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1 **Sub-problem learning and reversal of a multidimensional visual cue**  
2 **in a lizard: Evidence for behavioural flexibility?**

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## 26 ABSTRACT

27 Behavioural flexibility, the ability to adjust behaviour to environmental change by adapting  
28 existing skills to novel situations, is key to coping with, for example, complex social  
29 interactions, seasonal changes in food availability or detecting predators. We tested the tree  
30 skink (*Egernia striolata*), a family-living skink from eastern Australia, in a set-shifting  
31 paradigm of eight colour/shape discriminations including reversals, an intra-dimensional  
32 acquisition of a new colour/shape and extra-dimensional shift from colour to shape (and *vice*  
33 *versa*). Skinks could learn to discriminate between colour/shape pairs and reverse this initial  
34 stimulus-reward association; however, they showed no significant decrease in the probability  
35 of making a correct choice in the extra-dimensional shift suggesting that they did not form an  
36 attentional set. Subjects appear to have learnt each stage as a new problem instead of  
37 generalizing stimuli into specific dimensions (set-formation). In conclusion, tree skinks  
38 solved a discrimination reversal by focusing their attention towards visual stimuli and flexibly  
39 adjusting their choice behaviour accordingly. These lizards learned to use multidimensional  
40 visual stimuli to find a food reward, but did not generalise stimuli into dimensions.  
41 Furthermore, this study is the first to test for set-shifting in a lizard species and thereby  
42 allows us to extend set-shifting theory to a new taxon for comparison with primates, rodents,  
43 a bird and a turtle.

44

45 *Keywords:* cognition, discrimination learning, ID/ED task, reptile, set-shifting

46

## 47 INTRODUCTION

48 Social living has many benefits, but can also be a demanding environment in which  
49 interactions between individuals shape their social structure (Hinde, 1987). The resulting  
50 selective pressure is thought to have led to the evolution of extensive abilities in attention,  
51 memory and learning (Byrne, 1998; Byrne, 1994; Byrne & Whiten 1988), forming the  
52 foundation of the 'social intelligence hypothesis' (Humphrey, 1976). Complex cognition has  
53 been frequently investigated through behavioural flexibility—the ability to adjust behaviour to  
54 changes in the environment (Brown & Tait, 2015) by directing attention to essential stimuli  
55 (Dias, Robbins, & Roberts, 1996; Welsh & Pennington, 1988) and adjusting existing skills to  
56 a new problem (Manrique & Call, 2015). Behavioural flexibility can be a valuable tool in the  
57 social domain. To react flexibly to a change in the social environment (addition or removal of  
58 group members) and to selectively pay attention to interactions between individuals can be  
59 useful for tracking relationships within a social group (social monitoring; McNelis & Boatright-  
60 Horowitz, 1998). The insights gained can then be used to adjust behaviour directed towards  
61 conspecifics according to the current state of their inter-individual relationships (Byrne, 1998;  
62 McNelis & Boatright-Horowitz, 1998).

63 A common test for behavioural flexibility involves a test of attentional set-shifting  
64 which investigates the ability to apply an acquired attentional bias (by forming an attentional  
65 set) to novel situations (ID - intra-dimensional; Brown & Tait, 2015; Mukhopadhyay et al.,  
66 2008) and then to shift attention away from this established bias when relevance changes to  
67 a previously irrelevant stimulus aspect or dimension (ED - extra-dimensional; Brown & Tait,  
68 2015). It is possible to examine set-formation in a series of discriminations by quantifying  
69 acquisition speed and errors during each stage (Brown & Tait, 2015; Garner, Thogerson,  
70 Wurbel, Murray, & Mench, 2006). Perseverative errors to the former relevant dimension and  
71 a performance drop during a shift indicate a subject's level of behavioural flexibility (Brown &  
72 Tait, 2015; Garner et al., 2006).

73 A touch-screen test for ID/ED attentional set-shifting was first developed to compare

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74 human and non-human primates' attentional set-shifting ability (Dias, et al., 1996; Roberts,  
75 Robbins, & Everitt 1988). The task consists of multiple sequential visual discriminations  
76 (using shapes and lines as stimulus dimensions), designed to encourage an attentional set  
77 (through repeated exposure to consistently relevant and irrelevant information; Sutherland &  
78 Mackintosh, 1971) and then test the ability to shift away from that set. First, subjects learn a  
79 simple discrimination (SD) between stimuli of only one dimension. After reaching a  
80 predetermined learning criterion the stimulus-reward association is reversed and the other  
81 stimulus in the pair is reinforced. Next, stimuli of the irrelevant dimension are superimposed  
82 onto the SD stimuli, producing compound cues (CD), with the SD stimuli still associated with  
83 reward. After reaching criterion the reward associations are again reversed. Next, during the  
84 intra-dimensional acquisition (ID), new examples of shapes and lines are introduced. With  
85 dimensional relevance staying the same, subjects must maintain their attentional set and  
86 apply it to unfamiliar stimuli. After reaching criterion, the reward contingencies are again  
87 reversed. Finally, during the extra-dimensional shift (ED), again, unfamiliar shapes and lines  
88 are introduced. Contrary to the intra-dimensional acquisition, the reinforcement is now  
89 associated with the formerly irrelevant dimension. If set-formation occurred during earlier  
90 stages, performance in the extra-dimensional shift is expected to be worse compared to the  
91 intra-dimensional acquisition, since the previously established attentional set does not apply  
92 any more (Garner et al., 2006; Roberts et al., 1988). The extra-dimensional shift is again  
93 followed by a reversal.

94         The assumption that the number of trials to reach criterion during extra-dimensional  
95 shifting is higher than during the intra-dimensional acquisition (as a measure of attentional  
96 set-shifting) does not rely on absolute values. It is therefore possible to compare shift  
97 performance in different species (Table 1). For example, marmosets (*Callithrix jacchus*) can  
98 form an attentional set and shift to a previously irrelevant second dimension (Dias et  
99 al., 1996; Roberts et al., 1988); and similar results have been obtained in rhesus monkeys  
100 (*Macaca mulatta*; Weed, Bryant, & Perry, 2008; Baxter & Gaffan, 2007). Rodents, such as  
101 rats (e.g. Kim, Choi, Jeon, & Han, 2016; Hecht, Will, Schachtman, Welby, & Beversdorf,

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102 2014; McGaughy et al., 2014; McAlonan & Brown, 2003; Birrell & Brown, 2000) and mice  
103 (e.g. Janitzky et al., 2015; Bissonette, Lande, Martins, & Powell, 2012; Garner et al., 2006;  
104 Colacicco, Welzl, Lipp, & Wuerbel, 2002) also show the ability to form and shift attentional  
105 sets. Comparable findings in other taxa such as birds, fishes and reptiles are scarce (Table  
106 1). One study in great tits (*Parus major*) showed their ability to form an attentional set and  
107 shift to a new dimension (Titulaer, van Oers, & Naguib, 2012). Painted turtles (*Chrysemys*  
108 *picta*) show an improved performance during successive compound discrimination reversals,  
109 perhaps indicative of set-formation. However, without comparison between intra-dimensional  
110 and extra-dimensional stages, this improvement could equally be evidence of learning set-  
111 formation (training effect) instead of attentional set-formation (Cranney & Powers, 1983).

112 Most studies in lizards lack the details needed for a comprehensive comparison of  
113 attentional set-shifting because most focus on reversal performance only. For example,  
114 *Anolis evermanni* (Leal & Powell, 2012) were presented with two food wells covered by lids  
115 which animals had to dislodge to access a reward. Lizards learnt to open the food dishes  
116 using multiple methods and to discriminate between the two wells based on colour (blue and  
117 yellow); furthermore, two out of four individuals could reverse this learnt association showing  
118 flexibility in their use of visual information. A similar study investigated discrimination  
119 learning and reversal in hatchling three-lined skinks (*Bassiana duperreyi*). Almost all lizards  
120 (13/14) that learnt to displace lids could associate lid colour with reinforcement and eight  
121 showed flexibility by reversing this learnt association (Clark, Amiel, Shine, Noble, & Whiting,  
122 2014).

123 We tested tree skinks (*Egernia striolata*), which are viviparous, diurnal, family-living  
124 lizards found in arboreal as well as rocky habitats throughout eastern Australia (Wilson &  
125 Swan, 2008). Tree skinks show complex sociality in which lizards frequently live in family  
126 groups consisting of a socially monogamous parental unit and at least one generation of  
127 offspring (Whiting & While, 2017; Duckett, Morgan, & Stow, 2012; Chapple, 2003). They are  
128 visual foragers that eat plant material (including fruits) as well as insects such as  
129 cockroaches and grasshoppers (Chapple, 2003). As a diurnal, visual forager, *E. striolata* is a

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130 good model to investigate learning in a visual discrimination task. Furthermore, flexibly  
131 adjusting behaviour to changing conditions is beneficial for survival (Manrique & Call, 2015).  
132 Finally, complex sociality can select for enhanced cognitive abilities including flexibility in  
133 learning, attention, and memory (Byrne, 1998; Byrne, 1994; Byrne & Whiten 1988) which  
134 can be important tools within a social context as well as outside a social context (Byrne &  
135 Bates, 2007).

