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1 Group size, temperature and body size modulate the effects of social hierarchy on

2 basal cortisol levels in fishes

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- 9 *Corresponding author : <u>bgeffroy@ifremer.fr</u>
- 10
- 11 Abstract
- 12

13 Social rank in a structured society has been linked to basal levels of glucocorticoids in 14 various species, with dominant individuals generally presenting lower levels than 15 subordinates. The biotic and abiotic factors influencing glucocorticoids levels across 16 social ranks are still, however, unclear in fishes. We investigated the influences of group 17 size, fish size, sex, age, and reproduction type, plus water salinity and temperature, on the 18 basal levels of cortisol, the major stress hormone in fishes. A phylogenetically controlled 19 meta-analysis was performed on data from 72 studies over 22 species of fishes. As 20 expected, dominants generally exhibited lower levels of cortisol than subordinates. More 21 importantly, the strength of the correlation between cortisol and rank was modulated by 22 three main factors, group size, environmental temperature, and fish size. Differences in 23 basal cortisol between dominants and subordinates were significantly greater in small 24 groups (dyadic contexts) when compared to larger groups. Differences between 25 dominants and subordinates were also greater in temperate regions when compared to the 26 tropics, and in species with larger body size. These results help to decipher the links among hierarchy, stress and metabolism in fishes. 27

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- 29

30 Keywords: Stress; Aggressiveness; Sociality; Shoal; Meta-Analysis

31

32 **1. Introduction**

33

34 Many animal societies are organized into hierarchies. First described in the domestic 35 chicken (Schjelderup-Ebbe, 1922), a social hierarchy is a system where individuals 36 organize themselves into a particular, often linear, order of status from dominant to 37 subordinate individuals. Hierarchies are frequently established by aggression or by body 38 size and weight (Rubenstein and Abbot, 2017), and communicated visually (Campderrich 39 et al., 2017; Grosenick et al., 2007) and chemically (Bigiani et al., 2005). Socially 40 dominant individuals gain advantages that can increase their fitness, for instance they 41 have access to more food (Hahn and Bauer, 2008) or have lower risks of predation 42 (Teichroeb et al., 2015). Dominants also secure access to territory and partners for 43 reproduction (Holekamp and Strauss, 2020) and have enhanced access to helpers in 44 species with cooperative breeding (Dawson and Mannan, 1991). This apparently ideal 45 situation also, however, comes with some costs, especially because access to this specific 46 rank involves stressful situations, such as fights for dominance. Hence, gaining and 47 retaining status, but also being a subordinate, may all cause stress. Consequently, social 48 status has been found to be tightly linked to stress levels in various animal species, as 49 measured by tissue glucocorticoid concentrations (Abbott et al., 2003; Ejike and Schreck, 50 1980).

51 Fish represent a very interesting group to investigate the extent to which glucocorticoid 52 levels underscore social status, since they exhibit widely differing degrees of social 53 organization (Wilson, 2000). They vary from isolation with rare social contact (e.g. 54 swordfish Xiphias gladius (Palko et al., 1981)), to territorial individuals protecting an area 55 amid conspecifics (Sogawa et al., 2016), to loosely coordinated shoaling (e.g. stickleback 56 Gasterosteus aculeatus (Ginnaw et al., 2020)), to highly organized schooling (e.g. a 57 tropical damselfish Chromis viridis (Nadler et al., 2018)). Although this myriad of social 58 organization systems might be an advantage in studying links between physiology and 59 behavior, the fact that fish species greatly vary in their life history traits might also 60 obscure any underlying patterns. For example, Baker et al. (Baker et al., 2013) reported 61 higher basal glucocorticoid concentrations for dominant individuals in four tests but the 62 opposite in five other tests. The authors identified confounding factors that were fish 63 density, wild versus captive animals, and level of aggressiveness (Baker et al., 2013). 64 Even within a single species, differences in experimental design may impede the detection

of directional patterns. In addition, our understanding of the potential biotic and abioticfactors affecting patterns is limited.

67 Plasma cortisol is an established indicator of stress in fishes (Mommsen et al., 1999; Sadoul and Geffroy, 2019), produced by the Hypothalamus-Pituitary-Interrenal (HPI) 68 69 axis, it plays a key role in energy mobilization to cope with stressful events (Sadoul and 70 Vijayan, 2016) but it may also have multiple other impacts, notably on reproduction 71 (Rousseau et al., 2021) or sex determination (Geffroy and Douhard, 2019). Basal 72 circulating cortisol titer can vary in response to life-history (Ricklefs and Wikelski, 2002), 73 body condition (Breuner and Hahn, 2003) and season (Landys et al., 2006; Love et al., 74 2005). Since fishes are ectotherms, temperature is a major effector of their general 75 metabolism (Stevens and Fry, 2011), including the production of cortisol (Alfonso et al., 76 2020). Intra-specific differences in basal cortisol may be genetically driven (Samaras and 77 Pavlidis, 2018) and generally covary with differences in behavior, metabolism and life-78 history traits, as part of the well described 'coping style' (Koolhaas et al., 1999).

