1   Title: Learning for angling: an advanced learning capability for avoidance of angling gear in red

2   sea bream juveniles

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8

9   Abstract

10   Angling has been the cause of mortality for fish since ancient. The avoidance learning for angling

11   gear could be considered as a survival strategy against the mortality by angling. Whereas some

12   studies indicated the possibility of avoidance learning for angling gear, most studies investigated

13   the avoidance learning by using groups of fish, in which it is difficult to reveal the process and

14   mechanisms of the learning. The present study elucidated the avoidance learning for angling gear

15   by experiment of single fish in a tank using red sea bream *Pagrus major* juveniles. Individuals

16   with only once or twice of experience for angling avoided angling gear while showing the feeding

17   motivation for pellets, representing avoidance learning for the angling gear. Most of the

18   experienced individuals avoided the krill attached with a fishing line, but not krill and pellets near

19   the angling gear. Feeding rate for prey on a fishing line at two month after the angling trial

20   demonstrated that approximately half of fish kept the memory for angling gear. A series of

21   experiment for angling gear elucidated that red sea bream juveniles are equipped with

22   considerable learning capability for angling gear, suggesting a cognitive evolution for angling.

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24   Keywords: angling, fishing, cognitive ecology, preparedness, learning

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26 1. Introduction

27 Angling has been conducted as a major fishing method since ancient [1, 2]. Recent study indicated

28 the evidence that hooks for angling had been made at the 35,000–30,000 years before present [3].

29 Also, angling is one of popular leisure around the world. A study in Japan showed that one in

30 eleven people enjoy angling as a leisure [4]. It is predicted that angling continues to be central in

31 fishery societies and economies around the world.

32   On the other hand, the hooking and capturing during angling process is a matter of life

33 and death for fish. Some studies showed the evidence that angling leads to the mortality of fish

34 [5-7]. Even if an angling does not lead to death for fish, the angled fish would suffer from the

35 physiological damage, behavioral change and stress by the angling [8-10]. For example, large

36 mouth bass changed the nest site for parental care by multiple times of angled experiences [11].

37 Line fishing would accelerate the natural selection for fish which is targeted by fishing [12], and

38 thus they should equip the capability to avoid angling gear efficiently.

39   The avoidance learning for angling gear is considered as a survival strategy against the

40 angling, because fish should acquire the information of angling gear as a dangerous subject in

41 fishing ground where they frequently encounter angling gears. Past studies suggested that fish

42 can avoid the angling gear through the experience being angled [13-16]. Some studies showed

43 that the angling rate in a natural condition and fish pond is declined over time with angling [17-

44 20]. However, most of the past studies had investigated the avoidance learning for angling by

45 experiment using groups of fish in a large scale, e.g., ponds or large experimental tanks. There

46 are some problems to elucidate the learning of angling gear in such a scale of experiment. For

47 example, the feeding motivation should be confirmed for an individual avoiding the angling gear,

48 because the motivation may be lowered by the stress from angling experience for not only angling

49 gear but also prey itself [21,22]. Also, aversive experience often enhances the alertness of fish

50 [23]. If an individual to be angled enhances the alertness, the fish would be less vulnerable to

51 angling than naive fish, because bold fish often feed more aggressively than fish being cautious

52 [15,24]. Conversely, the competition with conspecific mates often prompts their feeding

53 motivations within a shoal [25,26]. Thus, fish may feed the angling gear in a group even though

54 they had learned the angling gear as a dangerous subject.

55   In particular, it is unclear for the performance and mechanism of avoidance learning for

56 angling gear, because it is difficult to elucidate these factors by experiments using groups of fish.

57 Animals should recognize appropriately and quickly essential information to survive, and they

58 often equip the cognitive ability fitted to the ecology [27, 28]. Learning performance for angling

59 should be evaluated in consideration of learning process and retention in individual basis, i.e.,

60 how many times of angling experience are required to learn the avoidance of angling gear, and

61 also how long it is retained on the memory for angling. Also, a pattern of an associative learning

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62   often depends on environmental factor they originate [29-32]. To understand the mechanism of

63   learning for angling, it is important how fish learn the angling gear as an aversive object.

