

1 Bold zebrafish (*danio rerio*) learn faster in an associative learning task

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10 **Abstract**

11 Animals differ in their ability to learn. One potential factor contributing to learning
12 differences is personality types. We investigated the relationship between learning and the bold-
13 shy continuum by testing learning performance of bold and shy zebrafish across two associative
14 learning tasks: conditioned place preference (CPP) and 2 choice. Bold fish learned significantly
15 faster than the shy fish but there were no differences in the magnitude of change in behavior
16 between the personality types in CPP. When tested in the 2 choice task, we found no clear
17 evidence of learning between personality types or controls. Overall, our study suggests that bold
18 fish tend to be faster learners when compared to shy fish. The lack of differences in the final
19 change in behavior suggests that the learning difference is due to neophobic tendencies and
20 resulting initial interactions with the learning stimulus.

21 **Keywords:** Personality, cognition, bold, shy, operant, classical, associative learning

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28 **Introduction**

29 As animals interact with their environment, how quickly they learn and recall these
30 interactions can vary between individuals (Boogert et al., 2018, Cauchoix et al., 2018). It has
31 been hypothesized that variation in learning between individuals can be explained in part by
32 differing personality types (Dingemanse & Wolf, 2010, Sih & Del Guidice, 2012, Sih et al.,
33 2004). Across many animal taxa, studies demonstrate that one common dimension of personality
34 is the bold-shy continuum (Réale et al., 2007). Bold individuals are characterized by displaying
35 lower neophobic and stress-related behaviors and have higher exploratory activity. In contrast
36 shy individuals tend to have opposing traits (Wilson et al., 1994, Sih et al., 2004, Baker et al.,
37 2018).

38 However, studies across taxa find a conflicting relationship between personality and
39 learning. Many studies showed that bold individuals learn faster than shy, in animals ranging
40 from mammals, birds, to teleost fish (Mazza et al., 2018, Guenther et al., 2014, Dugatkin &
41 Alfieri, 2002, DePasquale et al., 2014, Bensky et al., 2017, Daniel & Bhat, 2020, Kareklaš,
42 Elwood & Holland, 2017). Fewer studies either found the opposite (e.g. shy learn faster than
43 bold) or no relationship between personality and learning speed (Lermite, Peneaux & Griffin,
44 2016, Ferron et al., 2015, Sommer-Trembo & Plath, 2018, Baker & Wong, 2019).
45 Inconsistencies across studies suggest that other factors likely influence learning performance

46 beyond personality type. Aspects of the learning assay like the type of task (e.g. operant or
47 classical conditioning) or the context that the animal is tested in could affect the relationship
48 between personality and learning (Poirer et al., 2020, Dingemanse & Wolf, 2010).

49 Individuals of varying personality types likely interact with different learning task or
50 stimuli in different ways, which may influence learning performance (Sih & Guidice, 2012). For
51 example, different training paradigms will have the animal engage with the stimulus in different
52 ways. Some studies found that learning is not correlated across different training paradigms
53 (Guillette et al., 2015, Duceatz et al., 2014, Kassai et al., 2022, Poirer et al., 2020) and one found
54 that changing the difficulty of the learning task changed the relationship between personality and
55 learning speed (Chang et al., 2018). Similarly, a meta-analysis in non-human animals found a
56 low correlation between learning ability across cognitive tasks (Poirer et al., 2020). This suggests
57 the need for measurements of multiple learning tasks due to the potential for across task variation
58 (Griffin et al., 2015). Neophobia, associated with a shy personality, has been seen to affect
59 operant learning of a food reward due to higher latencies to approach (Stöwe et al., 2006). Thus,
60 comparing a passive (classical) task that does not require the animal to approach a novel object
61 to an active (operant) task that does require approach may produce different results.

62 In this study, we investigated the effect of personality type on learning performance
63 across two associative learning paradigms using zebrafish (*Danio rerio*). Using a within-subjects
64 and counter-balanced design we individually trained bold and shy zebrafish to associate a visual
65 stimulus with a food reward in both conditioned place preference and 2 choice tasks. We tested
66 the prediction that bold individuals will be faster learners compared to shy fish because of their
67 decreased neophobia. We also evaluated the prediction that there will be an interaction effect of
68 personality and training paradigm on learning speed. Given that the operant task requires fish to

69 actively make a choice, we expected that bold fish would learn faster in this task than shy fish
70 due to their decreased neophobia.