79 Consequently, an increase in cortisol production does not necessarily indicate a 80 physiological "stress" stricto sensu (Schreck and Tort, 2016), but could be the result of a 81 temperature-driven increase in metabolic rates (Alfonso et al., 2020). Intraspecific 82 differences in cortisol levels could also be the result of genetically-driven differences in 83 species overall metabolic rates. Therefore, distinguishing among stressful situations that 84 actually trigger an increase in cortisol level (Harris, 2020) from those that are a 85 consequence of differences in metabolic rates can be challenging. That is, differences in 86 basal cortisol within a social hierarchy may be caused by the stress imposed by a specific 87 social rank but, inversely, social rank might be the consequence of internal specificities 88 of an individual. For ease of description, we refer to the first situation as the "stress-89 enhanced hypothesis" and to the second as the "metabolic-enhanced hypothesis". 90 Providing more information about external and internal effectors that affect relationships 91 between basal cortisol level and social rank could ultimately provide support for these 92 two hypotheses.

Here we test whether group size and life history traits can partially explain contrasting results about how social rank influences plasma cortisol. We tested the effect of external and internal factors, and particularly the effect of climate zone (tropical *vs.* temperate), to assess whether differences in cortisol between social ranks could also be due to the environment. A phylogenetically controlled meta-analysis and meta-regression were conducted to verify these hypotheses, and also to estimate the extent to which life history 99 traits can modulate the relationship. In particular, beyond exploring how group size and 100 temperature would affect the correlation between basal cortisol production and social 101 rank (*i.e.* effect size), we also considered other moderators such as fish size, sex, age, 102 reproduction type and salinity.

103

104 **2. Methods**

105 (a) Literature search and study selection

We screened Web of Science for papers published before June 24th 2020 containing the terms "subordinate" OR "dominant" OR "dominance" AND "fish" AND "cortisol" AND "stress". We obtained 165 studies, from which we retained 73 that measured basal plasma cortisol in groups of fish with established hierarchies, in the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) checklist (Appendix A, supplementary Figure 1).

- 112
- 113 (b) Estimating effect sizes

114 Statistical results (r, t, F, χ^2 , z) were extracted directly from text or figures and, when not 115 directly available, results were converted into statistical tests (n=7). One study was 116 removed because direction of the effect was not known, leading to a total of 72 studies. 117 In most instances, a Pearson's product-moment correlation coefficient, r, was provided 118 and used as our measure of effect size, when cortisol values concerned at least three 119 individuals in the study. In all other cases, we followed a recognized methodology by 120 Rosenthal (1991) (Rosenthal, 1991) to estimate r using published statistics. This 121 calculation uses mean and variance of treatments, statistical results (t, F, χ^2 , z), and p-122 values reported with sample sizes, in respective order of preference. The calculation was 123 performed so that a positive relationship between cortisol values and rank order returned 124 a positive r, meaning that subordinates have higher cortisol than dominants. Generally, r-125 values above 0.5 are considered to reflect large effect sizes in ecology (Cohen, 1992; 126 Jennions and Møller, 2002). For further analyses, r-values were transformed to Fisher's 127 z, to improve normality (Koricheva et al., 2013).

128

129 (c) Meta-analyses using random effects

130 We used hierarchical linear mixed-effects meta-analysis to test for both overall effect 131 sizes and the importance of our moderators (Nakagawa and Santos, 2012) using the 132 package 'metafor' v. 2.1-0 (Viechtbauer, 2010) in R v. 3.5.2. The overall effect sizes (i.e., 133 mean of the effect sizes weighted by the inverse of their variance) were considered 134 significant if their 95% confidence intervals (CI) did not include zero (Koricheva et al., 135 2013). We used model selection based on the Akaike information criterion to determine 136 which random factors should be included in each meta-analysis (Nakagawa and Santos, 137 2012). Non-independence of effect sizes within and among studies was accounted for by 138 including i) the number of effect sizes collected "id" (which corresponds to the collected 139 values) and ii) the "studies" (which corresponds to the reviewed article itself) as random-140 factors. Phylogenetic non-independence across studies was handled by including 141 relationships among species (Appendix A, supplementary Figure 2) as a random effect 142 based on the Open Tree of Life database (Hinchliff et al., 2015), and Grafen's method 143 providing a correlation matrix from branch length (Grafen and Hamilton, 1989). The R 144 package 'rotl' v. 3.0.10 was used for this purpose (Michonneau et al., 2016)

145 Group size (dyadic contexts or group>2), reproduction type (monogamous, polygamous 146 and polygynous), habitat temperature (temperate vs tropical), time since grouping (h) and 147 fish weight (g, square-root transformed) were considered as moderators of effect size. We 148 tested for multi-collinearity among moderators and the highest variance inflation factor 149 was 2.26, below the recognized threshold of 3 (Zuur et al., 2010). The factors salinity 150 (freshwater n = 67, sea n = 7 and brackish n = 4 water) and sex (male n = 23, female = 7, 151 mixed group n=31 and ambisexual n=1) could not be considered owing to discrepancies 152 in the numbers of effect sizes. We used a multi-model inference approach based on 153 Akaike's criteria corrected for small sample sizes (AICc) (Burnham and Anderson, 2002). 154 We first assessed the relative strengths of each candidate model by calculating its Akaike 155 weight, to identify the most parsimonious model. A constant term (the intercept) was 156 included in all models. We estimated the importance of a predictor by summing the 157 Akaike weights of all models in which that candidate variable appeared, so ranking 158 predictors in order of importance (Burnham and Anderson, 2002). All multi-model 159 analyses were conducted using the MuMIn R package v. 1.40.0 (Barton, 2014).

