from adjacent VCZs because they had high P_{Free} values (see Section 5.6). Diagnostic sensitivity was assumed to be either 0.804 (i.e. allowing for the palpation- or necropsy-only inspection of some possums) or 1.00 (assuming that removal of possums compensates for less-than-perfect testing; Gormley & Nugent 2012)

Diagnostic sensitivity	*SSe mean*	*SSe low CI*	*SSe high CI*	*P_{Free} mean*	*P_{Free} low CI*	*P_{Free} high CI*
0.804	0.290	0.287	0.293	0.920	0.881	0.951
1.000	0.317	0.314	0.320	0.924	0.885	0.956

5.6 Assessment of TB freedom

5.6.1 Control history and prior P_{Free}

We condensed the complex history of possum control described by Coleman et al. (2003) and in Appendix 1 into a simple three-operation scenario beginning with an assumed modestly successful (80% reduction) aerial 1080 operation in 1996, a more intensive dual-pre-fed operation with 97% kill in 2004, and a ~10% reduction from the intensive ground survey that followed in 2005. Simulation of that control history in the SPM predicted the outcomes in Figure 9 and Table 2. The initial control operation reduced possum density somewhat, but the SPM predicted that by 2004 possum density had increased back to near pre-control levels, with TB predicted to be still present in all simulations. This is clearly conservative, in that ongoing ground control between 1996 and 2004 (not modelled), albeit patchy and not as successful as expected, appeared to have kept possum numbers in the central forested area well below 10% RTCI (Coleman et al. 2003). Similarly, the 2004 aerial operation resulted in a post-control RTCI of 0%, lower than the 0.07% attained by the 2014 operation documented above, so we consider that our assumption of a 97% kill in 2004 was also conservative. Further, the trapping effort in the 2005/06 ground control operation was almost three times higher than in our initial TCI survey in 2014 (which captured about 6% of the estimated 4000 possums in our study area), so our assumption of a 10% kill in 2005 also appears conservative. Supporting that, the predicted density in 2014 (~2.07 possums/ha) was far higher than that actually observed. Thus we consider our modelling is likely to substantially overstate possum abundance over much of the simulated period, and therefore underestimate the predicted prior P_{Free}.

Under the above control scenario, the SPM predicted that if TB had been widespread at a prevalence of ~3.2% in 1996, it would still have been present in 2004. However, the intensive 2004 operation had a 0.37 probability of eliminating TB within that year and (in conjunction with the subsequent ground control in 2005) 0.66 within 2 years, and the predicted prior P_{Free} had exceeded 0.90 by 2007/08. After 2004, all simulations predicted that very few TB+ve possums remained, and TB continued to disappear from all simulations over the five years after 2005 even though there was no further control and possum numbers were increasing (Figure 9, Table 2).

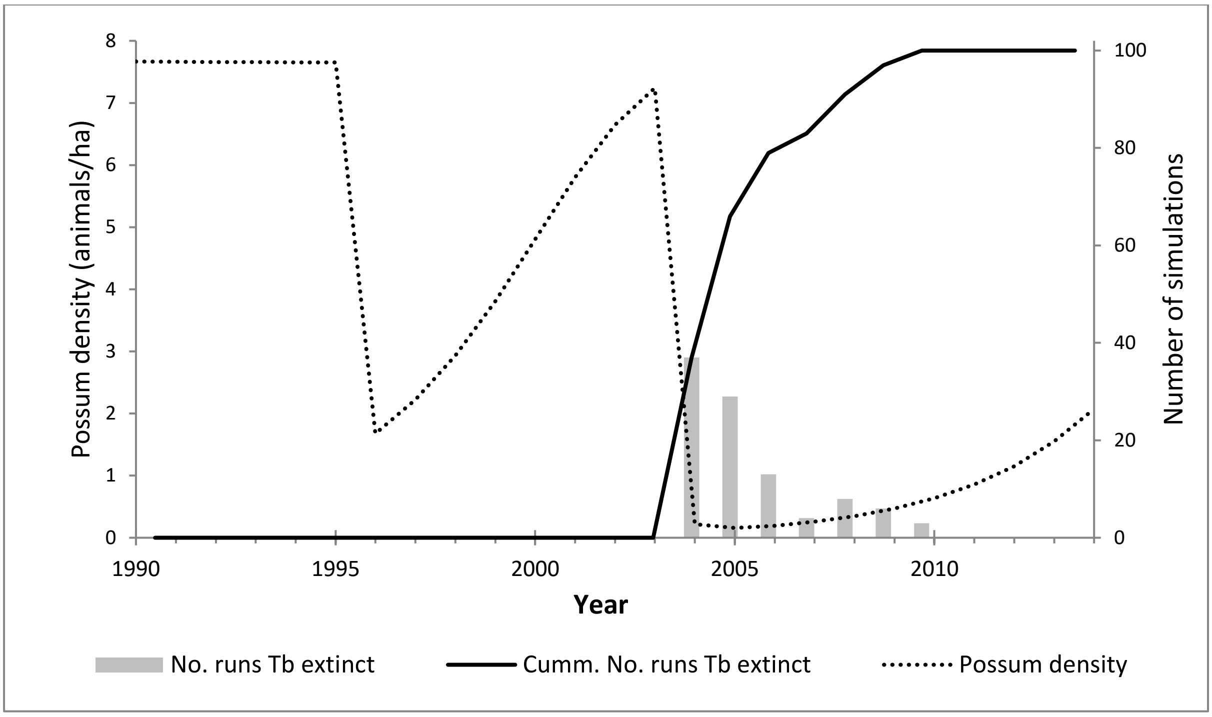

Figure 9 Predictions of the Spatial Possum Model for the 6567-ha Hokonui Hills VCZ, showing the changes in possum density (assuming a simplified control history involving an 80% reduction in 1996, 97% reduction in winter 2004 and 10% reduction in 2005), and in the annual and cumulative number of simulations (out of 100 simulations) in which TB is predicted to have disappeared.

Following TBfree New Zealand’s current standard protocol for Proof of Freedom, we assume a prior probability of 0.90 from 2007/08 onward (i.e. there was a >90% chance TB had already been eliminated from the Hokonui Hills VCZ before 2014).

Table 2 Predictions of the Spatial Possum Model for the 6552 -ha Hokonui Hills VCZ under default and assuming an 80% reduction in possum density in 1996, a 97% reduction in 2004 and a 10% reduction in 2005, showing the maximum and average number of infected possums across 100 simulations, and the percentage of simulations in which TB was predicted to still be present. Estimates are for the final 2-month season of each calendar year

Year	*Maximum no. TB+ve possums*	*Average no. TB+ve possums*	*% simulations with TB+ve possums*
1994	2118	1660	100
1995	2188	1680	100
1996	352	250	100
1997	228	156	100
1998	157	106	100
1999	138	79	100
2000	108	67	100
2001	111	61	100
2002	120	61	100
2003	138	62	100
2004	7	2	63
2005	6	2	34
2006	6	3	21
2007	3	2	17
2008	3	1	9
2009	1	1	3
2010	0	0	0
2011	0	0	0
2012	0	0	0
2013	0	0	0

5.6.2 Posterior P_{Free} in 2014

Assuming a pre-survey 2014 prior P_{Free} of 0.9 (as described in Section 5.6.1) and applying the PoF calculator to our combined 2014 surveys predicts a conventional (*indirect*) posterior P_{Free} of 0.920 (or 0.924 if perfect diagnostic sensitivity is assumed; Table 1)

Our most robust *direct* estimate of SS in 2014 was 0.103 (Section 5.5.1). We can be highly confident that far fewer than 100 TB+ve possums were present (Figure 10a). Assuming a 95.5% kill, the chance of all TB+ve possums being eliminated during the operation (P_E) falls from 95.5% if just a single possum was present to near zero if a 100 infected animals were present (Figure 10b).