71

72 **Methods**

73 *Animals*

74 We used zebrafish from selectively bred lines that exhibit shy (high stationary behavior,
75 HSB) or bold personality traits (low stationary behavior, LSB) (n = 48 per line). Across six
76 different stress and anxiety-like behavioral assays, the HSB line exhibits greater amount of
77 behaviors consistent with a shy personality type (e.g., freezing, less exploratory, higher cortisol
78 levels) than the LSB line (Wong et al., 2012, Baker & Wong, 2019). Additionally, the
79 exploratory behavior of the lines in an open field test is repeatable and reliable (Baker & Wong,
80 2019). The HSB line also shows faster release of cortisol under stress compared to the LSB line
81 (Wong et al., 2019). For simplicity, we will refer to the HSB and LSB lines as shy and bold
82 personality types, respectively. The fish used in this study were selectively bred for 13
83 generations from wild caught zebrafish. Before testing, we housed the fish together in 40L tanks
84 and fish were fed twice a day with Tetramin Tropical Flakes (Tetra, USA). One week prior to
85 testing we physically isolated fish into 3-liter tanks on a recirculating water system (Pentair
86 Aquatic Eco-Systems or Aquaneering) using UV and solid filtration on a 14:10 L/D cycle at a
87 temperature of 27 °C. Fish had visual and olfactory access to each other. Starting three days
88 before testing we withheld food from the fish to reduce the possibility of satiation while training.

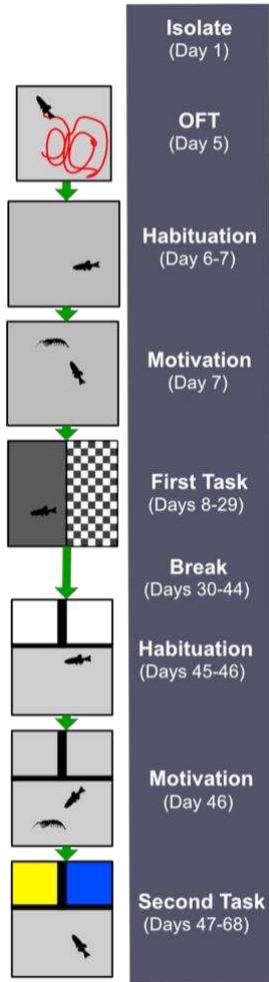


Figure 1. Overview of Experiment Timeline. Fish of all groups started with isolation on the first day then went through an open field test (OFT), habituation, motivation test, and training on the first task (conditioned place preference task in this illustration). After a break fish went through the same for the second task (2 choice discrimination task in this illustration). Our study design was counterbalanced and half of the fish began with the CPP task while the other half began with the 2 choice discrimination task.

89

90

91 *Behavioral Assays Overview*

92 We conducted four behavioral assays on each fish: an open field test (OFT), a test for
93 food motivation, a 2 choice discrimination task (operant conditioning), and a conditioned place
94 preference (CPP, classical conditioning) task. The OFT and food motivation test were performed
95 prior to training. Using a within-subjects design, we tested each fish in both associative learning
96 paradigms and counterbalanced the starting paradigm (Figure 1). We used frozen adult brine
97 shrimp (*Artemia* spp., San Francisco Bay Brand, USA) administered in liquid form as the food
98 reward. Half of the fish received distilled water instead of brine shrimp to serve as controls. We

99 started with four groups of 24 fish (bold control, shy control, bold treatment, and shy treatment).
100 All behavioral assays were performed between 3-8 hours after light onset. After 4 days of
101 isolation, we tested each fish in the open field test to validate behavioral phenotype. We then
102 habituated each fish for two consecutive days in the conditioning tank. We assessed for biases in
103 food motivation for the brine shrimp before starting baseline trials of the associative learning
104 assays. Each fish had a 14 day inter-assay testing interval to minimize influence of the tasks on
105 each other.

106 *Open Field Test*

107 We individually tested fish in an OFT in tank that was 31.75cm x 31.75cm x 10cm
108 containing 4L of water. Immediately after placing fish in the tank we video-recorded the
109 individual's behaviors for 5 minutes. We used Ethovision XT 17 (Noldus, Netherlands) to
110 quantify the amount of time that each individual spent frozen during the trial.

111 *Motivation Test*

112 This test was performed in the AD and LT models of the Zantiks semi-automated
113 behavioral units (Zantiks, Cambridge, UK). After 30 seconds for acclimation, the food reward
114 was administered 3 times at 30 second intervals. We quantified the time spent in a 9x12 cm
115 rectangle centered around the food administration tube. The time that was being measured started
116 immediately after the first brine shrimp administration until the end of the test to measure the
117 motivation of the fish for the food reward. We performed the test in both Zantik models but due
118 to the size and height of the tank in the larger LT unit, the food drifted outside the fish tracking
119 zone. Thus, we only used the data from the AD unit to assess motivation.