160

161 (d) Heterogeneity

We used the *P* index as a measure of heterogeneity in effect sizes. The index represents the proportion of total variation in the data that is not a consequence of sampling error 164 (Higgins et al., 2003). We used an extended version of P that partitions total heterogeneity 165 between the variation explained by the study identity and by the variation of the residuals. 166 The latter remains to be explained by our moderators (Nakagawa and Santos, 2012). We 167 calculated the degree of phylogenetic signal in our estimates of effect size using the 168 phylogenetic heritability index, H^2 , which is the variance attributable to phylogeny in 169 relation to the total variance in the data (Nakagawa and Santos, 2012). H^2 is equivalent to 170 Pagel's λ (Pagel, 1999), where higher values indicate stronger phylogenetic signals.

171

172 (e) Publication bias

We used Egger's regression to highlight any publication bias, because studies with low sample sizes tend to be rejected due to their lower probability of finding significant effects (Egger et al., 1997; Koricheva et al., 2013). The intercept of the regression is considered to demonstrate publication bias when its estimate significantly differs from zero (Egger et al., 1997). To overcome the non-independent nature of our data, we also applied the Egger's regression test on the meta-analytic residuals (Nakagawa and Santos, 2012).

- 179 **3. Results**
- 180

181 The final dataset comprised 78 effect sizes, across 22 species, from 72 studies. Of all 182 models tested (n=11), the most parsimonious (intercept-only) was the one with both 183 studies and number of effect sizes (categorical) as random factor (AICc= 144.41), while adding the phylogeny resulted in the second most parsimonious (AICc= 146.57). 184 185 Therefore, to retain phylogenetic relatedness in our models, we performed all analyses 186 using this latter model with three random factors. Overall and as predicted, the 187 hierarchical rank (higher for subordinates) was strongly and positively correlated with cortisol (Fisher's z = 0.4546, CI = 0.26 - 0.65, Figure 1). We found considerable variation 188 among studies ($I_{total}^2 = 88.5\%$, $P_{studies} = 84.6\%$, $P_{residual} = 4\%$), which was expected 189 190 because they were almost as numerous as effect sizes. The phylogenetic signal was weak 191 $(H^2 = 4.4\%)$. We found no evidence of publication bias (Egger's regression of effect sizes: 192 Intercept = 1.1158, P = 0.14; Egger's regression of meta-analytic residuals: Intercept =0.14, P = 0.85; Figure 2). Multi-model inference indicated that group size (pair or > 2), 193 194 habitat temperature (temperate or tropical) and body weight were the most important 195 predictors of the magnitude of rank-cortisol relationship (Table 1). When cortisol was 196 evaluated in a dyadic context, there was a stronger and more positive rank-cortisol

197 relationship than when evaluated in groups. For example, dominants in groups sometimes 198 had higher basal cortisol than subordinates (Table 1 and Figure 3A). The difference in 199 basal cortisol between dominant and subordinates was also stronger and more positive in 200 fishes from temperate compared to tropical regions (Table 1 and Figure 3B). Finally, 201 mean fish body weight was positively correlated to Fisher's z (Table 1 and Figure 3C), 202 such that the difference in basal cortisol across hierarchical ranks was highly and 203 positively correlated with fish body weight. Importantly, this was true when testing these 204 moderators alone or in the same model.

205

206 **4. Discussion**

207

Our data support the main hypothesis, that dominants generally exhibit lower basal cortisol level than subordinates. This effect is, however, modulated by group size, habitat temperature and fish size. We identified two main possible explanations for why cortisol varies in such cases, the 'stress-enhanced hypothesis' where this would be due to stress itself, or the 'metabolic-enhanced hypothesis' where this would be due to variation in metabolic rates.

