

1 **Seeing spots: Measuring, quantifying heritability, and**
2 **assessing fitness consequences of coat pattern traits in a**
3 **wild population of giraffes (*Giraffa camelopardalis*)**

4 Derek E. Lee,^{1*} Douglas R. Cavener,² and Monica L. Bond^{1#a}

5

6 ¹Wild Nature Institute, 15 North Main Street, Suite 208, Concord, New Hampshire, United
7 States of America

8 ²Department of Biology and the Huck Institute of Life Sciences, Pennsylvania State
9 University, University Park, Pennsylvania, United States of America

10 ^{#a}Current address: Institute of Evolutionary Biology and Environmental Studies, University
11 of Zürich, Zürich, Switzerland

12

13 * Corresponding author

14 E-mail: derek@wildnatureinstitute.org (DEL)

15 **Abstract**

16 Little is known about the heritability and fitness consequences of polymorphic
17 variation in mammalian coat pattern traits in wild populations. Understanding the evolution
18 of coat patterns requires reliably measuring traits, quantifying heritability of the traits, and
19 identifying the fitness consequences of specific phenotypes. Giraffe coat markings are highly
20 variable and it has been hypothesized that variation in coat patterns most likely affects fitness
21 by camouflaging neonates against predators. We quantified spot pattern traits of wild Masai
22 giraffes using image analysis software, determined whether spot pattern traits were heritable,
23 and assessed whether variation in heritable spot pattern traits was related to fitness as
24 measured by juvenile survival. The methods we described comprise a framework for
25 objective quantification of mammalian coat pattern traits based on photographic coat pattern
26 data, and spot trait measurements from individuals could be used as input to a cluster analysis
27 for taxonomic or other group classifications. We demonstrated that characteristics of giraffe
28 coat spot shape and color are heritable. We did not find evidence for fitness consequences of
29 variation in spot traits on juvenile survival, suggesting that spot traits are currently not under
30 strong directional or stabilizing selection for neonate camouflage in our study population.
31 This may be due to either reduced predation pressure in the study area, or because spot
32 variation may be more relevant to other components of fitness, such as adult survival or
33 fecundity.

34 **Keywords:** adaptation, coat pattern, heritability, microevolution, natural selection,
35 phenotypic selection, quantitative genetics

36

37 **Introduction**

38 Coats of various colors and patterns are found on many mammal species and these
39 phenotypic traits are hypothesized to play adaptive roles in predator and parasite evasion,
40 thermoregulation, and social communication (Cott 1940; Searle 1968; Waage 1981; Skinner
41 and Smithers 1990; Ortolani and Caro 1996). Pigmentation biology has played a prominent
42 role in the foundation of genetics and evolutionary biology with most work on mammals
43 focused on a few starkly different mouse color morphs (Hoekstra 2006). Studies of the
44 heritability and adaptation of complex animal skin patterns have largely concentrated on
45 fishes with their multiple chromatophores (Kalesh 2004). Researchers have proposed
46 mathematical models for pattern-formation mechanisms in animal markings (e.g. Murray
47 1981; Maini 1997, 2004; Garvie and Trenchea 2014), and hypothesized the genetic and
48 developmental mechanisms for the markings (Mills and Patterson 2009; Eizirik et al. 2010),
49 and although studies of heritability and adaptation in wild populations are becoming more
50 prevalent (Kruuk et al. 2008), we are aware of none that investigated the heritability and
51 fitness consequences of complex coat patterns in wild mammalian populations.

52 Understanding the evolution of a trait in a wild population requires measuring
53 individual variation in the trait, quantifying heritability as the proportion of observed
54 phenotypic variation of a trait that is passed from parent to offspring, and assessing the fitness
55 consequences of phenotypic variation (Lande and Arnold 1983; Falconer and Mackay 1996).
56 The fraction of variability in a phenotypic trait that is explained by genetic factors is the
57 broad-sense heritability, which can be estimated as the resemblance of the offspring to its
58 parents (Falconer and Mackay 1996; Roff 1997). Fitness of a trait can be assessed in many
59 ways, but because neonatal mortality is generally higher than any other age class (Lee and

60 Strauss 2017), one of the most direct measurements of phenotypic fitness is juvenile survival
61 (Paterson et al. 1998).