120

121 *Conditioned Place Preference*

122 We used a modified conditioned place preference protocol (Lau et al., 2006) in the
123 Zantiks LT unit. The testing tank (36 cm x 27 cm x 30 cm) was filled with 5.8 L of water. We
124 tested each fish in the CPP task for three weeks that consisted of 2 days of habituation, 1 day of
125 baseline testing, 11 days of conditioning, and 3 days of probe trials (Figure S1a). Probe trials
126 were done the day after a conditioning trial. To habituate each fish to the assay we placed the
127 fish in the tank for 10 minutes with no training stimulus lights. After habituation we determined
128 the baseline preference for the light stimuli (gray or checkered pattern) for each fish. Fish swam
129 freely for 10 minutes in the tank where one half was illuminated from the bottom with a gray
130 screen and the other half a checkered screen. We determined the conditioned and non-
131 conditioned stimuli as the stimuli where the fish spent the least and most amount of time,
132 respectively. During conditioning days we sequentially presented each stimulus for 5 minutes to
133 each fish. The non-conditioned stimulus was presented for the first five minutes followed by the
134 conditioned stimulus. One hundred microliters of brine shrimp or distilled water was
135 administered every minute during presentation of the conditioning and non-conditioning
136 stimulus, respectively. Food reward consisted of 11.4 grams of frozen brine shrimp in 30 mL of
137 distilled water. We fed control fish an equivalent amount of brine shrimp after each conditioning
138 trial. Probe trials were conducted after 3 days, 7 days, and 11 days after conditioning with a total
139 of 3 probe trials. Probe trial methods were the same as those used in the baseline preference step
140 where we quantified the time spent in each stimulus for each fish. The order of stimulus
141 presentation was consistent within a fish but random across fish for probe and baseline trials.

142

143 *2 Choice discrimination task*

144 We used a modified 2 choice discrimination task from an established protocol (Bilotta et
145 al., 2006). We used the AD model Zantiks unit (Zantiks, Cambridge, UK) with a 14cm x 20cm x
146 15cm tank filled with 2.5 L of water (Figure S1b). We habituated each fish for 20 minutes a day
147 for two consecutive days with white lights on in the wells as shown in Figure S1b. We tested each
148 fish every other day for a total of 10 testing days. Fish were fasted on non-testing days. In this
149 task the fish were presented with two 6.5 cm x 5.1 cm light stimuli (blue and yellow) from below
150 at one end of the tank. Prior studies show that with appetitive learning in zebrafish there is a bias
151 towards red compared to other colors such as blue and yellow (Spence & Smith, 2008, Kim et
152 al., 2017). For each fish, a color was randomly chosen at the start of testing to be the reinforced
153 stimulus where a food reward (brine shrimp) was administered at the other end of the tank when
154 the fish swam into the designated reinforced color. The food reward consisted of 5.7 grams of
155 frozen brine shrimp suspended in 30 mL of distilled water. Each trial began with an acclimation
156 period of two minutes with white lights in the two wells. After two minutes blue and yellow
157 lights were presented for 30 seconds. Swimming into the designated correct choice resulted in
158 the correct colored light staying on for an additional 30 seconds and we simultaneously
159 administered 25 μ l of the food reward. An incorrect choice resulted in all lights turning off for 30
160 seconds. This sequence ran for a total of 20 trials each day for each fish (i.e., one session consists
161 of 20 trials). The position of the yellow and blue lights (e.g., left or right) was randomly set for
162 each trial. There was an intertrial interval of 10 seconds. Control fish underwent the same
163 protocol with distilled water administered instead of brine shrimp and were fed brine shrimp
164 after each testing day. We compared the number of correct choices and the total number of
165 choices across sessions to assess learning.

166

167

168 *Statistical analysis*

169 We performed all statistical tests using R statistical (R 4.2.2 GUI 1.79 Big Sur ARM
170 build) software and Rstudio version 2022.12.0+353 (R Core Team, 2021). Due to fish mortality
171 during the experiment, the sample sizes for statistical analyses between the conditioned place
172 preference (bold control (n = 20), shy control (n = 20), bold treatment (n = 19), and shy treatment
173 (n = 19)) and 2 choice (bold control (n = 20), shy control (n = 17), bold treatment (n = 19), and
174 shy treatment (n = 20)) tasks differed. We conducted post-hoc tests using the emmeans (Lenth et
175 al., 2022) package and normality and assumptions were checked using base R. The lme4 package
176 (Bates et al., 2022) was used to test negative binomial linear mixed effect models. We obtained
177 simple statistics for all measures using the psych package (Revelle, 2022) (Table 1). Sex was
178 included in all models but was not significant and therefore removed. Normality and assumptions
179 were checked in R.