214 Since cortisol is recognized as the main stress hormone in fishes, the fact that dominants 215 generally produce less cortisol than subordinates was expected, as a consequence of the 216 "stress-enhanced hypothesis". Dominants are aggressive towards subordinates in order to 217 keep their status. All these aggressive acts are perceived as stressful events explaining the 218 difference in cortisol levels. Interestingly, we found that the cortisol differences between 219 dominant and subordinates were more prominent when groups are small. This is because 220 in most tests performed in dyadic contexts, dominants presented lower cortisol levels than 221 subordinates whereas, in tests performed in larger groups, dominants could actually 222 exhibited slightly higher cortisol levels than subordinates. This reveals the complexity 223 with which social hierarchies can elicit stress. Individuals in large groups may face more intense food competition (Borcherding et al., 2019), cannibalism (Naumowicz et al., 224 225 2017), susceptibility to capture (Thambithurai et al., 2018) and sex competition (Wacker 226 et al., 2017), for example. All of these processes may stress fishes, but dominants can use 227 their status to monopolise opportunities to improve fitness. In larger groups, such valuable 228 status could also result in more frequent challenges by conspecifics, increasing the costs 229 of retaining dominant status and resulting in higher relative stress levels. In addition, our 230 results could also indicate that subordinates are relatively less stressed in groups. This

231 would be the consequence of having the agressive behavior of the dominant fish shared 232 amongst multiple subordinates. Altogether, these results support the "stress-enhanced 233 hypothesis": holding a particular status drives your stress levels, indicated by cortisol 234 values. The metabolism of dominant individuals is often, however, higher than that of 235 subordinates (Geffroy et al., 2016; Metcalfe et al., 1995). Basal cortisol levels are also 236 positively correlated with metabolism (Mommsen et al., 1999), and we might therefore 237 expect that dominants would have higher cortisol values. The opposite was observed in 238 our study, further supporting the "stress-enhanced hypothesis".

239 Our findings relative to group-size also provide a plausible hypothesis to explain why 240 cortisol would show opposite results depending on the reproductive strategy of a species 241 (protandry or protogyny). In such species, cortisol has a key role in sex determination 242 (Geffroy and Douhard, 2019), notably in many hermaphroditic reef fishes where social 243 interactions shape the dynamics of sex-change (Goikoetxea et al., 2017). Indeed, a 244 contribution of cortisol was demonstrated in two protogynous species where big males 245 monopolize harems (large groups of females), and an increase in cortisol is necessary for 246 dominant females to change into the male (Nozu and Nakamura, 2015; Todd et al., 2019). 247 On the other hand, the female dominant of small groups of protandrous fish (e.g. 248 clownfish) may be less stressed, and subordinate males would then remain males because 249 of the stress imposed by the female, although this remains to be fully demonstrated 250 (Olivotto and Geffroy, 2017). The fact that mating system (monogamous, polygamous 251 and polygynous) was not included in the most parsimonious model does not permit us to 252 extrapolate our results, since most protandrous fish are monogamous and most 253 protogynous fish are polygynous. More studies focusing on hermaphroditic fishes are, 254 clearly, required.

In any case, the fact that the difference between basal cortisol in dominants and subordinates of large groups was lesser than in small groups argues for the 'stressenhanced hypothesis', where basal cortisol levels in dominants (and subordinates) is a consequence of a physiological stress, not merely of metabolic rate.

We found that habitat temperature (temperate versus tropical) also shapes the relationship between basal cortisol and social status. This would advocate for the "metabolicenhanced" hypothesis. In warmer habitats, fishes will generally have higher metabolic rates (Killen et al., 2016), reflecting direct thermodynamic effects on all biochemical and physiological processes (Cossins and Bowler, 1987). Rates of basal cortisol production are known to be stimulated by increased temperature within fish species (Goikoetxea et

265 al., 2021). High background rates of basal cortisol production in all individuals at high 266 temperatures may tend to obscure differences in production that reflect stress due to social 267 rank, especially if these are relatively subtle. The higher metabolism of fishes in warm 268 environments may also lead to higher overall activity levels and general aggressiveness 269 (Biro et al., 2010; Colchen et al., 2017), and reduce the stability of hierarchies (Kochhann 270 et al., 2015), which could also obscure inter-individual differences in basal cortisol 271 production due to social rank. That is, subtle differences in the level of stress due to social 272 rank might be easier to observe in fish with overall lower metabolic rates in cooler 273 temperate habitats (Alfonso et al., 2020), especially if differences in rank are associated 274 with greater relative differences in activity level or behavioral tendencies such as 275 boldness.

276 The finding that mean weight of fish was positively correlated to the Fisher's z might 277 simply indicate that a difference in size between dominant and subordinates becomes 278 more pronounced as mean size increases. Hence, cortisol differences between dominants 279 and subordinates also increase with this size difference. This could be due to the fact that 280 bigger dominant fish impose higher stress on subordinates. Size difference may also 281 reflect differences in life-history stage, another factor known to influence basal cortisol 282 (Love et al., 2005). Unfortunately, the exact size of fish at social ranks was rarely reported 283 in the articles reviewed, so this remains an interesting topic for future study.

After a dominance hierarchy is initiated by the formation of a group, cortisol should increase in all individuals. Cortisol levels are expected to stabilize and diverge once ranks are established. That led us to expect 'time since grouping' to be an important variable in our model but we did not observe this. This suggests that the hierarchy is established very readily in most instances and that differences in basal cortisol among social ranks are relatively consistent over time, or at least that the studies we used only began measuring cortisol once the hierarchy had been settled.

