1   **Do juvenile Atlantic salmon (*Salmo salar*) use chemosensory cues to detect and avoid**

2   **risky habitats in the wild?**

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19 **Abstract**

20   We examined whether juvenile Atlantic salmon (*Salmo salar*) in the wild adjust their

21 behaviour in response to chemical cues of predator activity during a four-week period after

22 emergence from gravel nests. In each of seven 75 m^2 sites in Catamaran Brook, New

23 Brunswick, we established three contiguous sections differing in perceived predator activity

24 by releasing stream water in control sections, conspecific alarm cues in risky sections, and

25 nothing in buffer sections in both 2006 and 2007. As predicted, the density of YOY salmon

26 tended to decrease in alarm-cue sections, while increasing in control and buffer sections. After

27 the two week manipulation in 2006, we switched treatments so that buffer sections became

28 alarm-cue sections and alarm-cue sections became buffer sections for an additional two-week

29 period. After the switch, the number of YOY increased least in the new alarm-cue sections

30 and most in control and new buffer sections. In contrast to YOY, the density of age 1+ parr

31 was not affected by the experimental treatments. Our results suggest that YOY salmon can use

32 chemical alarm cues to assess the predator activity of habitats in the wild.

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Introduction

Habitat selection has a profound influence on a number of vital processes including population regulation, species interactions, the assembly of ecological communities, and the origin and maintenance of biodiversity (Morris 2003). Given that habitats differ in growth potential and/or risk of predation, animals should prefer the habitat that maximizes their fitness (Dill 1978). However, the best areas for foraging are often the most dangerous, forcing individuals to trade off energy gain against safety from predators when deciding where to feed (Lima and Dill 1990). In perhaps the most elegant demonstration of this trade-off, Abrahams and Dill (1989) titrated the extra energy required to induce guppies (*Poecilia reticulata*) to forage in a more dangerous patch.

Prey capable of reliably assessing predation risk at the scale of whole habitats should presumably be at a selective advantage. To assess the safety of habitats, animals in the wild rely on various sources of information, including the direct assessment of the presence or abundance of predators via visual, chemosensory, auditory, and/or tactile cues (Lima and Dill 1990; Kelley and Magurran 2003). For example, fathead minnows (*Pimephales promelas*) avoid areas labelled with the faeces of predatory northern pike (*Esox lucius*) (Brown et al. 1995), and Hawaiian roof rats (*Rattus rattus*) avoid the fecal odours of their predators, the mongoose (*Herpestes auropunctatus*) and red fox (*Vulpes vulpes*) (Burwash et al. 1998).

Animals also use chemosensory cues to assess the level of immediate predation risk

57   (Lima and Dill 1990). Chemosensory cues released from the skin of injured fish provide

58   reliable information about current and past predation events (Smith 1992). Many freshwater

59   fishes, including fathead minnows, avoid areas that have been recently labelled with

60   conspecific alarm cues under laboratory and natural conditions (Mathis and Smith 1992;

61   Chivers and Smith 1998; Brown 2003). Similarly, terrestrial invertebrates avoid areas

62   containing dead congeners (Grostal and Dicke 1999; Dukas 2001; Nilsson and Bengtsson

63   2004).

64   Because it is often difficult to obtain reliable information about predator abundance

65   or activity, animals can use indirect measures such as habitat features to assess predation risk

66   (Lima and Dill 1990; Verdolin 2006). Deer mice (*Peromyscus maniculatus*) and white-

67   throated sparrows (*Zonotrichia albicollis*) prefer feeding stations close to or containing

68   abundant cover (Schneider 1984; Anderson 1986). Similarly, juvenile lingcod (*Ophiodon*

69   *elongatus*) and winter flounder (*Pseudopleuronectes americanus*) prefer habitats with

70   structure (rock, shell, or seagrass) compared to bare sand habitats (Petrie and Ryer 2006;

71   Pappal et al. 2009).

72   In stream-dwelling salmonids, studies of habitat selection typically focus on physical

73   variables, such as current velocity and depth, which affect foraging profitability (Girard et al.