180

181 *Open Field Test and Motivation*

182 We tested for differences between the bold and shy groups in the OFT and motivation
183 test using a Welch two-sample t-test. This test was used due to unequal variances between bold
184 and shy groups. We compared the duration of time frozen in the OFT between the bold and shy
185 personality types. To investigate difference in food motivation, we compared the duration of
186 time spent around the food administration tube between the bold and shy personality types.

187

188 *Conditioned Place Preference*

189 We modeled the duration of time spent in the conditioned stimulus for the last half (5
190 minutes) of the baseline and probe trials to test for a change in preference for the conditioned
191 stimulus across the task within the different groups. We did not include the first half (5 minutes)
192 in the analysis as that was time to allow fish to recover from handling. We performed a repeated
193 measures ANOVA to investigate the effects of treatment, personality type, and conditioning day
194 on the time spent in the conditioned stimulus with a linear mixed effects model with individual
195 as the random effect. We included all interactions in the model and used type II sums of squares.
196 We used Tukey post-hoc tests to evaluate differences in the response variables across trials for
197 each group and within trials between groups.

198

199 *2 Choice Discrimination Task*

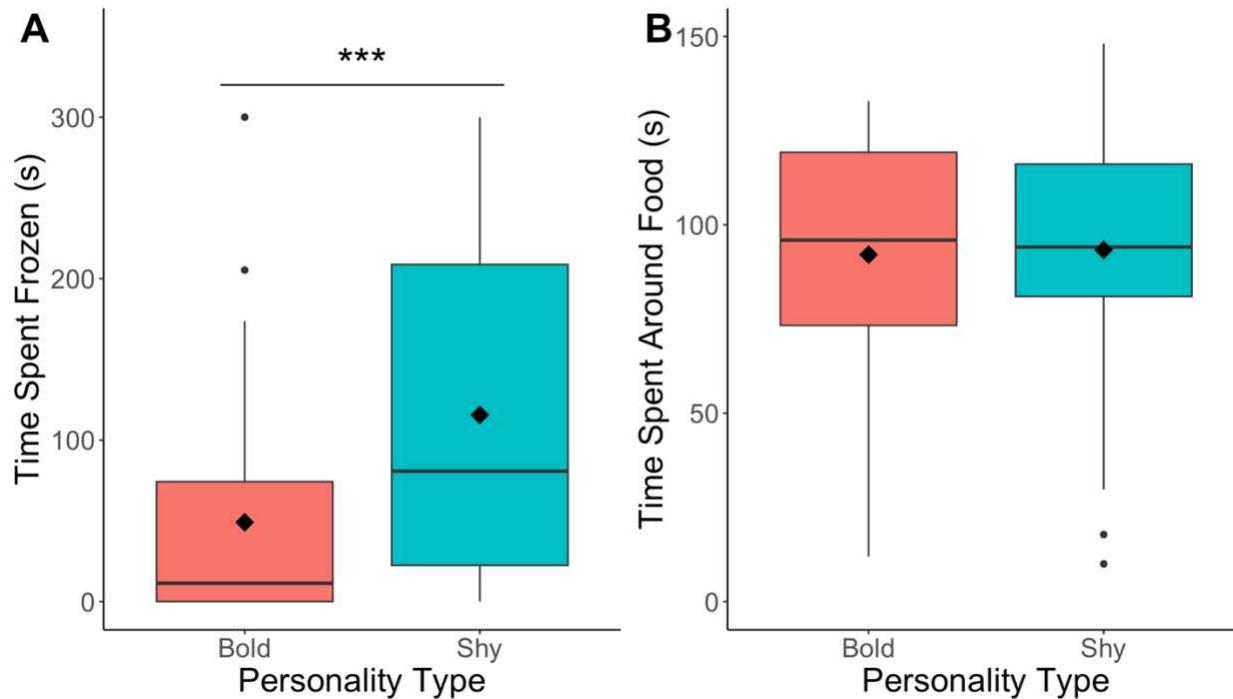
200 We modeled the number of correct choices over the conditioning days to examine
201 changes in correct choices over time within groups. We performed a negative binomial mixed
202 effect regression on the number of correct choices with treatment, personality type and session as
203 the fixed effects and ID as the random effect. Simple slopes were obtained to test for increases in
204 correct choices within each group using the interactions package in R and plotted using the same
205 package. Additionally, we performed a negative binomial mixed effect regression on the total
206 number of choices with treatment, personality type and session as the fixed effects and ID as the
207 random effect. We also obtained simple slopes for this model.

