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Rapid change in mammalian eye shape is explained by activity pattern

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7 **<u>Summary</u>**

8 The rate of morphological evolution along the branches of a phylogeny varies widely

9 [1-6]. Although such rate variation is often assumed to reflect the strength of historical

10 natural selection resulting in adaptation [7-14], this lacks empirical and analytical

11 evidence. One way to demonstrate a relationship between branchwise rates and

12 adaptation would be to show that rapid rates of evolution are linked with ecological

13 shifts or key innovations. Here we test for this link by determining whether activity

14 pattern – the time of day at which species are active – explains rapid bursts of

15 evolutionary change in eye shape. Using modern approaches to identify shifts in the

16 rate of morphological evolution [7, 13], we find that over 74% of rapid eye shape

17 change during mammalian evolutionary history is directly explained by distinct

18 selection pressures acting on nocturnal, cathemeral, and diurnal species. Our results

19 reveal how ecological changes occurring along the branches of a phylogeny can

20 manifest in subsequent changes in the rate of morphological evolution. Although

21 selective pressures exerted by different activity patterns have acted uniformly across

22 all mammals, we find differences in the rate of eye shape evolution among orders. The

23 key to understanding this is in how ecology itself has evolved. We find heterogeneity

24 in how activity pattern has evolved among mammals that ultimately led to differences

25 in the rate of eye shape evolution among species. Our approach represents an exciting

26 new way to pinpoint factors driving adaptation, enabling a clearer understanding of

27 what factors drive the evolution of biological diversity.

31 **Results and Discussion**

32 We test whether rapid shifts in the rate of morphological evolution can be linked to an

33 underlying ecological cause. We used the phylogenetic variable-rates regression

34 model [7] to test for variation in the rate of eye shape evolution across the mammal

35 phylogeny [15] whilst also estimating the relationship between corneal diameter (a

36 proxy for pupil size) and axial eye length (a proxy for focal distance). This relationship

37 has previously been used to summarize eye shape [e.g. 16, 17] (Figure 1). The

38 variable-rates regression model works within a Bayesian Markov chain Monte Carlo

39 (MCMC) framework to estimate a posterior distribution of the rate of evolution along

40 each individual branch of the phylogeny (r) and an underlying global background rate

41 of change (STAR Methods) [7, 13]. We define rate shifts where the posterior

42 distribution of estimated r for a branch exceeds 1 in ≥ 95% of the posterior distribution.

43 In these cases, the branch is evolving at a faster rate compared to the background

44 rate of evolution, and there is significant unexplained residual variance away from the

45 estimated underlying evolutionary relationship.

46 In our bivariate variable-rates regression between corneal diameter and axial length

47 (henceforth *simple eye shape model*), we find a significantly positive slope in the eye

48 shape relationship (judged by the proportion of the posterior distribution crossing zero

49 $[P_x] = 0$, Figure 2a, Table S1) and there is significant rate heterogeneity (Bayes Factor

50 [*BF*] = 520.438 compared to a regression model that estimates only a single

51 background rate, see STAR methods). We identify a total of 128 branches as

52   significant rate shifts (n_{rapid} = 128, Figure 2a) out of a total of 508 branches (n_{total} =

53   508, 25.2% of all branches have had rapid shifts in the rate of eye shape change).

54   These fall predominantly within carnivores (n_{rapid} = 74, modal r range = 5.16-10.20)

55   and anthropoid primates (n_{rapid} = 44, modal r range 3.49-6.95) but also along

56   branches leading to two pangolin species (n_{rapid} = 3, modal r range = 6.37-12.05),

57   the woodchuck (modal r = 6.59), the greater hedgehog tenrec (modal r = 4.04) and

58   three species of *Equus* (n_{rapid} = 5, modal r range = 5.62-6.42). 100% of branches

59   within carnivores and 54% within anthropoid primates are identified as rapid rate shifts.

60   Such rapid shifts in the rate of morphological evolution (Figure 2a) are often used to

61   identify episodes of exceptional change, where the magnitude of the rate shift is

62   implicitly associated with the strength of historical selection pressures [7-14]. However,

63   there is no current statistical evidence for this interpretation of rapid rates. One way to

64   demonstrate that branch-wise rates of morphological evolution reflect selection

65   pressures driving adaptation across millions of years would be to show that branches

66   undergoing rapid rates of morphological evolution are associated with shifts in

67   ecology, key innovations or increased ecological opportunity [14] (Figure 3). Here, we

68   use the phylogenetic variable rates regression framework to test whether activity

69   pattern (the time of day at which species are active) can explain shifts in the rate of

70   evolution in mammalian eye shape.

71   Among vertebrates, there is an established association between activity pattern and

72   eye shape [18-21]. Nocturnal vertebrates tend to maximize light sensitivity with larger

73   pupils [20-22], whereas diurnal species facilitate visual acuity with longer focal

74   distances (i.e. longer eyes relative to pupils) [20, 21, 23, 24]. Cathemeral species show

75   adaptations to unspecialised lifestyles, resulting in some intermediate eye shape [22,

76 25]. We expect activity pattern to be a primary driver of mammalian eye shape

77 evolution as it is in other vertebrates [25, 26], and it should be possible to detect this

78 using rates of evolution. In the variable-rates framework, rapid rates shifts arise as a

79 consquence of significant unexplained residual variance away from the estimated

80 underlying evolutionary relationship. If activity pattern was the primary selection

81 pressure on eye shape in the 128 branches we identify as rapid rate shifts (Figure 2a),

82 then including activity pattern as an additional explanatory factor into the simple eye

83 shape variable-rates regression model would result in all rate shifts disappearing

84 (Figure 3). This would be because activity pattern explains the exceptional deviations

85 away from the underlying eye shape relationship (i.e. the 128 rate shifts). That is,

86 activity pattern would reduce the previously unexplained phylogenetically structured

87 residual variance in eye shape (see STAR Methods and Figure 3).