74   2004; Rosenfeld and Taylor 2009). Although juvenile salmon prefer sites with an abundance

75   of cover (Culp et al. 1996; Dolinsek et al. 2007; Venter et al. 2008), little is known about how

76   they actually assess predator abundance or activity when selecting habitats. In the short term,

77   juvenile salmonids exhibit antipredator behaviour when exposed to visual and/or chemical

78   cues indicating the presence or activity of predators under laboratory and field conditions

79   (Leduc et al. 2006; Blanchet et al. 2007; Kim et al. 2009). While the short-term benefits

80   associated with antipredator behaviour are clear, whether and how juvenile Atlantic salmon

81   (*Salmo salar*) in the wild use chemical information to assess the relative predator activity of

82   alternative habitats at larger temporal and spatial scales are unknown.

83   The objectives of this study were to examine (1) whether wild juvenile Atlantic

84   salmon use chemical information to avoid habitats that have a higher perceived predator

85   activity when settling in a new habitat, and (2) how they respond to changes in perceived

86   predator activity after settling in a habitat and establishing a territory. Juvenile Atlantic

87   salmon are ideal subjects for our study because they are relatively sedentary (Steingrímsson

88   and Grant 2003; Breau et al. 2007), which allows us to manipulate the perceived predator

89   activity of small sections of habitat while monitoring abundance during the four weeks after

90   emergence from redds.

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92   **Materials and methods**

93   **Study site and species**

94   We conducted this study in the lower reach of Catamaran Brook, located in

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95   Northumberland County, New Brunswick, Canada (46°52'42"N, 66°06'00"W) from 17 June

96   to 22 July 2006 and 21 June to 25 July 2007. Catamaran Brook is a nursery stream for a

97   naturally reproducing population of anadromous Atlantic salmon (Cunjak et al. 1990). Young-

98   of-the-year (YOY) Atlantic salmon emerge from gravel nests in mid-June at about 2.6 cm in

99   fork length (Randall 1982). Upon emergence, juvenile salmon disperse from redds (gravel

100   nests) and begin defending foraging territories, even at 2-3 cm in length (Keeley and Grant

101   1995).

102   We selected seven sites of relatively shallow depth (< 50 cm) and slow current (0.2 –

103   $0.5 \text{ m} \cdot \text{s}^{-1}$), which are the preferred habitats for YOY Atlantic salmon in Catamaran Brook

104   (Girard et al. 2004). Within each site (mean width $\pm SD = 8.18 \pm 1.60$ m), we manipulated the

105   perceived activity of predators in three 5 X 5 m sections as follows: a risky section received

106   conspecific alarm cues; a control section received stream water; and, a buffer section received

107   nothing (Fig. 1a). Sections of this size (25 m^2) were chosen as an appropriate scale for our

108   study because YOY salmon typically defend a territory of about 1 m^2 (Steingrímsson and

109   Grant 2008) and typically move less than 5 m during their growing season (Steingrímsson and

110   Grant 2003). Consequently, YOY salmon respond to changes in habitat quality at spatial

111   scales as small as 6 m^2 (Dolinsek et al. 2007; Venter et al. 2008). To ensure that chemical

112   alarm cues from the risky section had no effect on the control section (Blanchet et al. 2008),

113   we always assigned the control to the upstream quadrat of the site (Fig. 1a). Furthermore, we

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114   added a buffer section between the control and risky sections so that the buffer and risky

115   sections could be switched during late settlement (Fig. 1b) and fish emigrating from the risky

116   section in an upstream direction could settle in habitat other than the control section during

117   early settlement (Fig. 1a). Because there were no barriers or enclosures, each site was also

118   exposed to the ambient risk of predation from potential predators, such as common merganser

119   (*Mergus merganser*), belted kingfisher (*Megaceryle alcyon*), brook charr (*Salvelinus*

120   *fontinalis*), and Atlantic salmon (Scott and Crossman 1973; Dolinsek et al. 2007). To

121   minimize the potential cumulative effects of chemical alarm cues dispersing from upstream to

122   downstream (Dionne and Dodson 2002; Kim et al. 2009), sites were at least 30 m (range: 30 -

123   93 m) apart.