208

209 **Results**

210 *Shy fish freeze more but had equal motivation to eat*

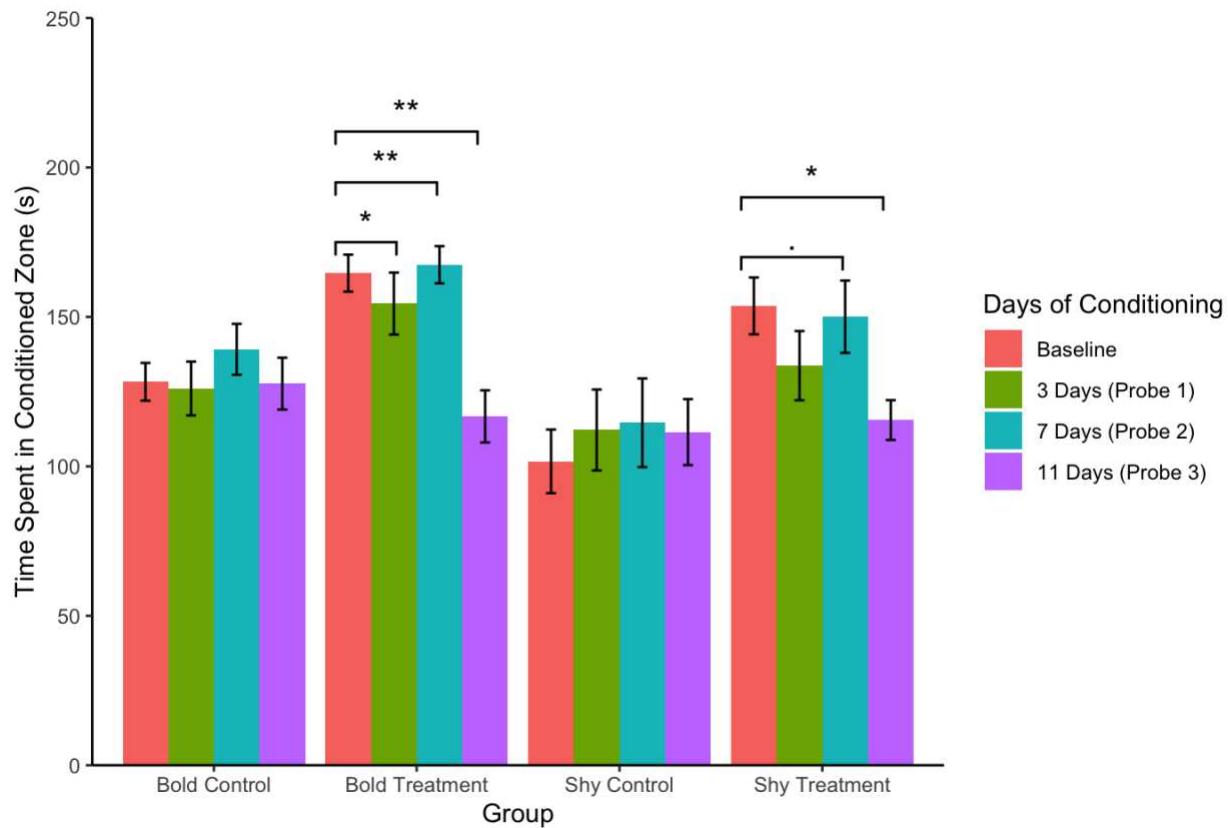
211 There was a significant effect of personality type on freezing time in the open field test
212 (Figure 2a). Shy fish spent significantly more time frozen than bold fish ($t = -3.55$, $df = 90$, $p =$
213 6.4×10^{-4}). There were no significant differences between personality types ($t = -0.19$, $df = 82$, $p =$
214 .85) in the amount of time spent around the food in the motivation task (Figure 2b).



215
216 **Figure 2.** A. Boxplot of time spent frozen in the open field test and B. boxplot of time spent around the food in the
217 motivation task. Bold fish are in red and shy fish are in teal. The diamond indicates the mean and the line is at the
218 median.* $p < .05$, ** $p < .01$ *** $p < .001$

219
220 *Bold Fish Change Their Behavior Before Shy Fish*
221 Treatment fish increased time spent in the conditioned stimulus in the CPP task, with
222 bold fish increasing time spent in the conditioned stimulus earlier in the task than shy fish
223 (Figure 3). In the full model (Table S1) the interaction effect between treatment and probe trial
224 was approaching significance ($F(3, 292) = 4.09$, $p = .09$). A Tukey post hoc test (Table S2)
225 revealed that there were no significant differences in the duration of time in the conditioned