64   The present study investigated the avoidance learning for angling gear by using red sea

65   bream *Pagrus major* juveniles. The species is one of the most important fishes for fisheries in

66   Japan, and hatchery-reared fish are available from commercial fish farms. The experiment was

67   conducted by a singular fish in experimental tanks to eliminate the effect of group. First, the

68   learning process and the retention of learned information for short period were investigated by

69   presentations of angling gear on consecutive 28 days. Second, we investigated what is the key of

70   avoidance learning by presenting angling gear separately. Third, we compared the angling rate

71   between experienced and control fish, which had never been angled, by blind test. Thereby, we

72   evaluated the effect of the learning for the avoidance of angling gear. Finally, the long term

73   retention of learned information was tested for each individual.

74

75   2.   Material and methods

76   (a) Materials

77   Twenty two hatchery reared red sea bream juveniles were used for experiment (details in electrical

78   supplemental material). Eleven blue polypropylene containers (width 45 × length 66 × depth

79   33cm, depth 25 cm) were used for the experimental tank (details in electrical supplemental

80   material). A fish was introduced in each tank, and each fish was acclimatized over three days with

81   feeding cycle in the morning (7:00-10:00) and evening (14:00-17:00) everyday. The experiment

82   was initiated when the fish foraged pellets within 30 s after feeding in the morning.

83

84   (b) Experiment 1: the process of learning for angling gear

85   An experimental trial consisted of a feeding motivation test and a presentation trial. The feeding

86   motivation was confirmed by providing pellets (three to four pellets), and when the fish ate the

87   pellets within 30 s, a presentation trial was immediately conducted. By confirming the feeding of

88   pellets before presenting angling gear for each trial, we evaluated whether the fish would avoid

89   the angling gear while having the motivation of feeding.

90   Individuals were divided into either of angling treatment and control (11 fish was used for

91   each treatment). Angling treatment fish was presented with the angling gear (details in electrical

92   supplemental information), and the feeding behavior for a krill attached with the angling gear was

93   observed for up to a maximum of 60 s. When fish took a krill with hook into the mouth, the fish

94   was quickly captured out of the water through the angling leader, i.e., the fish was angled. The

95   angled fish was kept in air for at least 30 sec, and then returned to the experiment tank after

96   removing the hook with the snout. When it was difficult to remove the hook from mouth, the

97   leader was cut; i.e., the hook was remained in mouth of the fish, but it did not affected feeding

4

98   motivation of fish. If fish did not feed a krill for 60 s, the presentation trial was ended as "no
99   angling". Also, biting times, i.e., the number of time that fish pecked the krill without intake by
100   mouth, were counted during the observation period. When the fish took the krill away without
101   being hooked during the observation, another krill was attached with the angling gear. Control
102   fish was provided with a gear of which hook end was cut by pliers; thus, they could feed a krill
103   attached with the gear without being hooked. An experiment trial for each treatment was
104   conducted during daytime before noon (7:00-11:00). To reduce the variability of hunger level for
105   each individual, sufficient pellets were fed for both angling and control fish every afternoon
106   (15:00-18:00). The experimental trial was repeated for consecutive 28 days. The experiment was
107   replicated for two cycles (1st cycle: angling treatment, n=6, control, n=5; 2nd cycle, angling
108   treatment, n=5, control n=6).