136 Our aim was to investigate behavioural flexibility in *E. striolata* by using a species-  
137 appropriate modified version of the widely-used set-shifting paradigm designed by Roberts  
138 et al. (1988). We presented individuals with visual compound cues consisting of two  
139 dimensions (colour and shape) across a series of stages including acquisitions and  
140 reversals and a final stage (extra-dimensional shift) in which the reward contingencies were  
141 shifted to the formerly irrelevant dimension. Based on this species' ecology and social  
142 structure, we predicted animals would learn the visual discriminations and show learning  
143 patterns indicative of set-formation and successful shift of attention.

144

145 **METHODS**146 *Study Animals and Husbandry*

147 We hand-captured 24 adult (snout-vent-length [SVL]  $\geq 100$  mm; Chapple, 2003) *E. striolata*  
148 (12 males and 12 females, mean SVL  $\pm$  standard deviation all:  $106.08 \pm 3.69$  mm; male:  
149  $105.58 \pm 4.14$  mm; female:  $106.58 \pm 3.29$  mm) near Albury, New South Wales ( $-35.980$  S,  
150  $146.970$  E), Australia, during April 2016. SVL, total length (TL), mass and sex (presence of  
151 hemipenes) were determined on site (Appendix Table A1). Additionally, each lizard was  
152 subcutaneously injected with a PIT-tag (Passive Integrated Transponder, Biomark, HPT8,  
153 8.4 mm; this method was chosen because animals do not show distinctive markings and it is  
154 preferable over toe-clipping) laterally 1.5 cm behind the front leg (no anaesthetic was  
155 applied), for individual identification. Skinks were transported to Macquarie University within  
156 two days of capture and transferred into individual plastic tubs (487 L x 350 W x 260 H mm)

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157 immediately after arrival. Lizards were housed indoors, with room temperature set at  $22.7 \pm$   
158  $1.9$  °C (mean  $\pm$  standard deviation, depending on season), relative humidity of 30-65% and  
159 a 12:12 h light:dark cycle. We installed heat cord underneath the enclosure to create a  
160 thermal gradient between 16-30°C ( $\pm 4$ °C). For the duration of the experiment, room  
161 temperature was monitored within enclosures using iButtons (Thermochron iButton model  
162 DS1921) which recorded temperature hourly. We used newspaper as a substrate and each  
163 enclosure had a hide, a small water bowl, and a wooden ramp. We fed lizards three times a  
164 week (Monday, Wednesday and Friday); twice with crickets powdered with vitamins  
165 (aristopet Repti-vite) and calcium (URS Ultimate Calcium) and once with baby food ( $2 \pm 0.1$   
166 g, Heinz™). During experiments, skinks were fed small amounts ( $0.15 \pm 0.01$  g) of baby food  
167 daily and crickets on Fridays; animals had *ad libitum* access to water. To ensure that  
168 animals had acclimated to the conditions of captivity, we kept them undisturbed for two  
169 weeks and made sure they were feeding consistently. All subjects were naïve and had never  
170 participated in any other cognition experiments.

171

172 *Learning experiment*

## 173 Habituation

174 To habituate the animals to the experimental setup, we transferred them to bigger tubs (683  
175 L x 447 W x 385 H mm) 12 days prior to the start of the experiment. Previous studies have  
176 shown that extensive handling and unfamiliar environments induce increased levels of stress  
177 which affects learning (Langkilde & Shine, 2006; Burghardt, 1978); therefore, animals were  
178 kept and tested in these enclosures for the entire experiment. They were identical to  
179 previous enclosures except that a second ramp was introduced (Figure 1). Baby food was  
180 presented on top of one ramp in a small white plastic saucer (3 cm in diameter) for two days  
181 (counterbalanced for side).

182

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## 183 Setup

184 During trials the newspaper substrate was taped down to prevent animals from crawling  
185 underneath and a small opaque food dish (12 mm high x 55 mm diameter, covered on the  
186 outside with black electrical tape) was placed on the top of each ramp. Both dishes  
187 contained a small amount of baby food ( $0.15 \pm 0.01$  g) and were covered with fine mesh  
188 screen; however, the reward was made accessible by a hole cut through the screen.  
189 Animals were not able to see into the feeding dishes from the starting position on the other  
190 side of the tub. Cue cards containing the stimuli (colour/shape) were fixed directly behind  
191 and as close as possible to the dishes. Half of the subjects ( $N = 12$ ) were first tested with  
192 colour as the relevant cue dimension, the other half ( $N = 12$ ) with shape (Figure 2). Within  
193 these two groups half of the subjects ( $N = 6$ ) started with stimulus 1 (Stages 1&2:  
194 triangle/light blue, stages 3&4: X/dark blue, stages 5&6: O/light orange, stages 7&8: H/light  
195 pink) and the other half ( $N = 6$ ) with stimulus 2 (second stimulus in the pair, Figure 2);  
196 making four stimulus groups and effectively counterbalancing the rewarded stimulus within  
197 the groups. All groups were counterbalanced for sex and mean SVL ( $\pm 0.1$  mm).

198

## 199 Stimuli

200 Cue cards with the stimuli (S) were made of pressed wooden coaster cards (rectangle  
201 coaster, Boyle Industries Pty Ltd, 11.3 L x 9.3 W x 0.3 H mm) and sprayed with differently  
202 coloured spray paint (Appendix Table A3). Shapes were drawn onto the sprayed cards by  
203 tracing a previously created pattern made of cardboard with a black waterproof marker  
204 (Figure 2; for information on area and circumference of the shapes see Appendix Table A5).  
205 Colour pairs were chosen to be easily discriminable based on lizards' perception  
206 (Fleishman, Loew, & Whiting, 2011) and shapes were made up of lines (e.g. X, O, H) or  
207 were solid. During experiments, left/right position was pseudo-randomly predetermined and  
208 counterbalanced for side, so that each stimulus was never on the same side more than twice  
209 in a row. For compound cues, the left/right position of each stimulus dimension varied



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210 independently of each other.

211

### 212 Experimental Procedure

213 At the start of a trial, each individual was ushered into its hide if not already in it and placed  
214 at the start position opposite the ramps. Next, both cue-cards were simultaneously fixed with  
215 putty (Bostik Blu-Tack) to the inner wall of the tub and immediately afterwards feeding  
216 dishes were placed on the ramps in the same manner in front of the stimuli. The order in  
217 which the subjects were set up was kept constant over the course of the study. After about  
218 three minutes of acclimation, the hide was removed and the trial lasted for 1.5 hours. We  
219 then returned the hide and removed feeding bowls and cues. Between trials both dishes  
220 were cleaned and rebaited, making sure that both bowls were touched. We made sure that  
221 cues and feeding dishes were never interchanged between individuals. Trials were  
222 conducted from May 2016 to March 2017. We tested subjects twice a day, between 08:00-  
223 12:30 h, five days a week (= 10 trials per week) with an inter-trial interval (ITI) of 40 minutes.  
224 All trials were videotaped (H.264 Digital Video Recorder, 3-Axis Day & Night Dome  
225 Cameras) and scored afterwards. Furthermore, during trials animals were left undisturbed to  
226 minimise stress caused by the inability to hide.

227       Choice (correct/incorrect) was scored as the first food dish an animal's snout passed  
228 over the edge of. Latency was scored as the time from first movement (directed,  
229 uninterrupted forward movement of the whole body ending in the examination of a food  
230 bowl; an interruption is defined as no movement for 10 s or more) to the first food dish  
231 examined. Animals were not actively corrected when making a wrong choice (non-correction  
232 method) and had ample opportunity to visit both stimuli and feeding dishes during trials. We  
233 used a learning criterion of 6/6 or 7/8 correct choices in consecutive trials. These criteria  
234 were chosen because they have shown to be good indicators of successful learning (Leal &  
235 Powell, 2012). To avoid overtraining, an animal was allowed no more than 100 trials for each  
236 stage. If a subject showed chance or below chance performance for at least six consecutive

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237 weeks (60 trials) or did not show criterion performance of 6/6 or 7/8 consecutive trials correct  
238 within the 100 trials, it was removed from the experiment ('non-learner'); as soon as an  
239 individual reached criterion, however, it moved on to the next stage.