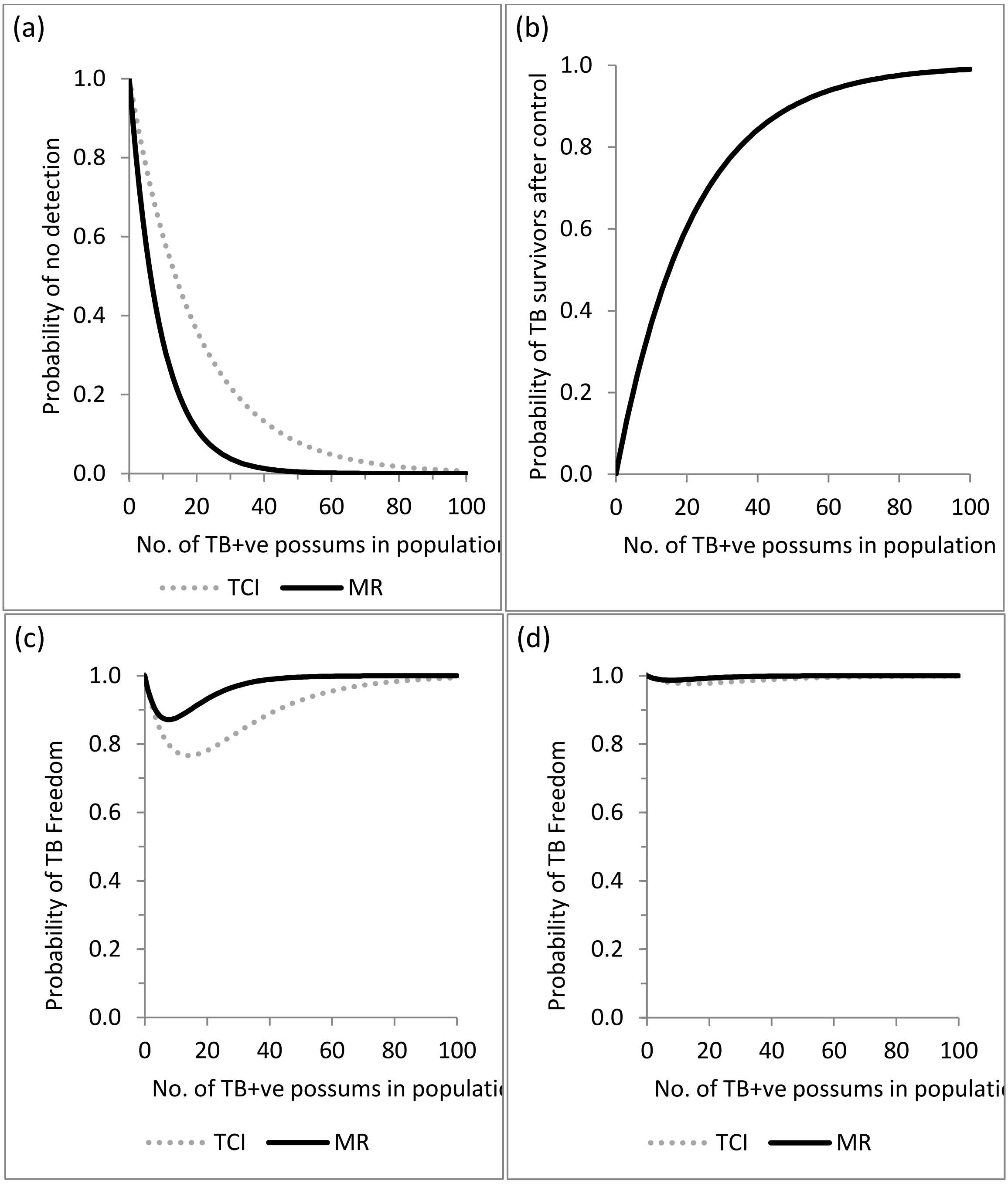

Figure 10 Direct estimation of the posterior probability of TB freedom after a single 'Survey then Control' operation in the Hokonui Hills in 2014, showing (a) the probability of getting the observed result (no TB+ve possums detected) if 1–100 TB+ve possums had been present, assuming a direct estimate of survey sensitivity derived from the percentage of the possum population sampled based on a mark–recapture (MR) estimate of possum density or on a density estimate extrapolated from a Trap-Catch Index (TCI); (b) the probability that at least one TB+ve possum survived; (c) the joint probability that no TB+ve possum survived undetected; and (d) the posterior probability of TB absence assuming a prior P_{Free} of 0.90.

The joint probability of a TB+ve possum surviving undetected therefore increases from 0.040 for a single TB+ve possum to a maximum of 0.129 if (worst case) eight TB+ve possums had been present. The converse of this is an empirical point-in-time estimate of P_{Free} (Figure 10c). With a prior P_{Free} set at 0.90, the posterior P_{Free} for this worst case is 0.985 (Figure 10d). If perfect diagnostic sensitivity is assumed, this increases to 0.99. These P_{Free} estimates are slightly reduced if the lower SS based on a higher density estimate is used (dotted lines, Figure 10c, d).

5.6.3 Probability of future persistence or re-establishment

Given the calculated high certainty that were far fewer than 100 TB+ve possums in the VCZ before control (Figure 10a), there would have been fewer than five TB+ve survivors in an estimated post-control population of about 180 possums (~0.03 possum/ha). From Figure 9 and Table 2, it is clear that the SPM would predict (given that possum density and a very small number of numbers of TB+ve possums) a near-zero chance TB could persist, let alone increase, over the next 10 years. Thus, if we assume, again very conservatively, that the probability of TB persistence or re-establishment during the next 10 years is <0.5, and combine that value with the StC posterior P_{Free} value estimated above, there is a >0.99 probability that TB is now functionally extinct in the Hokonui Hills VCZ – in other words, even in the highly unlikely event that TB was still present in possums it would be extremely unlikely to be able to persist or re-establish in the next 10 years.

5.7 2007–2014 ground control in Hokonui 'fringe' VCZs

5.7.1 Forest strata

The six fringe VCZs surrounding the central Hokonui Hills VCZ (Figure 1) were surveyed 3–7 times over the period 2007–2014 (Ben Bolt 3, Retreat 4, Glencoe 6, Repeater 6, Waitane 6, Valley 7; Appendix 2). A total of 89 604 trap sets were recorded for the 10 373 ha of forest in those six VCZs. There was an overall average of 43.2 trap nights/ha over the 7-year period (ranging from 30.7 in Ben Bolt to 60.6 in Glencoe). Overall, 3519 possum captures were recorded. The *all-area* all-survey mean OTCI for the period was 0.79%. The all-survey OTCI for each individual VCZ varied between 0.09% (Retreat) to 1.30% (Ben Bolt). The highest individual-survey OTCI of 1.94% was recorded in Ben Bolt in 2012/13, from a survey that was conducted after a 3-year gap in control.

Extrapolation from the estimated 3.9% kill (i.e. 164/4200) achieved from the trapping effort in the main necropsy survey in 2014 (Section 5.5.1; Appendix 2) provided predicted %kills for the individual 2007–2014 surveys of 26–63%, and between 36% and 63% for 2013/14. The lowest values are likely to reflect incomplete coverage (as a result of changing control boundaries or contracted exclusions of sub areas rather than contractor performance, such as the exclusion in 2013/14 of areas subject to aerial control).

Simulation of a simple and highly conservative control history using the SPM (50% reductions per year from 1996/97 to 1999/00 then just enough (~20%) per year until 2007 to keep density at or below 1 possum/ha) predicted TB extinction by 2007 in 89% of runs. Using this as a prior P_{Free}, and using the predicted %kill as a *direct* estimate of SS, the *direct* posterior P_{Free} increased from 0.89 in mid-2007 to between 0.978 and 0.996 (Figure 11a). Given the low OTCIs (Appendix 2), and therefore again assuming a <0.5 probability of TB persistence and/or re-establishment over the next 10 years (Section 5.6.3), there is a high probability (>0.98) that TB is now functionally extinct in the forest strata of the fringe VCZs.

Use of the PoF calculator to predict surveillance sensitivity from trapping effort produces *indirect* estimates of SS much higher than the *direct* values calculated from predicted %kills (Appendix 2). The possum data alone produced *indirect* SS estimates of >0.71 in all of the 32 full surveys conducted over the 7 years, and >0.90 in all but six of them. Because the SS values predicted from possum trapping alone were so high, inclusion of data from other sentinel species (Appendix 3) only increased the estimates slightly (Appendix 2).

Using the *indirect* SS estimates and assuming a 0.89 prior P_{Free} in 2007, the PoF calculator predicted that TB freedom (posterior P_{Free} >0.999) was attained in the forest strata of all six fringe VCZs by as early as either 2008/09 in Glencoe, Repeater, Retreat, and Valley or 2009/10 in Waitane, but not until 2012/13 in Ben Bolt (Figure 11b).

5.7.2 Farmland strata

For the six VCZs combined, the total farmland area (10 240 ha) contained just 1217 ha (~12%) of LCDBv4 possum habitat, but a much larger area (5852 ha; 57%) was trapped at least once over the 7 years, as illustrated in Figure 12 (Appendix 2).

A total of 33 638 trap sets were recorded, with an overall average of 28.7 trap nights per *trapped* hectare over the 7-year period (ranging from 22.7 in Retreat to 41.6 in Valley). Overall, 495 possum captures were recorded. The *all-area all-survey* mean OTCI for the period was 0.29%, 70% lower than for the adjacent forest strata. The highest individual-survey OTCI of 1.38% was recorded in Waitane in 2012/13, a survey conducted after a 2-year gap in control. All other individual-survey OTCIs were below 0.83%.

Predicted %kills for individual surveys were generally lower than for adjacent forest strata and varied between 15–47% for individual 2007-2014 surveys and 27–47% for 2013/14.

Using a prior P_{Free} = 0.89 for 2007/08, the direct posterior P_{Free} increased from 0.89 in mid-2007 to between 0.950 (Retreat) and 0.982 (Repeater, Figure 10c). Given the even lower OTCIs than for the forest area, and therefore assuming a <0.5 probability of TB persistence and/or re-establishment over the next 10 years (Section 5.6.3), there is a high probability (>0.97) that TB is now functionally extinct in the farmland strata of the fringe VCZs.

Again, the PoF calculator produced higher *indirect* estimates of SS than the *direct* estimates based on predicted %kill, but the indirect estimates were much lower than the indirect estimates for the forest strata (Appendix 2). The lowest indirect SS estimate was 0.415 (cf. 0.71 in forest surveys) and none exceeded 0.9 (cf. 26/32 in forest surveys).

Again assuming a 0.89 prior P_{Free} in 2007, the PoF calculator predicted a high probability of TB freedom (P_{Free} > 0.98) was attained in the farmland strata of all six fringe VCZs, but not as early as in the forest strata (Figure 11d).