124   To ensure that the three sections within a site were similar in habitat characteristics,

125   we measured the depth and current velocity at 40% of the water column depth, using a Marsh-

126   McBirney meter (Model 201D, Fredericton, MD, U.S.A.) at five locations along a transect

127   across each section. The depth (mean $\pm SD = 45.75 \pm 13.43$ cm) and current velocity ($0.40 \pm$

128   0.27 m $\cdot$ s^{-1}), the two key variables used in habitat selection (Girard et al. 2004), did not differ

129   significantly among treatments or sites (*P*-values all > 0.18).

130   For the purpose of this study, we defined the early settlement period for YOY Atlantic

131   salmon as June 15 – July 7, the time when salmon typically emerge and disperse from their

132   redds in Catamaran Brook (Randall 1982; Johnston 1997). During this period, YOY salmon

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133   select a suitable habitat and begin defending a territory (Armstrong and Nislow 2006). We

134   defined the late settlement period as July 8 – 21. During this late settlement period, fewer

135   YOY Atlantic salmon emerge from redds and more fish have settled into feeding territories

136   for the summer than during the early settlement period (Steingrímsson and Grant 2003).

137   **Collection of alarm cue**

138   Hatchery-reared Atlantic salmon parr (1+) from the Rocky Brook population of the

139   Miramichi watershed (2006, $n = 199$, mean ± SD, standard length = 8.57 ± 0.74 cm; 2007, $n =$

140   163, fork length = 10.64 ± 0.72 cm) were obtained from the Miramichi Salmon Conservation

141   Centre, South Esk, New Brunswick for use as skin donors. Skin donors were killed with a

142   single blow on the head in accordance with Concordia Animal Care Committee Protocol AC-

143   2005-BROW. Skin fillets from both sides were removed and immediately placed into an ice-

144   chilled container filled with stream water. Skin fillets were homogenized and diluted with

145   stream water. The resulting concentration (0.09 cm^2 of skin · ml^{-1}) of cue from this population

146   elicits a consistent anti-predator response in juvenile Atlantic salmon in Catamaran Brook

147   under natural conditions (Leduc et al. 2006; Leduc et al. 2007; Kim et al. 2009). Furthermore,

148   alarm cues are equally effective at eliciting anti-predator behaviour in field and laboratory

149   settings for a variety of prey fishes (Brown and Godin 1999; Wisenden et al. 2004; Wisenden

150   et al. 2010). The alarm cue was frozen in 50 ml aliquots at -20 °C until needed, whereas

151   stream water was obtained at the site. The frozen solutions were thawed 60 min prior to use.

152   For this study, 630 and 315 50-ml aliquots of alarm cue were used in 2006 and 2007,

153   sufficient for 30 and 15 days of the experiment, respectively. The remainder of the alarm cue

154   was used in other cognate studies.

155   **Experimental protocol**

156   *Early settlement*

157   We manipulated the long-term perceived predator activity in each site by releasing

158   either the alarm cue or stream water twice a day for 17 and 15 days in 2006 and 2007,

159   respectively (Fig. 1a). To coincide with the peak activity of YOY Atlantic salmon (Breau et al.

160   2007), we released either 50 or 100 ml of the chemical stimuli (alarm cue or stream water) at

161   1100 h (1030-1130) and 1700 h (1630-1730) for a total volume of 150 ml per day per section;

162   the amount released in the morning was alternated each day. Studies simulating a single

163   predation event typically use about 20 ml from 1.8 cm^2 of skin (Leduc et al. 2006; Kim et al.

164   2009), roughly equivalent to the size of a wound left by a predator (Smith and Lemly 1986).

165   Hence, the release of 150 ml per section per day would be equivalent to about eight predation

166   events, equivalent to the skin of one parr, being released in the 25 m^2 section per day. The

167   background predation rate in Catamaran Brook is estimated to be lower than 13 predation

168   events per section over eight weeks if fish density is 1 YOY $\cdot$ m^{-2} (Girard et al. 2004). We used

169   a 60-ml syringe to release the chemical stimuli continuously within 20 cm of the substrate,

170   while slowly walking across the site. To minimize the potential disturbance when releasing

171   the stimulus (alarm cue or stream water), we always entered the stream upstream of the

172   section receiving the stimulus. Hence, we walked across the stream upstream of the control

173   section to release stream water and across the buffer section to release the alarm cue in risky

174   sections. Furthermore, YOY Atlantic salmon do not seem to react to overhead movements,

175   including a person walking slowly in the stream (personal observation).