88 In a variable-rates regression model that allows each activity pattern to have a different

89 slope in the eye shape relationship (*activity pattern model*), we find that the

90 relationship is sharpest in nocturnal mammals (β = 0.904, Figure 2, Table S1). In line

91 with other vertebrates, [18-21, 26], the slope is shallowest is diurnal mammals (β =

92 0.810), and cathemeral species have a moderate slope (β = 0.698, Figure 2, Table

93 S1). This demonstrates a significantly increasing slope in the relationship between

94 corneal diameter and axial length with reducing amounts of daylight activity. That is,

95 nocturnal species increase their relative corneal size more with increasing eye length

96 than diurnal species across the same range of eye lengths (Figure 2B). That is, a

97 large-eyed diurnal species will have relatively clearer vision than a nocturnal species

98 with an eye of the same size – which will instead maximize image brightness.

99 In the activity pattern model, we still find significant rate heterogeneity (*BF* = 521.500),

100 but overall, there is a 74.2% reduction in the number of branches identified as rate

101   shifts (n_{rapid} = 33, see Table S2 for details) compared to the simple eye shape model

102   (Figure 2). Therefore, 95 branches have undergone what we will term *activity pattern*

103   *driven* episodes of rapid eye shape evolution, explained by the different evolutionary

104   slopes in the relationship between corneal diameter and eye length in the activity

105   pattern model (Figure 2).

106   Mammals have large overlap in eye morphology among species of different activity

107   patterns (Figure 1) and are often reported to have eyes similar to other nocturnal

108   vertebrates [16, 17]. This 'nocturnal' eye shape and an associated reduction in

109   morphological diversity among mammals is thought to have arisen (along with other

110   adaptations [16, 27, 28]) during a long period of life in the dark early in mammalian

111   history – a *nocturnal bottleneck.* This prolonged adaptation to nocturnality has led

112   some authors to suggest that changes in activity pattern later in evolution may not

113   have provided sufficient selection pressures to change eye shapes in the expected

114   way [16]. However, we find 95 activity pattern driven episodes of eye shape evolution

115   (Figure 1). Even in the case that incipient mammals underwent an early nocturnal

116   bottleneck, beyond their nocturnal origins there has been more than 160 million years

117   of independent eye shape evolution. The results of our variable rates regressions

118   reveal that during this time, over 74% of all branches with rapid rate shifts in eye shape

119   evolution can be directly explained by activity pattern.

120   Our results are consistent with predictions made by adaptive hypotheses, and provide

121   the first analytical evidence for the previously implicit idea [7-13] that intense and rapid

122   bursts of evolution can be attributed to historical natural selection.

123   Anthropoid primates are often heralded as unique in terms of their eye shape; they

124   have relatively reduced corneal diameters compared to other mammals and thus

125   relatively high visual acuity [e.g. 29, 30] (Figure 1). Notably, the branch leading to the

126 only nocturnal anthropoid primate, *Aotus*, is one of the activity pattern driven episodes

127 of rapid eye shape evolution we find here; owl monkeys rapidly changed their eye

128 shape in order to adapt to their exclusively reverted nocturnal niche. All other

129 anthropoid primates are diurnal. A transition to diurnality in combination with

130 behaviours heavily dependent on vision (such as visual predation) is commonly

131 invoked as an explanation for the origin of the unique anthropoid morphology [31-33].

132 This suggests that both diet and activity pattern might have driven rapid changes in

133 eye shape observed along the branch leading to anthropoid primates. Our variable

134 rates regression model demonstrates that activity pattern, at least, did play a key role

135 in this transition: there is a rapid shift in the rate of eye shape change observed along

136 the branch at the base of anthropoid primates that is completely explained by the eye

137 shape slope estimated for all diurnal mammals (Figure 2). However, although the

138 relatively reduced corneal sizes of anthropoids is associated with a shift to diurnality,

139 this group is not special or unique. With the exception of Papionini (drills, mangabeys,

140 and baboons) and the moustached tamarin (see Table S2, Figure 2), the reduction in

141 corneal diameter observed among anthropoid primates is *expected* given their

142 phylogenetic position and their activity pattern.

143 If activity pattern drives eye shape uniformly across mammals, then why do we

144 observe different patterns in the rate of eye shape change among orders? The key to

145 understanding this may be in how activity pattern itself has evolved. In order to

146 reconstruct the evolutionary history of mammalian activity pattern, we estimated

147 discrete transition rates among activity patterns (defined as the rate of switching

148 between different states along individual branches of a phylogenetic tree) using a

149 Continuous-time Markov transition model [34] allowing all transition rates to vary

150 implemented within a Bayesian framework [35] (henceforth referred to as *transition-*

151 *rates models*). Analyses of transition rates among mammalian activity patterns are

152 scant [cf. 36, 37, 38], and often limited in taxonomic scope [cf. 39]. We therefore

153 expanded our transition-rates models to include all mammals with available activity

154 pattern data (N = 3014, STAR Methods). Across all mammals, our results do not

155 support the recent suggestion that there has been no direct transitions between

156 nocturnal and diurnal lifestyles [39] (Figure 4a). Otherwise, transitions away from

157 cathemeral lifestyles occur more frequently than those towards cathemeral

158 (supporting recent results using a smaller dataset [39]).