291 Overall, our phylogenetic meta-analysis provided strong support for the hypothesis that 292 dominant fish generally display lower baseline cortisol than subordinates, where high 293 cortisol likely represents physiological stress. Nonetheless, it remains difficult to assess 294 whether social status is a cause (due to stress) or a consequence (due to metabolic rate) 295 of differences in basal cortisol levels among individuals. Our results seem to provide 296 support for both stress-enhanced and metabolism-enhanced hypotheses, indicating that 297 multiple contributing factors are probably at play. That is, there can be a contribution of 298 both biotic (sociality and fish size) and abiotic (habitat temperature) factors. The

- 299 establishment of a hierarchy triggers physiological stress but the degree of cortisol release
- 300 is accentuated or diminished by habitat temperature and its effects on metabolism. These
- 301 findings have major implications for our understanding of the ecology and sociality of
- 302 fishes.
- 303

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- 310
- 311 Bibliography
- 312
- 313 Abbott, D.H., Keverne, E.B., Bercovitch, F.B., Shively, C.A., Mendoza, S.P., Saltzman,
- 314 W., Snowdon, C.T., Ziegler, T.E., Banjevic, M., Garland, T., Sapolsky, R.M., 2003. Are
- subordinates always stressed? A comparative analysis of rank differences in cortisollevels among primates. Horm. Behav. 43, 67–82.
- 317 Alfonso, S., Gesto, M., Sadoul, B., 2020. Temperature increase and its effects on fish
- 318 stress physiology in the context of global warming. J. Fish Biol.
- 319 https://doi.org/10.1111/jfb.14599
- 320 Baker, M.R., Gobush, K.S., Vynne, C.H., 2013. Review of factors influencing stress
- 321 hormones in fish and wildlife. J. Nat. Conserv. 21, 309–318.
- 322 https://doi.org/10.1016/j.jnc.2013.03.003
- 323 Barton, K., 2014. MuMIn: Multi-model inference. R package version 1.40.0.
- 324 Bigiani, A., Mucignat-Caretta, C., Montani, G., Tirindelli, R., 2005. Pheromone
- 325 reception in mammals, in: Reviews of Physiology, Biochemistry and Pharmacology,
- 326 Reviews of Physiology, Biochemistry and Pharmacology. Springer, Berlin, Heidelberg,
- 327 pp. 1–35. https://doi.org/10.1007/s10254-004-0038-0
- 328 Biro, P.A., Beckmann, C., Stamps, J.A., 2010. Small within-day increases in
- 329 temperature affects boldness and alters personality in coral reef fish. Proc. R. Soc. B
- 330 Biol. Sci. 277, 71–77. https://doi.org/10.1098/rspb.2009.1346
- 331 Borcherding, J., Heubel, K., Storm, S., 2019. Competition fluctuates across years and
- 332 seasons in a 6-species-fish community: empirical evidence from the field. Rev. Fish
- 333 Biol. Fish. 29, 589–604. https://doi.org/10.1007/s11160-019-09567-x
- Breuner, C.W., Hahn, T.P., 2003. Integrating stress physiology, environmental change,
- and behavior in free-living sparrows. Horm. Behav. 43, 115–123.
- 336 https://doi.org/10.1016/S0018-506X(02)00020-X
- 337 Burnham, K.P., Anderson, D.R., 2002. Model selection and multimodel inference: a
- 338 practical information-theoretic approach, 2nd edn. ed. Springer, New York, NY.
- 339 https://doi.org/10.1016/j.ecolmodel.2003.11.004

