Relative effects of toe-clipping and pen-marking on short-term recapture probability of McCann's skinks (Oligosoma maccanni)

Christopher Jones & Trent Bell

Landcare Research, Alexandra, New Zealand

There has been much debate about the use and acceptability of toe-clipping as a means of permanently marking reptiles and amphibians during scientific surveys. Trapping studies of reptiles and amphibians are frequently, although not always, compromised by low probabilities of recapture of individuals following their initial capture and marking. Low recapture rates can influence the precision of estimates of population size and home range that are derived from trapping data. We investigated whether there was a difference in the short-term probability of recapture of toe-clipped McCann's skinks (*Oligosoma maccanni*) compared with those marked non-invasively with a pen. We pitfall-trapped skinks for seven days in a dry scrub habitat on the South Island of New Zealand. On first capture, skinks were individually marked by either toe-clipping or writing a code on their ventral surface with a silver marker pen. The resulting recapture histories for 118 skinks were analysed using Huggins' closed-population models in the MARK program. The best supported models indicated that recapture probability was significantly affected by sex (females > males) and previous toe-clipping experience (previously clipped > not). There was no support for an effect of marking method on short-term recapture probability, which is likely to be affected by other sampling factors such as trap spacing, social behaviour or response to handling. Females' higher recapture probability may reflect their increased energetic requirements during the breeding season, making them more likely to enter traps in search of food. We speculate that previous experience of toe-clipping is a surrogate for age and social dominance leading to increased access to traps.

Key words: marking methods, mark-recapture, Oligosoma spp.

INTRODUCTION

In investigations of animal social or foraging behav-Liour, or of population dynamics, the individual animal is the fundamental unit of study. It is therefore essential that researchers are able to identify an individual accurately before associations with behaviours or presence in a study area through time can be made. A variety of marking methods have been used (reviewed by Nietfeld et al., 1994; Murray & Fuller, 2000; Phillott et al., 2007), all of which depend on the implicit assumption that the marking method does not significantly affect the behaviour or demographic parameter under study. Information from a sample of marked individuals can therefore be assumed to apply to unmarked members of the same species or demographic group (Murray & Fuller, 2000). In demographic studies using capture-mark-recapture (CMR) methods, marking an animal is assumed to have no effect on its survival (Lebreton et al., 1992). Similarly, in closedpopulation CMR studies, violation of the assumption that marking has no subsequent effect on probability of recapture means that population estimates must be based on first captures only (model M_b of Otis et al., 1978). Any additional source of variation in capture rate (e.g. temporal effects or individual heterogeneity) can further bias estimates unless accounted for in constructing the analytical models to be used (Otis et al., 1978; White et al., 1982; Williams et al., 2002).

The most commonly used marking method in reptile and amphibian studies is toe-clipping (Ferner, 1979), which is permanent in most species and, depending on the number of toes clipped per foot, can be used to individually identify large numbers of animals. In recent years there has been debate over the technique's ethical and practical limitations (May, 2004; McCarthy & Parris, 2004; Phillott et al., 2007). While toe-loss is a natural occurrence for many lizard species (Hudson, 1996; Vervust et al., 2009) and has no effect on sprint speed in terrestrial lizards (Dodd, 1993; Borges-Landáez & Shine, 2003), it can compromise clinging ability in arboreal species (Bloch & Irschick, 2004). It may also compromise long-term survival in amphibians (Clarke, 1972; Parris & McCarthy, 2001; McCarthy & Parris, 2004), although it is difficult to distinguish between effects on recapture and survival rates in such studies because observed "survival" rates are dependent on both of these parameters. So far, no published studies have explicitly investigated whether toe-clipping induces a short-term behavioural ("trap-shy") response in marked individuals.