159 Estimating a single pattern of transition rates across all mammals in this way is fraught

160 with danger – when we estimate transition rates separately across all large orders of

161 mammals, we find substantial differences in not only the pattern of transitions (Figure

162 4b-d, Figure S1) but also the overall speed of activity pattern change [40] (Figure 4b-

163 d). This highlights that the emergent pattern in transitions across all mammals is likely

164 to be a meta-phenomenon which is difficult to interpret biologically. The previously

165 unappreciated non-uniformity in pattern and speed of activity pattern transitions is

166 interesting. While a formal analysis is beyond the scope of this study, it suggests that

167 the underlying drivers and mechanisms associated with these transitions are variable

168 – potentially associated with the varied environmental and ecological pressures facing

169 species within different mammalian orders.

170 With this in mind, direct transitions between nocturnality and diurnality are rare in

171 several orders (e.g. Lagomorpha and Eulipotyphla, Figure S1). This is in support of

172 the suggestion that transitions between diurnal and nocturnal lifestyles must pass

173 through an "intermediate" cathemeral phase [39]. However, although cathemeral eyes

174 are expected to have an "intermediate" shape between nocturnal and diurnal species

175 [22, 25], there is no particular reason to assume that it is impossible for species to

176 move from day- to night-living or vice versa. Such transitions are supported in both

177 carnivores and rodents (Figure 4). In general, heterogeneity in activity pattern

178 evolution such as that revealed by our transition rates analysis (Figure 3) may

179 ultimately be the underlying driver of heterogeneity in eye shape evolution (Figure 4).

180 Fundamental differences in ecology and how ecology has evolved among taxa has

181 the potential to explain why we observe different rates of continuous morphological

182 change among orders (in our variable rates regression models). Because eye shape

183 and activity pattern are linked (Figure 2), where activity pattern has evolved rapidly –

184 with many transitions between states in a short period of time (e.g. carnivores, Figure

185 3b inset) – it would necessarily result in rapid rates of eye shape evolution (Figure 4).

186 For now, there is a lack of approaches allowing us to characterize and incorporate

187 heterogeneity of transition rates among ecological characters within clades of

188 organisms – or even along individual branches of a phylogenetic tree – into our models

189 of discrete character evolution. Assuming simple directionality away from nocturnality

190 or allowing only a single pattern across all mammals [38, 39] in the face of this

191 heterogeneity (Figures 2, 3) can hinder our ability to infer ancestral forms, and so we

192 do not say anything about nor do we attempt to estimate the ancestral condition of

193 mammals here.

194 Fortunately, difficulties associated with ancestral state reconstruction or confirming

195 whether or not the earliest mammals were nocturnal has absolutely no bearing on the

196 selection pressures faced by different species as they evolved specializations and

197 adaptations beyond those faced by the first mammals millions of years ago.

198 Regardless of whether the ancestral mammal was nocturnal [16, 17, 38, 39] or as

199 some authors have recently suggested, cathemeral [37, 41, 42], as mammals evolved

200 and diversified, natural selection acted to sculpt their morphology in different and
201 important ways.

202 Here, we highlight a new way to determine which factors drive exceptional bursts of
203 phenotypic evolution. Although activity pattern can explain most rapid evolutionary
204 change in eye shape, there are 33 rapid shifts in the rate of mammalian eye shape
205 evolution that remain unexplained (Table S2). In these cases, other factors such as
206 brain size [43, 44], running speed [45], diet [24], or environment [46] must have
207 imposed different and more important selection pressures on eye shape. Fortunately,
208 the approach we describe here provides the potential to test for the influence of those
209 other factors as the data become available.

210 Beyond the mammalian eye, placing rates of continuous morphological change within
211 an explicitly ecological context provides a framework that offers researchers a way to
212 analyse links between ecology and morphology even in the absence of directional
213 change. Taken together, our approach provides the opportunity to obtain a deeper
214 understanding of what factors truly drive the evolution of biological diversity.

215 **Acknowledgements**

220 **Author Contributions**

221 Both authors contributed to all aspects of this work, including writing the paper.

222 **Declaration of Interests**

223 The authors declare no competing interests.

References

225   1.   Rabosky, D.L., Santini, F., Eastman, J., Smith, S.A., Sidlauskas, B., Chang, J.,
226   and Alfaro, M.E. (2013). Rates of speciation and morphological evolution are
227   correlated across the largest vertebrate radiation. Nat. Commun. *4*.

228   2.   Steeman, M.E., Hebsgaard, M.B., Fordyce, R.E., Ho, S.Y., Rabosky, D.L.,
229   Nielsen, R., Rahbek, C., Glenner, H., Sørensen, M.V., and Willerslev, E. (2009).
230   Radiation of extant cetaceans driven by restructuring of the oceans. Syst. Biol.
231   *58*, 573-585.

232   3.   Benson, R.B.J., Campione, N.E., Carrano, M.T., Mannion, P.D., Sullivan, C.,
233   Upchurch, P., and Evans, D.C. (2014). Rates of dinosaur body mass evolution
234   indicate 170 million years of sustained ecological innovation on the avian stem
235   lineage. PLoS Biol. *12*, e1001853.

236   4.   Benson, R.B.J., and Choiniere, J.N. (2013). Rates of dinosaur limb evolution
237   provide evidence for exceptional radiation in Mesozoic birds. Proc. R. Soc.
238   Lond. [Biol.] *280*, e20131780.

