
THE EFFECT OF TOE-CLIPPING ON THE SURVIVAL OF GECKO AND SKINK SPECIES

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Abstract.—Ethics committees that are required to oversee research activities involving the capture and handling of wild animals tend to take a cautious attitude because little has been published that quantifies their effects on animals. However, to address questions in ecology and evolution, it is often essential to be able to identify individual animals. Toe-clipping is one of the most commonly used marking techniques for individual identification of amphibians and reptiles. The effects of toe-clipping on survival have not been well studied. We used Cormack-Jolly-Seber mark-recapture models to estimate apparent survival (Φ) and the recapture probability (p) of an arboreal gecko species (*Gehyra variegata*) and a ground dwelling skink (*Morethia boulengeri*). We captured 551 geckos and 359 skinks over 12 y, individually marked them by clipping 1–7 toes, and we classified them as juvenile, sub-adult, or adult (stage). In *G. variegata*, the most parsimonious model included stage as the only factor affecting survival and year affecting capture probability. The best supported model that included the number of toes as a covariate was less than half as likely ($\Delta\text{QAICc} = 2.02$) but still had a weight of 0.2. Hence, there is a probability that the number of toes clipped had an effect on survival, with the number of toes negatively affecting survival in juveniles and subadults. In *M. boulengeri*, the most parsimonious model was constant apparent survival rates and capture probabilities. There was no evidence of an effect of the number of toes clipped on survival probability.

Key Words.—apparent survival; *Gehyra variegata*; Kinchega National Park; mark-recapture models; *Morethia boulengeri*

INTRODUCTION

To address questions in ecology and evolution, it is often necessary to be able to identify individual animals over long periods of time (Perry et al. 2011). A wide variety of techniques, such as toe-clipping, tags, tattoos and photo identification, have been used for this purpose (Henle et al. 1997). The choice of a suitable technique depends on the study species, the objectives and duration of the study, and the time and resources available to the researcher (Borges-Landaez and Shine 2003). Marking individuals can influence the physical and behavioral pattern of the study animal (Parris et al. 2010; Perry et al. 2011) and these disturbances should be minimized for scientific, as well as ethical and practical, reasons (Henle et al. 1997). Toe-clipping is a widely used method for permanently marking reptiles and amphibians in which a unique combination of digits is removed from each individual. Indeed, by clipping variable numbers of toes, thousands of combinations are possible (Henle et al. 1997; McCarthy and Parris 2004; Bloch and Irschick 2005).

Ethics committees charged with overseeing research activities on wild animals tend to take a precautionary approach because of insufficient data quantifying the

behavioral and life-history effects of capturing and handling animals (Henle et al. 1997; Wilson and McMahon 2006; Perry et al. 2011). Researchers themselves may be to blame for this lack of information, because they rarely address issues related to their own impacts and might assume that handling effects are negligible. Additionally, costs may also preclude handling effects studies, and appropriate controls on animals to test for the effects are often unavailable. Clearly, in the interests of animal welfare and sound research, there is a need for studies that objectively discuss and address both the ethical implications and scientific objectives of the research being undertaken (Wilson and McMahon 2006; Minter and Collins 2008; Parris et al. 2010).

A number of studies have tested the effects of marking methods on amphibians and have found that toe-clipping had a negative impact (McCarthy and Parris 2004; McCarthy et al. 2009; Waddle et al. 2008; Schmidt and Schwarzkopf 2010). Surprisingly, few studies have systematically evaluated the possibility of an effect of toe-clipping in reptiles (Perry et al. 2011). Most studies did not discover any effect on running performance and endurance, stress level, or survival in lizards, such as geckos and skinks (Borges-Landaez and Shine 2003;

Langkilde and Shine 2006; Jones and Bell 2010; Schmidt and Schwarzkopf 2010). Whereas Paulissen and Meyer (2000) did not find an influence on the clinging ability in the gecko *Hemidactylus turcicus*, the clinging performance of *Anolis carolinensis* was compromised by toe-clipping (Bloch and Irschick 2005). This indicates that there may be a species-specific response to toe-clipping and anyone using these techniques in mark-recapture should test for this effect.

There are several ways to investigate the impact of toe-clipping on the performance of amphibians and reptiles. Testing the clinging ability of arboreal species, such as geckos and *Anolis* lizards, is a common measure (Bloch and Irschick 2005). The ability of arboreal lizards to cling effectively to the surface can influence their ability to escape predators, find food, and defend territories (Bloch and Irschick 2005). Other measures are running performance and endurance (Borges-Landaez and Shine 2003; Schmidt and Schwarzkopf 2010), stress level (Langkilde and Shine 2006; Narayan et al. 2011), and capture probability and survival in mark-recapture field experiments (Davis and Ovaska 2001; Waddle et al. 2008; Jones and Bell 2010). Though running speed or clinging ability are often used as indicator for mortality risk, they may not be tightly related to it (Waddle et al. 2008; Jones and Bell 2010). Also, an effect of toe-clipping on survival violates the assumption that marked and unmarked individuals have the same survival probability, and thus leads to an underestimate of survival. Therefore, analyzing the effects of toe-clipping on survival is preferable in evaluating potentially negative impacts of toe-clipping on individual animals and at the population level.