109   We analyzed the effect of angling treatment for feeding behavior ("fed (including biting
110   without hooking)" or "not") by generalized linear mixed model (GLMM) with the "lme4" package
111   [33] for R statistical software. The error distributions of the response variables were fitted to the
112   binomial distribution, using restricted maximum likelihood parameter estimation. The fixed
113   factors were "treatment (angling or control)", "trial (1-28 trials)", and "interaction (treatment ×
114   trial)", whereas "individual" was treated as a random factor because the feeding behavior of each
115   individual was repeatedly measured during 28 trials. To evaluate the behavioral change of angling
116   treatment, the angling and feeding of only angling treatment fish was fitted to GLMMs (variable
117   factor: angling: "angled" or "not", feeding behavior: "fed" or "not", fixed factor for each variable
118   factor: "trial", random factor: "individual", error distribution: binomial). Biting time was also
119   fitted to GLMM with the error distribution of Poisson (fixed factor: "trial"). To evaluate the
120   feeding behavior while avoiding angling, angling rate focused on only the fish which showed the
121   feeding behavior for angling gear was also fitted to GLMM (fixed factor: "trial", error
122   distribution: binomial). We used the Wald test to evaluate the effect of the fixed factors for each
123   model.

124   The learning performance was investigated for each individual in the angling treatment.
125   When a fish avoided the angling even though it fed on pellets in the motivation test, the fish was
126   considered as leaned. The trials to be required until the learning was investigated for each
127   individual. After the learning, the retention of the learned information was evaluated by days to
128   be re-angled as a short duration memory.

129

130   (c) Experiment 2: the mechanism of learning for angling gear
131   Each individual was tested to investigate what of the angling gear was avoided by the learned fish
132   after one hour of the Experiment 1. In the test, both of angling and control treatment fish were
133   provided with following angling gears: "pellets and angling gear", "krill with the line", and "krill"

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134   (electronic supplemental material, figure 2a). The "pellets and angling gear" without krill was

135   presented to evaluate the decrease of the feeding motivation of fish by providing the angling gear.

136   In the "krill with the line" test, an angling gear was presented with a krill attached on a line without

137   a hook: i.e., the same gear as one of control treatment. The "krill" test was the presentation of a

138   krill without angling gear to confirm the avoidance for prey itself. The feeding motivation for

139   pellets was confirmed just before each test. These presentation tests were conducted sequentially

140   for each individual to reduce the number of fish used in the experiment. The order of presentation

141   test was fixed to decrease the effect of previous test. Each presentation test lasted for 60 s or until

142   fish foraged the pellets or krill. The feeding rate for each presentation was compared between

143   treatments and among presentation tests in each treatment by fisher's exact test. The latency to

144   feed was measured for each test, and compared between treatments by Student's t-test .

145

146   (d) Experiment 3: the effect of learning for angling gear

147   At one hour after the end of the Experiment 2, the angling test was blindly conducted by third

148   persons who were not informed about the experiment information. The blind tests were conducted

149   by eight participants in total. In the test, the angling gear attached with a hook and krill was

150   presented for 60 sec in the same manner as presentation trial of angling treatment in experiment

151   1. The feeding behavior was observed during the test, and then the latency until angling was

152   measured if the fish was hooked. The angling rate was compared between angling and control

153   treatment by Fisher's exact test, and the odds ratio of angling for angling treatment was estimated

154   against control. The fish was then captured after the test, transferred into the beaker, and was

155   photographed to measure the body length using Image J software (Open Source, Public Domain,

156   NIH). We confirmed that there was no difference of body length between treatments (angling

157   treatment, 117.8 ± 8.8mm (mean$\pm$standard deviation); control, 118.8 ± 12.6 mm; Student's t-test,

158   $t_{1, 20}$=-0.19 p>0.84). Then, individuals of angling treatment were introduced into rearing tanks

159   (200L transparent circular tank) for each experimental cycle, and they were reared in the same

160   manner as the stock tank until Experiment 4.

161

162   (e) Experiment 4: the long term retention of learning for angling gear

163   At about two month (87-98 days) after the Experiment 3, the retention of learning for angling

164   gear was investigated for each individual of angling treatment fish (n=10, because a fish of 2nd

165   cycle died before the test). On the day before testing, a fish was introduced in an experimental

166   tank to be acclimatized for a night. After a night, each fish was tested by presentation of "pellets

167   and angling gear" and "krill with the line" in the same manner as in the Experiment 2; the feeding

168   motivation for pellets was confirmed before each presentation. The feeding rate was compared

169   between presentation tests by Fishes' exact test.