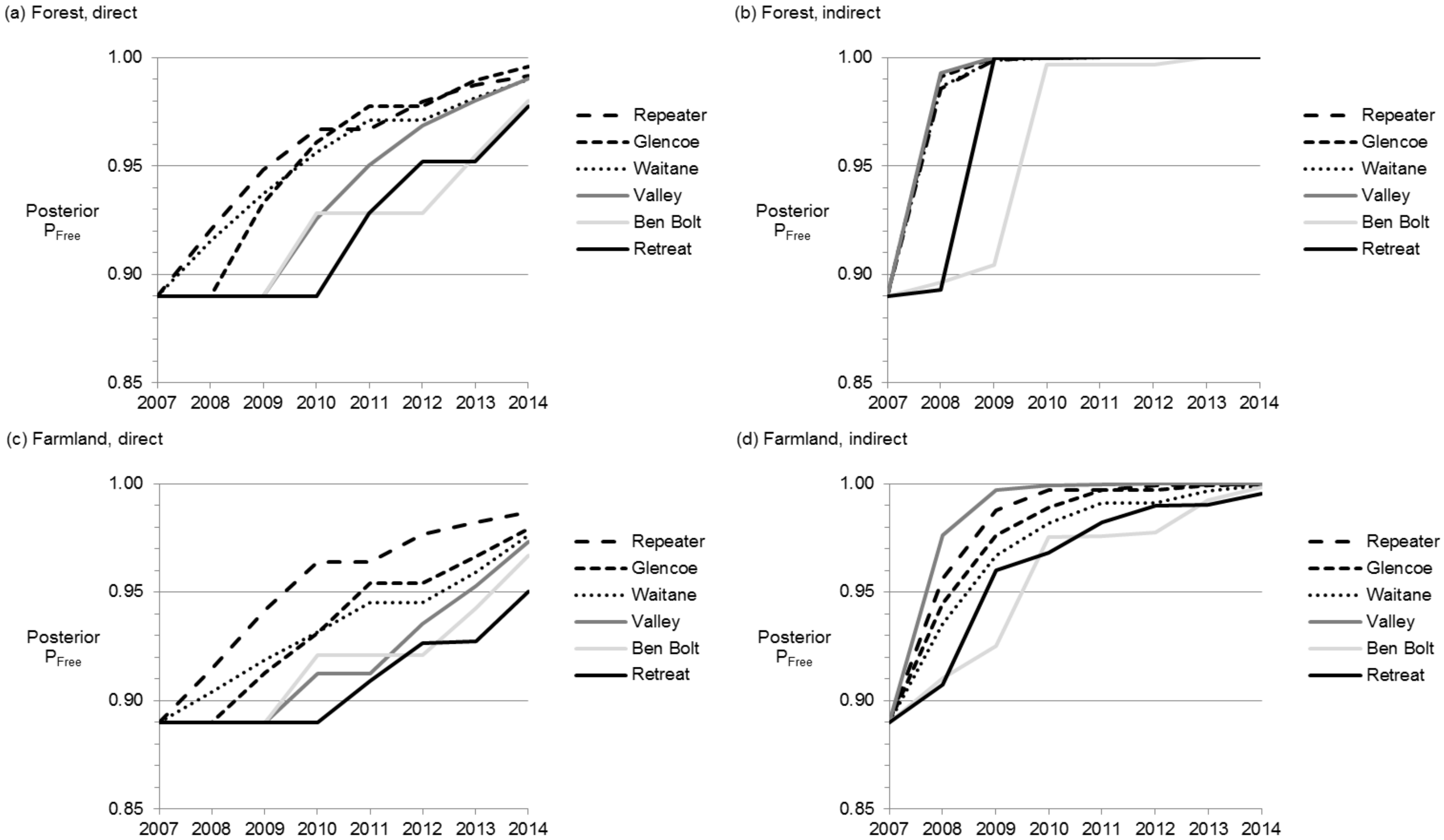

Figure 11 Trends in the annually updated posterior probabilities of TB freedom in possums over the 7-year period 2007–2014, assuming a 0.89 prior P_{Free} in 2007, and a zero probability of re-establishment. Posterior probabilities were calculated separately for each of six VCZs on the fringes of the main forest area of the Hokonui Hills and separately for the forest (a, b) and farmland (c, d) strata. See Figure 1 for VCZ locations and Appendix 2 for further details. The graphs on the left (a, c) show P_{Free} estimates calculated directly from estimates of %kill during surveys, whereas the graphs on the right (b, d) show P_{Free} estimates calculated indirectly from the possum trapping effort and number and type of non-possum sentinels killed during the surveys.

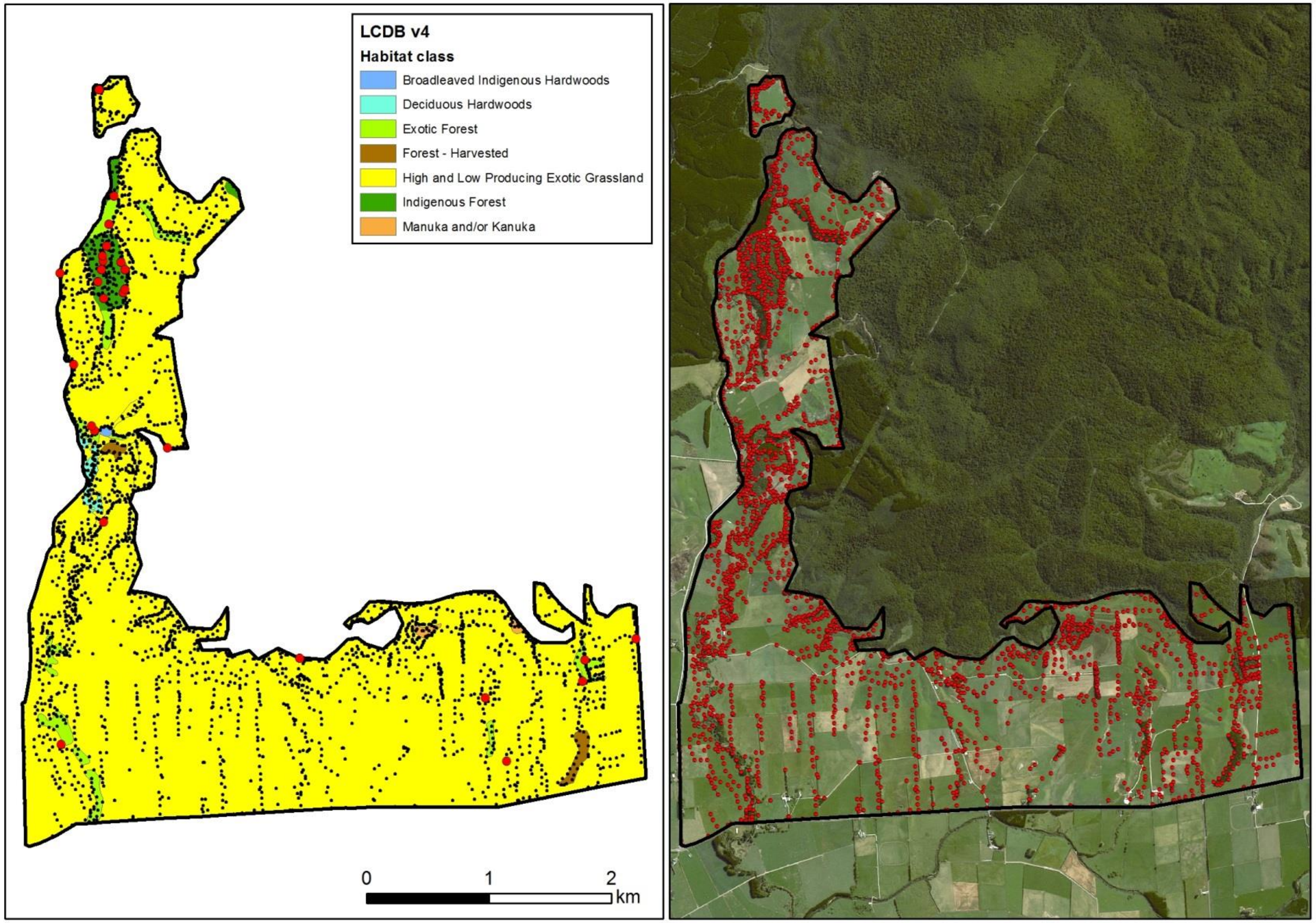

Figure 12 Illustration of possum trapping effort in farmland on the fringes of the main Hokonui forest area; Glencoe VCZ, 2011/13 survey, showing (left panel) Land Cover Data base (version 4; LCDBv4) habitat classes, trap locations (black dots) and possum capture sites (red dots), and (right panel) a satellite image with trap sites (red dots) overlaid.

6 Discussion

6.1 Demonstrating the StC approach

6.1.1 Overall outcomes of the trial

We have demonstrated that the logic of the StC approach can be translated into a practicable approach for rapid declaration of TB freedom in a heavily forested area of difficult terrain. The key innovation is recognition that if very few TB+ve possums are present, then high intensity control has a good chance of eliminating them all, and that calculation of the probability of eradication is straightforward if the % kill is known. By combining the probability of eradication with the probability of non-detection during pre-control surveillance, the resulting joint probability of a TB+ve possum surviving undetected (i.e. continued TB presence, the converse of P_{Free}) can be calculated for any plausible numbers of TB+ve possums potentially present. This obviates the need to specify design prevalence in advance. Instead, we simply assume the highest joint probability is a converse worst-case $P_{Free.}$ It is a worst-case scenario because it assumes that if TB was still present, then it would always just be present at the worst-case prevalence. In reality, the probability of a particular prevalence occurring will be less than 1.0, so our estimates are conservative.

A second innovation suggested and adopted here is the incorporation of a probability of future persistence given the outcome of control. As with the prior, this relies on the validity of the SPM, but it takes advantage of (we assume) control, rather than surveillance, being the final major action taken to prove freedom. Given the '40% of carrying capacity' TB persistence threshold predicted by Barlow (1991), it is unlikely that TB could persist or re-establish at the possum densities likely to prevail in and near the Hokonui Hills over the next 5-10 years, so our assumption of a probability of persistence of 0.5 is appropriate (and conservative). We note that at Hohotaka, holding an infected possum population at an overall RTCI of 7% was sufficient to eradicate TB within 5–6 years, despite initial control being uneven and leaving a small area of blackberry containing TB+ve possums uncontrolled for several years (Caley et al. 1999).

We suggest that a 10-year time frame for assessing probability of future persistence is appropriate for areas where TB might spill back from deer if the prior P_{Free} from possums was already 0.9 at the start of the assessment period (i.e. possum control started many years ago – this would easily encompass the 14 years of potential spillback risk identified by Barron et al. 2013). For areas without much risk of spillback from wild deer (or other hosts such as ferrets that are able to temporarily sustain infection independently of possums), we suggest a 5-year post-control time frame would be appropriate, based on Table 2.

6.1.2 Caveats and uncertainty

A key caveat not yet addressed during the development of the StC approach is that direct 'classical' estimation of SS from the proportion of the population sampled assumes random sampling. However if TB was present it would likely be highly clustered spatially, in which case the direct SS is likely to be biased high. Because 'Proof of Freedom' surveys are conducted only when it is likely that TB is already absent and possum densities are very low, the number of TB+ve possums per cluster will necessarily also be low, as suggested by the post-2004 predictions of the SPM model shown in Table 2. Where highly clustered transect-based sampling of possums is used, as in this study, the direct SS could perhaps be calculated as the likelihood of detecting a cluster of two, or possibly three, TB+ve possums. The mathematics of that have yet to be developed, but appear to be straightforward in principle, and intuitively should not make a large difference at the ‘worst-case’ *post hoc* design prevalence. An alternative that might help avoid or minimise this issue would be the use of wide-ranging sentinel species for estimation of SS. These would need to have ‘sampled’ the possum population during the 6-month period before the control operation (based on 6 months being the usual time-course of infection in possums).