62 Giraffe (*Giraffa camelopardalis*) skin pigmentation is uniformly dark grey (Dimond
63 and Montagna 1976), but the spots that make up their coat markings are highly variable in
64 color, roundness, and perimeter tortuousness, and this variation has been used to classify
65 subspecies (Lydekker 1904), and to reliably identify individuals (Foster 1966; Bolger et al.
66 2012). The variation in coat patterns and colors is consistent with the idea that these markings
67 are polymorphic, and if heritable they may be adaptive (Lydekker 1904; Mitchell and Skinner
68 2003). Dagg (1968) first presented evidence from a small zoo population that the shape,
69 number, area, and color of spots in giraffe coat patterns may be heritable, but analysis of spot
70 traits in wild giraffes, and tools for objectively measuring spot characteristics have been
71 lacking. It has been hypothesized that giraffe coat patterns evolved to camouflage neonates
72 whose primary defense against predation is concealment (Langman 1977; Mitchell and
73 Skinner 2003); thus the most likely fitness effects from variation in coat patterns should be
74 variation in juvenile survival. Alternative hypotheses about the adaptive value of giraffe coat
75 markings include thermoregulation (Skinner and Smithers 1990), and facilitation of
76 individual recognition (*sensu* Tibbetts and Dale 2007) and kin recognition (*sensu* Beecher
77 1982; Tang-Martinez 2001) in this social species with good visual sensory perception (Dagg
78 2014; VanderWaal et al. 2014).

79 The spot patterns of Masai giraffes (*G. c. tippelskirchii*) are particularly diverse
80 among giraffe populations, and the patterns of some Masai giraffes bear strong similarities to
81 other giraffe subspecies such as South African (*G. c. giraffa*), Rothschild's (*G. c. rothschildi*),
82 and reticulated (*G. c. reticulata*) giraffes (Dagg 1968, **Fig 1**). Indeed, some Masai giraffes
83 have spots that are almost indistinguishable from those of reticulated giraffes, the most
84 distinctively marked subspecies with spots that are nearly round with very smooth edges (low

85 tortuousness). Quantifying heritability and fitness consequences of phenotypic variation in
86 coat pattern traits of giraffes will both inform systematics for this species and contribute to
87 the understanding of the evolution of mammalian coat patterns. Our purpose in this study was
88 to 1) objectively quantify the spot pattern traits of wild Masai giraffes in northern Tanzania
89 using image analysis software, 2) determine whether spot pattern traits were heritable, and 3)
90 determine whether variation in heritable spot pattern traits was related to fitness as measured
91 by juvenile survival.

92

93 **Fig 1. Representative images of spot patterns of mother-calf pairs.** The blue rectangle
94 shows the area analysed using ImageJ to characterize spot pattern traits.

95 **Methods**

96 **Field data collection**

97 This study used data from individually identified, wild, free-ranging Masai giraffes in
98 a 1700 km² sampled area within a 4400 km² region of the Tarangire Ecosystem, northern
99 Tanzania, East Africa. We collected data during systematic road transect sampling for
100 photographic capture-mark-recapture (PCMR). We conducted 26 daytime surveys for giraffe
101 PCMR data between January 2012 and February 2016. We sampled giraffes three times per
102 year around 1 February, 1 June, and 1 October near the end of every precipitation season
103 (short rains, long rains, and dry, respectively) by driving a network of fixed-route transects on
104 single-lane dirt tracks in the study area. We surveyed according to a robust design sampling
105 framework (Pollock 1982; Kendall et al. 1995) with three occasions per year separated by a
106 4-month interval. Each sampling occasion was composed of two sampling events during

107 which we surveyed all road transects in the study area with only a few days interval between
108 events ($4.3 \text{ years} \times 3 \text{ occasions year}^{-1} \times 2 \text{ events occasion}^{-1} = 26 \text{ survey events}$).

109 During PCMR sampling events, the entire study area was surveyed and a sample of
110 individuals were encountered and either “sighted” or “resighted” by slowly approaching and
111 photographing the animal’s right side at a perpendicular angle (Canon 40D and Rebel T2i
112 cameras with Canon Ultrasonic IS 100 – 400 mm lens, Canon U.S.A., Inc., One Canon Park,
113 Melville, New York, 11747, USA). We identified individual giraffes using their unique and
114 unchanging coat patterns (Foster 1966) with the aid of pattern-recognition software Wild-ID
115 (Bolger et al. 2012). We attempted to photograph every giraffe encountered, and recorded sex
116 and age class based on physical characteristics. We categorized giraffes into four age classes:
117 neonate calf (0 – 3 months old), older calf (4 – 11 months old), subadult (1 – 3 years old for
118 females, 1 – 6 years old for males), or adult (> 3 years for females, > 6 years for males) using
119 a suite of physical characteristics (Strauss et al. 2015), and size measured with
120 photogrammetry (Lee et al. 2016a).