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170

171   Results

172   (a) Experiment 1: the process of learning for angling gear

173   All the fish, both angling treatment and control, accepted the presented pellets in the feeding

174   motivation test. Thus, lack of feeding with the presence of angling gear on a presentation trial

175   was considered as avoidance learning for angling gear, but not by loss of feeding motivation.

176   For the feeding behavior in both treatments, there were significant effects of treatment,

177   trial, and interaction (treatment × trial) in the GLMM analysis (all factors $P<0.001$; figure 1 &

178   electronic supplemental material, table S1a). For the angling treatment fish, there was a significant

179   effect of trial in each GLMM (feeding behavior $P<0.01$, angling $P<0.001$, biting times $P<0.05$,

180   electronic supplemental material, figure S2,3 & table S1b). Evaluating the angling of fish

181   representing feeding, there was a significant effect of trial for angling rate (table 1c, $P<0.001$;

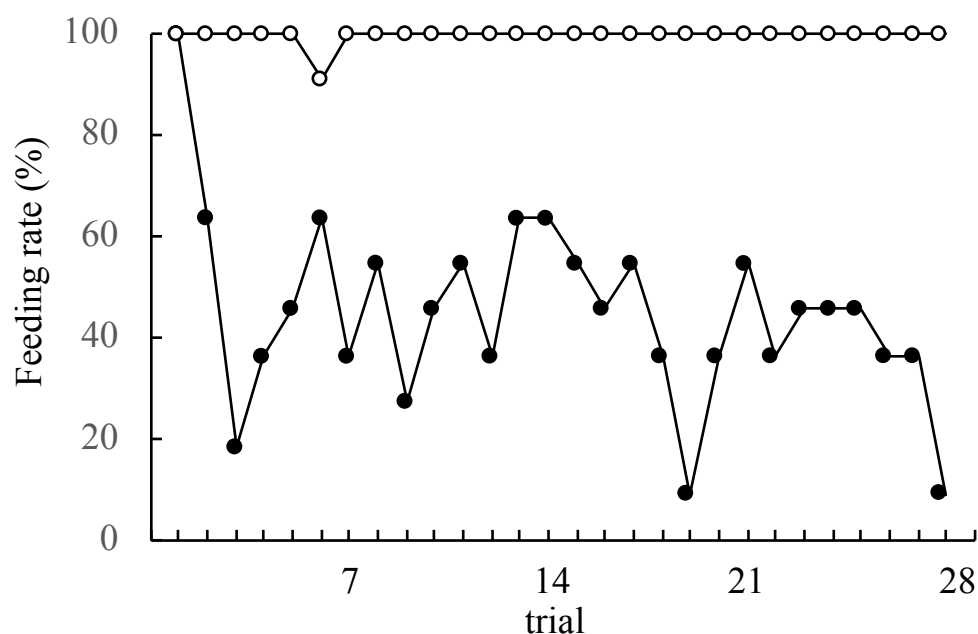182   electronic supplemental material, figure S4 & table S1c).

183

184

185   Fig1 The feeding rate of fish in the control (open circle) and angling (closed circle) treatment

186   during the 28 trials in Experiment 1

187   For individual analysis of angling treatment, all individuals avoided the angling gear

188   after one (n=5) or two angling trials (n=6); i.e., fish required 1.6 ± 0.5 (mean ± standard deviation)

189   trial to learn the avoidance of angling gear. Meanwhile, all fish was re-angled within 13 days after

190   the avoidance learning (electronic supplemental material, fig s5), and then 4.3 ± 3.3 days were

191   required until next angling. Times of angling during 28 days varied from three to eight among

192   individuals.

7

193

194   (b) Experiment 2: the mechanism of avoidance learning for angling gear

195   Angling treatment fish showed the feeding rate of 100 % (11/11 individuals) for "pellets and

196   angling gear", 9.1 % (1/11) for "krill with the line", and 100 % (11/11) for "krill", respectively

197   (figure 2b). The feeding rate of control fish was 100 % (11/11) for all the presentation test. The

198   feeding rate of "krill with the line" for angling treatment was significantly lowered than control

199   treatments (P<0.001), but not different from others (P=1.00). For angling treatment fish, the

200   feeding rate of "krill with the line" was significantly lower than other presentation (P<0.001).