We have not addressed uncertainty in our estimates of P_{Free}, because the mathematical and statistical development of the concept has not yet progressed that far. However, we have conducted a crude form of sensitivity testing by including an SS estimate based on the Ramsey et al. (2005) 5:1 RTCI: density calibration that recent work (Sweetapple & Nugent 2014) indicates is biased high (Figure 10). Even with what we consider is an unrealistically high estimate of possum density (and therefore a low-biased SS the probability that *no* TB+ve possums survived undetected was >0.70 (Figure 10c), more than the minimum 0.53 required to convert a prior P_{Free} of 0.90 to a posterior P_{Free} of 0.95.

6.2 TB freedom in the fringe VCZs

Our analyses suggest that TB has been absent from the Hokonui fringe VCZs for some years, reflecting a long history of intensive ground control. Even with the conservative assumption that ground-based possum control in the forested fringe areas in the early 2000s was just sufficient to maintain possum density at about 1 possum/ha (>5% RTCI), the SPM predicted a prior P_{Free} of 0.89 by 2007. That is consistent with no detections of TB-infected wildlife in those VCZs (or the Hokonui Hills VCZ) since early 2004. Given that prior, an SS of only 0.55 is required to reach the target posterior of 0.95. For the forest fringe VCZs, the SS predicted by the PoF calculator from trapping data exceeded that in every one of the 32 strata surveys conducted over the 2007–2014 period (Appendix 2). The same was true for most of the surveys in farmland strata. If a similar level of concurrent control and survey has been applied in the outer VCZs of the Hokonui Vector Risk Area, it is likely that analysis of current data would provide a high level of confidence that they are also free of TB in possums.

The conclusion that the fringe VCZs are almost certainly free of TB in possums is somewhat undermined by the clear indications that the PoF-calculated SS values must be biased high. The strongest indication of that is provided by the five pairs of surveys in the forest strata summarised in Table 3. The SS values for each survey exceed 0.95, which can only be true if almost all of the possums in the area have been checked for TB. As necropsy surveys required removal of possums, the implication is that virtually all possums would have been removed by the first survey in each pair, yet in all five cases substantial numbers were trapped a year later. Another indication of the apparent bias in the PoF-calculator estimates is the discrepancy between the PoF-predicted SS of 0.15 simulated for the main necropsy survey in 2014 (Section 5.1) compared with the observed SS of 0.04 (Section 5.5.1). One possible explanation for this apparent upward bias is that the PoF calculator may not adequately account for the decreasing trappability of possums as the percentage of the population removed during a single operation increases.

Despite this apparent bias in PoF-SS estimates, the conclusion of TB freedom in the fringe VCZs is supported by the alternative *direct* estimates of SS based on extrapolation from the empirical assessment of trapping effectiveness of the main necropsy survey in 2014. These direct SS estimates result in high posterior P_{Free} estimates by 2014 (Figure 11a, c).

Table 3 Surveillance sensitivity estimates predicted by the PoF calculator from possums only and possums plus sentinel species (ferrets and pigs) trapping efforts for five pairs of sequential surveys in forest strata of four Hokonui fringe VCZs

VCZ	*Financial year*	*Possums killed*	*Traps set*	*Trap nights/ha*	*PoF-SS possums only*	*PoF-SS all captures*
Ben Bolt	12/13	366	3779	7.3	0.984	0.984
	13/14	513	8405	12.9	0.999	0.999
Glencoe	08/09	61	1971	8.2	0.987	0.994
	09/10	76	2056	8.5	0.990	0.990
	12/13	222	2782	11.5	0.998	0.998
	13/14	97	4237	14.0	0.973	0.973
Repeater	11/12	221	2674	7.4	0.981	0.981
	12/13	161	2535	7.1	0.959	0.959
Valley	11/12	53	2282	7.1	0.953	0.954
	12/13	190	2311	7.2	0.949	0.950

6.3 Incidental findings

6.3.1 Fine-weather window for aerial poisoning

The cessation of movement within about 30 hours of toxic bait sowing by all of the radio-collared possums killed during the aerial 1080 operation (Figure 6) suggests that most if not possums were exposed to toxic bait on the first night post-sowing. The failure to kill any of the four survivors on the ensuing five nights of fine weather strongly suggests that the major cause of possum survival was not failure to encounter bait but, rather, failure to ingest a lethal dose of 1080. The regularity with which fieldworkers observed bait during fieldwork (four baits per hour; Section 5.4) suggests possums should have been easily able to find a lethal dose, implying that bait acceptance is probably the primary problem.

6.3.2 Radio-collaring for estimation of control effectiveness

We demonstrated that combining a pre-control assessment of possum abundance using the TCI method coupled with radio-collaring of possums for %kill estimation was feasible and provided a credible and more immediate estimate of control effectiveness than would convention post control RTCI monitoring. Multiplying the pre-control TCI by the %kill predicts an RTCI of 0.31%, 4–5 times higher than the 0.07% actually recorded for the main areas away from the small exclusion zone. There is empirical evidence from elsewhere of a substantial negative bias in post-control RTCI (Nugent et al. 2010), and the historical 2004–2006 Hokonui monitoring outcomes for the aerially poisoned area are consistent with that suggestion in that the post-control RTCI was 0% (from 58 traplines) yet an OTCI of 0.46% was recorded a year or so later (66 possums from 14436 trap nights; Appendix 2). The %kill of radio-collared possums could also be subject to bias – the bias could be positive if the most easily trapped possums are also the most easily poisoned, or negative if trapped and radio-collared possums become more neophobic as a result. However, in the absence of empirical evidence for such potential biases, we suggest that the TCI-based radio-collar monitoring of mortality can provide a more immediate, more readily understood, and more informative assessment of control effectiveness.

6.3.3 Possum movements

The maximum distance between capture site and kill site for non-dispersing possums was about 590 m but almost all were less than 400 m (Section 5.4). Assuming that 400 m approximates the diameter of a circular home range, it implies the largest home range size was about 12 ha. The relationship between mean or median capture–kill distances (160 and 130 m respectively) and mean home range radius is not known, but assuming approximate equivalence to either would suggest mean home ranges sizes of 8.0 and 5.3 ha, respectively. This is larger than is typical of forest-dwelling possums, although smaller than the very large home ranges recently recorded at extreme low density populations in North Island forest (P. Sweetapple, pers. comm.). We suggest that this tentative indication of an intermediate home range size would favour an RTCI: density/ha calibration intermediate between the 5:1 predicted by Ramsey et al. (2005) and the 15:1 ratio estimated for extremely low density populations by Sweetapple and Nugent (2014).

6.4 Conclusions

6.4.1 Hokonui Hills TB freedom

Our results indicate a very high probability that TB has been eradicated from the Hokonui Hills and the surrounding fringe VCZs. This implies that the area could be considered for revocation of Vector Risk Area status in 2015, which would enable TBfree New Zealand to claim eradication of TB from one area of difficult operational terrain well before the targeted date of 2026.

However, some other considerations may need to be taken into account. As one of the two ‘Proof of [Eradication] Concept’ areas specifically designated in the current National Pest Management Plan, there may be a greater need to ‘get it right’ in this area, because apparent failure (i.e. future re-emergence of TB in possums) could result in industry and government questioning the validity of the declaration-of-freedom process (even though the declaration is based on probabilistic rather than absolute data). Also, the process relies heavily on the predictions of the SPM, which not only makes a number of non-validated assumptions but also assumes that surveillance and control have been ‘properly’ (i.e. randomly and/or evenly) applied. If these uncertainties are considered to have substantial weight, then the declaration of freedom could be delayed for 3–5 years while a programme of annual low-intensity ‘assurance phase’ surveillance based on sentinel pig (and possibly deer) survey is implemented.

6.4.2 Proof of Freedom

Overall, the trial has demonstrated how the StC approach could be implemented to estimate TB freedom, and how it has the potential to speed up the declaration of TB freedom by effectively eliminating the ‘eradication’ phase (i.e. the time currently required to conduct TB surveillance after the ‘pre-eradication’ control phase has been completed). While substantial extra cost is incurred at the time of the final control operation that expenditure is likely to be substantially less than the cost of post-control monitoring and surveillance required to confirm TB freedom under the current strategic control-then-survey approach.

Further research and development of the StC concept is needed to incorporate uncertainty into the calculations, to account for non-random sampling of (presumably) spatially clustered TB+ve possums, and to identify which of the range of techniques explored here for density and SS estimation would be most reliable and suitable for standard operational use.

The overarching concept of extending the Proof of Freedom framework to include consideration of post-control persistence or re-establishment also has the potential to greatly reduce the time and cost required to enable declaration of TB freedom. If accepted, it would require a strategic reassessment of the posterior P_{Free} target because combining a prior P_{Free} of 0.9 with a 5-year persistence probability of <0.5 would predict a posterior of close to 0.95 (i.e. little or no further surveillance would be required).