- 340 Campderrich, I., Liste, G., Estevez, I., 2017. The looks matter; aggression escalation
- 341 from changes on phenotypic appearance in the domestic fowl. PLOS ONE 12,
- 342 e0188931. https://doi.org/10.1371/journal.pone.0188931
- 343 Cohen, J., 1992. A power primer. Psychol. Bull. 112, 155–159.
- 344 Colchen, T., Teletchea, F., Fontaine, P., Pasquet, A., 2017. Temperature modifies
- 345 activity, inter-individual relationships and group structure in a fish. Curr. Zool. 63, 175–
- 346 183. https://doi.org/10.1093/cz/zow048
- 347 Cossins, A.R., Bowler, K., 1987. The direct effects of temperature changes, in: Cossins,
- 348 A.R., Bowler, K. (Eds.), Temperature Biology of Animals. Springer Netherlands,
- 349 Dordrecht, pp. 23–60. https://doi.org/10.1007/978-94-009-3127-5_2
- 350 Dawson, J.W., Mannan, R.W., 1991. Dominance Hierarchies and Helper Contributions
- 351 in Harris' Hawks. The Auk 108, 649–660. https://doi.org/10.2307/4088105
- 352 Egger, M., Smith, G.D., Schneider, M., Minder, C., 1997. Bias in meta-analysis
- detected by a simple, graphical test. BMJ 315, 629–634.
- Ejike, C., Schreck, C.B., 1980. Stress and Social Hierarchy Rank in Coho Salmon.
- 355 Trans. Am. Fish. Soc. 109, 423–426. https://doi.org/10.1577/1548-
- 356 8659(1980)109<423:SASHRI>2.0.CO;2
- 357 Geffroy, B., Bolliet, V., Bardonnet, A., 2016. Kleptoparasitism and aggressiveness are
- influenced by standard metabolic rate in eels. Physiol. Behav. 157, 165–169.
- 359 Geffroy, B., Douhard, M., 2019. The Adaptive Sex in Stressful Environments. Trends
- 360 Ecol. Evol. 34, 628–640. https://doi.org/10.1016/j.tree.2019.02.012
- 361 Ginnaw, G.M., Davidson, I.K., Harding, H.R., Simpson, S.D., Roberts, N.W., Radford,
- A.N., Ioannou, C.C., 2020. Effects of multiple stressors on fish shoal collective motion are independent and vary with shoaling metric. Anim. Behav. 168, 7–17.
- 364 https://doi.org/10.1016/j.anbehav.2020.07.024
- 365 Goikoetxea, A., Sadoul, B., Blondeau-Bidet, E., Aerts, J., Blanc, M.-O., Parrinello, H.,
- 366 Barrachina, C., Pratlong, M., Geffroy, B., 2021. Genetic pathways underpinning
- 367 hormonal stress responses in fish exposed to short- and long-term warm ocean
- 368 temperatures. Ecol. Indic. 120, 106937. https://doi.org/10.1016/j.ecolind.2020.106937
- 369 Goikoetxea, A., Todd, E.V., Gemmell, N.J., 2017. Stress and sex: does cortisol mediate
- sex change in fish? Reproduction 154, R149–R160. https://doi.org/10.1530/REP-170408
- 372 Grafen, A., Hamilton, W.D., 1989. The phylogenetic regression. Philos. Trans. R. Soc.
- 373 Lond. B Biol. Sci. 326, 119–157. https://doi.org/10.1098/rstb.1989.0106
- 374 Grosenick, L., Clement, T.S., Fernald, R.D., 2007. Fish can infer social rank by
- observation alone. Nature 445, 429–432. https://doi.org/10.1038/nature05511
- 376 Hahn, S., Bauer, S., 2008. Dominance in feeding territories relates to foraging success
- and offspring growth in brown skuas Catharacta antarctica lonnbergi. Behav. Ecol.
- 378 Sociobiol. 62, 1149–1157. https://doi.org/10.1007/s00265-007-0543-7
- 379 Harris, B.N., 2020. Stress hypothesis overload: 131 hypotheses exploring the role of
- 380 stress in tradeoffs, transitions, and health. Gen. Comp. Endocrinol. 288, 113355.
- 381 https://doi.org/10.1016/j.ygcen.2019.113355
- 382 Higgins, J.P.T., Thompson, S.G., Deeks, J.J., Altman, D.G., 2003. Measuring
- inconsistency in meta-analyses. BMJ 327, 557–560.
- 384 https://doi.org/10.1136/bmj.327.7414.557
- 385 Hinchliff, C.E., Smith, S.A., Allman, J.F., Burleigh, J.G., Chaudhary, R., Coghill, L.M.,
- 386 Crandall, K.A., Deng, J., Drew, B.T., Gazis, R., Gude, K., Hibbett, D.S., Katz, L.A.,
- 387 Laughinghouse, H.D., McTavish, E.J., Midford, P.E., Owen, C.L., Ree, R.H., Rees,
- J.A., Soltis, D.E., Williams, T., Cranston, K.A., 2015. Synthesis of phylogeny and
- taxonomy into a comprehensive tree of life. Proc. Natl. Acad. Sci. 112, 12764–12769.