240

## 241 Coding

242 A subset (about 17% = 809) of trials randomly chosen from all subjects and stages was  
243 rated by two researchers (ML and PY) unfamiliar with the experiment and blind to the tested  
244 questions as well as the first author (BS). Inter-observer reliability was calculated based on  
245 Cohens kappa (Falissard, 2012), which estimates the inter-rater agreement between two  
246 independent raters; 100% agreement equals a kappa of 1, 0% agreement a kappa of 0. It  
247 was estimated at 0.92 and 0.94 between ML and BS and PY and BS, respectively.

248

## 249 Simple Discrimination and Reversal

250 We conducted a simple associative learning test with one stimulus (e.g. X) being positively  
251 reinforced ( $S^+$ ) and the other (e.g. triangle) being unrewarded ( $S^-$ ). This stage required  
252 subjects to associate one of the stimuli with a reward. After reaching the learning criterion  
253 they moved on to a reversal (SDR). Reversals incorporated the same stimulus pairs as the  
254 simple discrimination (SD), but with reward contingencies reversed so that the former  $S^-$   
255 became  $S^+$  and *vice versa* (e.g. the previously-unrewarded X was now rewarded, and the  
256 triangle was no longer rewarded, Figure 2).

257

## 258 Compound Discrimination and Reversal

259 As soon as subjects reached criterion on the simple discrimination reversal, they were tested  
260 on the compound discrimination (CD), introducing a second stimulus dimension. The initially  
261 trained stimulus dimension remained relevant (e.g. triangle and X), while the second  
262 dimension acted as an irrelevant distractor (e.g. background colour). Stimulus group 1 was  
263 presented with two different shapes (triangle and X) superimposed on the dark and light

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264 green background colours and stimulus group 2 with two different colours at the background  
265 (light and dark blue, Figure 2). To succeed at this stage, animals had to maintain their  
266 attention on the already learnt stimulus-reward association (e.g. X is rewarded regardless of  
267 background colour), while ignoring the new stimulus dimension. After the performance  
268 criterion was met on the CD, subjects moved on to a reversal (CDR), again changing the  
269 former unrewarded  $S^-$  to  $S^+$  and *vice versa* (e.g. triangle is rewarded regardless of  
270 background colour).

271

## 272 Intra-dimensional Acquisition and Reversal

273 After reaching the learning criterion on the compound discrimination reversal, we introduced  
274 new examples of shapes and colours, however, the relevant dimension (stimulus group 1:  
275 shape, stimulus group 2: colour) stayed the same as in previous stages (Figure 2). Test  
276 subjects had to apply their attentional set to novel pairs of stimuli (e.g. a square and circle),  
277 while still ignoring the second dimension (e.g. background colour). After reaching criterion at  
278 the intra-dimensional acquisition (ID), subjects were tested on a reversal (IDR), changing  
279 reinforcement to the alternate stimulus in a pair.

280

## 281 Extra-dimensional Shift and Reversal

282 As with the previous stage, we again introduced unfamiliar colours and shapes to the test  
283 subjects, however,  $S^+$  was shifted to one of the two stimuli within the former irrelevant  
284 dimension (stimulus group 1 to colours and stimulus group 2 to shapes; Figure 2). We  
285 introduced new stimuli to avoid any partial reinforcement effects (Shanab, & McClure, 1983),  
286 and to ensure the intra-dimensional (ID) and extra-dimensional (ED) stages were equivalent  
287 (save for the effects of the primed attentional set). Therefore, the reward contingencies  
288 changed so that the previously irrelevant dimension now contained the positive and negative  
289 stimuli (e.g. light and dark pink), whereas the former relevant dimension (shapes) became  
290 irrelevant to reinforcement. After reaching criterion at the ED, subjects were presented with a

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291 reversal (EDR)—the reward now associated with the formerly unreinforced stimulus in the  
292 new relevant dimension (e.g. ED: light pink was reinforced, EDR: dark pink was reinforced).

293

294 *Statistical Analyses*

295 All statistical analyses were performed in R version 3.2.4 (R Development Core Team, 2008)  
296 and reported p-values are two tailed (raw data files and R-code are available online through  
297 Zenodo, doi: 10.5281/zenodo.1162406). Prior to the start of the experiment, we measured  
298 snout-vent length (SVL; proxy for body size) to the nearest 1 mm using a plastic ruler and  
299 mass to the nearest 0.1 g using a digital balance. We compared body condition between  
300 learners and non-learners using a linear model (LM; Bates, Maechler, Bolker, & Walker,  
301 2015) with mass as the response variable and SVL and exclusion (categorical: yes or no) as  
302 fixed effects. Furthermore, we determined whether our learning criterion was robust by  
303 examining if the number of errors per stage differed between learners and non-learners  
304 (exclusion: yes or no) in a generalized linear mixed-effects model (GLMM; Bates et al.,  
305 2015). We performed this analysis to test the prediction that ‘non-learners’ make more errors  
306 than learners. Learners could have made many errors before reaching criterion and non-  
307 learners could have made few errors but never made enough correct choices in a row to  
308 reach the learning criterion. If our criterion was robust enough to detect learning, we would  
309 expect non-learners to make significantly more errors than learners.

310 To base estimates on as many data points as possible we included data from all  
311 animals that reached criterion in any given stage (excluding the stage they were removed) in  
312 the analysis. This means that the number of individuals decreases with stage (as they were  
313 removed after not reaching criterion) which can compromise statistical power. We applied  
314 Bayesian GLMMs (Hadfield, 2010) to test if the probability of choosing correctly increased  
315 with the number of trials (indicative of learning) each animal participated in, in each of the  
316 eight stages separately. We used the same approach to test if learning performance was  
317 influenced by sex or stimulus group as well as interactions between sex and stage, sex and  
318 stimulus group, and stimulus group and stage. However, non-significant interactions were

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319 subsequently dropped and the best-fitting model, based on DIC (deviance information  
320 criterion), presented (Appendix Table A4). The final global model included only sex and  
321 stimulus group as fixed effects. Our analysis did indicate a significant sex difference in one  
322 stage (SDR) and sex was therefore included in the analysis of this stage (Table 3). In all  
323 models, “trial” was z-transformed (mean centred and scaled by the standard deviation) for  
324 better interpretability of probabilities. Models also included individual level random slopes  
325 (trial) and intercepts to account for the possible autocorrelation between successive choices.

326         Instead of an attentional set, animals might have formed a learning set (training  
327 effect). To test this, we used a Bayesian approach, like the stage-by-stage analysis but  
328 including a random intercept for stage in the random effects term. Furthermore, we ran  
329 separate GLMMs to find out if performance on the extra-dimensional shift stage differed from  
330 the intra-dimensional acquisition stage (shift performance) as well as between acquisition  
331 and reversal stages of compound, intra-dimensional and extra-dimensional discrimination  
332 (reversal learning) by focusing only on data from stages of interest (either ID and ED, CD  
333 and CDR, ID and IDR or ED and EDR) (Table 3). Trial was included in the models as a fixed  
334 effect as a scaling variable to make estimates interpretable. Model diagnostics were  
335 performed on all models to ensure that no auto-correlation between samples of the posterior  
336 distribution occurred (correlation between lags  $< 0.1$ ; Hadfield, 2010) and that sufficient  
337 mixing took place (by visually inspecting plots of MCMC chains). We used a Heidelberg and  
338 Welch diagnostic tests to ensure that the chain was long enough. Lastly, to find out if  
339 animals made perseverative errors after the initial shift (extra-dimensional shift stage),  
340 indicative of the formation of an attentional set (Garner et al., 2006), we investigated their  
341 choice during the first 10 trials of the extra-dimensional shift stage using the binomial test.

342

343 *Ethical note*

344 Our study involved non-invasive observations of animal behaviour which were approved by  
345 the Macquarie University Animal Ethics Committee (ARA # 2013/031). Collection of skinks  
346 was approved by the New South Wales National Parks and Wildlife Service, Office of

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347 Environment and Heritage (License # SL101264). Skinks were captured individually by hand  
348 and placed in cloth bags until they could be transported by vehicle to Macquarie University  
349 from Albury, New South Wales, in an insulated box. If possible both animals in a pair were  
350 collected. All animals were euthanized by injecting Lethobarb, diluted 1:1 with saline buffer  
351 solution (100mg/kg) intrapleuroperitoneally at the end of the study to extract the brain for a  
352 comparative brain study.

353

## 354 RESULTS

355 Of the 24 animals tested, 15 were excluded because they did not reach the learning criterion  
356 in 100 trials: eight (five males and three females) during the simple discrimination; four (two  
357 males and two females) during the simple discrimination reversal, two females during the  
358 intra-dimensional reversal; and one female during the extra-dimensional shift. Motivation  
359 was high during the experiment: in a total of 4854 trials (sum of all 24 individuals) there were  
360 only two trials (one each for two subjects) during which the reward was not eaten.

