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Behavioural changes in an endangered grassland lizard resulting from simulated agricultural activities.

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Abstract

Agricultural modification of landscapes profoundly affect the habitat of endemic species. Most Australian native grasslands have now been taken over for agriculture activities, which have dramatically changed these grassland ecosystems. Now only tiny fragments of the once more continuous native grasslands remain, and this has had a negative impact on species that occupy this habitat. One important question is how agricultural activities have altered the behaviours of endemic species in these fragmented habitats. One such species is the endangered scincid lizard, *Tiliqua adelaidensis*, which is endemic to native grasslands in South Australia. Current population sites of this species are grazed by domestic stock. We found simulated grazing led to lower body mass increases in the lizards, increased the time that lizards spent basking at their burrow entrance, reduced the tendency of lizards to move outside of their burrow, or to move to a different burrow, but increased the tendency of lizards to disperse away from the patch of habitat provided. Simulated ploughing of the surrounding habitat led to a reduction in dispersal rates. These results suggest that heavy grazing would have adverse impacts on existing populations of *Tiliqua adelaidensis*. They confirm that lizards avoid ploughed substrate, perhaps explaining previous observations of extremely low gene flow between adjacent populations.

Key words: grazing, ploughing, lizard, behaviour, pygmy bluetongue lizard

1. Introduction

Habitat degradation because of human activity threatens the viability of many animal species (Pimm and Raven, 2000; Primack, 2010; Read and Cunningham, 2010). Agricultural activities such as grazing, ploughing and cropping are widespread forms of habitat degradation of native grasslands and woodlands (James, 2003; Pafilis et al., 2013). Continuous grazing by cattle or sheep with associated trampling effects, can reduce vegetation density, change microhabitats, and increase soil hardness, and these impacts affect the diversity of endemic invertebrates and vertebrates (Dorrough et al., 2012; Hoffmann and James, 2011; Smart et al., 2005). A major goal in the conservation of these already degraded habitats is to minimise the effect of such activities on the native species that still persist. One direct way is to stop agricultural activity in designated reserves, but further management is often required during habitat restoration. Other agricultural activities such as ploughing for cropping can have more serious effects on the microhabitat and on invertebrate diversity (Stašiov et al., 2010). Ploughing not only changes the environmental conditions, inhibiting occupancy for many species, but it can directly kill invertebrates and reduce their populations (Ivask et al., 2008; Thorbek and Bilde, 2004).

A specific question is how lower levels of these agricultural activities might affect the behaviour of native animal species, and the levels of disturbance that can be tolerated, to allow endemic biodiversity to persist away from reserves. Grazing that alters microclimates, and decreases cover, could cause animals to reduce their levels of activity, and ploughing might produce behavioural barriers to dispersal. We must identify and recognise these changes and evaluate how the agricultural habitats need to be managed to reduce impacts on the behaviour of native species.

To address these questions we chose to study an endangered Australian grassland skink, the pygmy bluetongue lizard (*Tiliqua adelaidensis*) now restricted to a few small isolated patches of the highly fragmented native grasslands of South Australia. This is a medium size (mean adult snout to vent length 107 mm), viviparous, scincid lizard. Previous research has shown this lizard uses single entrance vertical burrows constructed by mygalomorph and lycosid spiders both as refuges and as sites to ambush passing prey (Hutchinson et al., 1994). In their natural populations, lizards spend almost all of their time associated with their burrow, and rarely change burrows or move away from the immediate surrounding area, except for brief excursions to capture a passing prey item (Hutchinson et al., 1994; Milne et al., 2003b). Resident lizards rarely move more than a metre from their burrow, so their normal home ranges are very small, although lizards will sometimes move further while searching for new burrows or for mating partners, particularly in spring. Occupied burrows are vigorously defended against approaching conspecifics (Fenner and Bull, 2011) although neighbours are tolerated in burrows as close as 1 m apart (Milne et al., 2003b).