226 stimulus between trials for the control groups for either personality type ($p > .05$). In the bold
227 treatment group there was a significant difference between baseline and probe 1 (i.e., after 3 days
228 of conditioning; $t = -2.64$, $df = 296$, $p = .03$), probe 2 (i.e., after 7 days of conditioning; $t = -3.55$,
229 $df = 296$, $p = 2.8*10^{-3}$), and probe 3 (i.e., after 11 days of conditioning; $t = -3.35$, $df = 296$, $p =$
230 $4.7*10^{-3}$). In the shy treatment group there was no significant difference in time spent in the
231 conditioned stimulus between baseline and probe 1 ($t = -1.27$, $df = 296$, $p = .45$) but there was a
232 trend for a difference between baseline and probe 2 ($t = -2.42$, $df = 296$, $p = .07$) and at probe 3
233 shy treatment group spent significantly more time in the CS compared to baseline ($t = -2.67$, $df =$
234 296 , $p = .04$). No significant differences in duration of time in the CS between probe 1, 2, or 3
235 were detected in any of the groups ($p > .05$). There were no differences in time spent in the
236 conditioned zone at any of the time points between personality types ($p > .05$). Additionally,
237 there was no significant correlation between learning speed (change in CS time from baseline
238 after 3 days of conditioning) and final time spent in the conditioned stimulus in the CPP task for
239 the bold fish ($\rho = .19$, $p = .44$) or for the shy fish ($\rho = .22$, $p = .35$).



240

241 **Fig. 3** Time spent in the conditioned zone by group and day of conditioning in the CPP. Pink bars are at baseline,
242 green bars are after 3 days of conditioning (Probe 1), blue bars are after 7 days (Probe 2) and purple bars are after 11
243 days of conditioning (Probe 3). Error bars indicate standard error. $p < .1$, $*p < .05$, $**p < .01$.

244

245 *No Evidence of Learning in 2 Choice Discrimination Task with Correct Choices*

246 In the 2 choice discrimination task there was no significant difference in number of
247 correct choices between control and treatment fish (Table S3). There was only a significant main
248 effect of personality type such that bold fish made more correct choice compared to shy fish ($b =$
249 $-.49$, $t = -2.84$, $p = .01$) and a significant interaction between personality type and session ($b =$
250 0.03 , $t = 2.601$, $p = .01$). Testing for the simple slopes (Table S4, Figure S2), both shy control (m
251 $= 0.03$, $t = 3.15$, $p = 2.2*10^{-5}$) and shy treatment ($m = 0.04$, $t = 4.24$, $p = 4.4*10^{-6}$) groups had a

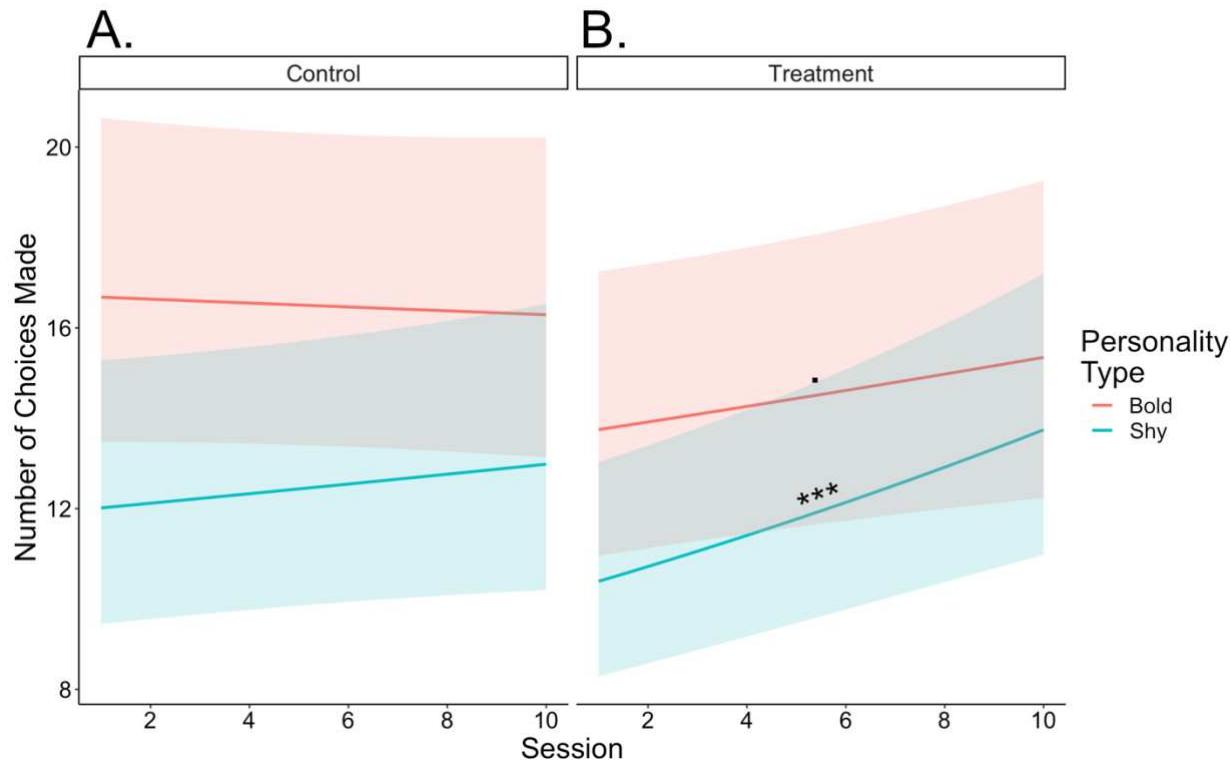
252 significant positive slope while bold control ($m = 0$, $t = -0.26$, $p = .79$) and bold treatment ($m =$
253 0.01 , $t = 0.98$, $p = .33$) have no significant relationship.