361         Body condition did not differ between learners and non-learners (LM: estimate = -  
362 0.49, std. error = 0.95,  $t = -0.52$ ,  $P = 0.610$ ). As predicted, animals that were removed (non-  
363 learners) during the experiment made significantly more errors compared to learners  
364 (GLMM: estimate = 0.16, std. error = 0.05,  $Z = 3.14$ ,  $P = 0.002$ ) showing that non-learners  
365 were actually performing badly. Additionally, the probability of choosing correctly increased  
366 with trial number, indicated by a positive value, for learners in each stage confirming the  
367 robustness of our learning criteria. Due to the decrease in samples size with each stage,  
368 statistical power decreases, and consequently, the width of confidence intervals increases to  
369 cross zero leading to non-significant results (Table 2). Neither stimulus group nor sex  
370 affected performance within any given stage except simple discrimination reversal, in which  
371 males' probability of choosing correctly was significantly higher than females (Table 2).  
372 Furthermore, animals did not show a shift cost (increase in number of trial to learn the extra-  
373 dimensional shift stage compared to intra-dimensional acquisition, Table 3 & Figure 3) or

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374 reversal cost (increased number of trials to learn between acquisition and reversal stages,  
375 Table 3 & Figure 3) and animals did not perseverate (base their choice on the previously  
376 reinforced dimension) on the formerly relevant dimension (Binomial test,  $N = 10$ ,  $P > 0.05$ ;  
377 Supplementary material Table 2) during the first 10 trials of the extra-dimensional shift stage.  
378 The probability of choosing correctly did not increase significantly with trial when controlling  
379 for stage as a random effect, showing that no learning set was formed either (GLMM, post.  
380 mean = 0.23, lower 95% Ci = -0.21, upper 95% Ci = 0.66,  $P = 0.275$ ).

381

382 **DISCUSSION**

383 Tree shrews learnt to discriminate between three pairs of either two shapes or two colours.  
384 Contrary to our predictions, however, animals did not show a significant decrease in the  
385 probability of choosing correctly between acquisition and reversal (no reversal cost:  
386 compound discrimination and reversal, intra-dimensional acquisition and reversal and extra-  
387 dimensional shift and reversal). Furthermore, animals learnt the extra-dimensional shift with  
388 the same level of performance as the intra-dimensional acquisition; showing no shift cost  
389 either. However, they did learn to use each new set of stimuli to find a reward and to reverse  
390 their initial association, indicating behavioural flexibility. Furthermore, the lack of evidence of  
391 attentional set-formation and the associated cost to set-shift cannot be based on our failure  
392 to reliably detect learning. Non-learners made more errors than learners during the trials  
393 they were given and our analyses show a positive effect of trial on choice performance for  
394 animals that did learn within a stage. Additionally, we found no effect of body condition or  
395 stimulus group on learning ability. Initially males were better at reversing the simple  
396 discrimination, but this difference disappeared as stages became more complex.

397         The attentional set-shifting task is designed to show attentional set-formation only if  
398 animals experience an increase in trials to criterion during the extra-dimensional shift relative  
399 to the intra-dimensional acquisition, after forming a set during the sequential progression  
400 from simple (in which an animal first learns what stimuli are relevant to find a reward; Baxter

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401 & Gaffan, 2007), then compound (in which the same stimuli plus a distractor in the form of a  
402 second dimension are presented; Birrell & Brown, 2000), to intra-dimensional discrimination  
403 (during which animals have to transfer previously acquired knowledge to unfamiliar stimuli;  
404 Brown & Tait, 2015; Dias et al., 1996). Subjects need to overcome this previously learnt  
405 attentional set, and shift their attention away from one dimension to the second, formerly  
406 irrelevant, dimension. Our data do not show evidence that the tested group of lizards formed  
407 an attentional set; therefore, we are not able to conclude that their performance at the extra-  
408 dimensional stage reflected an attentional shift. This stands in contrast to findings in  
409 primates, rodents and a bird which all showed a decrease in learning speed during the shift  
410 stage compared to the intra-dimensional acquisition (Table 1), whereas our lizards showed  
411 similar levels of learning in those stages.

412 In addition to the extra-dimensional shift stage, the standard set-shifting task includes  
413 reversal stages. During the acquisition (learning) of a discrimination, positive (rewarded  
414 stimulus) and negative (unrewarded stimulus) values are assigned to each stimulus (Wise,  
415 Murray, & Gerfen, 1996, cited by Manrique & Call, 2015) and the proportion of behavioural  
416 responses is increasingly directed towards the reinforced stimulus (learning). When a  
417 subject is confronted with a reversal it first must inhibit responding to the formerly positive  
418 stimulus and then form a new reward association with the formerly negative stimulus (Dias  
419 et al., 1996). Most of our lizards that could learn during acquisitions were able to reverse  
420 during the following stage, showing the ability to inhibit responding to an established  
421 stimulus-reward relationship and showing flexibility in their response behaviour.  
422 Furthermore, our lizards performed well during reversals showing no decrease in  
423 performance compared to the respective acquisition stages. This result stands in contrast to  
424 findings in rhesus monkeys (e.g. Weed et al., 2008), rats (e.g. McAlonan & Brown, 2003)  
425 and mice (e.g. Garner et al., 2006) which perform worse in reversals compared to  
426 acquisition.

427 During attentional set-formation, a subject first perceives both dimensions as equal  
428 and attention is increasingly directed towards the relevant dimension (Wise et al., 1996,



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429 cited by Manrique & Call, 2015). When an attentional set has formed, attention is focused on  
430 the relevant information and responses are directed towards the rewarded stimulus (Brown  
431 & Tait, 2015) within the relevant dimension. However, a shift to the second dimension  
432 requires subjects to inhibit responding to the whole dimension (Dias et al., 1996). Our  
433 animals were able to learn the dimensional shift without showing perseverative responses,  
434 but as to what strategy they used (e.g. attentional set-shift or learning of each compound  
435 cue as a distinct stimulus), and whether an attentional set was overcome, needs to be  
436 investigated in future studies. Additionally, based on our analysis, tree skinks also did not  
437 form a learning set (training effect), a predisposition to learn based on previous experience.  
438 It seems that animals treated each new version of the stimuli as a novel problem and  
439 subsequently learnt each acquisition stage individually without experiencing a training effect.  
440 The specific learning strategy used by our subjects is also unclear; our dataset is too small  
441 to permit any further analysis. Although both concepts, reversal learning and attentional  
442 shifts, are similar in the respect that they require some level of behavioural flexibility in  
443 responding to the change in stimulus relevance, attentional shifts are generally seen as  
444 more complex (Birrell & Brown, 2000; Colacicco et al., 2002).

445         In this study, colour pairs were chosen based on human-perceived brightness and  
446 shapes could be categorised into those made up of lines (e.g. X, O, H), and those that were  
447 solid (e.g. triangle, square, star). Combinations were chosen to be easily distinguishable  
448 based on the lizards' perceptual ability (they are tetrachromatic and have good visual acuity;  
449 Fleishman et al., 2011). Furthermore, skinks could have experienced a sensory bias towards  
450 one or more stimuli or a dimension. For example, rhesus monkeys perform differently when  
451 shifting according to the stimulus dimension they initially encountered. Monkeys had  
452 difficulty shifting from colour to shape but not *vice versa* (Baxter & Gaffan, 2007). During the  
453 first stage, we started each of four subsets of lizards (stimulus groups) with one of the four  
454 stimuli (light blue, dark blue, X or triangle). However, performance did not differ between  
455 stimulus groups, indicating that our dimensions were of similar difficulty to our test animals.  
456 There is a possibility that lizards used brightness instead of chroma or hue to learn the

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457 discrimination. As the order of presentation from simple discrimination to intra-dimensional  
458 reversal was bright-dark-dark-bright-bright-dark this can be seen as a sequence of simple  
459 acquisition and reversal stages. Therefore, if animals had used brightness, our data would  
460 show a decrease in trials to criterion since a reversal to a previously correct stimulus is  
461 easier than a reversal to a previously not-reinforced stimulus, but no such decrease was  
462 found. The same is true for shape solidity, area and circumference (line-solid-solid-line-solid-  
463 line, small-large-large-small-large-small and long-short-short-long-short-long). However,  
464 there is a small chance that animals used some other property of the cue cards (surface  
465 texture or minor imperfections due to the painting process) to learn the discrimination that  
466 was not visible to a human observer.