Although all known populations are on remnant fragments of native grassland, the sites are all on privately owned land, they all have been extensively invaded by exotic grasses and weeds, and they are mostly grazed by sheep (Souter, 2003). No populations are known from sites that have recently been ploughed or cropped. The previously reported persistence of populations in grazed grasslands suggests that the lizard can tolerate some level of agricultural grazing. A major question for conservation managers is to determine the impact of different levels of grazing, both to provide appropriate advice to conservation sympathetic landholders, and to develop specific management options for future reserved areas.

Previous studies have suggested that dispersing pygmy bluetongue lizards will accept artificial burrows inserted into the ground within native grassland patches (Milne et al., 2003a), but that they will not move into burrows in ploughed fields even if the burrows are immediately adjacent to occupied native grassland patches (Souter, 2003). Nor will they move into artificial burrows in patches of native grassland from which all of the above ground vegetation has been removed, simulating heavy grazing (Pettigrew and Bull, 2011). Other studies have explored the behaviour of lizards in the immediate surroundings of their burrow under different levels of simulated grazing as represented by different vegetation densities around the burrow (Pettigrew and Bull, 2014). Those studies, showing that lizards basked for longer with more of their body emerged from the burrow, and more frequently detected and attacked prey, when there was less grass around the burrow, suggested that a moderate level of grazing might benefit the lizards (Pettigrew and Bull, 2014). They suggested that benefits might be derived from less cover allowing more efficient thermal basking and allowing wider visual fields to observe potential prey (Pettigrew and Bull, 2012, 2014). In those previous experiments the simulated grazing was restricted to a small area immediately around the burrow entrance. That local reduction in vegetation density may have little impact on the density of invertebrate prey for the lizards, where real grazing over a wider area could significantly reduce prey availability. Questions that remain unanswered, and that will be addressed in the current paper, include whether lizard responses induced by localised vegetation reduction are similar in a more realistically wider area of simulated (or real) grazing, and whether other behaviours such as movement away from the resident burrow are also affected. Specifically, conservation managers will be concerned about dispersal behaviour. Dispersal has benefits in increasing gene flow within and among populations. But dispersal comes with risks for pygmy bluetongue lizards, both because it increases their exposure to predators while away from their burrow refuges (Fenner et al., 2008a; Fenner et al., 2008b), and because there is a chance they might disperse out of the small remnant fragment of native grassland habitat, and into the less suitable surrounding habitat. Dispersal away from population sites might reduce population densities, particularly if there is no balancing immigration from other sites.

Ultimately we will need to advise conservation managers and land holders about the impacts of variable levels of grazing pressure. In the current study we focus on comparing simulated, replicated grazed and ungrazed patches, but at a larger scale than previous studies. We also added a second agricultural practice, with simulated ploughing of the area surrounding a habitat patch. The aim of the study, was to confirm that previously reported behavioural responses to grazing are consistent at this larger scale of habitat manipulation, to explore the effects of grazing and ploughing on dispersal behaviour, and to develop more informed management options.

Our specific predictions were that:

i) Lizards would find less food in areas with low vegetation cover and would therefore gain weight at a lower rate, or even lose weight, relative to lizards with more vegetation cover.

ii) Lizards would spend more time at their burrow entrances waiting for prey when there was low vegetation cover

iii) Lizards would be more likely to disperse away from a habitat patch if there was less vegetation cover, because of lower feeding opportunity

iv) Dispersal would be inhibited by simulated ploughing that broke up the soil surface around the habitat patch