- 390 https://doi.org/10.1073/pnas.1423041112
- 391 Holekamp, K.E., Strauss, E.D., 2020. Reproduction Within a Hierarchical Society from
- a Female's Perspective. Integr. Comp. Biol. 60, 753–764.
- 393 https://doi.org/10.1093/icb/icaa068
- 394 Jennions, M., Møller, A.P., 2002. How much variance can be explained by ecologists
- and evolutionary biologists? Oecologia 132, 492–500. https://doi.org/10.1007/s00442 002-0952-2
- 397 Killen, S.S., Glazier, D.S., Rezende, E.L., Clark, T.D., Atkinson, D., Willener, A.S.T.,
- 398 Halsey, L.G., 2016. Ecological Influences and Morphological Correlates of Resting and
- 399 Maximal Metabolic Rates across Teleost Fish Species. Am. Nat. 187, 592–606.
- 400 https://doi.org/10.1086/685893
- 401 Kochhann, D., Campos, D.F., Val, A.L., 2015. Experimentally increased temperature
- 402 and hypoxia affect stability of social hierarchy and metabolism of the Amazonian
- 403 cichlid Apistogramma agassizii. Comp. Biochem. Physiol. A. Mol. Integr. Physiol. 190,
- 404 54-60. https://doi.org/10.1016/j.cbpa.2015.09.006
- 405 Koolhaas, J.M., Korte, S.M., De Boer, S.F., Van Der Vegt, B.J., Van Reenen, C.G.,
- 406 Hopster, H., De Jong, I.C., Ruis, M.A.W., Blokhuis, H.J., 1999. Coping styles in
- 407 animals: current status in behavior and stress-physiology. Neurosci. Biobehav. Rev. 23,
- 408 925–935. https://doi.org/10.1016/S0149-7634(99)00026-3
- 409 Koricheva, J., Gurevitch, J., Mengersen, K., 2013. Handbook of meta-analysis in
- 410 ecology and evolution. Princeton University Press, Princeton, New Jersey.
- 411 Landys, M.M., Ramenofsky, M., Wingfield, J.C., 2006. Actions of glucocorticoids at a
- 412 seasonal baseline as compared to stress-related levels in the regulation of periodic life
- 413 processes. Gen. Comp. Endocrinol. 148, 132–149.
- 414 https://doi.org/10.1016/j.ygcen.2006.02.013
- 415 Love, O.P., Chin, E.H., Wynne-Edwards, K.E., Williams, T.D., 2005. Stress Hormones:
- 416 A Link between Maternal Condition and Sex-Biased Reproductive Investment. Am.
- 417 Nat. 166, 751–766. https://doi.org/10.1086/497440
- 418 Metcalfe, N.B., Taylor, A.C., Thorpe, J.E., 1995. Metabolic rate, social status and life-
- 419 history strategies in Atlantic salmon. Anim. Behav. 49, 431–436.
- 420 https://doi.org/10.1006/anbe.1995.0056
- 421 Michonneau, F., Brown, J.W., Winter, D.J., 2016. rotl: an R package to interact with the
- 422 Open Tree of Life data. Methods Ecol. Evol. 7, 1476–1481.
- 423 https://doi.org/10.1111/2041-210X.12593
- 424 Mommsen, T.P., Vijayan, M.M., Moon, T.W., 1999. Cortisol in teleosts: dynamics,
- 425 mechanisms of action, and metabolic regulation. Rev. Fish Biol. Fish. 9, 211–268.
- 426 https://doi.org/10.1023/A:1008924418720
- 427 Nadler, L.E., Killen, S.S., Domenici, P., McCormick, M.I., 2018. Role of water flow
- 428 regime in the swimming behaviour and escape performance of a schooling fish. Biol.
- 429 Open 7. https://doi.org/10.1242/bio.031997
- 430 Nakagawa, S., Santos, E.S.A., 2012. Methodological issues and advances in biological
- 431 meta-analysis. Evol. Ecol. 26, 1253–1274. https://doi.org/10.1007/s10682-012-9555-5
- 432 Naumowicz, K., Pajdak, J., Terech-Majewska, E., Szarek, J., 2017. Intracohort
- 433 cannibalism and methods for its mitigation in cultured freshwater fish. Rev. Fish Biol.
- 434 Fish. 1, 193–208. https://doi.org/10.1007/s11160-017-9465-2
- 435 Nozu, R., Nakamura, M., 2015. Cortisol Administration Induces Sex Change from
- 436 Ovary to Testis in the Protogynous Wrasse, <i>Halichoeres
- 437 trimaculatus</i> Sex. Dev. 9, 118–124.
- 438 https://doi.org/10.1159/000373902
- 439 Olivotto, I., Geffroy, B., 2017. Clownfish, in: Calado, R., Olivotto, I., Oliver, M.P.,