To estimate survival parameters it is important to recapture individually marked animals periodically and to analyze the data quantitatively using capture-recapture models (CMR) that are available for estimation of survival rates (Lebreton et al. 1992). CMR models have the additional advantage that they allow modelling directly the effects of co-variables, here the number of toes clipped, on apparent survival and on detection probability. It should be mentioned that CMR methods usually cannot differentiate between mortality and permanent emigration; therefore, the estimates are called apparent survival (Lebreton et al. 1992).

Here, we report on a long-term demographic study estimating the influence of toe-clipping on the survival of individuals of the arboreal gecko *Gehyra variegata* and the ground-dwelling skink *Morethia boulengeri* in

Kinchega National Park, New South Wales (NSW), Australia. Many geckos have an extraordinary climbing ability, which is partially attributed to the fine structure of their toe pads (Zhao et al. 2008). The toes of the gecko *G. variegata* are all clawed except for the inner toe on each foot. These claws and toe pad structures may enable *G. variegata* to climb steep and vertical surfaces and their removal might have implications on climbing ability and survival. In contrast, the toe structure of terrestrial skinks that do not climb is less sophisticated. However, compared with arboreal geckos, the ground-dwelling skink *M. boulengeri* might be exposed to a wider range of predators including snakes and birds, which move rapidly (Henle 1989). As a result, toe-clipping might have a negative impact on the survival of both species. We used data from a mark-recapture study to determine whether increasing the number of toes removed had a negative effect on apparent survival and capture probability in *G. variegata* and *M. boulengeri*. We applied mark-recapture models (Williams et al. 2002) and tested the effect of the number of toes removed on survival and capture probability by including it as a predictor variable. We also investigated whether apparent survival and capture probability was similar between sexes and age-classes (stages), such as juveniles, sub-adults, and adults.

MATERIALS AND METHODS

Study site and sampling —The study was undertaken in Kinchega National Park from February 1996 until March 2007. The National Park is situated adjacent to the Darling River near the rural town Menindee in western New South Wales, Australia. The climate is characterized by highly variable and patchy rainfall, hot summers, and mild winters (Robertson et al. 1987). The study site for *G. variegata* contained 60 Black Box Trees (*Eucalyptus largiflorens*) in a plot of approximately 150 x 100 m. It was located in a long stretch of riverine woodland (RW I) along the Darling River. Within this habitat type, Black Box Trees are the only structure permanently occupied by *G. variegata*. A large tree is usually inhabited by two adult geckos and one to two juveniles or sub-adults, but smaller trees can entirely lack any resident geckos. The study site of *M. boulengeri* consisted of an 80 x 25 m plot in Riverine Woodland (RW II) bordering a red sand dune, approximately 300 m away from the gecko site. A dispersal study using drift fences showed that annual

emigration was very low (yearly emigration rate: 0.01–0.03) in 1986 and 1987 (Henle 1989). Trees covered approximately 50%, dwarf shrubs 20–40%, and herbs and grasses 0–60% of the area with the extent depending on rain (Henle 1989, 1990; Gruber and Henle 2004, 2008).

We sampled populations for 12–15 consecutive days each February between 1996 and 2007 using mark-recapture approaches. We sighted geckos via eye-shining at night using 130 lumen Phantom 130 head-torches (Petzl, Crolles, France) and captured them by hand. Each sampling session lasted until about two hours after sunset. We caught skinks by hand during two hours after sunrise and additionally in 24 pitfall traps placed in the centre of 10 x 10 m and 10 x 5 m grids within the study plot. We used 11 L aluminium containers without covers (as this best reflects the habitat) as pitfall traps.

We marked both geckos and skinks individually by toe-clipping for long-term identification. We made toe clips with sharp, sterilized scissors by removing the claw and a small part of the first phalange of each toe. We recorded snout-vent length (SVL), body mass, and age for all individuals; we also recorded sex for adult *G. variegata*. In *G. variegata*, sex can be accurately determined based on the shape of the tail base and the presence/absence of pre-anal pores in sexually mature individuals (Henle 1990). However, sex in *M. boulengeri* is difficult to determine in the field. We classified *G. variegata* into juveniles (born in the year of capture, mass: 0.42–0.49 g, SVL: 27–37 mm), sub-adults (born in the previous year, mass: 0.40–0.98 g, SVL: 38–49 mm) and adults (mature, mass: 1.9–3.9 g, SVL: 50–59 mm), and *M. boulengeri* into juveniles (born in the same year, mass: 0.19–0.36 g, SVL: 22–26 mm) and adults (mature, mass: 1.2–1.8 g, SVL: ≥ 40 mm; Henle 1989, 1990).

Demographic analysis.—We used the Cormack-Jolly-Seber model (CJS) as implemented in program MARK (White and Burnham 1999) to estimate overall apparent survival (Φ) and recapture rates (p). For this analysis we pooled the capture data each year across the 12–15 consecutive days of sampling. Thus, survival is estimated as the probability of surviving between successive years and capture probability is estimated as the probability of encountering an individual in a particular year. The CJS approach does not assume demographic closure and is suitable for estimating

demographic parameters given that there is an underlying stochastic process of birth, death, and emigration between sampling occasions (Cormack 1989; Lebreton et al. 1992). One of the major assumptions of the time-specific survival estimators of the CJS model is homogeneity of recapture likelihood within each specified group of individuals. This assumption is often addressed by including informative demographic or environmental covariates in a CJS model to account for differences in survival and/or recapture probabilities (Cormack 1989; Lebreton et al. 1992).