254

255 *Difference across Treatment and Control Only in Total Number of Choices in 2 Choice*

256 *Discrimination Task*

257 For the total number of choices, there was a significant difference between control and
258 treatment fish (Figure 4, Table S5). There was a main effect of personality type on total number
259 of choices ($b = -.34$, $t = -2.07$, $p = .04$) where bold fish had higher total number of choices than
260 shy. The interaction between session and treatment is approaching significance ($b = 0.15$, $t =$
261 1.69 , $p = .09$). Testing for the simple slopes, shy control ($m = 0.01$, $t = 1.22$, $p = .22$), and bold
262 control ($m = 0$, $t = -0.44$, $p = .66$) did not have a significant relationship (Figure 4a). Only shy
263 treatment ($m = 0.03$, $t = 4.59$, $p = .4.4*10-6$) had a significant positive slope (Figure 4b, Table
264 S6). In contrast, bold treatment had a slope approaching significance ($m = 0.01$, $t = 1.90$, $p = .06$)
265 (Figure 4b).



266 **Figure 4.** Regression lines of the number of choices made by personality type and treatment. 4A. shows the
267 regression lines for control fish and 4B. shows regression lines for treatment fish. The bold group is in red and the
268 shy group is in blue. Shaded regions indicate a 95% confidence interval. The simple slopes significance is indicated.
269 $p < .1$, $*p < .05$, $**p < .01$, $***p < .001$.

270 **Discussion**

271 Variation in learning performance can be due to complex interactions between intrinsic
272 (e.g., personality type) and extrinsic factors (e.g. learning task) (Sih & Guidice, 2012). We
273 investigated the effects of personality type and learning task by testing zebrafish of differing
274 personalities across two different associative learning assays. Overall, we found that learning
275 performance in one of the tasks was influenced by an animal's personality type.

276 Bold fish increased time spent in the conditioned stimulus earlier than shy fish in the
277 conditioned place preference task, which suggests that bold fish learned faster in this task. The
278 bold fish showed significant increases in time spent in the conditioned stimulus after just 3

279 conditioning days whereas it took shy fish an additional 8 days of conditioning to show a
280 significant change from baseline (Figure 3). These results are consistent with other studies
281 demonstrating that individuals with bold personality types learn faster than shy individuals
282 (Mazza et al., 2018, Guenther et al., 2014, Dugatkin & Alfieri, 2002, DePasquale et al., 2014,
283 Bensky et al., 2017, Daniel & Bhat, 2020, Kareklaas, Elwood & Holland, 2017). Differences in
284 learning speed between personality types in this task may be due to differences in behavior such
285 as stress reactivity, exploration, and neophobia (Sih & Guidice, 2012, Sommer-Trembo & Plath,
286 2018). Our observed differences in learning speeds between personality types cannot be
287 explained by differences in motivation for the food reward (Figure 2). Interestingly, there were
288 no differences in the amount of time spent in the conditioned stimulus between the personality
289 types after 11 days of conditioning, suggesting that individuals approach an asymptotic level of
290 performance. This suggests that both personality types are capable of changing their behavior
291 (e.g. learn) to similar extents and therefore differences in cognitive ability between personality
292 types is an unlikely explanation for differences in learning speed.