121 **Quantification of spot patterns**

122 We analysed spot traits of each animal within the shoulder and rib area by cropping
123 all images to a rectangle that fit horizontally between the anterior edge of the rear leg and the
124 chest, and vertically between the back and where the skin folded beneath the posterior edge
125 of the foreleg (**Fig 1**). We quantified spot characteristics of each animal’s pattern using the
126 Color Histogram and Analyze Particles procedures in Program ImageJ (Schneider et al 2012).
127 For color analysis, we used the entire analysis rectangle and full-color photos. For spot
128 measurements we analysed 8-bit greyscale images that we converted to bicolor (black and
129 white) using the Enhance Contrast and Threshold commands. To account for differences in
130 image resolution and animal size, we set the measurement unit of each image equal to the

131 number of pixels in the height of the analysis rectangle. Therefore all measurements are in
132 giraffe units (GU), where 1 GU = height of the analysis rectangle (**Fig 1**). We analysed
133 particles (spots) of all sizes, but excluded particles cut off by the edge of the analysis
134 rectangle to avoid the influence of incomplete spots. We also excluded spots whose area was
135 $< 0.00001 \text{ GU}^2$ to eliminate the influence of speckles.

136 We characterized each animal's spot pattern traits within the analysis rectangle using
137 the following twelve metrics: number of spots; mean spot size (area); mean spot perimeter;
138 mean angle between the primary axis of an ellipse fit over the spot and the x-axis of the
139 image; mean circularity ($4\pi \times [Area] / [Perimeter]^2$ with a value of 1.0 indicating a perfect
140 circle and smaller values indicating an increasingly elongated shape); mean maximum caliper
141 (the longest distance between any two points along the spot boundary, also known as Feret
142 diameter); mean Feret angle (the angle [0 to 180 degrees] of the maximum caliper); mean
143 aspect ratio (of the spot's fitted ellipse); mean roundness ($4 \times [Area]\pi \times [Major\ axis]^2$ or the
144 inverse of *aspect ratio*); mean solidity ($[Area] / [Convex\ area]$, also called tortuousness);
145 mean shade ($[(65536 \times r) + (256 \times g) + b]$ using RGB values from color histogram); and
146 mode shade.

147 **Heritability of spot traits**

148 Parent-offspring (PO) regression is one of the traditional quantitative genetics tools
149 used to measure heritability (Falconer and Mackay 1996). PO regression compares
150 phenotypic trait values in parents to those same trait values in their offspring, with the slope
151 of the linear regression line between the mean parent phenotype and the mean offspring
152 phenotype providing an estimate of the heritability of the trait. We assumed phenotypic
153 correlations provided a sufficiently accurate estimate of genetic correlation (Cheverud 1988;
154 Kruuk et al 2008). Advantages and disadvantages of the parent-offspring method to estimate

155 heritability compared to other methods such as the half-sibling method or the animal model
156 have been elucidated (Åkesson et al. 2008; de Villemereuil et al. 2017), with the primary
157 advantage when studying wild populations being that PO regression requires less information
158 about family structure and only one offspring per individual (Roff 1997; Lynch & Walsh
159 1998). While PO regression can have low statistical power when estimating heritability due
160 to environmental effects shared by related individuals (Wilson et al. 2010), this methodology
161 was the most appropriate for our study design because we were unable to identify fathers, and
162 our sample did not include any maternal siblings or half-siblings.

163 We identified mother-calf pairs by observing extended suckling behavior. Wild
164 female giraffes very rarely suckle a calf that is not their own (Pratt and Anderson 1979). We
165 examined all identification photographs for individuals in known mother-calf pairs, and
166 selected the best-quality photograph for each animal based on focus, clarity, perpendicularity
167 to the camera, and unobstructed view of the torso. For comparing mothers with calves, we
168 selected a photograph of the calf at >9 months of age, because calf fur of younger animals is
169 longer and obscures the spot edges. We found 31 known mother-calf pairs with high-quality
170 photographs of both animals. For comparison of spot characteristics between known mother-
171 calf pairs, we created a set of random cow-calf pairs using the same photographs by assigning
172 a random mother to each calf (without replacement and without pairing a mother with her
173 own calf).

174 We predicted spot pattern traits of a calf would be correlated with those of its mother
175 but not with a random cow. We tested this prediction for each spot characteristic using simple
176 linear regressions of calf values versus mother values, and calf values versus random cow
177 values. We performed statistical operations using the `lm` function in R (R Core Development
178 Team 2013).

179 **Fitness of spot patterns from juvenile survival**

180 We assembled encounter histories for all calves first observed as neonates for survival
181 analysis. For each calf we selected the best-quality youngest age photograph based on focus,
182 clarity, perpendicularity to the camera, and unobstructed view of the torso. For our survival
183 analysis, we used spot traits from the youngest-age photograph available in order to minimize
184 age-related bias in photographs of animals that survived and those that died young.