To achieve homogeneity of recapture likelihood, we distinguished three groups by classifying each of the 551 geckos and 359 skinks into stage (juveniles, sub-adults, and adults in *G. variegata*, juveniles and adults in *M. boulengeri*), sex (only for adult *G. variegata*), and number of clipped toes (toes). For each group we further included two covariates: time (year) and the number of toes clipped (Table 1 and 2). Subscripts indicated whether parameters were constant (e.g., Φ), time dependent (e.g., Φ_{year}), stage dependent (e.g., Φ_{stage}), or vary according to the numbers of toes clipped (e.g., Φ_{toes}). As it is difficult to determine the sex of juvenile *G. variegata* in the field, we could not investigate both the effect of sex and stage in the same model. An initial analysis of an effect of sex on survival showed no difference in overall survival between sexes and therefore we pooled adults into one group for subsequent analyses. We assessed potential effects of year, stage, and the number of toes on survival Φ and recapture probabilities p by evaluating the following combinations of models. In the survival terms, we allowed for an effect of year, stage, and toes, and interactive effects. We assumed that the numbers of toes clipped would likely have an additive effect on survival; hence animals that lost a greater number of toes due to marking potentially have a lower survival rate than those that lost fewer toes. The values of this covariate varied from 1–7 with a maximum of three toes per foot.

As the effect of number of toes clipped was our main concern, we studied both a constant toe clipping effect for all stages ($\Phi_{\text{stage} + \text{toes}}$) and a different effect of toes for each stage ($\Phi_{\text{stage} \times \text{toes}}$), which resulted in nine combinations, including the combination of survival being constant across year, stage, and no toe clipping effect. In the recapture term, we allowed for the following variation of recapture probability (constant, varying over years, age, and their interaction). This resulted in 9 x 4 combinations, whereas we excluded

TABLE 1. Capture frequencies (including recaptures) of *Gehyra variegata* per age class and year. Values in brackets indicate the average number of toes clipped across the individuals captured in each stage class and year.

Year	Juvenile	Subadult	Adult	Total
1996	9 (4.3)	14 (3.8)	55 (3.1)	78
1997	7 (5.1)	13 (4.8)	67 (3.9)	87
1998	12 (5)	8 (4.8)	78 (4.3)	98
1999	28 (4.2)	13 (4.3)	118 (4.4)	159
2000	16 (3.9)	25 (4.2)	94 (4.5)	135
2001	26 (4.3)	33 (3.9)	134 (4.2)	193
2002	26 (4.5)	27 (4.1)	117 (4.2)	170
2003	20 (5)	89 (4.7)	97 (4.4)	206
2004	22 (4.7)	21 (4.7)	94 (4.5)	137
2005	20 (4.8)	14 (5)	77 (4.6)	111
2006	14 (3.9)	35 (4.6)	74 (4.7)	123
2007	15 (3.9)	9 (3.8)	55 (4.7)	79
sum	215	301	1060	1576

models that would include non-identifiable parameters, such as $\Phi_{\text{year}} p_{\text{year}}$.

We used Program MARK (White and Burnham 1999) to test for the goodness of fit of the global model that included time, stage, and number of toes clipped (according to Anderson and Burnham 2002). We used quasi-likelihood adjusted Akaike’s Information Criterion

TABLE 2. Capture frequencies (including recaptures) of *Morethia boulengeri* per stage and year. Values in brackets indicate the average number of toes clipped across the individuals captured in each stage class and year.

Year	Juvenile	Adult	Total
1996	31 (3.9)	8 (4.3)	39
1997	45 (4.4)	12 (4.4)	57
1998	81 (4.2)	44 (4.2)	125
1999	75 (4.6)	59 (4.6)	134
2000	27 (4.3)	27 (4.1)	54
2001	100 (3.9)	32 (3.9)	132
2002	26 (4.2)	68 (3.8)	94
2003	4 (3.3)	62 (3.5)	66
2004	30 (3.6)	25 (3.4)	55
2005	58 (3.9)	44 (3.4)	102
2006	24 (3.8)	30 (3.6)	54
sum	501	411	912

(QAICc) corrected for small sample size and overdispersion for model comparison and for identification of the most parsimonious model from our candidate model set (Anderson et al. 1994; Burnham and Anderson 2002). We used QAICc weights as a measure of relative support for each model and present model averaged parameters calculated with MARK. We used the inverse logit link function to transform parameter estimates back to untransformed survival and recapture probabilities.

RESULTS

We caught from 78 to 206 *Gehyra variegata* from 1996 to 2007 (Table 1). Most lizards we caught were adults. Similarly, the numbers of *Morethia boulengeri* varied yearly with as few as 39 caught in 1996 and 134 caught in 1999 (Table 2). Unlike for *G. variegata*, we caught similar numbers of juvenile and adult *M. boulengeri* (Table 2). The number of toes clipped ranged from one to seven for both species, with a mean from 3.1 to 5.1 for *G. variegata* (Table 1) and 3.3 to 4.6 for *M. boulengeri* (Table 2).