293 When testing the same fish in the 2 choice discrimination assay, there was no significant
294 difference in the number of correct choices between treatment and control groups, which
295 suggests the fish did not learn in this task (Figure 4). However, there were differences across
296 personality types in which both shy treatment and control increased their number of correct
297 choices while the bold groups did not. The positive slope for the shy groups is likely due to an
298 overall increase in total choices with repeated exposure. When looking at the total number of
299 choices made over sessions, the control groups do not change over time while the treatment
300 groups increased the total number of choices made over sessions. This suggests that the treated
301 fish did not learn the color association but may instead have learned to go into the wells. Animals

302 can attend to several cues in discrimination learning and sometimes attend to unintentional or
303 general cues (Mackintosh, N., J., 1965). We also cannot rule out that rewarding the fish in a
304 different location than the stimuli may have decreased the strength of pairing between action and
305 reward (Murphy & Miller, 1958). While in the 2 choice discrimination task fish did not learn the
306 color association, the bold fish made more choices than shy fish in the first session. This is likely
307 due to decreased neophobia and increased exploration in the bold fish as demonstrated in the
308 open-field test (Sih et al., 2004, Wong et al., 2012).

309 Differences in neophobia (e.g. latency to approach novel objects) classically distinguish
310 bold and shy personality types (Carter et al., 2012, Sih et al., 2004, Wilson et al., 1994). In the
311 current study one potential explanation for bold fish learning quicker in the conditioned place
312 preference and making more initial choices in the 2 choice discrimination task relative to shy fish
313 are differences in neophobia between the personality types. The shy fish could have found the
314 colored lights in the 2 choice discrimination task initially aversive and increased their choices as
315 they habituated to the novel stimuli. Shy individuals tend to have increased neophobia and
316 habituate slower, which would result in the shy fish taking longer to make active choices (Carter
317 et al., 2012). The two days of habituation in the 2 choice discrimination task only allowed the
318 fish to experience the tank and lighted wells but at start of conditioning they were naïve to the
319 color of the lights and the changing stimulus. A similar effect was seen in *Gallus gallus* where
320 individuals that were less exploratory (i.e., shy) habituated slower to a loud sound than those that
321 were more exploratory (Dissegna et al., 2022). In terms of shy fish learning slower in the CPP
322 task, neophobia may also explain this due to shy fish being more stressed than the bold fish even
323 after habituation and so learned the positive association slower. Mollies (*Poecilia mexicana*) that
324 were desensitized to the lights and sounds used in the task showed no differences in learning

325 between bold and shy fish (Sommer-Trembo & Plath, 2018). Increasing familiarity with the task
326 environment and stimuli could explain why shy fish were slower to increase their preference for
327 the conditioned stimulus but ultimately reached a level of performance similar to bold fish after
328 11 days of conditioning. Bold individuals tend to make associations faster likely because they are
329 less neophobic and in a simple conditioned place preference task, this leads to them learning
330 faster but does not change the plateau (Dugatkin & Alfieri, 2002, DePasquale et al., 2014, Daniel
331 & Bhat, 2020).

332 The positive relationship between learning speed and bold personality type is not
333 consistent across all studies (Ferron et al., 2015, Lermite, Peneaux & Griffin, 2016). Potential
334 explanations are that the relationship between speed of learning and personality can depend on
335 aspects of the task such as learning stimulus valence or task complexity. Shy zebrafish trained in
336 a contextual fear learning paradigm showed faster learning than bold zebrafish (Baker et al.,
337 2019). As shy zebrafish have a faster glucocorticoid response to a novelty stressor than bold fish,
338 this may facilitate quicker learning of aversive stimuli (Wong et al., 2019, Rau et al., 2005,
339 Riggenbach et al., 2019) but inhibit learning of appetitive stimuli seen in current study. For task
340 complexity, a study looking at learning accuracy found that aggressive spiders (e.g. bold
341 personality type) were more accurate in a simple task but not in a more complex task (Chang et
342 al., 2018). One area to explore would be whether the same trend holds in a more complex
343 classical conditioning task. In a more complex task, bold fish may make incorrect associations
344 and not learn as quickly as shy fish.

345 Overall, we found support for differences in how bold and shy individuals interact with
346 two different learning tasks. These differences in performance could be explained by varying
347 neophobia between bold and shy individuals. In a task requiring an active behavioral response (2

348 choice discrimination task), there were differences in initial number of choices between
349 personality types, suggesting that the personality types naively interacted with the stimulus
350 differently. In the conditioned place preference task the bold fish learned faster than the shy fish,
351 potentially due to the shy fish habituating slower. Additionally, there were no differences in
352 overall change in magnitude of time spent in the conditioned stimulus between bold and shy fish.
353 We encourage future studies to test the performance of bold and shy individuals across different
354 tasks to compare performance both within and across tasks. Future work should also consider
355 explicitly measuring how individuals interact with the task environment, perhaps measuring
356 neophobia and motivation for the task.

357

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