201   There was no difference of feeding rate among presentation tests in control (P=1.00). The latency

202   to feed "krill with the line" of control was faster than the fish (11 sec) of angling treatment (one-

203   sample t-test, $t_{1, 10}$=-19.53, P<0.001; electronic supplemental material, fig s6). Meanwhile, there

204   was no difference of the feeding latency between treatments for "pellets and angling gear" and

205   "krill"("pellets and angling gear": $t_{1, 20}$=-1.33, P<0.20, "krill": $t_{1, 20}$=-1.04, P<0.31).

206   (a)
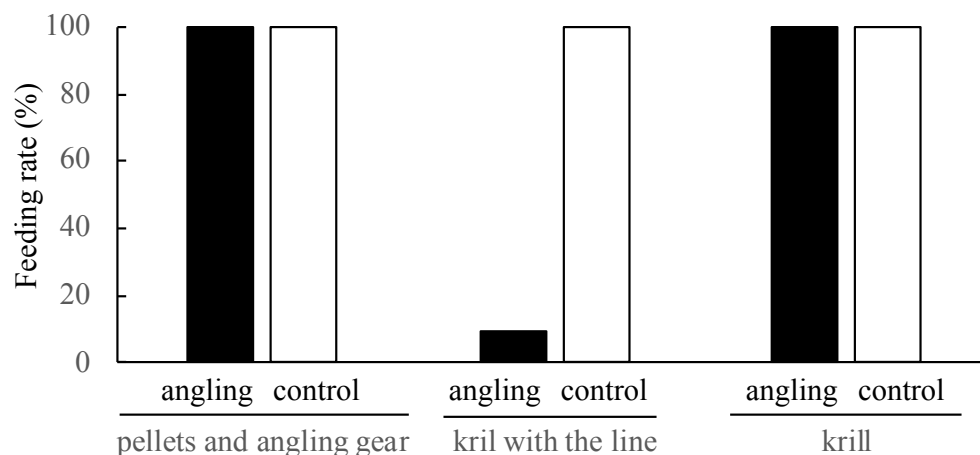

207

208   (b)

209
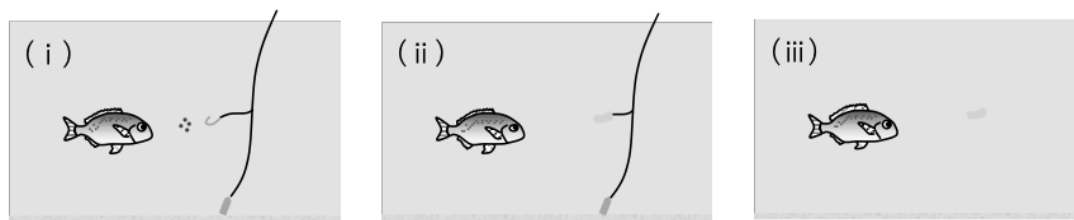
210   Fig2 (a) Schematic illustrations of test presentation in experiment 2: (i) pellets and angling gear,

211   (ii) krill with the line, and (iii) krill. (b) The feeding rate of fish in the angling (black column) and

212   control (white column) treatment when three types of gears or prey were presented in Experiment