185 We analysed survival using two methods: logistic regression and capture-mark-recapture. For
186 the first analysis, we determined whether heritable spot traits were related to first season
187 survival with logistic regression between survivors and those that were never sighted again
188 and presumed to have died, using the glm function in R (R Core Development Team 2013).
189 Based on our results from the PO regression method to quantify the heritability of spot traits,
190 we examined linear and quadratic relationships of circularity and solidity (tortuousness) on
191 juvenile survival to determine whether directional or stabilizing selection was occurring.

192 For the second survival analysis we estimated neonate survival during their first
193 season of life as a function of individual spot traits using Program MARK to analyse 258
194 complete capture-mark-recapture encounter histories of giraffes first sighted as neonates
195 (White and Burnham 1999). We analysed our encounter histories using Pollock's Robust
196 Design models to estimate age-specific survival (Pollock 1982; Kendall et al. 1995), with and
197 without spot covariates, and ranked models using AICc following Burnham and Anderson
198 (2002). We determined significance of spot trait covariates if the confidence interval of the
199 beta coefficient did not include zero, and/or if the covariate model was ranked higher than the
200 null model without any covariate.

201 **Results**

202 We were able to quantify twelve spot traits using ImageJ, and found the traits with
203 greatest individual heterogeneity as measured by the coefficient of variation (CV) were the
204 number and area of spots (negatively correlated traits), and mode shade (**Table 1**). Traits with
205 the least individual variation were solidity and roundness (**Table 1**).

206 We found no spot pattern traits that had significant PO regression coefficients
207 between calves and random cows, but two characters, circularity and solidity (tortuousness)
208 (**Fig 2**) were significantly correlated between calves and their mothers indicating heritability
209 (**Table 1**). The color characteristic of mode shade was nearly significant in the PO regression
210 (**Table 1**).

211

212 **Fig 2. Representative spot outlines from Masai giraffes in northern Tanzania and their**
213 **corresponding circularity and solidity values.** Ranges of spot values from 213 calves are
214 given in parentheses.

215

216 Our logistic regression survival analysis of spot traits between 161 neonates that
217 survived their first season and 87 that were never resighted after their first season and
218 presumed dead found no significant effects of traits on survival. Our survival analysis of 258
219 calves first encountered as neonates using Program MARK indicated there was little evidence
220 that individual covariates of spot traits significantly affected survival during the first season
221 of life, but model selection uncertainty was high (**Table 2**). No covariates had significant beta
222 coefficients, and the top-ranked model of survival was the null model with no spot covariates.

223 **Discussion**

224 Our photographic capture-recapture dataset spanning multiple years enabled us to
225 objectively quantify coat pattern traits of wild giraffes using image analysis software, and
226 demonstrate that giraffe coat pattern traits of spot shape and color are heritable from mother
227 to calf. The methods we described should serve as a framework for objective quantification
228 of mammalian coat pattern traits, and could also be useful for taxonomic classifications based
229 on photographic coat pattern data. We did not find strong evidence for fitness consequences
230 of individual variation in heritable spot traits on juvenile survival suggesting that spot pattern
231 traits are currently not under strong directional or stabilizing selection in our study
232 population.

233 One possible explanation for the lack of juvenile survival effects from spot variation
234 is the recent reduction in large predator density in our study area (Packer et al. 2011; Bauer et
235 al 2015). If the function of the coat markings is to provide anti-predation camouflage,
236 reduced predator densities due to trophy hunting and pastoralist retaliatory killings of
237 predators may have alleviated predation pressure on giraffe calves sufficiently to remove the
238 selection pressure for certain spot traits (Lichtenfeld 2005; Lee et al. 2016b). Alternatively,
239 the possibility remains that spot traits may serve adaptive functions such as thermoregulation
240 and/or social communication (Skinner and Smithers 1990; VanderWaal et al. 2014), and thus
241 may demonstrate associations with other fitness traits, such as survivorship in older age
242 classes or fecundity. Individual recognition, kin recognition, and inbreeding avoidance could
243 also play a role in the adaptation of spot patterns for individual recognition in giraffes
244 (Sherman et al. 1997; Tang-Martinez 2001; Tibbetts and Dale 2007).