Effect of toe-clipping on survival of *Gehyra variegata*.—The goodness-of-fit approach of the general CJS model using a bootstrap approach implemented in MARK indicated a slight over-dispersion ($\hat{c} = 1.31$). Therefore, we used QAICc for model evaluation. All models with ΔQAICc values < 2 should be taken into consideration (Burnham and Anderson 2002). Stage was found to be the most important factor affecting recapture probabilities, with a stage effect on survival and recapture being the most parsimonious model and all of the models with any AIC weight include stage as a factor (Table 3). The following two models with ΔQAICc of 2.02 and 2.03 included the covariate number of toes and were less than half as well supported as the best model but still had weights of 0.2. Hence, there is a probability that the number of toes clipped had an effect on survival. The model selection procedure revealed little support for an effect of time on apparent survival compared with otherwise identical candidate models excluding this factor. Furthermore, we found strong support for candidate models in which recapture probability varied over time (Table 3).

Model-averaged estimates of apparent survival and recapture probabilities with unconditional standard

TABLE 3. Candidate models for *Gehyra variegata* ranked according to the quasi-likelihood adjusted Akaike’s information criterion (Δ QAICc), their relative weights (QAICc weight), number of parameters (# par) and deviance. Models included possible effects of stage, toe-clipping (toes), time (year), or constant probabilities over years (.).

Model	Δ QAICc	QAICc weight	# par	Deviance
$\phi(\text{stage}) p(\text{year})$	0.00	0.56	14	1474.11
$\phi(\text{stage} \times \text{toes}) p(\text{year})$	2.02	0.20	17	1469.94
$\phi(\text{stage} + \text{toes}) p(\text{year})$	2.03	0.20	15	1474.08
$\phi(\text{stage}) p(\cdot)$	7.59	0.01	4	1502.08
$\phi(\text{stage}) p(\text{stage})$	8.90	0.01	6	1499.34
$\phi(\text{stage} \times \text{toes}) p(\cdot)$	9.15	0.01	7	1497.56
$\phi(\text{stage} + \text{toes}) p(\cdot)$	9.36	0.01	5	1501.82
$\phi(\text{stage} \times \text{toes}) p(\text{stage})$	10.53	0.00	9	1494.87
$\phi(\text{stage} + \text{toes}) p(\text{stage})$	10.68	0.00	7	1499.10
$\phi(\text{stage} \times \text{year}) p(\cdot)$	11.81	0.00	34	1443.96
$\phi(\text{stage} \times \text{year}) p(\text{stage})$	12.69	0.00	36	1440.56
$\phi(\text{stage} \times \text{toes}) p(\text{stage} \times \text{year})$	25.32	0.00	36	1453.19
$\phi(\text{stage} + \text{toes}) p(\text{stage} \times \text{year})$	25.40	0.00	34	1457.55
$\phi(\text{stage}) p(\text{stage} \times \text{year})$	29.73	0.00	36	1457.59
$\phi(\text{year}) p(\text{stage})$	40.82	0.00	14	1514.94
$\phi(\text{year} + \text{toes}) p(\text{stage})$	42.80	0.00	15	1514.85
$\phi(\cdot) p(\text{stage})$	51.01	0.00	4	1545.50
$\phi(\text{toes}) p(\text{stage})$	52.18	0.00	5	1544.64
$\phi(\text{year} \times \text{toes}) p(\text{stage})$	55.86	0.00	25	1507.10
$\phi(\text{year}) p(\cdot)$	69.15	0.00	12	1547.36
$\phi(\text{toes}) p(\text{stage} \times \text{year})$	69.48	0.00	33	1503.77
$\phi(\cdot) p(\text{year})$	70.87	0.00	12	1549.09
$\phi(\text{year} + \text{toes}) p(\cdot)$	71.04	0.00	13	1547.21
$\phi(\cdot) p(\text{stage} \times \text{year})$	71.98	0.00	34	1504.13
$\phi(\text{toes}) p(\text{year})$	72.43	0.00	13	1548.60
$\phi(\cdot) p(\cdot)$	76.95	0.00	2	1575.46
$\phi(\text{toes}) p(\cdot)$	77.94	0.00	3	1574.44
$\phi(\text{year} \times \text{toes}) p(\cdot)$	83.73	0.00	23	1539.16

errors were calculated based on model weights of all candidate models. The mean annual survival for adult and sub-adult geckos was 0.71 ± 0.02 and 0.66 ± 0.05 across years (using the mean number of toes clipped as value for the covariate toes), respectively. The mean annual survival of juvenile geckos was lower with 0.27 ± 0.04 across years. Recapture probability varied between years (mean = 0.69 ± 0.02).

Using the best-supported model that included the number of toes as a covariate (model 2), survival rates

declined with increasing number of toes clipped for juvenile and subadult geckos, but slightly increased for adult geckos. In juveniles the rate declined by 5.4% per clipped toe and in sub-adults by 6.2% (Fig. 1). This effect was weak and not significant for any stage class because the estimate of the effect included zero for all stages (95% confidence intervals of beta [toes] for juveniles = $-0.69-0.12$, sub-adults = $-0.83-0.22$, adults = $-0.087-0.22$).