We were alerted to this possible effect when we recorded low recapture rates of endemic *Oligosoma* skinks (snout-vent length (SVL) \leq 77 mm) in a recent pitfalltrapping study in the South Island of New Zealand (C. Jones, unpubl. data). Even after significantly reducing trap-spacing following a spatial power analysis using the program DENSITY (Efford et al., 2004), recapture probabilities remained low. Apparent behavioural responses to trapping and toe-clipping have also been suspected in other small lizards (Towns, 1975; Towns & Elliott, 1996; Freeman, 1997; Duncan, 1999, Wilson et al., 2007). It has not been possible to infer a robust causative relationship with the available literature because of variation in potentially important factors in the design of these studies (trap spacing, study species, number of toes clipped, duration of trap exposure, climate). We therefore decided to test explicitly whether marking by toe-clipping has a direct behavioural effect on short-term (i.e. within a seven-day trapping session) recapture probability of pitfall-trapped small skinks compared with a non-invasive method of marking with a marker pen. Because some of our study animals had been toe-clipped previously, we also investigated whether prior toe-clipping experience reduced subsequent capture or recapture probability.

MATERIALS AND METHODS

McCann's skinks (*Oligosoma maccanni*) are small (adult SVL \leq 73 mm) diurnal heliothermic lizards that are endemic and common in dry, rocky habitats over much of the eastern South Island of New Zealand (Patterson & Daugherty, 1995). We pitfall-trapped skinks in three established enclosures (50 × 50 m) in dry (annual mean rainfall 350 mm) scrubland dominated by introduced thyme (*Thymus vulgaris*) and sparsely distributed briar (*Rosa rubiginosa*) and radiata pines (*Pinus radiata*) at the Alexandra Airport Reserve, Central Otago, South Island, New Zealand (45°21'S 169°37'E). All enclosures faced north-west and had very similar topography. Each contained 91 pitfall traps arranged in a grid with 13 "columns" at 5 m intervals and seven "rows" at 10 m intervals. The traps consisted

of a plastic food container (depth 125 mm; base diameter 95 mm; rim diameter 120 mm) sunk into the ground so that the rim was flush with the ground surface. Each was covered with a square of plywood (150×150 mm), supported by a 60 mm nail at each corner, to protect trapped lizards from heat stress and avian predators. We baited the traps with a 1 cm³ cube of tinned pear and checked them at the same time each morning for seven days in late November 2006.

We captured 126 individual skinks over the seven days of trapping; eight of these were captured for the first time on the last day of trapping and so were released without being marked, and 10 skinks classified as juveniles (SVL \leq 44 mm) were excluded from further analyses. Twentyseven skinks (21%), distributed evenly across all three enclosures, appeared to be in poor body condition, with protruding spines and very thin tails; the majority of these were gravid females (24/27). Ventral pen marks remained clearly readable throughout the trapping period.

We assigned a marking method randomly by coin toss to skinks at their first capture. They were then marked either with a numeric code on the ventral surface with a xylene-free silver marker pen, the "pen" group (Wilson et al., 2007), or were marked permanently by removing one toe using small, sharp scissors that were cleaned with 70% ethanol between animals ("clip" group). Some skinks had suffered natural toe-loss or had been toe-clipped as part of previous studies, up to six months before the start of our work. We treated these as new animals, except that their existing pattern of toe loss was recorded and, if assigned to the clip group, the additional toe was not removed from a foot already subject to toe-loss. We measured all captured skinks (SVL), sexed them by examining the cloacal area for hemipenes, and recorded females' gravidity status.

Table 1. Summary of model selection results for probability of initial capture, p, and of recapture, c, of *Oligosoma maccani* in pitfall traps, Central Otago, New Zealand, November 2006. Models are ranked by delta AICc, the difference between each model and the best model, based on Akaike's information criterion (AICc; Akaike, 1973). Models are defined as follows: (.) parameter constant; (trt) treatment (marking method) effect on parameter; (sex) sex effect on parameter; (prev) effect of prior toe-clipping on parameter; (t) parameter varies with trapping occasion (day of capture); (g) parameter varies according to group (defined by marking method, sex and whether previously toe-clipped); + indicates an additive model.