245 Masai giraffe spot patterns are particularly diverse among giraffe populations, and
246 there are spot patterns in northern Tanzania that bear strong similarities to other giraffe
247 subspecies or species elsewhere in Africa (Dagg 1968, **Fig 1**). Two recent genetic analyses of
248 giraffe taxonomy both placed Masai giraffes as their own species (Brown et al. 2007;

249 Fennessy et al. 2016), but the lack of quantitative tools to objectively analyze coat patterns
250 for taxonomic classification may underlie some of the confusion that currently exists in
251 giraffe systematics (Bercovitch et al. 2017). We expect the application of image analysis to
252 giraffe coat patterns will provide a new, robust dataset to address taxonomic hypotheses. We
253 hope the framework we have described using imageJ to quantify spot characteristics will
254 prove useful to future efforts at quantifying animal markings, and suggest the resultant trait
255 measurements could be useful in a formal cluster analysis to classify subspecies or other
256 groups based on variation in markings (Kaufman and Rousseeuw 2009).

257 Our analyses highlighted two aspects of giraffe spots that were most heritable and
258 which may have adaptive significance. Circularity describes how close the spot is to a perfect
259 circle, and solidity describes how smooth and entire versus tortuous, ruffled, lobed, or incised
260 the edges are. These two characteristics could form the basis for quantifying spot patterns of
261 giraffes across Africa, and gives field workers a new quantitative lexicon for describing spots
262 (**Fig 2**). It is interesting to consider that the roundness and smooth versus rough edge traits we
263 found heritable in giraffe spots brings to mind the smooth and wrinkled peas of Mendel
264 (Bateson and Mendel 1913) and the rough/smooth bacterial colonies in Griffith's (1928)
265 discovery of the "transforming principle" which led to the discovery that DNA was the
266 heritable material (Avery et al. 1944). Our mode shade measurement was a crude metric, but
267 even this rough approximation of spot color showed evidence of heritability. Color is greatly
268 affected by lighting conditions, and we suggest standardization of photographic methods to
269 control for lighting if color is to be analyzed in future studies.

270 Mammalian patterned coats are hypothesized to be formed by two distinct processes:
271 a spatially oriented developmental mechanism that creates a species-specific pattern of skin
272 cell differentiation and a pigmentation-oriented mechanism that uses information from the
273 pre-established spatial pattern to regulate the synthesis of melanin (Eizirik et al. 2010). The

274 giraffe skin has more extensive pigmentation and wider distribution of melanocytes than most
275 other animals (Dimond and Montagna 1976). Future work on the genetics of coat patterns
276 will hopefully shed light upon the mechanisms of coat pattern variation.

277 Our study revealed that several spot traits are heritable and, hence, able to respond to
278 selection in giraffe populations. However, given that these spot characteristic traits did not
279 significantly affect neonatal survival, the main theory for the adaptive nature of giraffe spots
280 (Langman 1977; Mitchell and Skinner 2003), we conclude that spot patterns are currently of
281 minor importance for the adaptation of our study population, although this may be a recent
282 consequence of fewer predators. Other aspects of spot variation may prove to be more
283 relevant to fitness, such as social effects of individual recognition or kin recognition, or
284 thermoregulation, and deserve further investigation.

285 **Acknowledgements**

286 This research was carried out with permission from the Tanzania Commission for Science
287 and Technology (COSTECH), Tanzania National Parks (TANAPA), the Tanzania Wildlife
288 Research Institute (TAWIRI), African Wildlife Foundation, and Manyara Ranch
289 Conservancy. Financial support for this work was provided by Sacramento Zoo, Columbus
290 Zoo, Tulsa Zoo, and Cincinnati Zoo.

291 **References**

292 Åkesson, M., Bensch, S., Hasselquist, D., Tarka, M., and Hansson, B. 2008. Estimating
293 heritabilities and genetic correlations: comparing the ‘animal model’ with parent-
294 offspring regression using data from a natural population. *PLoS ONE* 3(3):e1739.
295 doi:10.1371/journal.pone.0001739

- 296 Avery, O.T., MacLeod, C.M., and McCarty, M. 1944. Studies on the chemical nature of the
297 substance inducing transformation of pneumococcal types: induction of
298 transformation by a desoxyribonucleic acid fraction isolated from pneumococcus type
299 III. *Journal of Experimental Medicine* 79:137-158.
- 300 Beecher, M.D. 1982. Signature systems and kin recognition. *American Zoologist* 22:477-490.
- 301 Bateson, W. and Mendel, G. 1913. *Mendel's principles of heredity*. University Press,
302 Cambridge.
- 303 Bauer, H., Chapron, G., Nowell, K., Henschel, P., Funston, P., Hunter, L.T., Macdonald,
304 D.W., and Packer, C. 2015. Lion (*Panthera leo*) populations are declining rapidly
305 across Africa, except in intensively managed areas. *Proceedings of the National*
306 *Academy of Sciences* 112:14894-14899.
- 307 Bercovitch, F.B., Berry, F.S.M., Dagg, A., Deacon, F., Doherty, J.B., Lee, D., Mineur, F.,
308 Muller, Z., Ogden, R., Seymour, R., *et al.* (2017). How many species of giraffe are
309 there? *Current Biology* 27:R136–R137.
- 310 Bolger, D.T., Morrison, T.A., Vance, B., Lee, D. and Farid, H. 2012. A computer-assisted
311 system for photographic mark-recapture analysis. *Methods in Ecology and Evolution*
312 3:813-822.
- 313 Brown, D.M., Brenneman, R.A., Koepfli, K.-P., Pollinger, J.P., Mila, B., Georgiadis, N.J.,
314 Louis, E.E., Jr., Grether, G.F., Jacobs, J.K., and Wayne, R.K. 2007. Extensive
315 population genetic structure in the giraffe. *BMC Biology* 5:57.
- 316 Burnham, K.P., and Anderson, D.R. 2002. *Model selection and multimodel inference: a*
317 *practical information-theoretical approach*. Springer-Verlag, New York.
- 318 Cott, H.B. 1940. *Adaptive coloration in animals*. Methuen Publishing, London.