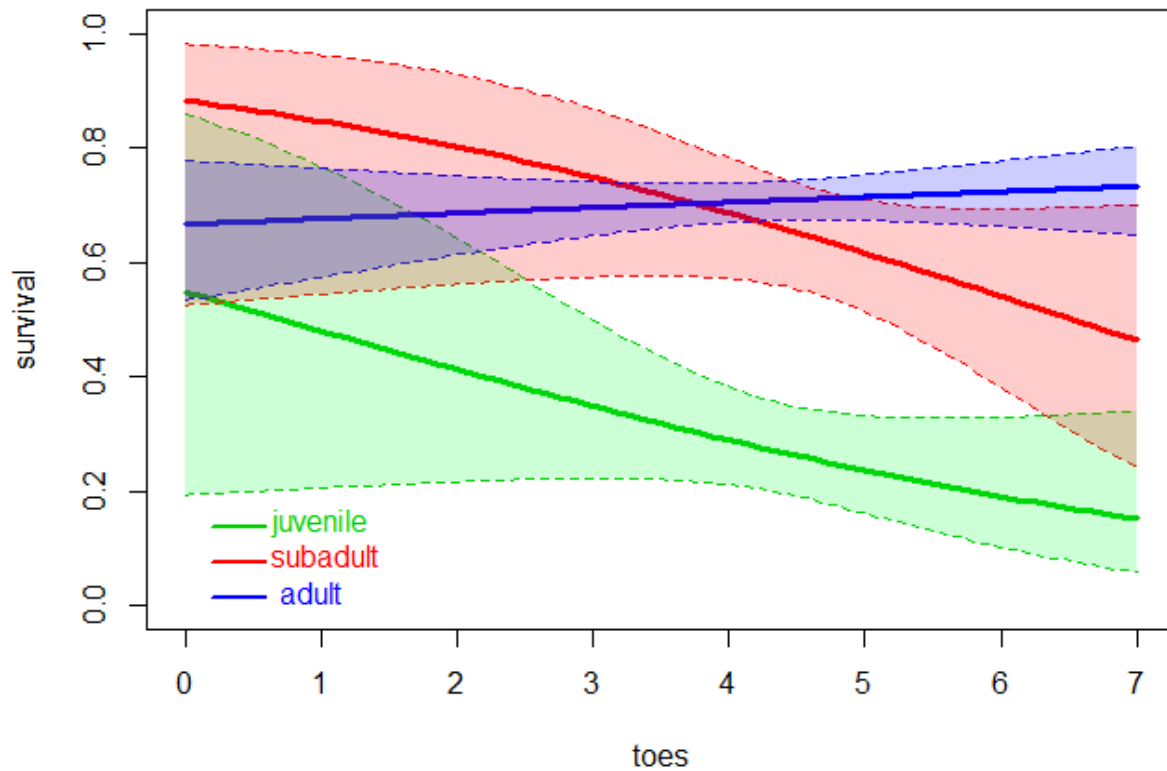


FIGURE 1. Relationship between the number of toes clipped (toes) and the survival rate including upper and lower 95% confidence intervals of juvenile, sub-adult, and adult *Gehyra variegata* under the most parsimonious model that included a toe-clipping effect (second best of all models) [Table 3, model 2, $\Phi(\text{stage} \times \text{toes}) p(\text{stage})$]. Juveniles, sub-adults, and adults are indicated by green, red, and blue colored lines, respectively.

Effect of toe-clipping on survival of *M. boulengeri*.—The goodness-of-fit approach of the general CJS model using the bootstrap approach implemented in MARK, indicated no over-dispersion ($\hat{c} = 1.05$) and therefore no adjustment was necessary. The most parsimonious model was constant apparent survival rates (Table 4). There was no evidence of the covariate toes within in the most parsimonious models (up to a threshold of 2 for ΔAIC) and cumulative weights of all models with a toe effect, including models with low support ($\Delta\text{AIC} > 5$), were only 0.28. The best model with an effect of the number of toes had low support ($\Delta\text{AIC} = 2.03$). The model selection procedure revealed support for candidate models in which recapture probability is constant over time, but also models with a stage specific capture rate had some support (Table 4).

Mean apparent survival was constant between years and the same for adult and juvenile skinks (0.34 ± 0.04). Recapture probability was also constant and the same for adults and juveniles (0.48 ± 0.08). When using the best supported model that included the number of toes as a

covariate [model 4, $\Phi(\text{toes}) p(\cdot)$], the effect of the number of toes on survival was minor (Fig. 2). The estimate on the effect included zero for both groups (95% confidence intervals of beta [toes] for juveniles and adults = -0.209 – 0.219).

DISCUSSION

Ethical and scientific concerns impose increasing pressure for marking methods to be selected for minimum impact; the adoption of such methods is becoming mandatory because, in many countries (e.g., the USA, Australia, the European Union), any marking methods requires special permits. Therefore, it is necessary to provide information about marking methods used in the field to enable comparisons of these methods and to determine if they impact individual survival (Henle et al. 1997; Lindner and Fuelling 2002; Perry et al. 2011). As a contribution to this debate and to provide useful information, we evaluated the effect of toe-clipping on the survival of the gecko *G. variegata* and

the skink *M. boulengeri* during a long-term field study. We did not detect effects of toe-clipping on apparent survival or capture probabilities in adults of either species. However, in juvenile and sub-adult geckos, the survival rate may have decreased by 5.4% or 6.2% per clipped toe. As this result was not significant and the support for this model was less than half of that for the best model, we conclude that we could not find an effect of toe-clipping on survival. As sample sizes were smaller for these two groups, it warrants further experimental study with larger numbers of juveniles and subadults.