Model	Delta			
	AICc	AICc weights	Parameters	Deviance
p(.) c(sex + prev)	0.00	0.564	4	695.73
p(.) c(sex + trt + prev)	0.66	0.406	5	694.36
p(.) c(g)	7.52	0.013	9	693.06
p(.) c(sex)	8.70	0.007	3	706.45
p(.) c(sex + trt)	9.56	0.005	4	705.28
p(.) c(g+t)	10.76	0.003	14	685.98
p(.) c(prev)	13.29	0.001	3	711.03
p(.) c(trt + prev)	13.33	0.001	4	709.06
p(.) c(trt)	24.07	0.000	3	721.82

Table 2. Relative effects of sex, marking method and previous history of toe-clipping on short-term recapture probability of *O. maccanni* based on output from the model p(.) c(sex + trt + prev), where p = probability of initial capture; c = probability of recapture; sex = effect of being female relative to male; trt = effect of toe-clipping relative to pen-marking; prev = effect of being previously toe-clipped relative to not; SE = standard error; LCI and UCI are lower and upper 95% confidence intervals, respectively. Beta estimates are logit-transformed linear regression coefficients.

Parameter	Beta estimate	SE	LCI	UCI
p intercept	-0.2705938	0.1546608	-0.573729	0.0325413
c intercept	-3.2629743	0.3726247	-3.9933186	-2.5326299
Sex	1.0297322	0.2832847	0.4744941	1.5849702
Trt	0.2972235	0.2549547	-0.2024878	0.7969349
Prev	0.9700305	0.3147726	0.3530763	1.5869847

We used Huggins' closed-CMR modelling procedures (Huggins, 1989, 1991) implemented in MARK (White & Burnham, 1999) to compare the influence of the two marking methods on the probability of recapture. Huggins' models are a parsimonious approach to estimating capture and recapture probabilities because data are not used to simultaneously estimate population size, as is the case with classical closed-population models (Otis et al., 1978). Data were pooled across grids. Our candidate set of models included time, treatment and sex effects on probability of recapture, but initial capture probability was held constant in all models. Juveniles were not included due to low sample sizes. Because some of our study animals had suffered prior toe-loss, we also examined whether previous experience of toe-clipping led to lower probability of recapture in our study. We began by classifying skinks into eight groups defined by combinations of marking method (toe-clip or pen), sex and prior toe-clip status and then constrained the models to look for linear effects of these factors on a logit scale on probability of recapture. Our starting model contained time and group effects, which were then decomposed into factors. We sequentially reduced the number of factors in our models to compare model fit with and without the factor being tested. Models were ranked based on Akaike's information criterion, adjusted for sample size (AICc: Akaike, 1973; Burnham & Anderson, 2002), and the relative support for each model was compared using the weighting approach outlined in Burnham & Anderson (2002).

RESULTS

Two models from our candidate set received most of the support from the data. These included both sex and previous toe-clipping effects on recapture probability (Table 1). The second-ranked model also included an effect of marking method, but examination of the logittransformed regression coefficients (beta estimates) for this model showed this to be a relatively weak positive effect of toe-clipping on recapture probability, with 95% confidence intervals that included zero (Table 2). The beta estimates for the effects of sex and previous toe-clipping suggested that probability of recapture increased for females compared with males and previously toe-clipped compared with unclipped animals.

DISCUSSION

Our analyses showed no evidence that clipping a single toe from skinks had a negative effect on their subsequent probability of recapture compared with the relatively noninvasive method of marking with a pen. Some authors have reported very low recapture rates of small skinks in studies where pitfall-trapping and toe-clipping were used. Towns & Elliott (1996) reported that 75-80% of a range of small skink species were never recaptured and Freeman (1997) failed to recapture 96% of marked O. nigriplantare polychroma. Similarly, 82% of O. lineoocellatum trapped and toe-clipped by Duncan (1999) were captured only once. Although this may seem to indicate a general pattern, any link between toe-clipping and low recapture rates may be correlational or coincidental rather than causative. Both Dixon (2004) and Wilson et al. (2007) recorded low within-session recapture rates of small Oligosoma skinks without using toe-clipping to mark individuals.