- 319 Cheverud, J.M. 1988. A comparison of genetic and phenotypic correlations. *Evolution*
320 42:958–68.
- 321 Dagg, A.I. 1968. External features of giraffe. *Mammalia* 32: 657-669.
- 322 Dagg, A.I. 2014. *Giraffe biology, behavior and conservation*. Cambridge University Press,
323 New York.
- 324 Dimond, R.L. and Montagna, W. 1976. The skin of the giraffe. *The Anatomical Record*,
325 185:63-75.
- 326 Eizirik, E., David, V.A., Buckley-Beason, V., Roelke, M.E., Schäffer, A.A., Hannah, S.S.,
327 Narfström, K., O’Brien, S.J., and Menotti-Raymond, M. 2010. Defining and mapping
328 mammalian coat pattern genes: multiple genomic regions implicated in domestic cat
329 stripes and spots. *Genetics* 184:267-275.
- 330 Falconer, D.S., and Mackay, T.F.C. 1996. *Introduction to quantitative genetics*. 4th edition.
331 Pearson/Prentice Hall, New York.
- 332 Foster, J.B. 1966. The giraffe of Nairobi National Park: home range, sex ratios, the herd, and
333 food. *African Journal of Ecology* 4:139-148.
- 334 Fennessy, J., Bidon, T., Reuss, F., Kumar, V., Elkan, P., Nilsson, M.A., Vamberger, M.,
335 Fritz, U., and Janke, A. 2016. Multi-locus analyses reveal four giraffe species instead
336 of one. *Current Biology* 26:2543–2549.
- 337 Garvie, M.R. and Trenchea, C. 2014. A three level finite element approximation of a pattern
338 formation model in developmental biology. *Numerische Mathematik* 127:397-422.
- 339 Griffith, F. 1928. The significance of pneumococcal types. *Epidemiology & Infection* 27:113-
340 159.

- 341 Hoekstra, H.E. 2006. Genetics, development and evolution of adaptive pigmentation in
342 vertebrates. *Heredity* 97:222-234.
- 343 Kaufman, L. and Rousseeuw, P.J. 2009. *Finding groups in data: an introduction to cluster*
344 *analysis*. John Wiley & Sons.
- 345 Kelsh, R.N. 2004. Genetics and evolution of pigment patterns in fish. *Pigment Cell &*
346 *Melanoma Research* 17:326-336.
- 347 Kendall, W.L., Pollock, K.H., and Brownie, C. 1995. A likelihood based approach to
348 capture–recapture estimation of demographic parameters under the robust design.
349 *Biometrics* 51:293–308.
- 350 Kruuk, L.E., Slate, J., and Wilson, A.J. 2008. New answers for old questions: the
351 evolutionary quantitative genetics of wild animal populations. *Annual Review of*
352 *Ecology, Evolution, and Systematics* 39:525-548.
- 353 Lande, R. and Arnold, S.J. 1983. The measurement of selection on correlated characters.
354 *Evolution* 37:1210–1226.
- 355 Langman, V.A. 1977. Cow-calf Relationships in Giraffe (*Giraffa camelopardalis giraffa*).
356 *Ethology* 43:264-286.
- 357 Lee, D.E., Bond, M.L., Kissui, B.M., Kiwango, Y.A., and Bolger, D.T., 2016a. Spatial
358 variation in giraffe demography: a test of 2 paradigms. *Journal of Mammalogy*
359 97:1015-1025.
- 360 Lee, D.E., Kissui, B.M., Kiwango, Y.A., and Bond, M.L. 2016b. Migratory herds of
361 wildebeests and zebras indirectly affect calf survival of giraffes. *Ecology and*
362 *Evolution* 6:8402–8411. doi: [10.1002/ece3.2561](https://doi.org/10.1002/ece3.2561)