467 In conclusion, tree skinks can learn to distinguish between two visual cues made up of either  
468 two colours and/or two shapes. Furthermore, they can reverse an initial stimulus reward  
469 association and show behavioural flexibility which can be a beneficial trait while coping with  
470 environmental and social challenges. The pattern of learning suggests, however, that they  
471 neither formed an attentional set (establishment of a rule set on which subsequent choices  
472 are based) nor a learning set (animals' performance increases based on extensive training).  
473 From our results, it is unclear if the tested species is unable to establish dimensionality in  
474 compound visual stimuli, or if our methodology was not suitable to test attentional set-  
475 formation and shifting in this reptile species. It is most likely that they viewed each new pair  
476 of stimuli as a distinct problem and learnt to discriminate the stimuli as a whole, instead of  
477 generalizing to a dimension. Our study revealed new insights into visual discrimination  
478 learning in lizards that will help design future studies investigating learning in non-avian  
479 reptiles. Furthermore, adding evidence on set-shifting in species with a varying degrees of  
480 sociality will help understand the relationship between sociality and behavioural flexibility in  
481 lizards.

482

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## 489 REFERENCES

- 490 Bates, D., Maechler, M., Bolker, B., & Walker, S. (2015). Fitting Linear Mixed-Effects Models  
491 Using lme4. *Journal of Statistical Software*, 67(1), 1-48. doi:10.18637/jss.v067.i01.
- 492 Baxter, M. G., & Gaffan, D. (2007). Asymmetry of attentional set in rhesus monkeys learning  
493 colour and shape discriminations. *The Quarterly Journal of Experimental Psychology*,  
494 60(1), 1-8. doi:10.1080/17470210600971485
- 495 Birrell, J. M., & Brown, V. J. (2000). Medial frontal cortex mediates perceptual attentional  
496 set-shifting in the rat. *The Journal of Neuroscience*, 20(11), 4320-4324.
- 497 Bissonette, G. B., Lande, M. D., Martins, G. J., & Powell, E. M. (2012). Versatility of the  
498 mouse reversal/set-shifting test: Effects of topiramate and sex. *Physiology &*  
499 *Behavior*, 107(5), 781-786.
- 500 Brown, V. J., & Tait, D. S. (2015). Behavioral flexibility: attentional shifting, rule switching  
501 and response reversal. In I. P. Stolerman & L. H. Price (Eds.), *Encyclopedia of*  
502 *Psychopharmacology* (pp. 264-269). Berlin, Germany: Springer-Verlag.
- 503 Burghardt, G. M. (1978). Learning Processes in Reptiles. In C. Gans & D. W. Tinkle (Eds.),  
504 *Biology of the Reptilia. Volume 7. Ecology and Behaviour A* (Vol. 7, pp. 555-681).  
505 London, New York: Academic Press.
- 506 Byrne, R. W. (1994). *The evolution of intelligence*. Cambridge, GB: Cambridge University  
507 Press.
- 508 Byrne, R. W. (1998). Machiavellian Intelligence. *Evolutionary Anthropology*, 5(5), 172-180.
- 509 Byrne, R. W., & Bates, L. A. (2007). Sociality, evolution and cognition. *Current Biology*,  
510 17(16), R714-723. doi:10.1016/j.cub.2007.05.069
- 511 Byrne, R. W., & Whiten. (1988). *Machiavellian Intelligence: Social Expertise and the*  
512 *Evolution of Intellect in Monkeys, Apes, and Humans*. New York, NY: Clarendon  
513 Press/Oxford University Press.
- 514 Chapple, D. G. (2003). Ecology, Life-History, and Behavior in the Australian Scincid Genus

**Running head: SET-SHIFTING IN EGERNIA STRIOLATA**

- 515           Egernia, with Comments on the Evolution of Complex Sociality in Lizards.  
516           *Herpetological Monographs*, 17, 145-180.
- 517 Clark, B. F., Amiel, J. J., Shine, R., Noble, D. W. A., & Whiting, M. J. (2014). Colour  
518           discrimination and associative learning in hatchling lizards incubated at 'hot' and  
519           'cold' temperatures. *Behavioral Ecology and Sociobiology*, 68(2), 239-247.  
520           doi:10.1007/s00265-013-1639-x
- 521 Colacicco, G., Welzl, H., Lipp, H.-P., & Wuerbel, H. (2002). Attentional set-shifting in mice:  
522           Modification of a rat paradigm, and evidence for strain-dependent variation.  
523           *Behavioural Brain Research*, 132, 95-102.
- 524 Cranney, J., & Powers, A. S. (1983). The effects of core nucleus and cortical lesions in  
525           turtles on reversal and dimensional shifting. *Physiological Psychology*, 11(2), 103-  
526           111. doi:10.3758/BF03326779
- 527 Dias, R., Robbins, T. W., & Roberts, A. C. (1996). Primate Analogue of the Wisconsin Card  
528           Sorting Test- Effects of Excitotoxic Lesions of the Prefrontal Cortex in the Marmoset.  
529           *Behavioral Neuroscience*, 110(5), 872-886.
- 530 Duckett, P. E., Morgan, M. H., & Stow, A. J. (2012). Tree-Dwelling Populations of the Skink  
531           Egernia striolata Aggregate in Groups of Close Kin. *Copeia*, 2012(1), 130-134.  
532           doi:10.1643/ce-10-183
- 533 Falissard, B. (2012). psy: Various procedures used in psychometry. R package version 1.1.  
534           <https://CRAN.R-project.org/package=psy>
- 535 Fleishman, L. J., Loew, E. R., & Whiting, M. J. (2011). High sensitivity to short wavelengths  
536           in a lizard and implications for understanding the evolution of visual systems in  
537           lizards. *Proceedings of the Royal Society B: Biological Sciences*, 278(1720), 2891-  
538           2899. doi:10.1098/rspb.2011.0118
- 539 Garner, J. P., Thogerson, C. M., Wurbel, H., Murray, J. D., & Mench, J. A. (2006). Animal  
540           neuropsychology: validation of the Intra-Dimensional Extra-Dimensional set-shifting

**Running head: SET-SHIFTING IN EGERNIA STRIOLATA**

- 541 task for mice. *Behavioral Brain Research*, 173(1), 53-61.  
542 doi:10.1016/j.bbr.2006.06.002
- 543 Hadfield, J. D. (2010). MCMC methods for Multi-response Generalised Linear Mixed Models:  
544 The MCMCglmm R Package. *Journal of Statistical Software*, 33(2), 1-22.
- 545 Hecht, P. M., Will, M. J., Schachtman, T. R., Welby, L. M., & Beversdorf, D. Q. (2014). Beta-  
546 adrenergic antagonist effects on a novel cognitive flexibility task in rodents.  
547 *Behavioural Brain Research*, 260, 148-154. doi:10.1016/j.bbr.2013.11.041
- 548 Hinde, R. A. (1987). *Themes in the social sciences. Individuals, relationships & culture:*  
549 *Links between ethology and the social sciences*. New York: Cambridge University  
550 Press.
- 551 Humphrey, N. (1976). The social function of intellect. In P. P. G. Bateson & R. A. Hinde  
552 (Eds.), *Growing Points in Ethology* (pp. 303-317). Cambridge: Cambridge University  
553 Press.
- 554 Janitzky, K., Lippert, M. T., Engelhorn, A., Tegtmeier, J., Goldschmidt, J., Heinze, H. J., &  
555 Ohl, F. W. (2015). Optogenetic silencing of locus coeruleus activity in mice impairs  
556 cognitive flexibility in an attentional set-shifting task. *Frontiers in Behavioral*  
557 *Neuroscience*, 9, 286. doi:10.3389/fnbeh.2015.00286
- 558 Kim, D. H., Choi, B. R., Jeon, W. K., & Han, J. S. (2016). Impairment of intradimensional  
559 shift in an attentional set-shifting task in rats with chronic bilateral common carotid  
560 artery occlusion. *Behavioural Brain Research*, 296, 169-176.  
561 doi:10.1016/j.bbr.2015.09.007
- 562 Langkilde, T., & Shine, R. (2006). How much stress do researchers inflict on their study  
563 animals? A case study using a scincid lizard, *Eulamprus heatwolei*. *Journal of*  
564 *Experimental Biology*, 209(6), 1035-1043. doi:10.1242/jeb.02112
- 565 Leal, M., & Powell, B. J. (2012). Behavioural flexibility and problem-solving in a tropical  
566 lizard. *Biology Letters*, 8(1), 28-30. doi:10.1098/rsbl.2011.0480
- 567 Manrique, H. M., & Call, J. (2015). Age-dependent cognitive inflexibility in great apes. *Animal*  
568 *Behaviour*, 102, 1-6. doi:10.1016/j.anbehav.2015.01.002