7 Recommendations

TBfree New Zealand should consider:

- Funding further theoretical development and broadening of the StC concept, giving priority to incorporation of uncertainty, particularly in relation to:
    - the surveillance sensitivity of non-random sampling when TB infection in possums is highly clustered in space
    - the estimation of surveillance sensitivity of slaughterhouse inspection of livestock, for both estimation of TB freedom in herds and for use of livestock as sentinels for possum TB
    - the development of logic and theory for replacing the ‘worst case’ approach with a realistic and less conservative method for identifying which design prevalence is most appropriate.
- Inclusion of a probability of persistence and/or potential for re-establishment (based on SPM modelling) within the Proof of Freedom framework. This would enable faster declaration of TB freedom if historical control has reduced possum densities (and immigration rates) to extremely low levels, a factor that is not fully accounted for within the current framework.
- Undertaking further operational field trials to:
    - Replicate and streamline the application of the StC approach in contexts similar to that in the Hokonui Hills (i.e. undertaking surveillance in conjunction with the last major aerial operation scheduled for an area – effectively aiming to convert the end of the ‘pre-eradication’ phase into a very short single-year eradication phase). This should include development and testing of a field survey design in

  which trend monitoring and post-control RTCI performance monitoring are both replaced by pre-control TCI monitoring that is coupled with radio-collaring for %kill estimation and necropsy survey of TB in possums. Where sentinels are available, intensive sentinel surveillance should be undertaken in the period before the aerial control operation and could be extended to include sampling after the operation if sentinels born after the control are excluded.

  - Extend the application of the approach from the ‘final aerial control’ context explored here to application of surveillance in conjunction with either the second, or even the first, of the three aerial operations usually planned for the pre-eradication phase. Provided no TB is detected, this should shorten the duration of control and surveillance needed to declare TB freedom. We consider the early use of the StC approach would be most appropriate in VCZs at the fringes of VRAs, as, in many of these, TB+ve possums may have never been present.
  - Extend the application of the approach to ground-based ‘performance’ or ‘output’ contracting, again by switching monitoring resources from post-control RTCI assessment of achievement to simultaneous pre-control TCI measurement, radio-collaring and TB survey. This would require performance target to be couched in terms of %kill of radio-collared possums, and would also require recovery for necropsy of some (but nowhere near all) of the possums killed by the contractor.
- Revisiting the default parameters currently used in the PoF calculator, specifically to account and adjust for (1) the indications of overestimation of SS from high-intensity input surveys documented in this study in the fringe VCZs’ forest strata and (2) the indications of possible underestimation of SS from input surveys documented in this study in the fringe VCZs’ farmland strata that we suspect reflects inclusion of substantial areas not used by possums.
- Beginning the process for declaring TB freedom in possums and livestock in the Hokonui Proof of Concept area. This could include extending the analyses of historical control in the fringe VCZs presented in Section 5.6 to the outer ring of VCZs in the Hokonui Vector Risk Area, to determine whether declaration of TB freedom in those VCZs could be considered from historical data alone.
- Whether a 2-day window of fine weather (rather than the current stipulation of 3 days) is sufficient to allow for full exposure of the possum population to the 1080 baits during aerial poisoning.
- Switching to the use of radio-telemetry (coupled with pre-control TCI monitoring) as an alternative approach for assessing the effectiveness of control.

8 Acknowledgements

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Appendix 1 – History of TB presence and control in the Hokonui area

History of TB

Throughout New Zealand, tuberculin skin-testing of beef cattle was voluntary until 1969, after which compulsory testing was progressively implemented between 1970 and 1975 (Livingstone et al. 2015). As a result, the number of herds tested in the Hokonui area increased from just 11 in 1969 to 92 in 1975 and 1976 (Table 4).[1] In the first seven years, 26–58% of herds tested had TB reactors and 1–5% of cattle reacted to the test. This was much higher than elsewhere in Southland, where, by 1975, only 0.5% of herds had reactors and only 0.02% of cattle reacted to the skin test (Table A1).

Table A1 Early history of TB infection in livestock in the Hokonui area, 1969–1976, and comparative data for the whole of the Southland Region for 1975, and for the Hokonui area at some more recent times. Sources: New Zealand Archives file, Hokonui TB, copy provided by Dr P Livingstone, TBfree New Zealand; and unpublished data from TBfree New Zealand databases provided by G. Knowles, Area Disease Manager, TBfree New Zealand. *Estimate correcting an error in the source data.

Year	*Source*	*Herds tested*	*Cattle tested*	*Reactor herds*	*%*	*Reactors*	*% reactors*
				Hokonui area			
1969	NZ Archive	11	1242	4	36%	44	3.54%
1970	NZ Archive	24	2856	14	58%	140	4.90%
1971	NZ Archive	45	7990	20	44%	212	2.65%
1972	NZ Archive	26	7321	11	42%	50	0.68%
1973	NZ Archive	26	6826	12	46%	124	1.82%
1974	NZ Archive	60	9007	18	30%	108	1.20%
1975	NZ Archive	92	20682	24	26%	55	0.27%
1976	NZ Archive	92	11972	12	13%	46	0.38%
1980	G Knowles			0			
1995	G Knowles			16			
2002	G Knowles			21			
				All of Southland			
1975		2188	165574	35	1.6%	84	0.05%
				Southland excluding Hokonui area			
1975		2076*	144892	11	0.5%	29	0.02%

[1]New Zealand Archives file, Hokonui TB, copy provided by Dr P Livingstone, TBfree New Zealand.

Following an intensive pre-fed aerial 1080 carrot-bait poisoning operation in 1976 and complementary ground control by the NZ Forest Service or the local Pest Destruction Board, the number of infected herds in the Hokonui area fell to zero by 1980. The Pest Destruction Board continued to maintain the possum population at a low level for a few years after the operation (Coleman et al. 2003). However, presumably as a result of the total or near-total reduction in TB+ve possum control activity during the 1980s, the number of infected herds in the area increased to near previous levels by about 1996 (Table A1). After that, re-intensification and modernisation of possum control resulted in the number of infected herds in Southland declining substantially (Knowles et al. 2005), falling to zero by 2008 followed by a brief re-emergence of TB in a few herds in 2011–2013 (Figure A1).

Most of the infected herds detected in Southland during 1996–2008 were linked to the Hokonui Hills – in March 2003, for example, 17 of the 22 cattle and deer herds infected in the whole of Southland were located in Hokonui and, in 2001–2003, 33 of 69 TB breakdowns in livestock recorded throughout Southland were infected with the distinctive TB strain recorded in wildlife (and livestock) in the Hokonui Hills (Coleman et al. 2003). We note the sharp final drop in the number of infected herds between 2004 and 2006 was coincident with the particularly intensive possum control implemented in the Hokonui Forest area during that time (see History of possum control below).

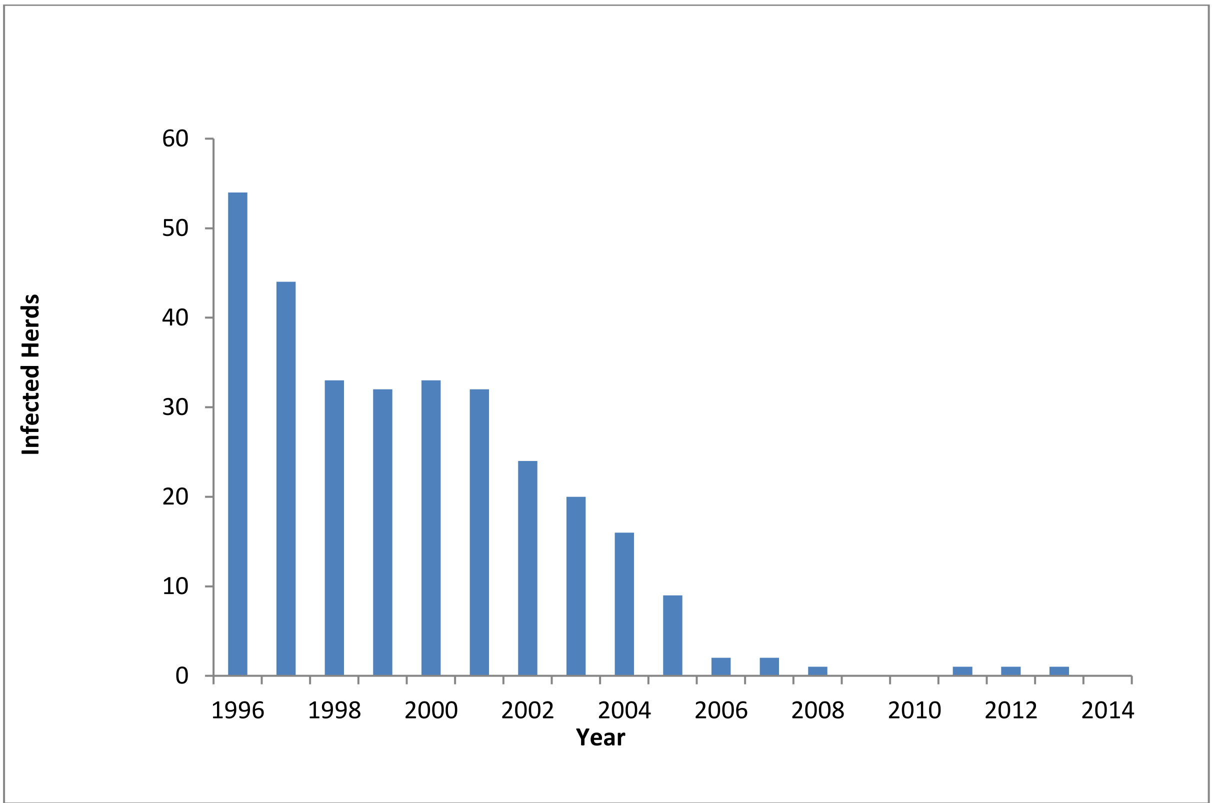

Figure A1 Trend in the number of infected herds in the whole of the Southland Region, including the Hokonui Hills, from 1996 to 2014. Source: Unpublished data from TBfree New Zealand databases supplied by G. Knowles, Area Disease Manager, TBfree New Zealand.

The high incidence of TB in cattle in the Hokonui area in the early 1970s (Table A1), combined with the detection of TB in possums and feral pigs elsewhere in New Zealand in the 1960s (Ekdahl et al.1970), was likely the prompt for an early investigation of Hokonui wildlife as a possible TB source. In 1973, 170 possums from the area were inspected but no evidence of TB was found. However, in May 1974, four of six pigs from the western end of the area and one from the extreme eastern end were confirmed infected. A subsequent survey by the NZ Forest Service in that year resulted in infection being identified in 4/13 pigs, 0/2 sheep, 0/8 deer, 0/14 hares, 1/2 stoats, and 11/887 (1.24%) possums, with the TB+ve possums and pigs distributed over the whole area (Figure A2). A likely TB+ve possum was recorded in 1986 and TB+ve possums were again found in 1987 (Coleman et al. 2003).