239   5.   Puttick, M.N., Thomas, G.H., and Benton, M.J. (2014). High rates of evolution
240   preceded the origin of birds. Evolution *68*, 1497-1510.

241   6.   Rabosky, D.L., and Adams, D.C. (2012). Rates of morphological evolution are
242   correlated with species richness in salamanders. Evolution *66*, 1807-1818.

243   7.   Baker, J., Meade, A., Pagel, M., and Venditti, C. (2016). Positive phenotypic
244   selection inferred from phylogenies. Biol. J. Linn. Soc. *118*, 95-115.

245   8.   Baker, J., Meade, A., Pagel, M., and Venditti, C. (2015). Adaptive evolution
246   toward larger size in mammals. Proc. Natl. Acad. Sci. U.S.A. *112*, 5093-5098.

247   9.   Kratsch, C., and McHardy, A.C. (2014). RidgeRace: Ridge regression for
248   continuous ancestral character estimation on phylogenetic trees. Bioinformatics
249   *30*, i527-i533.

250   10. Kutsukake, N., and Innan, H. (2013). Simulation-based likelihood approach for
251   evolutionary models of phenotypic traits on phylogeny. Evolution *67*, 355-367.

252   11. Kutsukake, N., and Innan, H. (2014). Detecting phenotypic selection by
253   Approximate Bayesian Computation in phylogenetic comparative methods. In

254   Modern Phylogenetic Comparative Methods and Their Application in
255   Evolutionary Biology, L.Z. Garamszegi, ed. (Berlin: Springer-Verlag), pp. 409-
256   424.

257   12.   Rabosky, D.L. (2014). Automatic detection of key innovations, rate shifts, and
258   diversity-dependence on phylogenetic trees. PLoS ONE *9*, e89543.

259   13.   Venditti, C., Meade, A., and Pagel, M. (2011). Multiple routes to mammalian
260   diversity. Nature *479*, 393-396.

261   14.   Duchen, P., Leuenberger, C., Szilágyi, S.M., Harmon, L., Eastman, J.,
262   Schweizer, M., and Wegmann, D. (2017). Inference of evolutionary jumps in
263   large phylogenies using Lévy processes. Syst. Biol. *66*, 950-963.

264   15.   Hedges, S.B., Marin, J., Suleski, M., Paymer, M., and Kumar, S. (2015). Tree
265   of life reveals clock-like speciation and diversification. Mol. Biol. Evol. *32*, 835-
266   845.

267   16.   Heesy, C.P., and Hall, M.I. (2010). The nocturnal bottleneck and the evolution
268   of mammalian vision. Brain Behav. Evol. *75*, 195-203.

269   17.   Hall, M.I., Kamilar, J.M., and Kirk, E.C. (2012). Eye shape and the nocturnal
270   bottleneck of mammals. Proc. R. Soc. Lond. [Biol.] *279*, 4692-4968.

271   18.   Motani, R., and Schmitz, L. (2011). Phylogenetic versus functional signals in
272   the evolution of form–function relationships in terrestrial vision. Evolution *65*,
273   2245-2257.

274   19.   Schmitz, L., and Motani, R. (2010). Morphological differences between the
275   eyeballs of nocturnal and diurnal amniotes revisited from optical perspectives
276   of visual environments. Vision Res. *50*, 936-946.

277   20.   Hall, M., and Ross, C. (2007). Eye shape and activity pattern in birds. J. Zool.
278   *271*, 437-444.

279   21.   Hall, M.I. (2008). Comparative analysis of the size and shape of the lizard eye.
280   Zoology *111*, 62-75.

281   22.   Walls, G.L. (1942). The vertebrate eye and its adaptive radiation, (Bloomfield
282   Hills (MI), Cranbrook Institue of Science: Hafner Publishing Company).

283 23.   Kiltie, R.A. (2000). Scaling of visual acuity with body size in mammals and birds.
284   Funct. Ecol. *14*, 226-234.

285 24.   Veilleux, C.C., and Kirk, E.C. (2014). Visual acuity in mammals: effects of eye
286   size and ecology. Brain Behav. Evol. *83*, 43-53.

287 25.   Kirk, E.C. (2006). Eye morphology in cathemeral lemurids and other mammals.
288   Folia Primatol. *77*, 27-49.

289 26.   Kirk, E.C. (2004). Comparative morphology of the eye in primates. Anat. Rec.
290   A Discov. Mol. Cell. Evol. Biol. *281*, 1095-1103.

291 27.   Lovegrove, B.G. (2017). A phenology of the evolution of endothermy in birds
292   and mammals. Biol. Rev. Camb. Philos. Soc. *92*, 1213-1240.

293 28.   Crompton, A., Taylor, C.R., and Jagger, J.A. (1978). Evolution of homeothermy
294   in mammals. Nature *272*, 333-336.

295 29.   Ross, C.F. (2000). Into the light: the origin of Anthropoidea. Annu. Rev.
296   Anthrop. *29*, 147-194.

297 30.   Ross, C.F., and Kirk, E.C. (2007). Evolution of eye size and shape in primates.
298   J. Hum. Evol. *52*, 294-313.

299 31.   Cartmill, M. (1992). New views on primate origins. Evolutionary anthropology:
300   Issues, news, and reviews *1*, 105-111.

301 32.   Heesy, C.P. (2008). Ecomorphology of orbit orientation and the adaptive
302   significance of binocular vision in primates and other mammals. Brain Behav.
303   Evol. *71*, 54.