Our results also showed that the survival rates in general are lower for juvenile (27%) than for adult geckos (71%), while there is no difference in survival rates for adult and juvenile skinks (34%). In most lizards, mortality among juveniles in the first year of life exceeds annual mortality of adult animals (Rogovin and Semenov 2004). The size difference between adult and juvenile geckos is larger than for skinks, and juvenile *M. boulengeri* reach adult size quickly (within 6–12 mo; Henle 1989). This might explain the difference in the age-(in) dependency of survival rates. The difference in survival rates between the two species can be explained by a large number of predator species for the diurnal and terrestrial *M. boulengeri* but few for the nocturnal and arboreal *G. variegata*, especially adults (Henle 1989, 1990). Concomitantly, *M. boulengeri* produced about three times as many offspring per year as *G. variegata* even in high density years that preceded a population crash (Henle 1989, 1990). Recently, there have been speculations that the survival rates of juveniles might be biased and misleadingly low as a result of the difficulty of capturing and recapturing small individuals (Pike et al. 2008). However, in our study, recapture probabilities for juvenile geckos were not significantly different from those for adults.

Evaluation of mark-recapture methods for estimating survival.—In numerous studies, mark recapture data are used to estimate survival probability of long-lived species (Lebreton et al. 1992; Cormack 1993; Chaloupka and Limpus 2005; Dodd et al. 2006; Pike et al. 2008). The oldest individuals of *G. variegata* and of *M. boulengeri* in our study had ages of 28 y and 4 y (in 2014), respectively. For studies of long-lived species, reliable estimation of adult survival probability

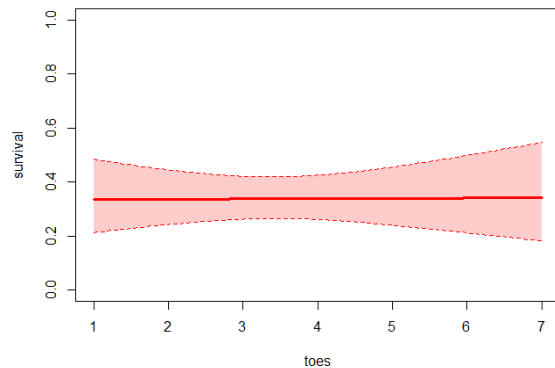


FIGURE 2. Relationship between the number of toes clipped (toes) and the survival rate including upper and lower 95% confidence intervals of *Morethia boulengeri* under the most parsimonious model that included a toe-clipping effect (fourth best of all models [Table 4, model 4, $\Phi(\text{toes})$ p(.)]).

can be crucial in understanding population dynamics (Doak et al. 1994) and hence for effective management of a population. It has long been recognized that the reliability of survival probability can be affected by variation among capture probabilities of individuals, called capture heterogeneity (Fletcher et al. 2012). However, the heterogeneity among individuals in our species is small and differences between age groups have been tested. In addition, our sampling effort was high across years, based on an average population size estimate of around 56 adult geckos over the years (Gruber and Henle 2008).

Toe-clipping effects on the survival of reptiles and amphibians.—Our results are consistent with the finding

of other studies on geckos and skinks (Paulissen and Meyer 2000; Borges-Landaez and Shine 2003; Langkilde and Shine 2006). In the gecko *Hemidactylus turcicus*, clipping of four toes had no effect on the species' ability to cling and run along vertical walls (Paulissen and Meyer 2000). Eastern Water Skinks (*Eulamprus quoyii*) were run in a race track experiment and neither the average nor maximum running speeds of lizards were affected by clipping three toes (Borges-Landaez and Shine 2003). Clipping of three toes also did not induce significant increases in corticosterone levels in the skink *E. heatwolei*, suggesting that these stimuli generated relatively little stress (Langkilde and Shine 2006). A mark-recapture study on McCann's Skink (*Oligosoma macanni*) showed no evidence that

TABLE 4. Candidate models for *Morethia boulengeri* ranked according to the quasi-likelihood adjusted Akaike’s information criterion ($\Delta AICc$), their relative weights (AIC weight), number of parameters (# par) and deviance. Models included possible effects of stage, toe-clipping (toes), time (year), or constant probabilities over years (.).