Toe-loss is a natural occurrence in many terrestrial lizard species (Schoener & Schoener, 1980; Middelburg & Strijbosch, 1988; Hudson, 1996; Vervust et al., 2009) and has been shown to have no effect on running speed in three independent studies (Huey et al., 1990; Dodd, 1993; Borges-Landáez & Shine, 2003). Furthermore, Langkilde & Shine (2006) showed that the removal of three toes from *Eulamprus heatwolei* resulted in no significant increase in stress response, as measured by plasma corticosterone levels, compared with a control group of unclipped skinks.

In our study, female skinks were more likely to be recaptured than males. We suggest that this resulted from hunger for the pear bait overcoming any trap-shy response. Around two-thirds of trapped females were gravid and the majority of these were in poor condition, with severely depleted caudal lipid stores. This depletion was probably related to the unusually dry winter and spring that preceded our study and its effect on food availability. In a study of the closely related *O. n. polychroma*, Barwick (1959) noted that the "usually round tail becomes emaciated and square in cross-section in lizards deprived of their normal food supply". Although some species of skink may be resistant to fluctuations in food supply postovulation (Doughty & Shine, 1998), little is known about the energetics of pregnancy in *O. maccanni* and how these processes are related to temporal variations in food supply.

Surprisingly, those skinks that had been toe-clipped during previous studies were more likely to be recaptured during our study than previously unmarked animals. Although we can offer no definitive explanation for this effect, lizards that were marked prior to this study had a greater mean SVL than those not previously marked (t=76.53; df=123; P<0.001), which would imply a greater average age. We suspect that the effect could be linked to territorial behaviour, with newly captured skinks perhaps being more transient, or to dominance of older, more established lizards affecting access to traps, similar to previous reports of aggressive behaviour between individual *O. maccanni* based around defence of specific sites (Patterson, 1992).

The low recapture rates recorded in some studies of small terrestrial lizards may be due to other factors, such as trap spacing. Little is known about the home-range sizes of these species, and published estimates vary greatly. Patterson (1985) estimated home range at 330 m², which would give a diameter of twice our largest trap spacing. Barwick (1959) estimated home-range diameter to be 5.5 m for O. n polychroma. Low recapture rates have been recorded by Dixon (2004) and Duncan (1999) for traps spaced at 4 and 5 m, respectively. Alternatively, trap-shy responses may be induced by confinement in a pitfall trap. One of the highest stress responses recorded by Langkilde & Shine (2006) resulted from confining skinks in an unfamiliar enclosure. In contrast, hand-caught Lacerta vivipara showed no behavioural response to capture, even though the lizards were marked by toe-clipping (Massot et al., 1992).

Although we found no evidence of a short-term reduction in recapture rate due to toe-clipping, we removed only one toe per lizard whereas many researchers clip combinations of two or three toes to mark individuals. Whether removing more toes induces a "trap-shy" behavioural response should also be tested by assigning animals randomly to treatments based on the number of toes clipped. This is rarely done, as most researchers attempt to minimize the degree of mutilation by first assigning codes that require fewest toes to be clipped, thus inducing a strong temporal autocorrelation in their data. Researchers must also consider the longer term effects of toe-clipping on individuals. Studies of amphibians suggest that recapture rates decrease with increasing numbers of toes removed (Parris & McCarthy, 2001; McCarthy & Parris, 2004), and, while it is difficult to distinguish between the relative effects on survival and trappability, a precautionary approach is advisable until further clarification is available. There is also evidence that toe-clipping may have an immediate impact on climbing ability in arboreal species (Bloch & Irschick, 2004). All available evidence should be considered carefully before using a technique that may have the potential to influence behaviour and thereby lead to bias in the estimation of corresponding parameters.

ACKNOWLEDGEMENTS

This work was completed under Landcare Research Animal Ethics Committee permit number 04/07/01 and was funded by a New Zealand Foundation for Research, Science and Technology Postdoctoral Fellowship (to CJ) and the Mainstream Programme (to TB). We thank Andrea Byrom, David Fletcher, Wendy Ruscoe and Deb Wilson for commenting on earlier drafts of the manuscript.