213   2

214   (c) Experiment 3: the effect of learning for angling gear

215   The angling rate in blind test was 18.2% (2/11) for angling treatment and 81.82 % (9/11) for

8

216   control, respectively, and there was a significant difference of angling rate between the treatments
217   (P<0.01). The latency for angling was 18s and 52s for angling treatment, and 9.4 ± 10.2s for
218   control, respectively, and there was a significant difference between treatments (one-sample *t*-test
219   "control vs 18s"; $t_{1, 8}$=-2.37, P<0.05). The cause of not being captured in angling treatment was
220   either they did not feed at all (in two individuals) or showed only biting (in other seven
221   individuals). For control fish, two fish showed the biting for the angling gear but were not
222   captured. The odds ratio for angled fish in the angling treatment was 0.05 against control.
223
224   (d) Experiment 4: long term retention of learning for angling gear
225   The feeding rate was 100 % (10/10) with the presentation of "pellets and angling gear" and 50%
226   (5/10) with that of "krill with the line". The foraging rate of "krill with the line" was significantly
227   lower than "pellets and angling gear" on the two month later test (P<0.04).
228
229   Discussion
230   All individuals including both treatments of angling and control fed the krill attached with angling
231   gear and were angled on the first trial. Meanwhile, angling treatment fish decreased to feed from
232   the angling gear with the experience of angling, although they had always demonstrated the active
233   feeding on the pellets in the motivation tests prior to the angling trial. This result suggests that the
234   fish recognized the angling gear as an object to avoid feeding through the experience of angling;
235   i.e., avoidance learning for angling gear. Whereas past studies using groups of fish suggested the
236   avoidance learning for angling gears [13-16], these studies have not confirmed the feeding
237   motivation of fish during experiment. The present study verified the avoidance learning for
238   angling gear to elucidate the fact that fish desired prey at the presentation of angling gear.
239   The angling rate in the angling treatment decreased during 28 days, suggesting that
240   learning progressed gradually as the experience of angling. The major factor for the decrease of
241   angling would be the refusing to feed from the angling gear, because the feeding rate also decrease
242   with trials during angling treatment. Meanwhile, the feeding for angling gear was maintained to
243   a certain extent even after the learning progressed. Or rather, biting times, i.e., pecking krill
244   attached with the angling gear without being hooked, had increased during learning process. This
245   means that fish getting experience of angling might become not only to avoid angling gear, but
246   also to steel prey on the angling gear without being hooked. In fact, the angling rate regarding
247   only feeding fish decreased remarkably with trials. The present study indicates that red sea bream
248   juveniles improve the feeding behavior of angling gear while avoiding to be angled. Improvement
249   of feeding skill is found in fishes [34, 35]; e.g., the shooting accuracy in archer fish was improved
250   rapidly through the experience of target movement [36]. However, in our knowledge, present
251   study is the first to have elucidated the improvement of feeding for angling gear.

9

252   All individuals of angling treatment learned to avoid angling gear through only once or
253   twice experience of angling. Red sea bream juveniles were able to quickly learn the angling gear
254   as a dangerous object. Similarly, past studies suggested that the fish can learn angling gear by
255   only a few times of angling experience [14,15]. For example, Beukma [37] found in carp by an
256   experiment on artificial lake that many individuals became invulnerable to angling once being
257   angled; however, this study could not rule out the possibility of reduced feeding motivation.
258   Avoidance learning for angling gear may be a simple task for fish, at least for some species which
259   are subject to angling. Essential information in an ecology is often learned rapidly with only a
260   few experience in fishes [38, 39]. For example, predator information is learned by a single
261   conditioning [40]. Shanny, which form the nest in rock tide pool, can learn the location of shelter
262   with only one trial [41]. The remarkable cognitive ability for angling gear in red sea bream implies
263   that the learning for angling gear is important in their ecology.

264   Although angling treatment fish quickly learned to avoid angling gear, all individuals
265   were re-captured after the learning; 4.3 days on average were required to be re-angled. This means
266   that by no means fish can avoid completely the angling gear after the learning of angling. Fish
267   may be angled in a place where anglers are abundant, such as a major fishing ground or fishing
268   pond, even if the fish can quickly learn the angling gear as dangerous subject. Some studies have
269   shown the re-angling of fish that had previously experienced angling [42, 43]. For example,
270   Tsuboi & Morita [44] using white-spotted charr found that some of fish were angled repeatedly
271   during research period. The study also showed that the fish with a greater experience of being
272   angled were more angled; that is, there were individual variations of learning for angling. In the
273   present study, angling times varied among individuals of angling treatment, and some fish were
274   vulnerable to angling. In the following experiment of the present study, the vulnerability of
275   angling is related to the boldness of individuals (Takahashi, unpublished); i.e., bold fish were
276   angled more frequently than shy ones. The individual difference of learning should be
277   investigated in more detail. On the other hand in the present study, the fish is confined in a small
278   tank, and then they were obliged to be encountered with angling gear after the learning. Thus,
279   they might have fed the prey on the angling gear knowing that it was dangerous; the increased
280   biting times of angling gear during angling trials supports this speculation.