304 33.   Williams, B.A., Kay, R.F., and Kirk, E.C. (2010). New perspectives on
305   anthropoid origins. Proc. Natl. Acad. Sci. U.S.A. *107*, 4797-4804.

306 34.   Pagel, M. (1994). Detecting correlated evolution on phylogenies: a general
307   method for the comparative analysis of discrete characters. Proc. R. Soc. Lond.
308   [Biol.] *255*, 37-45.

309 35.   Pagel, M., and Meade, A. (2006). Bayesian analysis of correlated evolution of
310   discrete characters by reversible-jump Markov chain Monte Carlo. Amer. Nat.
311   *167*, 808-825.

312 36. Roll, U., Dayan, T., and Kronfeld-Schor, N. (2006). On the role of phylogeny in
313   determining activity patterns of rodents. Evol. Ecol. *20*, 479-490.

314 37. Gerkema, M.P., Davies, W.I., Foster, R.G., Menaker, M., and Hut, R.A. (2013).
315   The nocturnal bottleneck and the evolution of activity patterns in mammals.
316   Proc. R. Soc. Lond. [Biol.] *280*, 20130508.

317 38. Anderson, S.R., and Wiens, J.J. (2017). Out of the dark: 350 million years of
318   conservatism and evolution in diel activity patterns in vertebrates. Evolution *71*,
319   1944-1959.

320 39. Maor, R., Dayan, T., Ferguson-Gow, H., and Jones, K. (2017). Temporal niche
321   expansion in mammals from a nocturnal ancestor after dinosaur extinction. Nat.
322   Ecol. Evol. *1*, 1889-1895.

323 40. Pagel, M., and Meade, A. (2018). The deep history of the number words. Philos.
324   Trans. R. Soc. Lond. B Biol. Sci. *373*.

325 41. Davies, W.I.L., Collin, S.P., and Hunt, D.M. (2012). Molecular ecology and
326   adaptation of visual photopigments in craniates. Mol. Ecol. *21*, 3121-3158.

327 42. Davies, W.I.L., Tamai, T.K., Zheng, L., Fu, J.K., Rihel, J., Foster, R.G.,
328   Whitmore, D., and Hankins, M.W. (2015). An extended family of novel
329   vertebrate photopigments is widely expressed and displays a diversity of
330   function. Genome Res. *25*, 1666-1679.

331 43. Barton, R.A. (2004). Binocularity and brain evolution in primates. Proc. Natl.
332   Acad. Sci. U.S.A. *101*, 10113-10115.

333 44. Garamszegi, L.Z., Møller, A.P., and Erritzøe, J. (2002). Coevolving avian eye
334   size and brain size in relation to prey capture and nocturnality. Proc. R. Soc.
335   Lond. [Biol.] *269*, 961-967.

336 45. Heard-Booth, A.N., and Kirk, E.C. (2012). The influence of maximum running
337   speed on eye size: a test of Leuckart's Law in mammals. Anat. Rec. *295*, 1053-
338   1062.

339 46. Mass, A.M., and Supin, A.Y.A. (2007). Adaptive features of aquatic mammals'
340   eye. Anat. Rec. *290*, 701-715.

341 47. Pagel, M. (1999). Inferring the historical patterns of biological evolution. Nature
342 *401*, 877-884.

343 48. Ho, L.S.T., and Ané, C. (2014). Intrinsic inference difficulties for trait evolution
344 with Ornstein-Uhlenbeck models. Methods Ecol. Evol. *5*, 1133-1146.

345 49. Xie, W., Lewis, P.O., Fan, Y., Kuo, L., and Chen, M.-H. (2010). Improving
346 marginal likelihood estimation for Bayesian phylogenetic model selection. Syst.
347 Biol. *60*, 150-160.

348 50. Raftery, A.E. (1996). Hypothesis testing and model selection. In Markov Chain
349 Monte Carlo in Practice, W.R. Gilks, S. Richardson and D.J. Spiegelhalter, eds.
350 (London, Great Britain: Chapman & Hall), pp. 163-187.

351 51. Meade, A., and Pagel, M. (2017). BayesTraits.
352 http://www.evolution.rdg.ac.uk/BayesTraitsV3.0.1/BayesTraitsV3.0.1.html.

353 52. Ross, C.F., Hall, M.I., and Heesy, C.P. (2007). Were basal primates nocturnal?
354 Evidence from eye and orbit shape. In Primate origins: adaptations and
355 evolution, M.J. Ravosa and M. Dagosto, eds. (New York: Springer), pp. 233-
356 256.

357 53. Plummer, M., Best, N., Cowles, K., and Vines, K. (2006). CODA: convergence
358 diagnosis and output analysis for MCMC. R news *6*, 7-11.

359 54. Jones, K.E., Bielby, J., Cardillo, M., Fritz, S.A., O'Dell, J., Orme, C.D.L., Safi,
360 K., Sechrest, W., Boakes, E.H., Carbone, C., et al. (2009). PanTHERIA: a
361 species-level database of life history, ecology, and geography of extant and
362 recently extinct mammals. Ecology *90*, 2648.

363 55. Bennie, J.J., Duffy, J.P., Inger, R., and Gaston, K.J. (2014). Biogeography of
364 time partitioning in mammals. Proc. Natl. Acad. Sci. U.S.A. *111*, 13727-13732.

365 56. Organ, C.L., Janes, D.E., Meade, A., and Pagel, M. (2009). Genotypic sex
366 determination enabled adaptive radiations of extinct marine reptiles. Nature
367 *461*, 389-392.

368 57. Shultz, S., Opie, C., and Atkinson, Q.D. (2011). Stepwise evolution of stable
369 sociality in primates. Nature *479*, 219-222.