2. **Methods**

We established four 15 m diameter enclosures at Monarto Zoo (35°06′S; 139°09′E), near Adelaide in South Australia. Cages were located in a line, 5 m apart. They had 1 m high galvanized iron walls and bird wire roofs to prevent avian predation. Each cage was divided into three areas, a central 4 m diameter circular area, with artificial burrows (see below), where lizards were released, surrounded by a matrix, consisting of a 5 m wide ring of bare ground, with no burrows, that was considered unsuitable lizard habitat, and a 0.5 m wide ring, lightly grassed and with artificial burrows provided, around the inner perimeter of the cage. Artificial burrows for lizards were constructed from 30 cm lengths of 3 cm diameter wooden dowling with the central 2 cm drilled out. These were hammered vertically into augured holes in the ground until their entrance openings were flush with the ground surface. Pygmy bluetongue lizards readily use these in the same way as natural burrows (Ebrahimi and Bull, 2012; Ebrahimi et al., 2012; Milne et al., 2003a). We placed 41 artificial burrows in the central area of each cage, one in the centre and 40 in three concentric rings with 8, 16 and 16 burrows spaced 65-75 cm apart. We added another 30 burrows, spaced evenly around the perimeter ring, to act as refuges for any lizard that dispersed from the central area across the matrix, and to allow us to monitor that dispersal.

We planted tussocks of wild oats *Avena barbata* (mean height 20.8 ± 0.03 cm) at a density of 93 tussocks/ m^2 in the central area of two cages. *Avena barbata* is an exotic weed but it grows densely among the native grasses within natural population sites of pygmy bluetongue lizards (Souter et al., 2007). In the other two cages all grass was cut to ground level in the central region. Thus there were two treatment cages with grass (with *Avena*) and two with no grass. Then in two cages, one with and one without grass, we ploughed a 2 m wide ring in the matrix around the central area, leaving a 3 m wide unploughed ring of the matrix before the perimeter region of the cage. We used a hand-held shovel, and broke up the matrix soil surface to a depth of about 30 cm.

Sixteen pygmy bluetongue lizards (8 male and 8 female) were captured from two populations near Burra in South Australia (33º42´S; 138º56´) in Sept 2009. They were kept in individual plastic boxes (52.5 x 38 x 31cm) in a room with ambient light and temperature, and fed every day with mealworms or crickets.

We ran three trials starting at 0700 h on Oct 20, Nov 16 and Nov 26, 2010. In each trial we released four lizards, two males and two females, chosen at random from the 16 lizards, into the central region of each cage. We confined them to that region for 24 h, using a temporary, 20 cm high, black plastic wall (Ebrahimi and Bull, 2013) to allow them to become familiar with the release environment. We then removed the wall and observed lizard behaviour in each cage over the next four days.

We observed lizard behaviour, with four surveillance cameras (Longse: LICS23Hf, 3.5 mm lens), attached at 1.5 m above ground level to four star pickets in each cage, with a combined field of view that covered the 4 m diameter central area of that cage. The cameras recorded all lizard activity in the central area of each cage during the daylight hours 0700 – 1800 h on each day of the experiment, on a 16 channel h.264 DVR (ESW26), powered by four 12 V batteries.

No supplementary food was provided in the cages, although lizards could prey on natural invertebrate fauna. Video images were not detailed enough to allow us to document feeding, particularly in the grass treatment.

We derived seven behavioural parameters for each lizard from the video recordings. 1) Activity time was defined as the total time in a day from when the head of the lizard first emerged from its burrow until the last time it retreated into a burrow on that day. 2) We considered a lizard to be basking if it was sitting partially or fully emerged at its burrow entrance, and we calculated basking time for a lizard each day, in minutes per hour, by dividing the total time it was observed to be basking by 11, the total hours of video recording for that day. 3) We defined a movement if a lizard fully emerged and walked away from its burrow entrance, and then retreated back into the same burrow. Movements could include walking around the burrow area, moving to bask away from the burrow entrance, emerging to prey on passing invertebrates, or leaving the burrow for defecation. 4) We defined a burrow change if a lizard emerged from one burrow and subsequently entered another burrow within the central region that could be seen on the video recordings. 5) We defined a movement to the perimeter if a lizard was observed on the video recording to have left the central area. We counted the number of movements, the number of burrow changes in the central area, and the number of moves to the perimeter, for each lizard on each day. 6) Where a lizard changed burrows within the central region, we measured the direct line distance between the old burrow and the new one. 7) During the experiment lizards sometimes contacted each other, and this agonistic encounter always resulted in a brief scuffle, or one lizard running away from the other. For the seventh behavioural parameter, we recorded the number of these "fights" per lizard per day.