- 363 Lee, D.E. and Strauss, M.K.L., 2016. *Giraffe demography and population ecology*. Reference
364 module in earth systems and environmental sciences. Elsevier, Amsterdam.
- 365 Lichtenfeld, L.L. 2005. *Our shared kingdom at risk: Human–lion relationships in the 21st*
366 *century*. PhD Dissertation, Yale University, New Haven, CT.
- 367 Lydekker, R. 1904, June. On the Subspecies of *Giraffa camelopardalis*. In *Proceedings of the*
368 *Zoological Society of London* (Vol. 74, No. 1, pp. 202-229). Blackwell Publishing
369 Ltd.
- 370 Lynch, M. and Walsh, B., 1998. *Genetics and analysis of quantitative traits* (Vol. 1, pp. 535-
371 557). Sunderland, MA: Sinauer.
- 372 Maini, P.K. 1997. Bones, feathers, teeth and coat markings: a unified model. *Science*
373 *Progress* 80:217-229.
- 374 Maini, P.K. 2004. Using mathematical models to help understand biological pattern
375 formation. *C.R. Biologies* 327:225-234.
- 376 Mills, M.G. and Patterson, L.B. 2009. Not just black and white: pigment pattern development
377 and evolution in vertebrates. In: *Seminars in cell & developmental biology* (Vol. 20,
378 No. 1, pp. 72-81). Academic Press.
- 379 Mitchell, G. and Skinner, J.D. 2003. On the origin, evolution and phylogeny of giraffes
380 *Giraffa camelopardalis*. *Transactions of the Royal Society of South Africa* 58:51-73.
- 381 Murray, J.D. 1981. On pattern formation mechanisms for lepidopteran wing patterns and
382 mammalian coat markings. *Philosophical Transactions of the Royal Society of*
383 *London Series B*. 295:473-496.

- 384 Ortolani, A. and Caro, T.M. 1996. The adaptive significance of color patterns in carnivores:
385 phylogenetic tests of classic hypotheses. *Carnivore behavior, ecology, and evolution*
386 2:132-188.
- 387 Packer, C., Brink, H., Kissui, B. M., Maliti, H., Kushnir, H., and Caro, T. 2011. Effects of
388 trophy hunting on lion and leopard populations in Tanzania. *Conservation Biology* 25:
389 142-153.
- 390 Paterson, S., Wilson, K., and Pemberton, J.M. 1998. Major histocompatibility complex
391 variation associated with juvenile survival and parasite resistance in a large
392 unmanaged ungulate population (*Ovis aries* L.). *Proceedings of the National Academy*
393 *of Sciences* 95:3714-3719.
- 394 Pollock, K.H. 1982. A capture–recapture design robust to unequal probability of capture.
395 *Journal of Wildlife Management* 46:752–757.
- 396 R Core Development Team. 2013. *R: a language and environment for statistical computing*.
397 R Foundation for Statistical Computing, Vienna, Austria.
- 398 Pratt, D.M. and Anderson, V.H. 1979. Giraffe Cow-Calf Relationships and Social
399 Development of the Calf in the Serengeti. *Ethology* 51:233-251.
- 400 Roff, D.A. 1997. *Evolutionary Quantitative Genetics*. Chapman & Hall, New York, USA.
- 401 Searle, A.G. 1968. *Comparative genetics of coat color in mammals*. Logos Press, London.
- 402 Schneider, C.A., Rasband, W.S., and Eliceiri, K.W. 2012. NIH Image to ImageJ: 25 years of
403 image analysis, *Nature Methods* doi: [doi:10.1038/nmeth.2089](https://doi.org/10.1038/nmeth.2089) [C].
- 404 Sherman, P.W., Reeve, H.K., and Pfennig, D.W. 1997. Recognition systems. In: *Behavioural*
405 *ecology: an evolutionary approach* (Ed. by J. R. Krebs & N. B. Davies), pp. 69–96.
406 Oxford: Blackwell Scientific.