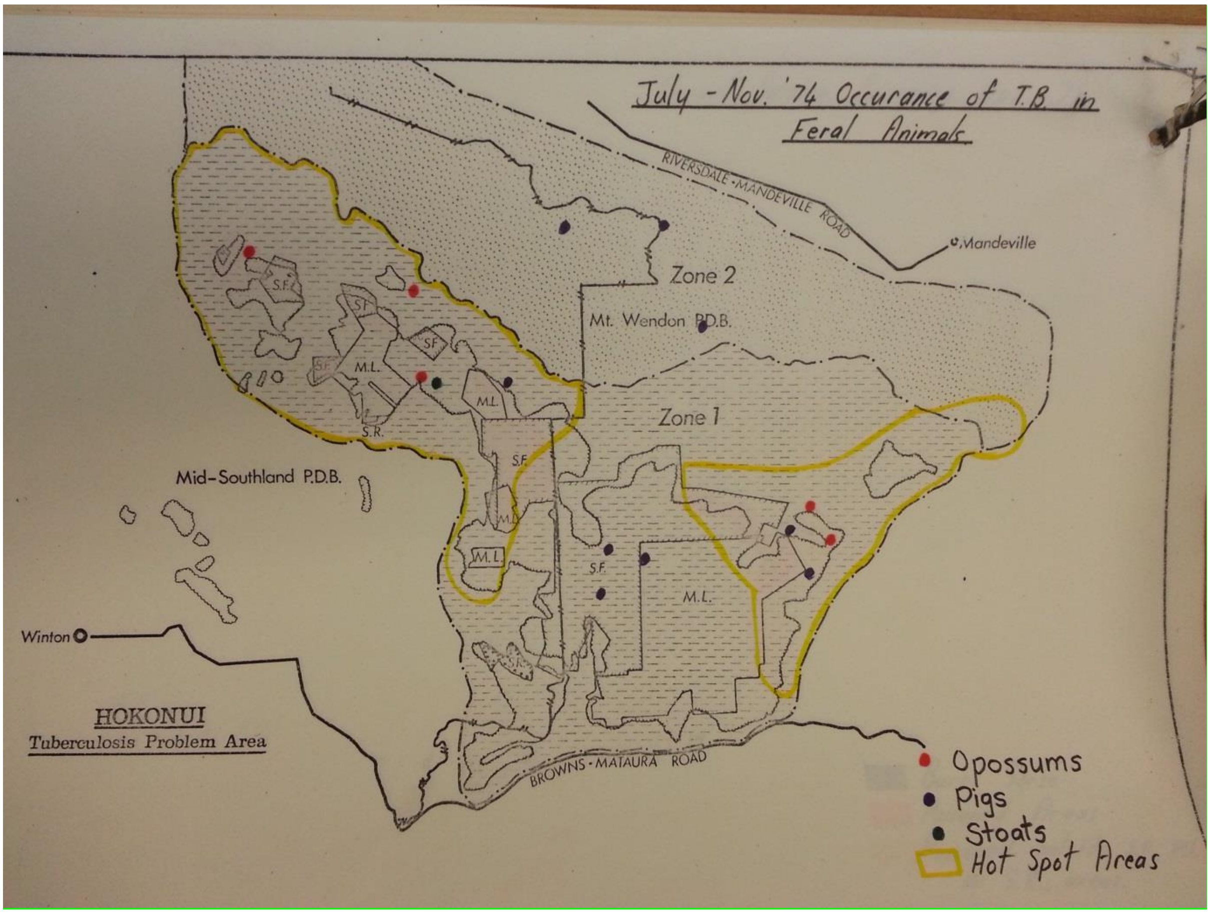

Figure A2 Distribution of known TB infection in wildlife in 1974. Source: New Zealand Archives file, Hokonui TB, copy provided by Dr P Livingstone, TBfree New Zealand.

Data extracted from TBfree New Zealand databases by K. Crews, TBfree New Zealand National Disease Manager, indicates TB+ve possums were also recorded in the 1990s, but none have been recorded since before 1999. From 1999, however, TB has been identified in 40 wild animals from the Hokonui Vector Risk Area (specifically 3 cats between 2000 and 2002, 32 ferrets between 1999 and 2008, and five pigs between 2001 and 2004). Only one of the cats and two of the pigs were recorded as being lesion positive.

The last cases of TB in Hokonui wildlife (two TB+ve ferrets identified in 2007/08) were not in VCZs directly contiguous with the main central forest area (i.e. were not in the Hokonui

Hills VCZ or the six surrounding fringe VCZs contiguous with it). The last known cases in or near the main central forest area were a number of ferrets in 2004–2006 (all from adjoining farmland) and a pig within the forest in 2003/04.

History of possum control

Large-scale intensive control of possums in the Hokonui area was implemented soon after the discovery of TB in possums, with planning documents for a 1976 operation (NZ Archives file, Hokonui TB) indicating an intention to cover ~18,000 ha of forested land for an estimated cost of $114,516 (~$878,000 or ~$47/ha in 2014 terms). The specification for that operation included 11 kg/ha of prefeed (non-toxic carrot bait) and 28 kg/ha of toxic carrot with 0.1% 1080. The possum kills achieved by that operation and the complementary integrated ground control operations are not known, but the rapid decline in the number of infected herds in the Hokonui area to zero by 1980 (Table A1) provides a strong indication that these operations were highly successful in reducing the levels of TB in wildlife.

Throughout New Zealand, spending on TB-related possum control was reduced from previous during the 1980s (Livingstone et al. 2015). The subsequent resurgence to high levels in the numbers of livestock herds infected with the Hokonui TB strain suggests that the limited possum control conducted during the 1980s was not sufficient to prevent the possum populations, and the disease, from some resurgence. It is not clear when possum control resumed in the Hokonui area, but by 1996 another aerial poisoning operation was conducted by Environment Southland. The specifications and possum control outcomes of that operation are not known.

Subsequently, until 2004, ground-based possum control was implemented by Environment Southland (the Southland regional council) covering the whole area in most years (Coleman et al. 2003). The performance targets (couched in terms of RTCI means and maximum numbers of captures per line) for these contracted operations varied between areas depending on perceived achievability, but often proved difficult to achieve in practice, prompting research to determine whether this difficulty was the result of the Hokonui forest possum population having some unusual characteristics such as bait-shyness or generalised neophobia (Coleman et al. 2003). However, the research found no evidence of such problems, but instead indicated that ease of access for ground control was the primary determinant of contractor effectiveness.

Most of the main forested area was aerially poisoned in 2004, using what was at that time a high-intensity specification (specifically dual prefeeding, and increase in the toxic bait sowing rate from the usual 2–3 kg/ha to 5 kg/ha) over 7794 ha of bush (Figure A3). The operation was highly successfully, with standard Residual Trap-Catch Index (RTCI; NPCA 2011) possum trapping on 58 traplines for ~1740 trap nights catching no possums in the aerially poisoned and exclusion zones.

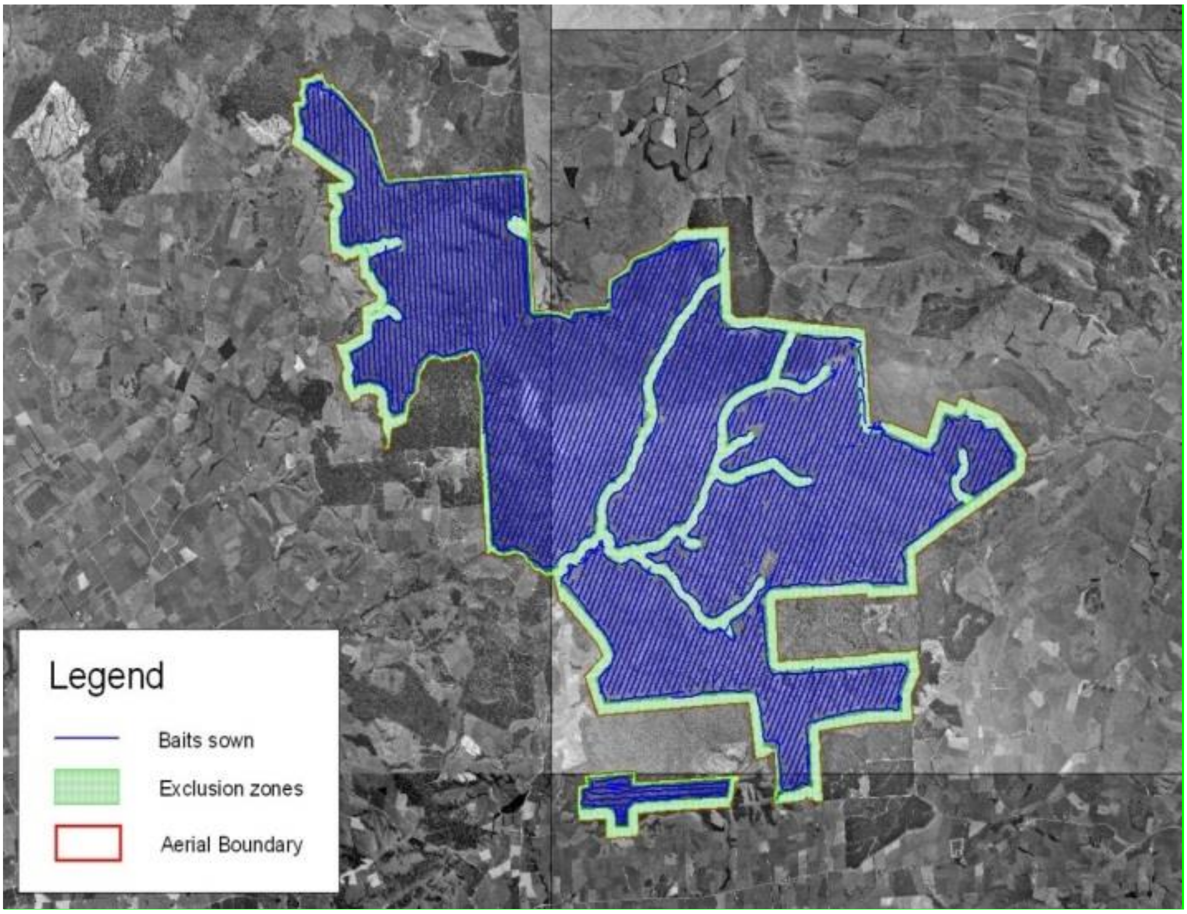

Figure A3 Location and boundaries of the 2004 aerial poisoning operation in the Hokonui Hills, showing the flight paths along which bait was sown, and the associated exclusion around and within the poisoned area in which simultaneous ground-based control was undertaken.