**Running head:** SET-SHIFTING IN EGERNIA STRIOLATA

- 569 McAlonan, K., & Brown, V. J. (2003). Orbital prefrontal cortex mediates reversal learning and  
570 not attentional set-shifting in the rat. *Behavioural Brain Research*, *146*(1-2), 97-103.  
571 doi:10.1016/j.bbr.2003.09.019
- 572 McGaughy, J. A., Amaral, A. C., Rushmore, R. J., Mokler, D. J., Morgane, P. J., Rosene, D.  
573 L., & Galler, J. R. (2014). Prenatal malnutrition leads to deficits in attentional set-  
574 shifting and decreases metabolic activity in prefrontal subregions that control  
575 executive function. *Developmental Neuropsychology*, *36*(6), 532-541.  
576 doi:10.1159/000366057
- 577 McNelis, N. L., & Boatright-Horowitz, S. L. (1998). Social monitoring in a primate group: the  
578 relationship between visual attention and hierarchical ranks. *Animal Cognition*, *1*, 65-  
579 69.
- 580 Mukhopadhyay, P., Dutt, A., Kumar Das, S., Basu, A., Hazra, A., Dhibar, T., & Roy, T.  
581 (2008). Identification of neuroanatomical substrates of set-shifting ability - evidence  
582 from patients with focal brain lesions. *Progress in Brain Research*, *168*, 95-104.
- 583 R Development Core Team (2008). R: A language and environment for statistical computing.  
584 R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL  
585 <http://www.R-project.org>.
- 586 Roberts, A. C., Robbins, T. W., & Everitt, B. J. (1988). The effects of intradimensional and  
587 extradimensional shifts on visual discrimination learning in humans and non-human  
588 primates. *The Quarterly Journal of Experimental Psychology Section B*, *40*(4), 321-  
589 341. doi:10.1080/14640748808402328
- 590 Shanab, M. E., & McClure, F. H. (1983). Age and sex differences in discrimination learning.  
591 *Bulletin of the Psychonomic Society*, *21*(5), 387-390. doi:10.3758/bf03329988
- 592 Sutherland, N. S., & Mackintosh, N. J. (1971). Mechanisms of animal discrimination learning.  
593 New York: Academic Press.
- 594 Titulaer, M., van Oers, K., & Naguib, M. (2012). Personality affects learning performance in  
595 difficult tasks in a sex-dependent way. *Animal Behaviour*, *83*(3), 723-730.

**Running head: SET-SHIFTING IN EGERNIA STRIOLATA**

- 596           doi:10.1016/j.anbehav.2011.12.020
- 597 Weed, M. R., Bryant, R., & Perry, S. (2008). Cognitive development in macaques: attentional  
598 set-shifting in juvenile and adult rhesus monkeys. *Neuroscience*, *157*(1), 22-28.  
599           doi:10.1016/j.neuroscience.2008.08.047
- 600 Welsh, M. C., & Pennington, B. F. (1988). Assessing frontal lobe functioning in children:  
601 Views from developmental psychology. *Developmental Neuropsychology*, *4*(3), 199-  
602 230. doi:10.1080/87565648809540405
- 603 Wiley, R. H. (2013). Specificity and multiplicity in the recognition of individuals: implications  
604 for the evolution of social behaviour. *Biological Reviews of the Cambridge*  
605 *Philosophical Society*, *88*(1), 179-195. doi:10.1111/j.1469-185X.2012.00246.x
- 606 Wilson, S., & Swan, G. (2008). *A Complete Guide to Reptiles of Australia*. Sydney: New  
607 Holland.
- 608 Wise, S. P., Murray, E. A., & Gerfen, C. R. (1996). The frontal cortex basal ganglia system in  
609 primates. *Critical Reviews in Neurobiology*, *10*, 317e356.
- 610 Whiting, M., & While, G. (2017). Sociality in Lizards. In D. Rubenstein & P. Abbot (Eds.),  
611 *Comparative Social Evolution* (pp. 390-426). Cambridge: Cambridge University  
612 Press. doi:10.1017/9781107338319.014
- 613



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614 **Table 1.** Literature comparison between studies incorporating the described methodology.

Species	Age	Methodology	Dimensions	ED>ID	Study
<b>Primates</b>					
Common Marmoset	Sub-adult	CANTAB ID/ED	Visual	Yes	Dias et al., 1996
Common Marmoset	Sub-adult	CANTAB ID/ED	Visual	Yes	Roberts et al., 1988
Rhesus Monkeys	Juvenile	CANTAB ID/ED	Visual	Yes	Weed et al., 2008
Rhesus Monkeys	Adult	CANTAB ID/ED	Visual	Yes	
Rhesus Monkeys	Adult	CANTAB ID/ED	Visual	Partly validated	Baxter & Gaffan, 2007
<b>Rodents</b>					
Wistar rats	Adult	ID/ED	Olfactory/touch	Yes	Kim et al., 2016
Sprague Dawley rats	Adult	ID/ED	Olfactory/touch	Yes	Hecht et al., 2014
Long-Evans hooded rats	Adult	ID/ED	Olfactory/touch	Yes	McGaughy et al., 2014
Lister hooded rats	Adult	ID/ED	Olfactory/touch	Yes	McAlonan & Brown, 2003
Lister hooded rats	Adult	ID/ED	Olfactory/touch	Yes	Birrell & Brown, 2000
Mice (B6.Cg-Tg(Th-cre)1Tmd/J)	Adult	ID/ED	Olfactory/touch	No	Janitzky et al., 2015
Mice (C57BL/6)	Adult	ID/ED	Olfactory/touch	Yes	Bissonette et al., 2012
Mice (C57BL/6)	Adult	ID/ED	Olfactory/touch	Yes	Garner et al., 2006
Mice (C57BL/6J)	Adult	ID/ED	Olfactory/touch	Yes	Colacicco et al., 2002
<b>Birds</b>					
Great tits	Adult	Reversals and shift	Visual/spatial	Yes	Titulaer et al., 2012
<b>Reptiles</b>					
Painted turtle	adult	Series of ED and REV	Visual	No direct comparison	Cranney & Powers, 1983

615 Findings in primates, rodents (some examples), birds and reptiles including species tested, age of subjects, methodology and stimuli used are  
 616 listed as well as if set-formation impaired performance during the extra-dimensional shift. ID – intra-dimensional acquisition, ED – extra-  
 617 dimensional shift, REV – reversal, CANTAB ID/ED - Cambridge Neuropsychological Automated Test Battery ID/ED attentional set-shifting test.

618

619 **Table 2.** Summary of parameter estimates and test statistics calculated for each stage.

Parameter	posterior Mean	Lower 95% Confidence interval	Upper 95% Confidence interval	<i>p</i> MCMC
Simple Discrimination				
Intercept	0.424	0.055	0.813	0.0228
Trial	<b>0.373</b>	<b>0.068</b>	<b>0.704</b>	<b>0.0183</b>
Simple Discrimination Reversal				
Intercept	1.383	-0.562	4.363	0.1225
Trial	1.286	-0.754	4.179	0.1743
Sex	<b>1.704</b>	<b>0.453</b>	<b>3.015</b>	<b>0.0083</b>
Compound Discrimination				
Intercept	1.147	0.069	2.368	0.0151
Trial	<b>1.209</b>	<b>0.091</b>	<b>2.538</b>	<b>0.0149</b>
Compound Discrimination Reversal				
Intercept	1.023	-0.136	2.521	0.0455
Trial	0.781	-0.288	2.094	0.1058
Intra-dimensional Discrimination				
Intercept	0.915	0.083	1.896	0.0200
Trial	0.589	-0.114	1.389	0.0735
Intra-dimensional Discrimination Reversal				
Intercept	1.196	-0.199	2.904	0.0585
Trial	0.996	-0.418	2.733	0.1242
Extra-dimensional Shift				
Intercept	1.241	-0.175	2.971	0.0539
Trial	0.757	-0.625	2.306	0.2251
Extra-dimensional Shift Reversal				
Intercept	1.363	0.023	2.872	0.0320
Trial	1.107	-0.267	2.463	0.0953
Global model				
Intercept	0.170	-0.034	0.380	0.1000
Sex	0.223	-0.082	0.532	0.1440

620 Learning performance (probability of correct choices) was analysed separately for each of  
621 the eight stages of the experiment including a global model based on data of all stages to  
622 investigate the effect of sex on performance. Sample sizes decreased with stage due to  
623 animals being removed as non-learners, they are as follows:  $SD_N = 24$ ,  $SDR_N = 16$ ,  $CD_N =$   
624  $12$ ,  $CDR_N = 12$ ,  $ID_N = 12$ ,  $IDR_N = 12$ ,  $ED_N = 10$ ,  $EDR_N = 9$ . Significant parameters are  
625 indicated in bold. *p*MCMC – significance of parameter based on Bayesian modelling.