The 16 lizards were randomly assigned to new groupings of four lizards in between trials, with each lizard in a different cage in each of the three trials. The trials were treated as independent replicates of the four combinations of treatments, although the groups of lizards exposed to each treatment were not entirely independent of each other. Between trial 1 and trial 2 we replanted the central regions of the "grass" treatment cages, and re-ploughed the 2 m wide circle for the "ploughed" treatment cages.

For analyses we derived one overall mean per cage for each behavioural parameter, from all four lizards over all four days (or fewer if the lizard left the central area) in each trial. These mean values were used as the dependent variable in two way ANOVAs testing the impact of central treatment (grass/ no grass) and matrix treatment (plough/no plough). The results from these analyses need to be treated with some caution because the groups of lizards exposed to each treatment were not entirely independent of each other. Permit conditions for this endangered lizard did not allow us to catch enough lizards for a completely independent experimental design.

Behavioural parameter values are presented as means with standard errors.

3. Results

There were no significant interactions between the central treatment (grass/ no grass) and the matrix treatment (plough/ no plough) for any behavioural parameter, allowing us to examine the main effects separately. Five of the seven behavioural parameters showed a significant effect of the central grass/ no grass treatment (Table 1). In cages with no grass, lizards were active for longer in each day and basked for more minutes each hour (total activity time 3.56 ± 0.04 hour day^{-1}; basking time 20.03 ± 0.14 min h^{-1}) than in cages with grass (2.0 ± 0.05 min day^{-1}; 14.0 ± 0.16 min h^{-1}). In cages with no grass lizards moved around their burrows less often than in cages with grass (No grass: 0.66 ± 0.03 moves per lizard per day; grass: 1.35 ± 0.04 moves per lizard per day), but they moved to the perimeter significantly more often in cages with no grass (Fig 1). Although there was no effect of either treatment on the number of moves to a new burrow within the central area (overall mean 1.42 ± 0.03 moves per lizard per day), the distance moved between burrows in those moves was significantly further when there was no grass (127.4 ± 0.46 cm) than when grass was present (23.9 ± 0.20 cm). Lizard body mass increased significantly more in cages with grass (+1.73 ± 0.18 g) than in cages with no grass (-0.80 ± 0.20 g) (Table 1; Fig 2).

There was only one significant main effect of ploughing. Lizards moved significantly less often through the matrix to the perimeter when the matrix had been ploughed (Fig 1). There were no fights recorded during this experiment.

4. Discussion

Our results showed a significant influence of vegetation density on the behaviour of the pygmy bluetongue lizard. When there was no grass within the central region of the cages, lizards were active over a longer period of each day, and they basked for a higher proportion of time, than in cages with grass. When there was no grass, they also moved about on the surface close to their burrows less often, and changed burrows within the central area less often, but were more likely to disperse away from the central area than when there was grass. If this behaviour is replicated in natural populations, then reducing grass levels, for instance by heavy grazing, could lead to increased dispersal and reduction of population size.

Although we introduced lizards to entirely novel habitats in these trials, the results are completely consistent with patterns shown by Pettigrew and Bull (2012) who described lizard behaviour when grass was clipped away from a small area around natural pygmy bluetongue lizard burrows. That study reported that when there was less grass, lizards spent longer periods basking, and spent more time fully emerged than when there was more grass. Similarly, they reported in another study (Pettigrew and Bull, 2011) that lizards were less likely to move into artificial burrows if there was simulated grazing around those burrows, consistent with the current result that lizards were more likely to disperse away from treatments with no grass. In the current study, however, we were comparing simulations of no grazing with simulations of very heavy grazing pressure, involving the complete removal of grass from the no grass treatment cages. We cannot use these results to comment on any potential advantages or disadvantages to lizards with a less severe level of grazing. Some grazing might be beneficial to the lizards if it were to open up the habitat without completely removing grass cover.