- 407 Skinner, J.D. and Smithers, R.H.N. 1990. *The mammals of the Southern African Sub-region*.
408 University of Pretoria, 2nd Edition, pp. 604-607.
- 409 Tang-Martinez, Z. 2001. The mechanisms of kin discrimination and the evolution of kin
410 recognition in vertebrates: a critical re-evaluation. *Behavioural Processes* 53:21-40.
- 411 Tibbetts, E.A. and Dale, J. 2007. Individual recognition: it is good to be different. *Trends in*
412 *Ecology & Evolution* 22:529-537.
- 413 VanderWaal, K.L., Wang, H., McCowan, B., Fushing, H. and Isbell, L.A. 2014. Multilevel
414 social organization and space use in reticulated giraffe (*Giraffa camelopardalis*).
415 *Behavioral Ecology* 25:17-26.
- 416 de Villemereuil, P., Gimenez, O., and Doligez, B. 2013. Comparing parent-offspring
417 regression with frequentist and Bayesian animal models to estimate heritability in
418 wild populations: a simulation study for Gaussian and binary traits. *Methods in*
419 *Ecology and Evolution* 4:260-275.
- 420 Waage, J.K. 1981. How the zebra got its stripes – biting flies as selective agents in the
421 evolution of zebra coloration. *Journal of the Entomological Society of South Africa*
422 44:351-358.
- 423 White, G.C. and Burnham, K.P. 1999. Program MARK: survival estimation from populations
424 of marked animals. *Bird Study* 46(Supplement):120–138.
- 425 Wilson, A.J., Réale, D., Clements, M.N., Morrissey, M.M., Postma, E., Walling, C.A.,
426 Kruuk, L.E.B., and Nussey, D.H. 2010. An ecologist's guide to the animal model.
427 *Journal of Animal Ecology* 79:13-26.
- 428

Table 1. Summary statistics for parent-offspring regressions of spot traits of Masai giraffes in northern Tanzania. Mean trait values, SD (standard deviation), and CV (coefficient of variation), PO slope coefficients (heritability), F-statistics, P values, and r-squared values are provided. Significantly heritable traits are in bold.

	Number	Area	Perimeter	Angle	Circularity	Maximum Caliper	Feret Angle	Aspect Ratio	Roundness	Solidity	Mean Shade	Mode Shade
mean	18.9	0.04	0.99	87.96	0.51	0.29	88.2	1.69	0.63	0.84	7799280	6924050
SD	7.5	0.01	0.25	15.39	0.08	0.06	14.5	0.15	0.04	0.04	1985064	3930565
CV	0.40	0.39	0.25	0.17	0.15	0.19	0.16	0.09	0.06	0.05	0.25	0.57
PO Slope coefficient	0.20	0.20	0.27	0.04	0.52	0.21	-0.15	0.19	0.08	0.53	0.16	0.44
F _{1,29}	0.76	0.87	2.27	0.04	9.97	1.01	0.91	1.11	0.19	9.73	0.55	4.16
P value	0.39	0.36	0.14	0.84	0.004	0.32	0.35	0.30	0.66	0.004	0.47	0.051
r squared	0.03	0.03	0.07	0	0.26	0.03	0.03	0.04	0.010	0.25	0.02	0.13

Table 2. Model selection results for giraffe calf survival as a function of spot trait covariates. No covariate model had a significant beta coefficient, and the top-ranked model was the null model of no covariate effects. Full model in all cases was $\{S(A + \dots) g''(\cdot) g'(\cdot) p(t) c(t)\}$ with covariate structure in survival.

Covariate Model	$\Delta AICc$	W	k
Null	0	0.20	38
Number of spots	0.61	0.15	39
Aspect Ratio	1.22	0.11	39
Roundness	1.53	0.09	39
Solidity	1.73	0.08	39
Max Caliper	1.87	0.08	39
Circularity	2.03	0.07	39
Area	2.18	0.07	39
Feret Angle	2.29	0.06	39
Perimeter	2.96	0.05	39

Pair#

Mother

Calf

1



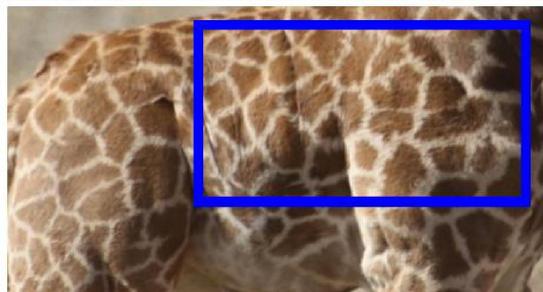
2



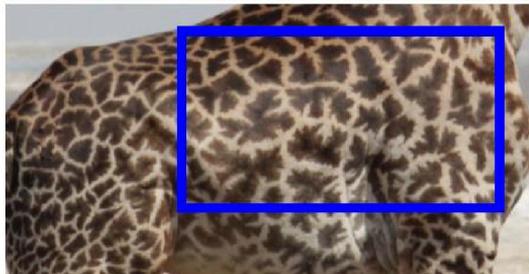
3



4



5



Shape

Circularity
(0.10—0.94)

Solidity
(0.52—0.96)



0.15

0.66



0.13

0.60



0.88

0.94



0.37

0.76



0.40

0.83



0.57

0.82



0.44

0.76



0.12

0.53



0.83

0.96