- 440 Holt, G.J. (Eds.), Marine Ornamental Species Aquaculture. John Wiley & Sons, Ltd, pp.
- 441 177–199. https://doi.org/10.1002/9781119169147.ch12
- 442 Pagel, M., 1999. Inferring the historical patterns of biological evolution. Nature 401,
- 443 877–884. https://doi.org/10.1038/44766
- 444 Palko, B.J., Beardsley, G.L., Richards, W.J., 1981. Synopsis of the biology of the
- 445 swordfish, Xiphias gladius Linnaeus. NOAA Tech. Rep.
- 446 Ricklefs, R.E., Wikelski, M., 2002. The physiology/life-history nexus. Trends Ecol.
- 447 Evol. 17, 462–468. https://doi.org/10.1016/S0169-5347(02)02578-8
- 448 Rosenthal, R., 1991. Meta-analytic procedures for social research, Rev. ed, Meta-
- 449 analytic procedures for social research, Rev. ed. Sage Publications, Inc, Thousand Oaks,
- 450 CA, US. https://doi.org/10.4135/9781412984997
- 451 Rousseau, K., Prunet, P., Dufour, S., 2021. Special features of neuroendocrine
- 452 interactions between stress and reproduction in teleosts. Gen. Comp. Endocrinol. 300,
- 453 113634. https://doi.org/10.1016/j.ygcen.2020.113634
- 454 Rubenstein, D.R., Abbot, P., 2017. Comparative Social Evolution. Cambridge455 University Press.
- 456 Sadoul, B., Geffroy, B., 2019. Measuring cortisol, the major stress hormone in fishes. J.
- 457 Fish Biol. 94, 540–555. https://doi.org/10.1111/jfb.13904
- 458 Sadoul, B., Vijayan, M.M., 2016. 5 Stress and Growth, in: Carl B. Schreck, L.T.,
- 459 Anthony P. Farrell and Colin J. Brauner (Ed.), Fish Physiology, Biology of Stress in 460 Eich Eich Physiology, Academia Press, pp. 167, 205
- 460 FishFish Physiology. Academic Press, pp. 167–205.
- 461 Samaras, A., Pavlidis, M., 2018. Regulation of divergent cortisol responsiveness in
- 462 European sea bass, Dicentrarchus labrax L. PLOS ONE 13, e0202195.
- 463 https://doi.org/10.1371/journal.pone.0202195
- 464 Schjelderup-Ebbe, T., 1922. Contributions to the social psychology of the domestic
- 465 chicken. Repr. Z. Fuer Psychol. 88, 225–252.
- 466 Schreck, C.B., Tort, L., 2016. 1 The Concept of Stress in Fish, in: Schreck, C.B., Tort,
- 467 L., Farrell, A.P., Brauner, C.J. (Eds.), Fish Physiology, Biology of Stress in Fish.
- 468 Academic Press, pp. 1–34. https://doi.org/10.1016/B978-0-12-802728-8.00001-1
- 469 Sogawa, S., Ota, K., Kohda, M., 2016. A dear enemy relationship in a territorial cichlid:
- 470 evidence for the threat-level hypothesis. Behaviour 153, 387–400.
- 471 https://doi.org/10.1163/1568539X-00003351
- 472 Stevens, E.D., Fry, F.E.J., 2011. Heat transfer and body temperatures in non-
- 473 thermoregulatory teleosts. Can. J. Zool. https://doi.org/10.1139/z74-152
- 474 Teichroeb, J.A., White, M.M.J., Chapman, C.A., 2015. Vervet (Chlorocebus
- 475 pygerythrus) Intragroup Spatial Positioning: Dominants Trade-Off Predation Risk for
- 476 Increased Food Acquisition. Int. J. Primatol. 36, 154–176.
- 477 https://doi.org/10.1007/s10764-015-9818-4
- 478 Thambithurai, D., Hollins, J., Leeuwen, T.V., Rácz, A., Lindström, J., Parsons, K.,
- 479 Killen, S.S., 2018. Shoal size as a key determinant of vulnerability to capture under a
- 480 simulated fishery scenario. Ecol. Evol. 8, 6505–6514. https://doi.org/10.1002/ece3.4107
- 481 Todd, E.V., Ortega-Recalde, O., Liu, H., Lamm, M.S., Rutherford, K.M., Cross, H.,
- 482 Black, M.A., Kardailsky, O., Graves, J.A.M., Hore, T.A., Godwin, J.R., Gemmell, N.J.,
- 483 2019. Stress, novel sex genes, and epigenetic reprogramming orchestrate socially
- 484 controlled sex change. Sci. Adv. 5, eaaw7006. https://doi.org/10.1126/sciadv.aaw7006
- 485 Viechtbauer, W., 2010. Conducting meta-analyses in R with the metafor package. J.
- 486 Stat. Softw. 36, 1–48.
- 487 Wacker, S., Ness, M.H., Östlund-Nilsson, S., Amundsen, T., 2017. Social structure
- 488 affects mating competition in a damselfish. Coral Reefs 36, 1279–1289.
- 489 https://doi.org/10.1007/s00338-017-1623-4

490 491 492 493 494 495	Wilson, E.O., 2000. Sociobiology: The New Synthesis, Twenty-Fifth Anniversary Edition. Harvard University Press. Zuur, A.F., Ieno, E.N., Elphick, C.S., 2010. A protocol for data exploration to avoid common statistical problems. Methods Ecol. Evol. 1, 3–14. https://doi.org/10.1111/j.2041-210X.2009.00001.x
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505	Figure Captions
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507	Figure 1. Forest plot of the social rank-cortisol effect sizes. Effect sizes are shown in
508	ascending order. Filled circles with horizontal lines represent effect size \pm 95%
509	confidence intervals. The overall and significant effect size is represented by a diamond.
510	
511	Figure 2. Funnel plots of the meta-analyse using both the effect sizes and the meta-
512	analytic residuals of the best model.
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514	Figure 3. Effects of (A) group size, (B) temperature and (C) mean fish size on the social
515	rank-cortisol relationship (Fisher's z). Plots (A) and (B) show mean \pm 95% confidence
516	intervals. The number of species tested at each factor level is shown in the bottom of
517	plots.
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Table 1. Summary of the multi-model inference conducted to explain variation in the

535 social rank-cortisol in fish

Predictor	Levels	Estimate	S.E.	z-value
Intercept		0.5	0.34	1.49
Group size	Paired	0.28	0.14	1.98*
Species body size		0.028	0.014	2.12*
Environment	Tropical	-0.67	0.32	-2.08*

536 Estimates are average coefficients of the model, their associated standard error (S.E). *

- 537 indicates a p-value < 0.05.