The only inconsistency from previous results concerns the greater weight gain in cages with grass than in cages with no grass for the lizards in the current study. Weight gain presumably reflects the ability of lizards to find and capture invertebrate prey. Previous research had showed lizards captured prey more frequently in treatments with less grass (Pettigrew and Bull, 2012). The current results may simply reflect that there was more natural prey available in the cages when there was grass, rather than any effect on the efficiency of prey capture. Other studies have also shown that grazing can decrease the available prey for lizards (Fair and Henke, 1997; Pafilis et al., 2013). Reduced numbers of available prey in our experimental cages with no grass, could also explain why lizards bask for longer when grass density is lower, a consistent observation across this and previous studies. In another experiment, Ebrahimi and Bull (2012) showed that lizards basked less when they were fed supplementary food. Although we used the term “basking” for the behaviour of sitting partially exposed at the burrow entrance, Ebrahimi and Bull (2012) suggested that basking lizards are also keeping watch for passing prey items, and that lizards that have fed less remain exposed for longer at their burrow entrance to increase their chance of encountering and capturing some prey items. This increased basking time, while allowing the potential for more prey captures, might come with the increased risk of longer exposure to predation.

Pygmy bluetongue lizards are preyed upon by both snakes and avian predators (Fenner et al., 2008b). The conflicting demands of detecting prey and avoiding predators might influence a range of other behaviours. Thus when grass was present, and there was more cover from predation, lizards completely emerged, and moved on the surface around their burrows more often. They also changed burrows within the central area more often, perhaps reflecting a lower chance of their movements being detected. On the other hand, when there was no grass, lizards were more likely to attempt to leave the central area of the cage, perhaps because of a perception of higher predation risk, or of lower prey availability in the habitat. Ebrahimi and Bull (2012) similarly reported that lizards without supplementary food were more likely to disperse. In this study it is difficult to differentiate between whether the dispersal response was a direct consequence of the altered vegetation structure or an indirect consequence of the altered vegetation changing the food levels.

Our study is not unique in exploring the impacts of grazing on endemic lizards. Several other studies have compared arid or semi-arid lizard populations in sites with heavy and with light grazing by domestic stock. The results have not been consistent. Jones (1981) reported that heavily grazed sites had lower reptile abundance and species diversity than lightly grazed sites. James (2003) found a similar trend for most lizards although one gecko species was more abundant in heavily grazed habitats. Castellano and Valone (2006) found two species increased in abundance when grazing was reduced, apparently because of lower predation rates, while a third species decreased in abundance with grazing. And Read (2002) reported that most of the lizard species in his plots showed no response to altered grazing regime, although one agamid increased in abundance with heavy grazing. While these results generally support the idea that grazing influences lizard populations, it emphasises the need to consider each species and habitat separately, especially if conservation decisions need to be made.

The disruption of the soil surface, by simulated ploughing in the matrix around the central areas of the cages in our experiments had no significant influence on either the behaviour of lizards within the central area, or their weight gain, but it did significantly reduce the tendency for lizards to disperse across the matrix. This is consistent with previous unsuccessful attempts to use artificial burrows to encourage lizards to colonise previously ploughed fields, immediately adjacent to existing population sites (Souter, 2003). It is also consistent with genetic studies that have suggested very little recent gene flow across ploughed fields, between adjacent populations within 1 km of each other (Smith et al., 2009).

The experiments confirmed two established recommendations for sustainable management of pygmy bluetongue lizard populations in agriculturally modified habitats. These are that population sites should not be subjected either to heavy grazing or to ploughing. They also provided new insights into the behavioural responses of this lizard to reduced vegetation density and specifically into the balance between the advantages (increased opportunities to see passing prey) and disadvantages (increased risk of being detected by a predator) of exposure at their burrow entrances, and of moving around on the surface.