REFERENCES

- Akaike, H. (1973). Information theory as an extension of the maximum likelihood principle. In *Second International Symposium on Information Theory*, 267–281. Petrov, B.N & Csaki, F. (eds). Budapest: Akademiai Kiado.
- Barwick, R.E. (1959). The life history of the common New Zealand skink Leiolopisma zelandica (Gray, 1843). Transactions of the Royal Society of New Zealand 86, 331– 380.
- Bloch, N. & Irschick, D.J. (2004). Toe-clipping dramatically reduces clinging performance in a pad-bearing lizard (*Anolis* carolinensis). Journal of Herpetology 37, 293–298.
- Borges-Landáez, P.A. & Shine, R. (2003). Influence of toeclipping on running speed in *Eulamprus quoyii*, an Australian scincid lizard. *Journal of Herpetology* 37, 592–595.
- Burnham, K.P. & Anderson, D.R. (2002). Model Selection and Multimodel Inference: A Practical Information-Theoretic Approach, 2nd edn. New York: Springer.
- Clarke, R.D. (1972). The effect of toe-clipping on survival in Fowler's toad (*Bufo woodhousei fowleri*). Copeia 1972, 182–185.
- Dixon, K.M. (2004). Biodiversity along a Gradient of Modification: Plant, Invertebrate and Reptile Diversity in Mid-altitude Tall Tussock (Chionochloa rigida) Grasslands, Eastern Central Otago, New Zealand. PhD thesis. Dunedin, New Zealand: University of Otago.
- Dodd, C.K. (1993). The effects of toe clipping on sprint performance of the lizard *Cnemidophorus sexlineatus*. *Journal of Herpetology* 27, 209–213.
- Doughty, P. & Shine, R.(1998). Reproductive energy allocation and long-term energy stores in a viviparous lizard (*Eulamprus tympanum*). *Ecology* 79, 1073–1083.
- Duncan, P. (1999). *Behavioural and Ecological Factors Affecting the Trappability of Two Skink Species in Nelson*. MSc thesis. Dunedin, New Zealand: University of Otago.
- Efford, M.G., Dawson, D.K. & Robbins, C.S. (2004). DENSITY: software for analysing capture–recapture data from passive capture–mark–recapture arrays. *Animal Biodiversity and Conservation* 27, 217–218.
- Ferner, J.W. (1979). A Review of Marking Techniques for Amphibians and Reptiles. Herpetological Circular 9. Oxford, Ohio: Society for the Study of Amphibians and Reptiles.
- Freeman, A.B. (1997). Comparative ecology of two *Oligosoma* skinks in coastal Canterbury: a contrast with Central Otago. *New Zealand Journal of Ecology* 21, 153–160.
- Hudson, S. (1996). Natural toe loss in southeastern Australian skinks: implications for marking lizards by toe-clipping.

Journal of Herpetology 30, 106-110.