626

627 **Table 3.** Summary of parameter estimates and test statistics to investigate reversal and shift  
 628 performance.

Parameter	Posterior Mean	Lower 95% Confidence interval	Upper 95% Confidence interval	<i>p</i> MCMC
Shift performance				
Intercept	<b>0.663</b>	<b>0.116</b>	<b>1.261</b>	<b>0.0181</b>
Stage	0.009	-0.480	0.505	0.9692
Trial	0.391	-0.110	0.885	0.1027
Interaction Stage & Trial	-0.145	-0.640	0.348	0.5633
Reversal learning in Compound stages				
Intercept	<b>0.690</b>	<b>-0.005</b>	<b>1.391</b>	<b>0.0315</b>
Stage	-0.133	-0.579	0.311	0.5667
Trial	0.581	-0.046	1.259	0.0512
Interaction Stage & Trial	-0.144	-0.639	0.353	0.5680
Reversal learning in Intra-dimensional stages				
Intercept	<b>0.886</b>	<b>0.195</b>	<b>1.671</b>	<b>0.0105</b>
Stage	-0.148	-0.575	0.278	0.4952
Trial	<b>0.626</b>	<b>0.014</b>	<b>1.289</b>	<b>0.0285</b>
Interaction Stage & Trial	-0.115	-0.556	0.343	0.6126
Reversal learning in Extra-dimensional stages				
Intercept	<b>0.958</b>	<b>0.125</b>	<b>1.860</b>	<b>0.0226</b>
Stage	-0.108	-0.525	0.317	0.6160
Trial	0.545	-0.221	1.378	0.1452
Interaction Stage & Trial	-0.126	-0.539	0.287	0.5493

629 Shown is the difference of the probability of a correct choice between the intra-dimensional  
 630 and extra-dimensional shift stage (shift performance) as well as acquisition and reversal  
 631 stages. Significant parameters are indicated in bold. *p*MCMC – significance of parameter  
 632 based on Bayesian modelling.

633

634 **Table A1.** Summary of measurements and stimulus group composition.

PIT	Sex	SVL (mm)	TL (mm)	Mass (g)	Learnt	Stimulus group
1469228	F	110	189	24.5	No	Shape 1
1469674	M	108	210	29.8	No	Shape 1
1469711	F	105	210	29.6	Yes	Shape 1
1469738	M	103	221	25.8	No	Shape 1
1469743	F	105	179	24.1	Yes	Shape 1
1469675	M	105	215	27.4	No	Shape 1
1469662	F	106	201	25.6	No	Shape 2
3366149	M	108	214	28.1	No	Shape 2
1469657	F	111	209	26.3	No	Shape 2
1469715	M	108	204	31.4	Yes	Shape 2
1469722	F	103	154	20.5	No	Shape 2
1469708	M	101	219	23.7	Yes	Shape 2
1469735	F	105	170	25.2	No	Colour 1
1469677	M	103	185	27.6	Yes	Colour 1
1468492	F	109	199	24.7	Yes	Colour 1
1469685	M	103	206	26.5	Yes	Colour 1
1469744	F	106	198	24.3	Yes	Colour 1
1469705	M	106	187	27.5	No	Colour 1
1469667	F	110	186	24.9	No	Colour 2
1469719	M	105	165	23.2	No	Colour 2
1469713	F	100	193	23.2	No	Colour 2
1469742	M	101	160	23.2	No	Colour 2
1469655	F	109	203	26.4	No	Colour 2
1469709	M	116	190	28.0	Yes	Colour 2

635 Measurements from time of capture as well as if the lizard finished the eight stages of the

636 task (learnt) and which stimulus group each animal belonged to. PIT – animal ID, SVL –

637 Snout Vent Length, TL – Total length.

638

639 **Table A2.** Perseverative errors.

PIT	Sex	Stimulus group	Perseverative trials (out of 10)	Stimulus	Binomial significance level
1469711	F	Shape 1	6/4	Star/H	0.7539
1469743	F	Shape 1	4/6	Star/H	0.7539
1469715	M	Shape 2	5/5	Star/H	1
1469708	M	Shape 2	4/6	Star/H	0.7539
1469677	M	Colour 1	5/3	DP/LP	0.7266
1469492	F	Colour 1	6/4	DP/LP	0.7539
1469685	M	Colour 1	3/7	DP/LP	0.3438
1469744	F	Colour 1	5/5	DP/LP	1
1469709	M	Colour 2	5/5	DP/LP	1

640 Number of perseverative errors during the first 10 trials (677 received only eight trials) of the  
641 extra-dimensional shift stage of the nine individuals reaching criterion during this stage. The  
642 table includes PIT (animal identification), sex of subjects, stimulus group subjects were in,  
643 number of errors to the previously reinforced dimension, stimulus perseverative errors were  
644 focused on and significance based on a two-tailed binomial test. DP – dark pink, LP – light  
645 pink.

646

647 **Table A3.** Spray paints used to create the colour dimension on the cue cards.

Stage	Colour	Brand	Colour name
SD/SDR/CD/CDR colour group	Light blue	White Knight, Squirts	Gloss Sky Blue
	Dark blue	White Knight, Squirts	Gloss Bermuda Blue
SD shape group	Beige	Fiddly Bits	Ivory (discontinued)
	Light green	British Paints, Spray Easy	Lime Green
CD/CDR shape group	Dark green	White Knight, Squirts	Gloss Bright Green
	Light orange	White Knight, Squirts	Gloss Golden Yellow
ID/IDR	Dark orange	White Knight, Squirts	Gloss Orange X15
	Light pink	White Knight, Squirts	Gloss Pink
ED/EDR	Dark pink	White Knight, Squirts	Gloss Fuchsia

648 List includes the stage cards were used in, the colour, the name of the paint brand and  
 649 name of the colour as per manufacturer. SD – simple discrimination, SDR – simple  
 650 discrimination reversal, CD – compound discrimination, CDR – compound discrimination  
 651 reversal, ID – intra-dimensional acquisition, IDR – intra-dimensional reversal, ED – extra-  
 652 dimensional shift, EDR – extra-dimensional reversal.

653

654 **Table A4.** Order of stepwise model simplification.

Order	Model	DIC	Reason for removal of terms
1	Correct ~ Sex + Stimulus group + Sex : Stage + Sex : Stimulus group + Stimulus group : Stage	4750.95	Important interactions Stimulus group:Stage in CD, ID and ED are not significant (interaction dropped)
2	Correct ~ Sex + Stimulus group + Sex : Stage + Sex : Stimulus group	4736.71	Sex:Stimulus group interaction was only partly estimable due to insufficient data (interaction dropped)
3	Correct ~ Sex + Stimulus group + Sex : Stage	4736.92	Only one interaction significant (Sex:Stage – SDR), added to stage analysis (interaction dropped)
4	Correct ~ Sex + Stimulus group	4736.35	Neither Sex nor stimulus group are significant
5	Correct ~ Sex	4735.07	Best fitting model based on DIC
6	Correct ~ Stimulus group	4736.81	Model 5 explains the data better

655 Models were run to explore the effect of sex, stimulus group, stage and interactions between  
656 the three effects on the probability of choosing correctly during the whole experiment (all  
657 stages). Reasons for removal of terms is given as well as the DIC (deviance information  
658 criterion). The final model was selected based on DIC.

659

660 **Table A5.** Shape area and circumference.

Shape	Stage	Area (cm <sup>2</sup> )	Circumference (cm)
X	Simple & compound discrimination and reversal	14.08	36.80
Triangle	Simple discrimination and reversal	22.01	21.30
Circle	Intra-dimensional acquisition and reversal	13.85	30.47
Square	Intra-dimensional acquisition and reversal	26.01	22.00
H	Extra-dimensional shift and reversal	13.28	35.00
Star	Extra-dimensional shift and reversal	17.43	28.00