Finally, because we were placing our lizards into a novel habitat, our results provided some new insights into possible short-term responses of lizards to translocation. Modelling has suggested that some form of translocation will be essential for the ultimate persistence of viable populations of this species (Fordham et al., 2012). For any future translocation program we need to understand what will encourage lizards to remain close to where they are released. This is to ensure that they do not move away from areas of preferred habitat, but also to ensure that translocated individuals can locate conspecifics for mating. In our trials, at least over a few days, lizards were less likely to disperse from the release area if there was more vegetation. Irrespective of whether that was a response to more cover or more food, the management implications are the same. Release sites should not be heavily grazed.

The fact that “ploughing” inhibited dispersal may be less useful as a translocation management tool. Although this disruption of the habitat may keep released lizards in one place, it will also reduce the overall habitat quality for later population expansion into the surrounding landscape. Nevertheless it suggests that some form of matrix manipulation might provide a short term reduction of dispersal from releases, although subsequently patch connectivity at a landscape scale will be very important for maintenance of populations. Any dispersal barriers that are maintained for the long-term might reduce abundance and population growth rates, leaving isolated populations with a higher risk of local extinction (Fahrig, 2007; Leavitt and Fitzgerald, 2013; Letcher et al., 2007; Lowe and Bolger, 2002).

An implication for conservation management on land that is primarily agricultural could be to recommend that the land should not be heavily grazed or left without grass because that could decrease prey and increase dispersal for native species like the pygmy bluetongue lizard. Similar recommendations have emerged from parallel studies of other lizards (Pafilis et al., 2013; Stašiov et al., 2010; Vitt et al., 1998).

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Table 1. Two way analyses of variance for behavioural parameters in response to centre treatment (grass or no grass) and matrix treatment (ploughed or not ploughed). Values in bold indicate significant effects ($p < 0.05$).

		Activity time		Basking time		No. moves		No. moves to perimeter		Distance moved		Body weight change	
	df	*F*	p	*F*	p	*F*	p	*F*	p	*F*	p	*F*	p
Plough	1, 8	0.697	0.428	0.158	0.701	2.246	0.172	5.887	**0.041**	0.207	0.661	0.823	0.391
Grass	1, 8	6.600	**0.033**	24.91	**0.001**	8.393	**0.020**	5.887	**0.041**	23.21	**0.001**	8.692	**0.018**
Plough x Grass	1, 8	0.015	0.904	0.009	0.926	0.902	0.370	2.148	0.181	3.540	0.097	0.026	0.875