370   58.   Opie, C., Atkinson, Q.D., Dunbar, R.I.M., and Shultz, S. (2013). Male infanticide

371   leads to social monogamy in primates. Proc. Natl. Acad. Sci. U.S.A. *110*,

372   13328-13332.

373

Figure Titles and Legends

375 **Figure 1: Mammalian eye shape.** A bivariate plot depicting mammal eye shape (n =

376 266) as the relationship between corneal diameter (a proxy for pupil size) and axial

377 eye length (a proxy for focal distance). Colours indicate activity pattern (see legend).

378 Anthropoid primates are shown as squares; all other species are shown as circles.

379 This plot must be interpreted with caution; data points are not independent owing to

380 shared ancestry.

381 **Figure 2: The effect of activity pattern on the rate of eye shape evolution**.

382 Branches of the mammal phylogeny (n = 266) along which there have been rapid rate

383 shifts (*r* >1 in ≥95% of the posterior distribution) in the simple eye shape model (a) and

384 the activity pattern model (b) are stretched to represent their median rate of evolution

385 (i.e. longer branches have faster rates) and are coloured by group. The branch leading

386 to anthropoid primates is marked with an arrow. All other branches are measured in

387 millions of years. The posterior predicted phylogenetic slopes are shown in (a, inset)

388 for the simple eye shape model and in (b, inset bottom) for the activity pattern model

389 – the median predicted slope is highlighted. Pairwise comparisons between the

390 magnitudes of each slope are given in (b, inset top) as the posterior distributions of

391 differences between two estimated β parameters. The nocturnal slope is significantly

392 different to both the cathemeral ($P_{x[diff]} = 0.045$) and the diurnal slopes ($P_{x[diff]} =$

393 0.003). The diurnal slope is the shallowest and is significantly shallower than the

394 cathemeral slope ($P_{x[diff]} = 0.031$). See also Table S1 for parameter values and Table

395 S2 for details on rate shifts that remain unexplained by activity pattern.

396 **Figure 3: A schematic of how we can reveal the underlying causes of rate**

397 **variation.** (a) A phylogeny with branches measured in millions of years. (b) Tests for

398   rate heterogeneity on this phylogeny in combination with eye shape data for species

399   at the terminal branches reveals multiple rate shifts along individual branches in the

400   tree (exceptionally rapid rates of evolutionary change arising from significant

401   unexplained phylogenetic residual variance in the eye shape relationship). These

402   branches are coloured black and are stretched according to their rate of evolution

403   (longer branches = faster rates). All other branches have evolved as expected given

404   their length in time i.e. they are encompassed within the variation explained by the

405   underlying regression relationship in combination with the overall background rate of

406   eye shape change acting across all mammals. We show two potential scenarios with

407   extreme outcomes of including activity pattern into tests for rate variation (yellow =

408   diurnal, green = cathemeral, blue = nocturnal). (c) In the first scenario, natural

409   selection on eye shape has been driven exclusively by activity pattern. All rapid bursts

410   of change in eye shape evolution – all rate shifts – can therefore be explained by the

411   inclusion of activity pattern into the model i.e. no branches remain stretched. (d) In the

412   second scenario, activity pattern is randomly distributed with regards to eye shape and

413   so all rate shifts remain identified as instances of significant and substantial

414   unexplained variation in eye shape (black, stretched branches). That is, activity pattern

415   does not explain any of the unexplained phylogenetic residual variance in eye shape

416   that manifests as rapid rate shifts. Note that here, eye shape variation is represented

417   by pupil size– in reality, it is *relative* pupil size that is important.

418

419   **Figure 4: Transition rates amongst activity patterns in mammals and the three**

420   **largest orders.** The results of our discrete transition analyses across all mammals (n

421   = 3014). In all cases, pairwise transitions between activity patterns are indicated by

422   the directions of the arrows and each transition rate is shown as a density distribution

423   in a corresponding colour. Activity patterns are indicated by the letters and coloured

424   boxes where N (blue) = nocturnal, C (green) = cathemeral/crepuscular and D (yellow)

425   = diurnal. Each arrow is shaded to match the corresponding distribution of estimated

426   transition rates. Results are shown for a model run across (a) all mammals, n = 3014

427   (b) carnivores, n = 236 (c) primates, n = 301 and (d) rodents, n = 1098. Inset for each

428   of the three individual orders is a posterior distribution of the global rate of activity

429   pattern evolution, comparing the overall speed at which transitions between activity

430   patterns have occurred along the branches of the phylogenetic tree during the course

431   of each group's evolution. The global rates are estimated simultaneously with the

432   patterns of pairwise transition rates – see STAR Methods for more details. See also

433   Figure S1 for results from other mammal groups.

STAR Methods

METHOD DETAILS

The variable-rates regression model

We used the variable-rates regression model [7, 13] to simultaneously estimate phylogenetic regression parameters whilst identifying the position and magnitude of rate shifts in the phylogenetically structured residual variance of the eye shape relationship (see below). The variable-rates model partitions the underlying Brownian variance (σ^2) of a continuously varying generalized least squares model of trait evolution [e.g. 47] into two components: (1) a background rate (σ_b^2) and (2) a set of rate scalars r defining branch-specific shifts. Note that this background rate σ_b^2 measures the instantaneous variance of change (i.e. change per unit time) acting along each individual branch of the phylogenetic tree. Together, σ_b^2 and r estimate an optimized variance for each branch ($\sigma_v^2 = \sigma_b^2 r$), and identify where branches have evolved faster ($r > 1$) or slower ($0 \leq r < 1$) than the background rate. A gamma prior ($\alpha = 1.1$, β rescaled to give a median of 1) is placed on each scalar parameter, ensuring an even number of rate increases and rate decreases are proposed. Importantly, contrary to what has previously been reported [48] there is no prior placed on the number of rate parameters, i.e. the reversible-jump procedure flexibly allows for anywhere between 0 and n scalars to be estimated (where n is the number of nodes, including tips, in the phylogeny).