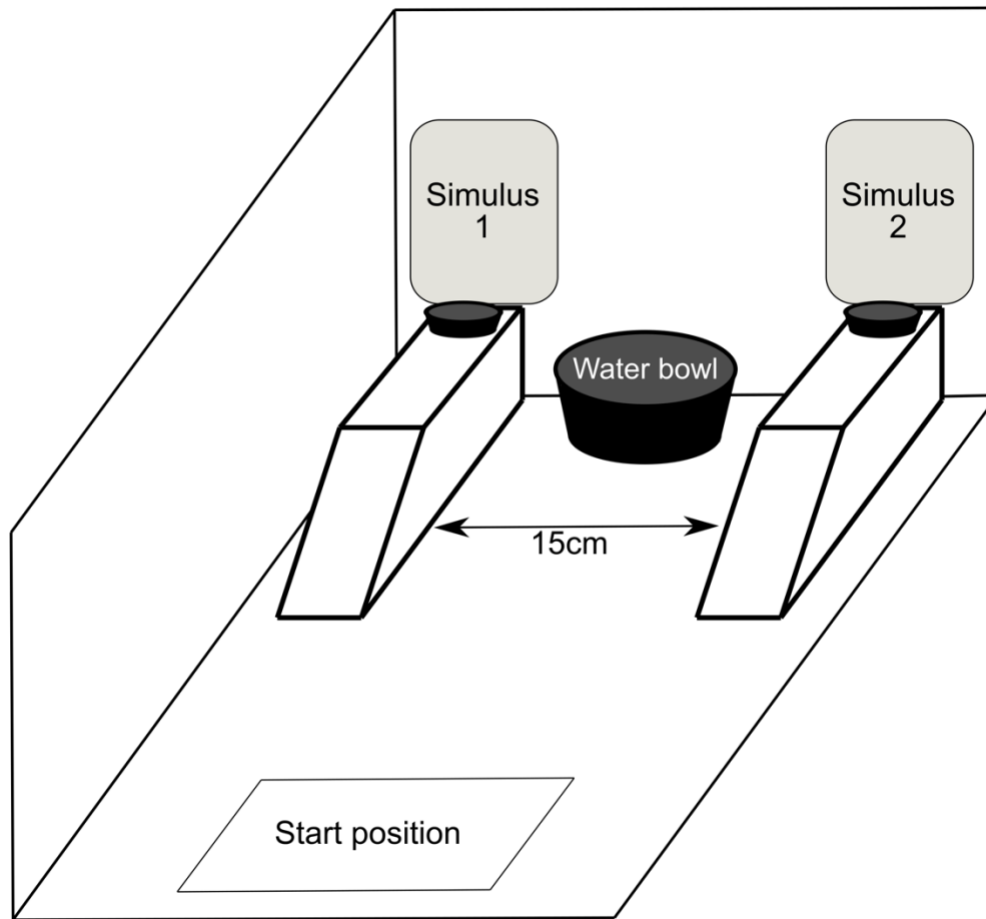
661 Amount of black area and circumference of the different shapes used during the set-shifting

662 experiment. Shapes are given in writing and as symbols, as well as what stages they were

663 used in and their area and circumference.

664



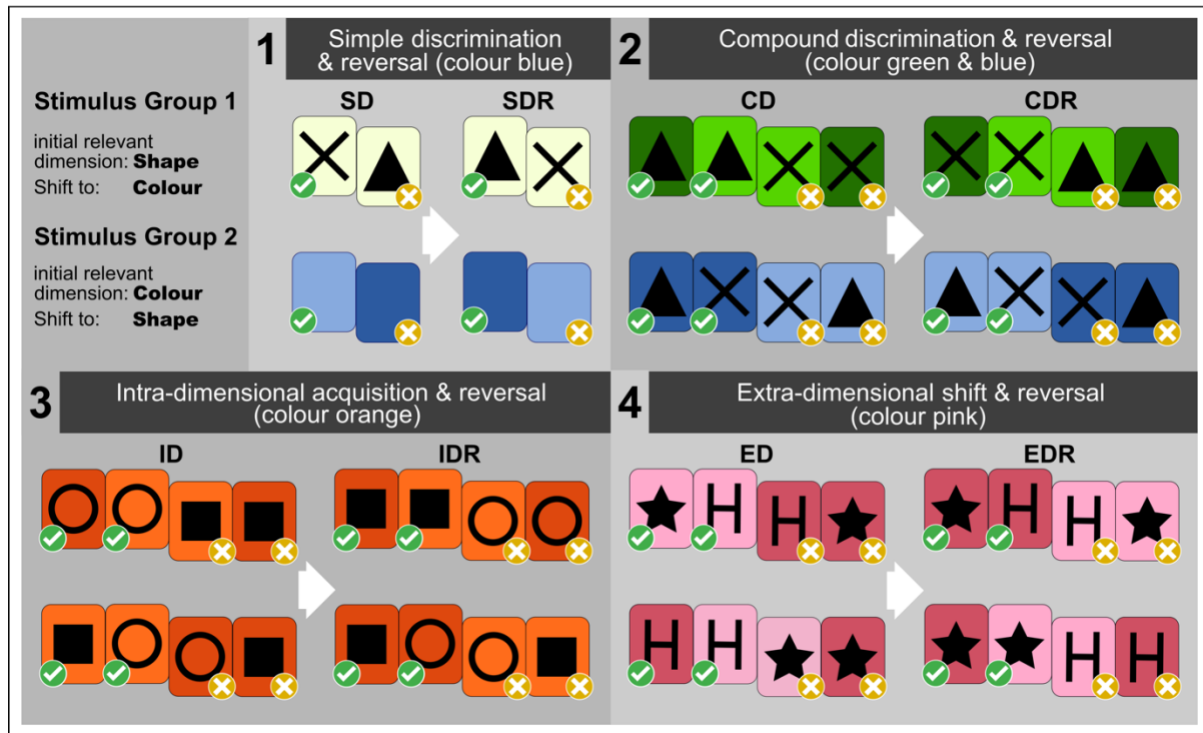


665

666 **Figure 1.** Schematic representation of the setup used during the set-shifting experiment.

667 Two ramps are placed at one end of the tub (approximately 15 cm apart, the water bowl in  
668 between) with the cue cards containing the stimuli attached to the inner wall of the enclosure  
669 at the top end of the ramp and the food dishes containing the reward directly in front of the  
670 cards. The start position indicates the position from where animals started in each trial.

671



672

673 **Figure 2.** Stimulus pairs and order of presentation (1 to 4) for both stimulus group 1 (which

674 started with shape as the positive stimuli, top row) and stimulus group 2 (which started with

675 colour as the positive stimuli, bottom row) during the eight stages of the set-shifting task.

676 Tick marks indicate the rewarded (correct) choice during each stage, whereas Xs indicate

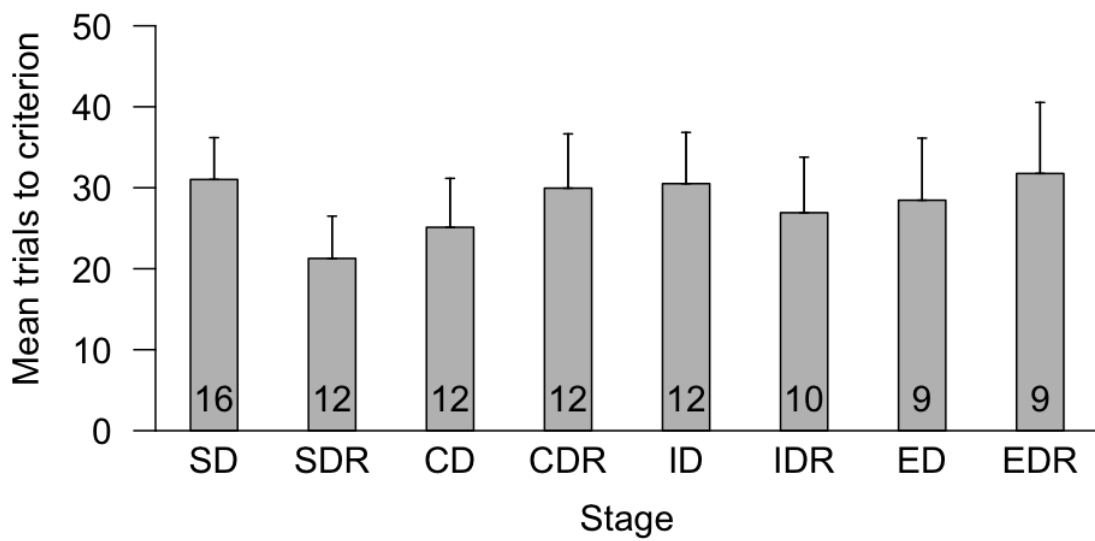
677 that access to the reward was blocked (incorrect choice). SD – simple discrimination, SDR –

678 simple discrimination reversal, CD – compound discrimination, CDR – compound

679 discrimination reversal, ID – intra-dimensional acquisition, IDR – intra-dimensional reversal,

680 ED – extra-dimensional shift, EDR – extra-dimensional reversal.

681



682

683 **Figure 3.** Mean + SE trials to criterion (including criterion trials) for each stage of the  
684 experiment. Sample sizes are given within bars. SD – simple discrimination, SDR – simple  
685 discrimination reversal, CD – compound discrimination, CDR – compound discrimination  
686 reversal, ID – intra-dimensional acquisition, IDR – intra-dimensional reversal, ED – extra-  
687 dimensional shift, EDR – extra-dimensional reversal. Stages were not significantly different  
688 from each other.

689

690 APPENDIX A

691