281   Although angling treatment fish was re-angled in a few days after the fish had once
282   learned angling (Experiment 1), the fish developed the cognition for angling by repeated
283   presentation of angling gear. At two months after the 28 consecutive angling presentations,
284   approximately a half of angling treatment fish avoided a krill attached with a line (experiment 4).
285   This suggests that the angling treatment fish, at least a part of them, retained the learning for
286   angling gear even if they had not encountered the angling gear for this period. Similar suggestions
287   of long term memory for angling are also found in experiment which was conducted in a large

288 pond [37]; the experiment suggests that fish kept the memory of being hooked for at least a year.

289 The present study has verified the long term memory of avoidance learning for angling gear by a

290 small scale experiment. Retention of learned information can be often related to the ecology of

291 animal. For example, food-storing birds have better spatial memory than birds that do not store

292 food [45]. In fish, the retention of information for predator is shaped by individual growth rate as

293 a behavioral tactics [46]. For memory retention of red sea bream, Kaneko [47] showed that most

294 fish were not able to retain the learned information of feeding area for 60 days. It might be

295 important for red sea bream to keep the information of angling gear for a long term, because they

296 would often encounter the angling gear in their lives after the learning for angling.

297 In Experiment 2, all of learned fish fed pellets under the presence of angling gear. It is

298 expected that the feeding motivation was not lowered by the vigilance against the presence of

299 angling gear. Also, all fish fed a krill without the line and hook. Some study found that fish avoid

300 prey containing aversive substance, such as toxic or unpalatable food [48,49]. However, the fish

301 in the present study did not recognize the krill itself as a prey which should be avoided. The result

302 indicates that the avoidance learning for the angling gear is regarded as distinct from food

303 aversion learning. Aversion learning for prey is often affected by combination between

304 conditioned stimulus and unconditioned stimulus; for a classical example of rat, the aversion for

305 prey can be conditioned with lithium chloride, but not with electrical shock [50]. The aversion

306 learning for angling in the present study would be difficult to be associated with prey itself in red

307 sea bream.

308 Meanwhile, almost all fish of the angling treatment avoided to feed a krill with a line;

309 the feeding latency of a feeding fish was longer than control feeding fish. This means that red sea

310 bream juveniles evaluated the risk of angling gear with the presence of fishing line attached with

311 a prey. Whereas past studies suggested that fish can avoid angling gears through a learning effect,

312 the mechanisms have not been clear [16]. The experiment elucidated that a prey attached with a

313 fishing line is the key factor of the learning for angling gear. The result in experiment 2 indicates

314 that they could feed a prey they had once eaten in angling process unless the prey is attached with

315 a fishing line. Angled fish are often returned to fishing ground, either being released intentionally

316 by angles or the fish being able to escape during the angling process. The fish after the release

317 must avoid a prey which lead to be angled, because repeated angling would increase the risk of

318 mortality [5-7]. However, while avoiding the dangerous prey, the fish need to take prey for their

319 lives. If fish learn to avoid the prey itself previously angled, such as food aversion learning, the

320 fish would lose a chance to get the prey even when the prey is safe without angling line. It would

321 be essential to take a prey discriminating correctly whether the prey is dangerous. The making

322 decision of feeding a prey, i.e., estimating a presence of angling line, must be useful for fish to

323 survive in a fishing ground.