Model	$\Delta AICc$	AICc weight	# par	Deviance
$\phi(.) p(.)$	0.00	0.35	2	462.86
$\phi(.) p(\text{stage})$	1.78	0.14	3	462.61
$\phi(\text{stage}) p(.)$	1.83	0.14	3	462.66
$\phi(\text{toes}) p(.)$	2.03	0.13	3	462.86
$\phi(\text{stage}) p(\text{stage})$	3.80	0.05	4	462.59
$\phi(\text{toes}) p(\text{stage})$	3.81	0.05	4	462.60
$\phi(\text{stage} + \text{toes}) p(.)$	3.86	0.05	4	462.65
$\phi(\text{stage} \times \text{toes}) p(.)$	5.53	0.02	5	462.27
$\phi(\text{stage} + \text{toes}) p(\text{stage})$	5.84	0.02	5	462.58
$\phi(\text{stage} \times \text{year}) p(.)$	6.40	0.01	18	435.50
$\phi(\text{stage} \times \text{toes}) p(\text{stage})$	7.54	0.01	6	462.22
$\phi(\text{stage} \times \text{year}) p(\text{stage})$	8.42	0.01	19	435.31
$\phi(\text{year}) p(.)$	8.85	0.00	11	453.06
$\phi(.) p(\text{year})$	9.40	0.00	11	453.61
$\phi(\text{stage}) p(\text{stage} \times \text{year})$	9.57	0.00	17	440.86
$\phi(\text{toes}) p(\text{stage} \times \text{year})$	9.57	0.00	17	440.87
$\phi(\text{year}) p(\text{stage})$	10.76	0.00	12	452.85
$\phi(\text{year} + \text{toes}) p(.)$	10.89	0.00	12	452.98
$\phi(\text{stage}) p(\text{year})$	11.38	0.00	12	453.47
$\phi(\text{toes}) p(\text{year})$	11.52	0.00	12	453.61
$\phi(\text{stage} + \text{toes}) p(\text{stage} \times \text{year})$	11.76	0.00	18	440.86
$\phi(\text{year} + \text{toes}) p(\text{stage})$	12.83	0.00	13	452.78
$\phi(\text{stage} + \text{toes}) p(\text{year})$	13.51	0.00	13	453.46
$\phi(\text{stage} \times \text{toes}) p(\text{stage} \times \text{year})$	13.94	0.00	19	440.84
$\phi(\text{stage} \times \text{toes}) p(\text{year})$	15.22	0.00	14	453.02
$\phi(.) p(\text{stage} \times \text{year})$	18.42	0.00	21	440.87
$\phi(\text{year} \times \text{toes}) p(.)$	21.97	0.00	21	444.42
$\phi(\text{year} \times \text{toes}) p(\text{stage})$	24.05	0.00	22	444.26

clipping a single toe had a negative effect on their short-term survival compared with the relatively non-invasive method of marking with a pen (Jones and Bell 2010). However, clipping 1–4 toes had a negative effect on the clinging ability observed among anole species (Elstrott and Irschick 2004; Bloch and Irschick 2005), and clipping of four toes had a negative impact on the immediate endurance and running speed of the skink *Carlia pectoralis* (Schmidt and Schwarzkopf 2010). Our study is the first long-term study of the effects of toe-

clipping on survival of a reptile species. It did not show any effect on *M. boulengeri* nor on adult *G. variegata*, but it might have had a small effect on juveniles and subadults, though the effect was not significant and the model had less support than models with no effect.

In contrast to lizards, a number of studies observed an effect of toe-clipping on the survival of amphibians (Waddle et al. 2008; Schmidt and Schwarzkopf 2010; Perry et al. 2011). For example, jump distance decreased in *Litoria nasuta* frogs immediately after

marking, and toe-clipped individuals (four toes) jumped less far relative to their original jump distance (Schmidt and Schwarzkopf 2010). After two weeks, there was an increase in jump distance of the toe-clipped frogs, but for all groups, performance was lower than at the start of the experiment. Mark-recapture studies on the Green Tree Frog (*Hyla cinerea*) revealed a significantly decreased survival of 5% and 11.2% with three and four toes removed, respectively, compared with individuals with just two toes removed (Waddle et al. 2008). Mark-recapture studies on salamanders suggested a large effect on recapture rate (14–17%), but only a negligible effect on survival rate (0–2% when removing three toes; McCarthy et al. 2009). In another study, toe-clipped (three toes) salamanders gained less body mass than the control group, suggesting that the foraging abilities of salamanders might be suffering (Davis and Ovaska 2001). This might affect the quantity of stored energy reserves required for survival over dry periods in summer (Davis and Ovaska 2001).

Furthermore, a number of studies in amphibians have also observed infection and inflammation of feet and limbs following toe-clipping (McCarthy and Parris 2004). While amphibians have a robust immune system, as aquatic species, injury might result in a greater risk of infections and inflammation than for reptiles (McCarthy and Parris 2004). McCarthy and Parris (2004) also demonstrated that “absences of statistically significant effects in some previously published studies could be attributed to a lack of statistical power rather than absences of actual effects.”

Alternatives to toe-clipping.—Toe-clipping is one of the most practical permanent marking techniques for long-term studies in lizards (Perry et al. 2011). It is extremely easy, fast, and inexpensive to perform and large numbers of animals can be identified individually. In contrast, Passive Integrated Transponder (PIT) tags (12 x 2 mm) and electronic readers are expensive, especially if a large number of animals have to be identified. The use of PIT tags is feasible for adult geckos, but is impractical for marking juveniles because of their small size (< 0.30 g at hatching; Henle et al. 1997; Paulissen and Meyer 2000). To evaluate the stress imposed by research practices, plasma corticosterone levels have been measured in a skink (*Eulamprus heatwolei*; Langkilde and Shine 2006) and another lizard species (*Zootoca vivipara*; Le Galliard et al. 2011). The implantation of PIT tags induced a major rise in

corticosterone levels in the skink and the stress level lasted for at least 14 d. Implanted PIT tags caused irritation, hindered movement, and occasionally migrated around under the skin or into the body cavity (Roark and Dorcas 2000; Langkilde and Shine 2006). PIT tag injection in *Z. vivipara* caused no observable increase in plasma corticosterone levels over 5 d and no negative effects on long-term growth and survival (Le Galliard et al. 2011). However, PIT-tag injection also had negative effects on locomotor activity during at least 7 d, possibly implying pain (Le Galliard et al. 2011). There are very small pit tags (6 x 1 mm) now available (Nonatec. Available from <http://www.nonatec.net> [Accessed 20 January 2015]). To our knowledge, they have not been tested in reptiles and amphibians, but have been used to monitor mice during laboratory experiments (Howerton et al. 2012).