- Huey, R.B., Dunham, A.E., Overall, K.L. & Newman, R.A. (1990). Variation in locomotor performance in demographically known populations of the lizard *Scleropus merriami*. *Physiological Zoology* 63, 845–872.
- Huggins, R.M. (1989). On the statistical analysis of capture experiments. *Biometrika* 76, 133–140.
- Huggins, R.M. (1991). Some practical aspects of a conditional likelihood approach to capture experiments. *Biometrics* 47, 725–732.
- Langkilde, T. & Shine, R. (2006). How much stress do researchers inflict on their study animals? A case study using a scincid lizard, *Eulamprus heatwolei*. Journal of Experimental Biology 209, 1035–1043.
- Lebreton, J.-D., Burnham, K.P., Clobert, J. & Anderson, D.R. (1992). Modeling survival and testing biological hypotheses using marked animals: a unified approach with case studies. *Ecological Monographs* 62, 67–118.
- Massot, M., Clobert, J., Pilorge, T., Lecomte, J. & Barbault, R. (1992). Density dependence in the common lizard: demographic consequences of a density manipulation. *Ecology* 73, 1742–1756.
- May, R.M. (2004). Ethics and amphibians. Nature 431, 403.
- McCarthy, M.A. & Parris, K.M. (2004). Clarifying the effect of toe clipping on frogs with Bayesian statistics. *Journal of Applied Ecology* 41, 780–786.
- Middelburg, J.J.M. & Strijbosch, H. (1988). The reliability of the toe-clipping method with the common lizard (*Lacerta vivipara*). *Herpetological Journal* 1, 291–293.
- Murray, D.L. & Fuller, M.R. (2000). A critical review of the effects of marking on the biology of vertebrates. In *Research Techniques in Animal Ecology: Controversies and Consequences*, 15–64. Boitani, L. & Fuller, T.K. (eds). New York: Columbia University Press.
- Nietfeld, M.T., Barrett, M.W. & Silvy, N. (1994). Wildlife marking techniques. In *Research and Management Techniques for Wildlife and Habitats*, 140–168. Bookhout, T.A. (ed.). Bethesda, MD: The Wildlife Society.
- Otis, D.L., Burnham, K.P., White, G.C. & Anderson, D.R. (1978). Statistical inference from capture data on closed animal populations. *Wildlife Monographs* 62, 1–135.
- Parris, K.M. & McCarthy, M.A. (2001). Identifying effects of toe clipping on anuran return rates: the importance of statistical power. *Amphibia–Reptilia* 22, 275–289.
- Patterson, G.B. (1985). The Ecology and Taxonomy of the Common Skink Leiolopisma nigriplantare maccanni in

Tussock Grasslands in Otago. PhD thesis. Dunedin, New Zealand: University of Otago.

- Patterson, G.B. (1992). The ecology of a New Zealand grassland lizard guild. *Journal of the Royal Society of New Zealand* 22, 91–106.
- Patterson, G.B. & Daugherty, C.H. (1995). Four new species and one new subspecies of skink, genus *Leiolopisma* (Reptilia: Lacertilia: Scincidae) from New Zealand. *Journal of the Royal Society of New Zealand* 20, 65–84.
- Phillott, A.D., Skerratt, L.F., McDonald, K.R., Lemckert, F.L., Hines H.B., Clarke, J.M., Alford, R.A. & Speare, R. (2007). Toe-clipping as an acceptable method of identifying individual anurans in mark recapture studies. *Herpetological Review* 38, 305-308.
- Schoener, T.W. & Schoener, A. (1980). Ecological and demographic correlates of injury rates in some Bahamian *Anolis* lizards. *Copeia* 1980, 839–850.
- Towns, D.R. (1975). Ecology of the black shore skink, Leiolopisma suteri (Lacertilia: Scincidae), in boulder beach habitats. New Zealand Journal of Zoology 2, 389–407.
- Towns, D.R. & Elliott, G.P. (1996). Effects of habitat structure on distribution and abundance of lizards at Pukerua Bay, Wellington, New Zealand. *New Zealand Journal of Ecology* 20, 191–206.
- Vervust, B., Van Dongen, S., Grbac, I. & Van Damme, R. (2009). The mystery of the missing toes: extreme levels of natural mutilation in island lizard populations. *Functional Ecology* 23, 996–1003.
- White, G.C., Anderson, D.R., Burnham, K.P. & Otis, D.L. (1982). Capture–Recapture and Removal Methods for Sampling Closed Populations. LA-8787-NERP. Los Alamos, NM: Los Alamos National Laboratory.
- White, G.C. & Burnham, K.P. (1999). Program MARK: survival estimation from populations of marked animals. *Bird Study* 46, 120–138.
- Williams, B.K., Nichols, J.D. & Conroy, M.J. (2002). Analysis and Management of Animal Populations. San Diego, CA: Academic Press.
- Wilson, D.J., Wiedemer, R.L. & Clark, R.D. (2007). Sampling skinks and geckos in artificial cover objects in a dry mixed grassland-shrubland with mammalian predator control. *New Zealand Journal of Ecology* 31, 169–185.

Accepted: 13 July 2010