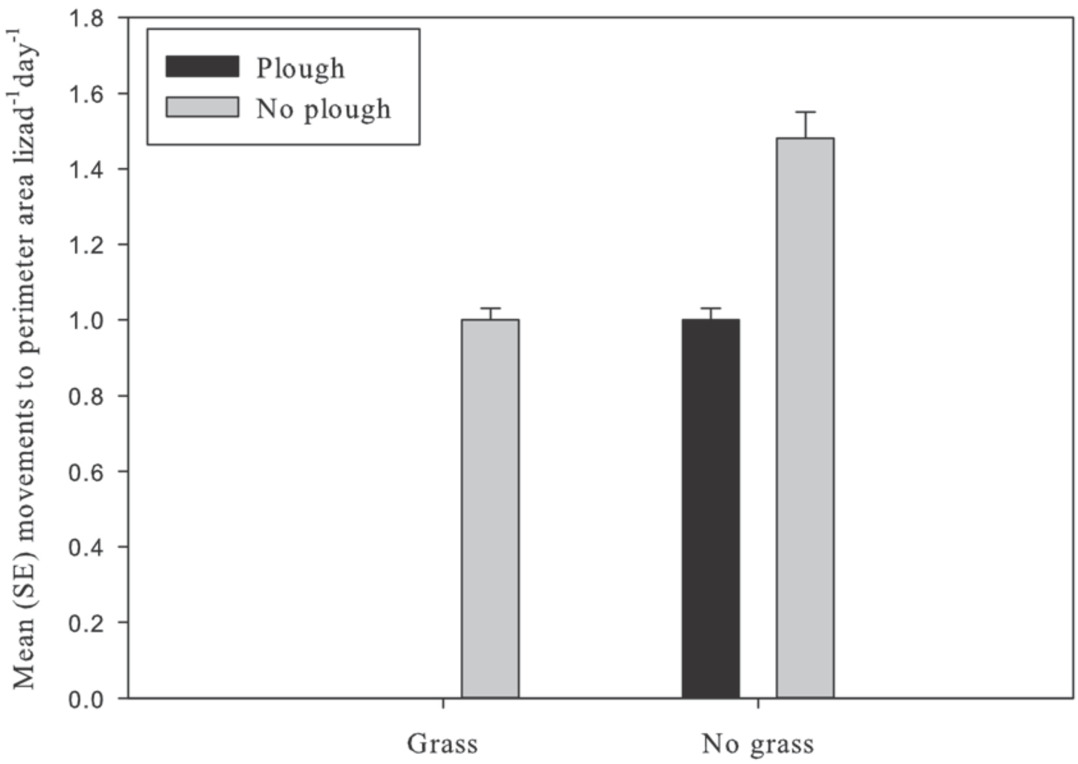

Figure 1. Mean (SE) number of lizards per day that moved to the perimeter area in each treatment combination. Note that there were no dispersal movements to the perimeter area when there was grass and the surrounding area was ploughed.

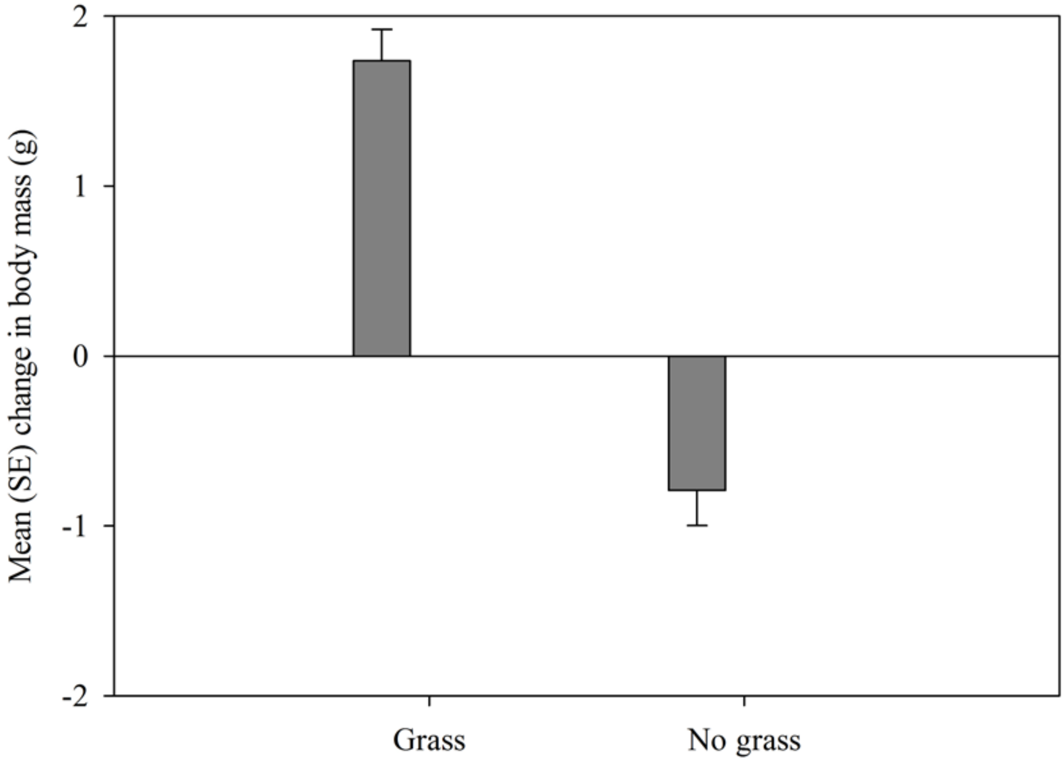

Figure 2. Mean (SE) change in body mass over the four days for lizards presented with grass and no grass. N = 8 lizards in each category.