457 The presence of rate heterogeneity can be identified using Bayes factors (BF),

458 calculated as $BF = -2\log_e[m_1/m_0]$, where m_0 and m_1 are the marginal likelihoods of

459 a single-rate Brownian motion regression model and the variable-rates regression

460 model respectively. Marginal likelihoods are estimated using a stepping stone sampler

461 [49], where values are drawn from a beta-distribution (α = 0.4, β = 1) [49]. Where BF

462 > 2 it is regarded positive support for rate variation [50].

463 The variable-rates regression model is implemented within a Bayesian Markov chain

464 Monte Carlo (MCMC) reversible-jump framework and was introduced by Venditti *et al*

465 (2011) [13] and Baker et al 2016 [7]. It is run using BayesTraits V3 (see below for link

466 to software download).

467 *The transition-rates model*

468 We estimated discrete transition rates (the rate of switching between different states

469 along individual branches of a phylogenetic tree) among activity patterns using a

470 Continuous-time Markov transition model implemented within a Bayesian framework

471 [34, 35]. The model seeks to estimate the values of a transition matrix that define the

472 instantaneous rate of switching between each pair of states (i.e. from nocturnal to

473 diurnal, diurnal to nocturnal, etc.). The model we use is implemented in a reversible-

474 jump framework which allows the dimensionality of the estimated transition rate matrix

475 to be reduced where required to avoid over parameterization [35]. This allows two or

476 more rates in the matrix to take the same value (if supported by the data) – or even

477 for all rates to have different values. More details about the Markov transition model

478 and its implementation in the reversible-jump framework can be found in Pagel and

479 Meade (2006) [35].

480   We also implement a recently published variant of the Continuous-time Markov

481   transition model [40] which allows for normalization of the estimated transition rate

482   matrix. That is, the model simultaneously estimates the transition rates among states

483   (as in the standard reversible-jump model [35]) alongside a *global rate* of evolution.

484   The pattern of transition rates is still inferred, but the rate parameters are not directly

485   interpretable. Instead, the global rate describes the overall speed at which transitions

486   between states have occurred along the branches of the phylogenetic tree during the

487   course of a group's evolution. That is, rates can be interpreted as deviations from a

488   generalized rate acting across any set of data [40]. Therefore, estimating a global rate

489   for the evolution of a single character among multiple different groups facilitates

490   comparisons between the overall rates of change of a character regardless of the

491   patterns of transition rates. Details of how the normalization constant is calculated can

492   be found in Pagel & Meade, 2018 [40].

493   We use BayesTraits V3 [51] to run all discrete character transition models (see below).

494   **QUANTIFICATION AND STATISTICAL ANALYSIS**

495   *Modelling the eye shape relationship*

496   Mammal eye shape was described using the previously described relationship

497   between corneal diameter and axial length [17, 26, 52] for $n = 266$ species spanning

498   29 mammalian orders (figure 1). All measurements were taken from Hall *et al,* 2012

499   [17], matched to the recently published time tree of life [15], and $\log_{10}$-transformed.

500   For the 266 species with eye shape data, we obtained activity patterns from the same

501   source [17], where species are defined as nocturnal (typically active at night),

502   cathemeral (active at both day and night), or diurnal (typically active at day). Sample

503   sizes for all models are recorded in the figure captions of the main text; all data and

504   sources can be found in Table S3.

505   Significance of regression parameters was assessed by the proportion of the posterior

506   distribution that crosses zero (P_x). Where $P_x <$ 0.05, that variable can be considered

507   significantly different from zero. To compare parameters amongst different activity

508   patterns, we compared the estimated slopes for each state using pairwise

509   comparisons between the differences of two parameters at each iteration and

510   assessed the proportion of the posterior distribution of differences crossing zero

511   ($P_{x[diff]}$). Where $P_{x[diff]} <$ 0.05, two parameters are considered distinct. For our

512   regression models, we summarize the median parameter values and their variance in

513   Table S1, and visualize parameters and their differences in Figure 2.

514   All MCMC chains were run for a total of 200 million iterations, sampling every 100,000

515   iterations after convergence and were repeated multiple times to ensure convergence.

516   Uniform priors ranging between -10 and 10 were placed on all estimated regression

517   coefficients. We ensured that the effective sample size for all estimated parameters

518   was greater than 750, calculated using R package coda [53].

519   *Identifying rate shifts*

520   We defined significant *rate shifts* where there was significant unexplained residual

521   variance away from an estimated underlying evolutionary relationship (see below for

522   details of what relationships were studied). Where the posterior distribution of

523   estimated r for a branch exceeded 1 in ≥ 95% of the posterior distribution, that branch

524   was defined as a significant rate shift – it is evolving at a significantly faster rate to the

525   background rate (note that rate decreases could also be identified where $r < 1$ in 95%

526   of the posterior). Although significance is identified across the posterior sample, we

527 summarize r for individual branches using modes (calculated using kernel density

528 estimation across the posterior distribution) and for clades comprised of multiple

529 branches, we report the range of branchwise modes of r (modal r range).