Visual implant elastomers (VIE) are marking tags that are mainly used for marking fish (Olsen and Vollestad 2005; Sutphin et al. 2007) and amphibians (Bailey 2004; Heemeyer et al. 2007; Moosman and Moosman 2006). They consist of a biocompatible two-part fluorescent silicone elastomer material, which is mixed and injected using a hypodermic needle for subcutaneous injection. Some experience by the researcher is required to maximize tag visibility and the success of injections (Olsen and Vollestad 2005). In a study on the salamander *Eurycea bislineata wilderae*, mass gain did not differ between implanted and control individuals (Bailey 2004), but misidentification rates were very high in many studies on amphibians (Bailey 2004; Moosman and Moosman 2006; Heemeyer et al. 2007). To our knowledge this technique has only rarely been used in reptiles (Penney et al. 2001), but a recent study on skinks (*Carlia pectoralis*) showed that VIE reduced the skinks short-term endurance and running speed. However, after two weeks skinks were fully recovered (Schmidt and Schwarzkopf 2010).

Another variant for long-term marking of lizards is the technique of using coloured beads, which are attached by a surgical operation at the base of the tail of a lizard (Fisher and Muth 1989). Galdino et al. (2014) claim to have improved the technique, describing a less invasive way to attach the beads to the tail. Beads have potentially the same advantage as elastomers, but are cheaper and can be recognized from longer distance as PIT tags. A number of studies have used beads for animals > 40 mm SVL (Martins 1991, Halloy and Robles 2002, Paterson 2002), which seems to be the

recommended minimum size. As our juvenile geckos and skinks are below this recommended threshold, we could not use this technique.

Tattooing or branding (Clark 1971) are likewise precluded by the difficulty of finding implements small enough to mark juvenile geckos without seriously harming them (Klawinski et al. 1994). While recent technical advances allow microbranding (2 mm x 1 mm sized spots), and there was no evidence of branding-related mortality for marked New Zealand Common Geckos (*Woodworthia maculata*) and Copper Skinks (*Oligosoma aeneum*), brands took more than three months to heal and then faded rapidly (Hitchmough et al. 2012). Painting with small bands or spots of colored, non-toxic paint can be used on all sizes of geckos, but the marks are lost several times a year through ecdysis. The geckos must therefore be recaptured frequently for remarking (Klawinski et al. 1994), which is impractical and subjects individual lizards to stress and an increased risk of injury from repeated handling (Paulissen and Meyer 2000).

Photo identification is a promising, non-invasive method that does not alter the appearance, behavior, or survivorship of individuals (Henle et al. 1997; Sacchi et al. 2010). Its application to ecological studies is mainly limited by the time consuming comparison of pictures within large datasets and the huge variation of ornamentation patterns among different reptile species (Sacchi et al. 2010). In the case of *G. variegata*, the variegation pattern on the head and back consists of a number of uniquely shaped scales, which do not vary over the lifetime of an animal (Klaus Henle, unpubl. data). This may be suitable for photo identification. In *M. boulengeri*, a combination of head scale pattern and dorsal color pattern may also allow photographic identification, but this has not yet been evaluated and the small size of juveniles provides a challenge for

photographic identification. Advances in digital and computer technologies improve the speed of photo identification. For example, the Interactive Individual Identification System (I[3]S) has been used to reliably identify 98–99% of recaptured individuals in the lacertid lizard species *Podacris muralis* using a database of 1,043 images (Sacchi et al. 2010).

Conclusions.—Our study shows that there is no effect of toe-clipping on the skink *M. boulengeri* and on adult *G. variegata*, but there may be a small effect (not significant) on juvenile and subadult geckos. Due to the fairly high sample size (we marked 88 juvenile geckos with 215 recaptures), we believe there to be sufficient statistical power to detect an effect should it exist (McCarthy and Parris 2004). Most studies show little impact of toe-clipping on herptiles with the exception of toe-clipped amphibians (Perry et al. 2011). Notwithstanding, a change in the survival rate of marked individuals following toe-clipping invalidates one of the basic assumptions of mark-recapture studies, unless this effect is known and accounted for in subsequent analyses (Donnelly and Guyer 1994). Therefore, we recommend that mark-recapture analyses estimating recapture and survival probabilities should incorporate the effects of toe-clipping by including the number of toes clipped as a covariate (McCarthy and Parris 2004), if the effect of toe clipping has not been studied.

Acknowledgements.—The study was carried out under permits S10935 and A478 of New South Wales (NSW) National Parks and Wildlife Service, Australia. The study was approved by the animal ethics committee of the Helmholtz Centre for Environmental Research (UFZ), Germany. We thank NSW National Parks and Wildlife Service for logistic support and Wendy Dimond for comments on an earlier manuscript.

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