Following this operation, a large-scale ground-based operation was undertaken in 2005/06 to determine whether the mix of aerial and ground-based control had delivered uneven or 'patchy' control, resulting in identification of a few locations in which possum densities may have been high enough for TB to persist locally (G. Knowles, pers. comm.). Parallel east–west traplines were established spanning all or most of the forested area, and spaced 700 m apart, with traps spaced 50 m apart along these transects set for six fine nights. A total of 5332 traps (31 992 trap nights) were set and resulted in capture of 214 possums; an overall Operational Trap-Catch Index (OTCI) of 0.67%. For the aerially poisoned area, an OTCI of 0.46% was recorded (66 possums from 14 436 trap nights), lower than in the exclusion zones (0.64%; 29 possums from 4524 trap nights), and in the ground-control areas surveyed (0.91%; 119 possums from 13 022 trap nights). The higher OTCI recorded 1–2 years after the poisoning operation than the 0% RTCI recorded immediately afterward provided further evidence that immediate-post-control RTCIs were biased low (Nugent et al. 2010) and indicated that the 2004 operation had not been quite as successful as first thought.

The bias is likely to reflect selective survival of possums with very small home ranges at the time of the poisoning operation (Monks & Ramsey 2005), but is also accentuated by estimates derived 1–2 years later being inflated by home-range-size expansion in response to reduced density (Sweetapple & Nugent 2014). We therefore estimate that the 'true' OTCI (i.e. comparable with pre-control measures) in 2005/06 was likely to be about 0.25%.

Adjusting that for 1–2 years of population growth at either a ‘conventional’ (0.35) or a high rate (0.59; Sweetapple & Nugent 2009), the post-control OTCI would have been about 0.08–0.14%. That suggests that the operation achieved a >97% reduction in possum density, as RTCIs in deep-bush strata in 2001/02 averaged more than 2.8% (Coleman et al. 2003) and, as a result of population growth, would have been even higher immediately before the aerial control operation.

Appendix 2 – Surveillance effort and outcomes for the six 'fringe' VCZs surrounding the central Hokonui Hills VCZ

Summary of the known possum trapping effort and captures (and associated predicted densities and probabilities of TB freedom) for six VCZs surrounding the Hokonui Hills VCZ for each financial year from 2004/05 to 2013/14

Results are presented separately for the forest (bush) and farmland strata in each VCZ. Because stratum boundaries changed over this period, traps were assigned to the VCZ boundaries shown in Figure 1. Traps were set for 5 nights in the operations between 2007/08 and 2012/13, but for 4 nights in the 2013/14 operations.

The first column shows the total area. For the bush strata, the whole area is assumed to be trappable possum habitat. For the farmland strata, however, the area (H) of LCDBv4 habitat and an estimate of the trapped area (T) are also shown. The trapped area T includes all LCDBv4 habitat plus a circular buffer of 50-m radius around every trap set from 2007/08 to 2013/14. The predicted kill (Pred %kill) was calculated by expressing the trapping effort (trap nights per hectare of habitat in bush strata or per hectare trapped in farmland strata; TN/ha) as multiples (N) of the trapping effort in the main Hokonui Hills survey conducted as part of this study in 2014. In that survey, an effort of 1.7 trap nights/ha resulted in the capture of 164 possums (~4% of the estimated 4000 possums present; Section 5.2.2; N = (TN/ha)/1.7; Pred %kill = 0.04^N).

A direct surveillance sensitivity (SS, see Methods) was calculated from the predicted %kill assuming a worst-case design prevalence of 0.03% (i.e. just one TB+ve possum present) and assuming a diagnostic sensitivity of 0.95. A direct P_{Free} was calculated from the direct SS. An indirect possum SS was calculated using the PoF calculator (Anderson 2011) to spatially model the likelihood that the trapping effort would have detected the presence of one or more TB+ve possums in a single 1-ha cell. An indirect 'possum' P_{Free} was calculated solely from the indirect possum SS, while an 'all captures' indirect P_{Free} was calculated by also including pigs, deer, and ferrets. Proof of Freedom calculations were based on simple annual Bayesian updating of an assumed 0.89 prior P_{free} for 2007/08 (from Section 5.7), with no allowance for uncertainty (i.e. outside the PoF calculator). The risk of TB reintroduction and re-establishment was assumed to be zero because all OTCIs were below 2.2%, which is considered well below the 5–10% TCI threshold at which TB can persist (Nugent et al. 2014). ND = no data.

Forest (bush) strata

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free}, possums*	*Indirect P_{Free}, all captures*
Ben Bolt	04/05	25	143	3.5%	0.3								
2597 ha	05/06	129	ND										
	06/07	0	ND										
	07/08	0	51		0.1				0.061	0.066	0.890	0.891	0.884
	08/09	0	69		0.1				0.077	0.083	0.890	0.898	0.893
	09/10	160	3629	0.88%	7.0	39%	0.16	0.373	0.968	0.968	0.928	0.996	0.996
	10/11	0	0						0.003	0.003	0.928	0.996	0.996
	11/12	0	0						0.003	0.021	0.928	0.996	0.996
	12/13	366	3779	1.94%	7.3	41%	0.35	0.385	0.984	0.984	0.956	1.000	1.000
	13/14	513	8405	1.53%	12.9	60%	0.33	0.573	0.999	0.999	0.981	1.000	1.000
Total	***All***	***1039***	***15933***	***1.30%***	***30.7***	***89%***							
Glencoe	04/05	50	339										
1209 ha	05/06	105	ND										
	06/07	0	ND										
	07/08	0	1537		6.4				0.929	0.929	0.890	0.991	0.990
	08/09	61	1971	0.62%	8.2	44%	0.11	0.419	0.987	0.994	0.933	1.000	1.000
	09/10	76	2056	0.74%	8.5	45%	0.14	0.432	0.990	0.990	0.961	1.000	1.000
	10/11	99	2078	0.95%	8.6	46%	0.18	0.436	0.925	0.925	0.978	1.000	1.000
	11/12	0	0						0.000	0.000	0.978	1.000	1.000
	12/13	222	2782	1.60%	11.5	56%	0.33	0.532	0.998	0.998	0.989	1.000	1.000
	13/14	97	4237	0.57%	14.0	63%	0.13	0.601	0.973	0.973	0.996	1.000	1.000
Total	***All***	***555***	***14661***	***0.76%***	***60.6***	***99%***							

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free}, possums*	*Indirect P_{Free}, all captures*
Hokonui	04/05	40	2011	0.40%	1.5								
Hills	05/06	34	ND										
6567 ha	06/07	2	ND										
	07/08	0	292		0.2				0.063	0.074	0.890	0.888	0.890
	08/09	5	477	0.21%	0.4	3%	0.03	0.024	0.112	0.186	0.892	0.899	0.909
	09/10	15	253	1.19%	0.2	1%	0.17	0.013	0.070	0.070	0.894	0.906	0.914
	10/11	9	266	0.68%	0.2	1%	0.10	0.014	0.060	0.060	0.895	0.911	0.919
	11/12	11	287	0.77%	0.2	2%	0.11	0.015	0.081	0.081	0.896	0.917	0.925
	12/13	24	219	2.19%	0.2	1%	0.31	0.011	0.078	0.078	0.897	0.923	0.931
	13/14	179	2869	1.56%	1.7	12%	0.23	0.111	0.321	0.321	0.908	0.947	0.952
Total	***All***	***243***	***4663***	***1.04%***	***3.6***	***22%***							
Repeater	04/05	15	358	0.84%	1.0								
1797 ha	05/06	5	ND										
	06/07	119	ND										
	07/08	23	1914	0.24%	5.3	32%	0.04	0.300	0.878	0.883	0.890	0.983	0.984
	08/09	105	2483	0.85%	6.9	39%	0.15	0.370	0.919	0.923	0.928	0.999	0.999
	09/10	81	2521	0.64%	7.0	39%	0.11	0.374	0.921	0.919	0.954	1.000	1.000
	10/11	0	41		0.1				0.028	0.027	0.954	1.000	1.000
	11/12	221	2674	1.65%	7.4	41%	0.30	0.391	0.981	0.981	0.971	1.000	1.000
	12/13	161	2535	1.27%	7.1	40%	0.23	0.376	0.959	0.959	0.982	1.000	1.000
	13/14	55	2802	0.49%	6.2	36%	0.09	0.341	0.710	0.710	0.988	1.000	1.000
Total	***All***	***646***	***14970***	***0.86%***	***41.6***	***95%***							