Detecting the drivers of rate shifts

531 We first identified rate shifts in eye shape evolution using a bivariate regression

532 between corneal diameter and axial eye length (*simple eye shape model,*). We then

533 compared the subset of branches identified in this model to those identified as

534 significant rate shifts in a model allowing for different slopes and intercepts in the

535 relationship for each of the three activity patterns *(activity pattern model)*. Note that

536 these models estimate both regression parameters and rate scalars simultaneously.

537 Branches identified as rate shifts in the bivariate linear model represent significant

538 unexplained variance in eye shape. If this unexplained variance can be explained by

539 the differential slopes in the eye shape relationship faced by mammals of different

540 activity patterns – i.e. differences in the slope of the relationship between corneal

541 diameter and axial eye length as has previously been reported in birds [20] – we would

542 observe a reduction in the number of identified branches in our activity pattern model

543 (Figure S1). This is because activity pattern will explain the previously exceptional

544 deviations away from the underlying eye shape relationship that manifested as bursts

545 of rapid evolution by reducing the phylogenetically structured residual variance in eye

546 shape; i.e. activity pattern explains the previously unexplained residual variance

547 (Figure S1).

548 In the (unlikely) scenario in which activity pattern has not exerted sufficient selection

549 pressure to change eye shape, then incorporating activity pattern into our tests for

550 selection would result in no reduction in the number of branches identified as having

551 rapid bursts of eye shape change along them (Figure S1). This is because there would

552 be no link between the rate of eye shape change and activity pattern: beyond the

553 underlying regression relationship and the overall background rate of eye shape

554 change across all mammals, activity pattern explains no additional variation. The only

555 way to explain bursts of eye shape change without including additional possible

556 explanatory factors into our model would be to increase the rate of evolution along

557 branches leading to changes in eye shape; we would therefore continue to detect rapid

558 evolutionary change in eye shape (Figure S1).

559 As with any regression framework, it is important to recognize that factors should be

560 tested using a hypotheses-driven approach to avoid variation being explained by

561 chance. Here, we have strong a priori reasons for using activity pattern as an

562 explanatory factor (see Results & Discussion).

563 *Modelling activity pattern evolution*

564 In order to reconstruct the evolution of activity pattern, we estimated discrete transition

565 rates of activity pattern evolution across all mammals (N = 3014, supplementing our

566 original dataset [17] with activity pattern classifications from the literature [54, 55],

567 Table S3). Crepuscular species, those that are active in twilight hours [55] are, on

568 average, presumed to experience similar light levels to cathemeral species and so

569 here we collapse these species into a single category as in previous classifications

570 [54] and in order to match the three-state classification used in our main variable rates

571 regression analyses.

572 To estimate transition rates among activity patterns, we use a Continuous-time Markov

573 transition model allowing all transition rates to vary implemented within a Bayesian

574 framework [35]. To investigate potential different patterns present across the mammal

575   tree of life, we also ran an additional model estimating transition rates separately for

576   all large orders of mammals: carnivores (N = 236), primates (N = 301), rodents (N =

577   1098, cetartiodactyls (N = 209), insectivores (N = 249), and lagomorphs (N = 79). We

578   also analyse marsupials (N = 252) as a single group. Note that although bats are also

579   one of the largest orders (N = 533 with activity pattern data), we do not estimate

580   transition rates separately for this group owing to the fact that they are predominantly

581   nocturnal with very few exceptions (Table S3).

582   We implemented all models in a reversible-jump framework [35], effectively reducing

583   the dimensionality of the estimated transition rate matrix where required to avoid over

584   parameterization. This allows two or more rates in the matrix to take the same value

585   (if supported by the data). We used a hyper-prior approach [35] to reduce inherent

586   uncertainty and biases in prior choice [35, 56]. We placed an exponential distribution

587   as the prior on transition rates (seeding the mean from a uniform distribution ranging

588   between 0 and 2) [56-58]. Alternative prior distributions produce qualitatively identical

589   results. All chains were run for 10 million iterations, sampling every 10,000 iterations

590   after convergence. We repeated the analysis with multiple MCMC chains to ensure

591   convergence.

592   Finally, for the three largest individual mammalian groups we present in the main text,

593   we additionally ran models that normalized the estimated transition rate matrix [40].

594   This estimated a *global rate* of activity pattern evolution, describing the overall speed

595   at which transitions between activity patterns have occurred along the branches of the

596   phylogenetic tree making it possible to determine whether activity patterns were

597   evolving at faster or slower rates in different groups regardless of their overall patterns

598   of change.

599   **DATA AND SOFTWARE AVAILABILITY**

600 The full dataset of eye shape measurements and activity patterns used in our main

601 analysis is already published and available in Hall *et al*, 2012 [17]. In Table S3, we

602 provide this dataset where we have matched taxa names to the recently published

603 time tree of life [15]. For our multi-state activity pattern analysis, we aimed to

604 incorporate all available data for all mammals (N = 3,014). This additional data was

605 obtained from published literature and all sources and data are documented in Table

606 S3.

607 We use BayesTraits V3 [51] to implement the variable-rates regression models [7] and

608 discrete transition rates analyses [35, 40]. The code for this program is open-source

609 and is freely available to download from the following website:

610 http://www.evolution.rdg.ac.uk/BayesTraitsV3.0.1/BayesTraitsV3.0.1.html.

Supplemental Item Titles and Legends

Table S3: Eye shape, activity pattern, and diet data for mammals. **Related to STAR Methods.** All data used in our analyses is recorded here, along with its original published source.