11

324   In the blind test after the angling treatment (experiment 3), the angling rate of angling

325   treatment fish was remarkably lower than control fish; twenty times difference of the vulnerability

326   for angling between treatments. The experiment showed that the vulnerability for angling is

327   markedly improved by repeated angling experiences. There are some possibilities for the decline

328   of angling in large scale experiment. For example, the recapture rate of angled fish decreases if

329   fish leave the fishery ground [51]. Stålhammar [22] found that the feeding behavior for a prey of

330   pike was delayed by an angling experience. Meanwhile, angling treatment fish in the present

331   study were not able to go away from the angling gear in the experiment tank, and had the active

332   feeding motivation for prey just before the angling test. Thus, it is predicted that the angling

333   vulnerability was improved by the avoidance learning for angling gear. Re-angling of learned fish

334   should have occurred less in a natural fishing ground than in a limited space on the present study.

335   Furthermore, the learning efficiency is often enhanced by the social learning for various contents

336   [52,53]. Whereas it is not still clear for the social learning of angling gear, if the social learning

337   for angling gear is established in groups of fish, the invulnerability of angling would be prompt

338   in natural condition.

339   Animals often have a cognitive ability adapted to their ecology [27,28], and the adaptive

340   cognition is often prepared innately in them [29]. For example, tropical poeciliid in higher

341   predation pressure exhibit an innate behavioral character adapted with predatory environment

342   [54]. Also, hatchery-reared jack mackerel juveniles under rearing condition develops

343   ontogenetically the learning capability to fit with the habitat shift during life history in nature [55].

344   The red sea bream juveniles in the present study were hatchery-reared, and thus had never

345   encountered angling gear in their lives before the experiment. Nevertheless, they demonstrated

346   the advanced learning capability for angling gear. This suggests that the cognition for angling

347   gear is innately equipped in the red sea bream juveniles. Red sea bream at the adult stage is

348   targeted for fishery of pole-and-line fishing and recreational angling in Japan [56]. The juveniles

349   are released for stock enhancement around Japan to coastal zone and frequently captured by

350   recreational angling, but forced to release for regulation of angling size by fishery adjustment rule

351   in   each   prefecture   [57]   (see   also   Kanagawa   prefecture   Web   page,

352   http://www.pref.kanagawa.jp/index.html). It is predicted that they repeatedly encounter angling

353   gears during the life. The advanced cognition for angling would help them to survive after the

354   release.

355   In Experiment 2, the juveniles learned angling gear depending on a line attached with a

356   prey. It is considered that such learning would not be expected in their lives except for angling

357   process, because a prey with a line would not be present in the natural environment without

358   angling gear. The avoidance learning in red sea bream might be formed under a cognitive

359   mechanism specialized in angling. Angling has been a common fishing method since ancient

360   times [1-3], and then fish had been exposed to the risk of angling on their evolution. The cognition

361   for angling gear might be evolved in red sea bream under the selection for angling. Fishing has

362   the potential to induce evolutionary change in traits in fish populations [12, 58, 59]. For example,

363   faster-growing genotypes which may be more vulnerable to fishing depletion were more

364   frequently harvested than slow-growing fish [60]. In largemouth bass, recreational angling

365   induced evolutionary changes in various physiological and behavioral traits after only four

366   generations [61]. The adaptive cognition for angling gear in red sea bream might have occurred

367   through the natural selection by the angling. The present study suggests the cognitive evolution

368   for a fishing activity. Whereas the avoidance learning for angling gear is suggested for various

369   fish species, past studies are limited to the experiment in large scale [13-16]. These studies

370   focused on only the targeted species for angling, including the present study. It is unclear for the

371   learning capability of angling in non-targeted species. In future, comparative study between

372   targeted and non-targeted species would elucidate the cognitive evolution for angling in fishes.

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379   Author contribution

380   Experiment and writing paper was conducted by Kohji Takahashi, and the experiment was

381   directed by Reiji Masuda.

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387   Ethical notes

388   All experiments were performed according to the regulations on animal experimentation of Kyoto

389   University. A minimum number of fish was used to test the hypothesis. After the experiment, the

390   fish were donated to local aquarium.

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392   Conflict of interests. We have no competing interests.

393   Data accessibility. Data have been submitted as electronic supplementary material.

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