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free}, possums*	*Indirect P_{Free}, all captures*
Retreat	04/05	10	344	0.58%	1.7								
1026 ha	05/06	8	ND										
	06/07	0	ND										
	07/08	0	10		0.0				0.028	0.028	0.890	0.881	0.888
	08/09	0	2716		13.2	61%			0.998	0.998	0.890	1.000	1.000
	09/10	0	49		0.2				0.129	0.130	0.890	1.000	1.000
	10/11	18	1450	0.25%	7.1	40%	0.04	0.376	0.962	0.963	0.928	1.000	1.000
	11/12	13	1301	0.20%	6.3	36%	0.03	0.346	0.948	0.948	0.952	1.000	1.000
	12/13	0	18		0.1				0.036	0.036	0.952	1.000	1.000
	13/14	8	3087	0.06%	12.0	58%	0.01	0.547	0.993	0.993	0.978	1.000	1.000
Total	***All***	***39***	***8631***	***0.09%***	***42.0***	***95%***							
Valley	04/05	35	687	1.02%	2.1								
1603 ha	05/06	51	ND										
	06/07	2	ND										
	07/08	0	1932		6.0				0.943	0.943	0.890	0.992	0.992
	08/09	0	3397		10.6				0.992	0.992	0.890	1.000	1.000
	09/10	55	2075	0.53%	6.5	37%	0.09	0.352	0.940	0.940	0.926	1.000	1.000
	10/11	49	2044	0.48%	6.4	37%	0.08	0.347	0.916	0.916	0.950	1.000	1.000
	11/12	53	2282	0.46%	7.1	40%	0.08	0.378	0.953	0.954	0.969	1.000	1.000
	12/13	190	2311	1.64%	7.2	40%	0.29	0.382	0.949	0.950	0.980	1.000	1.000
	13/14	44	4437	0.25%	11.1	55%	0.05	0.519	0.862	0.862	0.990	1.000	1.000
Total	***All***	***391***	***18478***	***0.42%***	***57.7***	***98%***							

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free}, possums*	*Indirect P_{Free}, all captures*
Waitane	04/05	45	594	1.52%	1.4								
2141 ha	05/06	66	ND										
	06/07	1	ND										
	07/08	77	1845	0.83%	4.3	26%	0.14	0.252	0.889	0.889	0.890	0.985	0.986
	08/09	90	2058	0.87%	4.8	29%	0.14	0.276	0.870	0.899	0.918	0.998	0.999
	09/10	90	2424	0.74%	5.7	33%	0.13	0.316	0.905	0.905	0.942	1.000	1.000
	10/11	158	2752	1.15%	6.4	37%	0.20	0.350	0.925	0.926	0.962	1.000	1.000
	11/12	2	48	0.83%	0.1	1%	0.12	0.008	0.037	0.040	0.962	1.000	1.000
	12/13	219	2808	1.56%	6.6	37%	0.27	0.355	0.971	0.971	0.975	1.000	1.000
	13/14	213	4996	1.07%	9.3	49%	0.20	0.462	0.758	0.758	0.986	1.000	1.000
Total	***All***	***849***	***16931***	***1.00%***	***39.5***	***94%***							
All 16 940 ha		***3762***	***94267***	***0.80%***	***27.8***	***86%***							
Excluding Hokonui Hills 10 373 ha		***3519***	***89604***	***0.79%***	***43.2***	***95%***							

Farmland strata

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free} possums*	*Indirect P_{Free}, all captures*
Ben Bolt	04/05	0	28		0.1								
3066 ha	05/06	8	ND										
H 298 ha	06/07	0	ND										
T 1712 ha	07/08	0	266		0.8				0.194	0.205	0.890	0.902	0.907
	08/09	0	246		0.7				0.174	0.179	0.890	0.917	0.922
	09/10	12	1866	0.13%	5.4	32%	0.02	0.306	0.679	0.686	0.921	0.972	0.974
	10/11	0	61		0.2				0.030	0.030	0.921	0.973	0.975
	11/12	0	4		0.0				0.010	0.060	0.921	0.973	0.976
	12/13	37	1751	0.42%	5.1	31%	0.07	0.290	0.672	0.674	0.943	0.991	0.992
	13/14	43	3688	0.29%	8.6	46%	0.05	0.436	0.774	0.774	0.967	0.998	0.998
Total	***All***	***92***	***7882***	***0.23%***	***23.0***	***81%***							
Glencoe	04/05	0	12		0.1								
1105 ha	05/06	3	ND										
H 58 ha	06/07	0	ND										
T 726 ha	07/08	0	487		3.4				0.527	0.527	0.890	0.940	0.942
	08/09	2	556	0.07%	3.8	24%	0.01	0.227	0.534	0.588	0.913	0.971	0.975
	09/10	7	555	0.25%	3.8	24%	0.04	0.227	0.534	0.534	0.931	0.986	0.988
	10/11	3	931	0.06%	6.4	37%	0.01	0.349	0.750	0.749	0.954	0.997	0.997
	11/12	0	3		0.0				0.066	0.044	0.954	0.997	0.997
	12/13	11	718	0.31%	4.9	30%	0.05	0.283	0.675	0.679	0.967	0.999	0.999
	13/14	13	1317	0.25%	7.3	40%	0.04	0.384	0.749	0.749	0.979	1.000	1.000
Total	***All***	***36***	***4567***	***0.16%***	***31.5***	***89%***							

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free} possums*	*Indirect P_{Free}, all captures*
Repeater	04/05	0	112		0.3								
2722 ha	05/06	1	ND										
H 609 ha	06/07	74	ND										
T 1670 ha	07/08	8	1404	0.11%	4.2	26%	0.02	0.246	0.626	0.629	0.890	0.952	0.954
	08/09	44	2037	0.43%	6.1	35%	0.07	0.335	0.722	0.726	0.924	0.986	0.987
	09/10	25	2510	0.20%	7.5	42%	0.04	0.394	0.752	0.757	0.953	0.997	0.997
	10/11	0	11		0.0				0.009	0.009	0.953	0.997	0.997
	11/12	92	2215	0.83%	6.6	38%	0.15	0.358	0.716	0.718	0.969	0.999	0.999
	12/13	44	1445	0.61%	4.3	27%	0.10	0.252	0.610	0.609	0.977	1.000	1.000
	13/14	27	1818	0.37%	4.4	27%	0.06	0.254	0.568	0.568	0.982	1.000	1.000
Total	***All***	***240***	***11440***	***0.42%***	***34.3***	***91%***							
Retreat	04/05	1	84	0.24%	0.5								
1903 ha	05/06	2	ND										
H 169 ha	06/07	2	ND										
T 869 ha	07/08	0	138		0.8				0.173	0.173	0.890	0.898	0.902
	08/09	0	1151		6.6				0.577	0.592	0.890	0.954	0.957
	09/10	0	164		0.9				0.194	0.210	0.890	0.963	0.966
	10/11	6	541	0.22%	3.1	20%	0.03	0.189	0.446	0.447	0.909	0.979	0.981
	11/12	5	606	0.17%	3.5	22%	0.03	0.209	0.447	0.444	0.927	0.988	0.989
	12/13	1	27	0.74%	0.2	1%	0.10	0.010	0.022	0.022	0.927	0.988	0.990
	13/14	12	1323	0.23%	6.1	35%	0.04	0.335	0.556	0.556	0.950	0.995	0.995
Total	***All***	***24***	***3950***	***0.12%***	***22.7***	***80%***							

VCZ (Area)	*FY*	*Possums killed*	*Traps set*	*OTCI*	*Trap nights /ha*	*Pred %kill*	*Possums /ha*	*Direct SS*	*Indirect SS, possums*	*Indirect SS, all captures*	*Direct P_{Free}*	*Indirect P_{Free} possums*	*Indirect P_{Free}, all captures*
Valley	04/05	3	44	1.36%	0.6								
485 ha	05/06	2	ND										
H 49 ha	06/07	0	ND										
T 394 ha	07/08	0	369		4.7				0.791	0.802	0.890	0.972	0.974
	08/09	0	496		6.3				0.858	0.877	0.890	0.996	0.997
	09/10	4	300	0.27%	3.8	24%	0.04	0.226	0.728	0.739	0.913	0.999	0.999
	10/11	0	452		5.7				0.816	0.816	0.913	1.000	1.000
	11/12	3	385	0.16%	4.9	29%	0.03	0.279	0.790	0.808	0.936	1.000	1.000
	12/13	4	386	0.21%	4.9	29%	0.03	0.280	0.700	0.700	0.953	1.000	1.000
	13/14	6	890	0.17%	9.0	47%	0.03	0.451	0.877	0.877	0.973	1.000	1.000
Total	***All***	***17***	***3278***	***0.10%***	***41.6***	***95%***							
Waitane	04/05	12	115	2.09%	1.2								
958 ha	05/06	9	ND										
H 34 ha	06/07	0	ND										
T 481 ha	07/08	7	215	0.65%	2.2	15%	0.10	0.140	0.415	0.426	0.890	0.927	0.927
	08/09	4	265	0.30%	2.8	18%	0.05	0.170	0.462	0.493	0.907	0.959	0.962
	09/10	8	267	0.60%	2.8	18%	0.09	0.171	0.442	0.442	0.922	0.977	0.978
	10/11	7	335	0.42%	3.5	22%	0.07	0.209	0.504	0.504	0.937	0.988	0.989
	11/12	0	1		0.0				0.002	0.010	0.937	0.988	0.989
	12/13	31	450	1.38%	4.7	28%	0.23	0.270	0.630	0.630	0.953	0.996	0.996
	13/14	29	988	0.73%	8.2	44%	0.14	0.421	0.780	0.780	0.972	0.999	0.999
Total	***All***	***86***	***2521***	***0.68%***	***26.2***	***85%***							
All 10240 ha, H 1217 ha, T 5852 ha		***495***	***33638***	***0.29%***	***28.7***	***87%***							

Appendix 3 – Sentinel species killed in the Hokonui Hills and six fringe VCZs, 2007/08 to 2013/14

The following data were supplied by K Crews, TBFree New Zealand, and were sourced from TBFree New Zealand databases. There are some known omissions, specifically some tens of resident and released pigs killed as part of a 'released sentinel pig' trial conducted in the area in about 2008-2009.

VCZ	*Strata*	*Ferrets*							*Pigs*	
		07/08	*08/09*	*09/10*	*10/11*	*11/12*	*12/13*	*13/14*	*07/08*	*08/09*
Ben Bolt	Bush	1	1	3		2	1	1		
	Farmland	5	2	9		15	1			
Glencoe	Bush	2						1		7
	Farmland	1					2			
Hokonui Hills	Bush								1	2
Repeater	Bush							1		
	Farmland		2	2		5				
Retreat	Bush									
	Farmland		10	3		3				
Valley	Bush		6	4		2	1	2		
	Farmland	4	12	2		4				
Waitane	Bush				2	1				3
	Farmland	1				1				