176   To determine how long the chemical stimulus remained in a section, we released

177   samples of either milk or salt water ($n = 3$ for milk and $n = 4$ for salt water) in similar stream

178   reaches ($n = 7$; current velocity $= 0.70$ m $\cdot$ s^{-1}, depth $= 0.44$ m). The initial plume of milk or

179   salt water, detected by eye and using a conductivity meter, respectively, took an average of

180   11.5 sec to reach 5 m downstream of the point of release. However, milk could be detected in

181   the 5 m-section for up to 20 sec and the salt water for up to 34 sec after release (J.-W. Kim,

182   unpublished data).

183   Juvenile salmon can minimize their predation risk by either settling in alternative

184   habitats or hiding in the substrate of risky habitats. To estimate both components of avoidance,

185   we recorded the number, age class (only for Atlantic salmon), and species of all visible fishes

186   in each site via snorkelling. Snorkelling is a useful technique for estimating the abundance of

187   active fishes with a minimum of disturbance (Breau et al. 2007), particularly when

188   electrofishing is ineffective, such as in small areas of stream or at night (Gries et al. 1997;

189   Mullner et al. 1998; Dolinsek et al. 2007). In response to an increase in predation risk, salmon

190   parr often become more active at night (Gries et al. 1997; Imre and Boisclair 2004; Breau et al.

191   2007). Hence, we estimated density during both the day and night. For all surveys, the

192   snorkeler moved slowly upstream, completing each 1-m subsection by moving from the left

193   bank to the right bank, taking approximately 30 - 40 min to complete one 15-m site during

194   either the day or night. The daytime surveys of all sites were completed within the same day.

195   For night surveys, we used a waterproof handheld flashlight (20 watt) to illuminate fish,

196   which were typically lethargic and unresponsive when approached. Night surveys of all sites

197   were completed either in one or two consecutive nights.

198   For the initial counts, three density surveys were conducted on day 1 and 2 for each

199   section: two during the day (between 1200 h and 1600 h) and one at night (between 2300 and

200   0300 h). All daytime surveys were conducted at least one hour after or before the release of

201   the chemical stimuli at 1100 and 1700, respectively. For the final counts, we conducted three

202   density surveys on day 17 and 18 in 2006, and on day 14 and 15 in 2007: two during the day

203   and one at night. We had planned to release chemical stimuli for 14 consecutive days during

204   early settlement period. In 2006, however, heavy rain increased the water level and turbidity

205   of all sites on day 14. Hence, to minimize any potential effects of this rainfall, we continued

206   the treatments for a total of 17 days.

207   *Late settlement*

208   To investigate whether juvenile Atlantic salmon that have likely established territories

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209   respond to changes in the long-term riskiness of their local habitat, the 17-day experiment was

210   extended for another 13 days in 2006. The treatment protocol was similar to the 17-day

211   experiment except the buffer section during the early settlement now received the alarm cue

212   making it the risky section and the risky section now received nothing making it the buffer

213   section (see Fig. 1b). We expected the density to decrease the most in the new risky sections

214   (formerly the buffer sections), where the perceived predator activity increased the most. All

215   sections received a total of 150 ml of chemical stimulus (alarm cue or stream water) twice

216   daily for 13 days on July 8-21 in 2006 (Fig. 1b). The density survey protocol was similar to

217   that used in early settlement.

218   **Statistical analysis**

219   Because YOY are primarily day-active (Breau et al. 2007), whereas 1+ parr are active

220   during the day and night (Imre and Boisclair 2004; Dolinsek et al. 2007), we analyzed the

221   data separately for each age class. In addition, because of different diel activity patterns of

222   YOY and 1+ parr, we analyzed the YOY data separately for day and night, whereas we used

223   the average of the mean daytime densities and night-time densities for 1+ parr. To test for the

224   effects of perceived predator activity on the population density of YOY and 1+ parr Atlantic

225   salmon during the early settlement periods, a repeated measures two-way ANOVAs (three

226   treatments by two years with the early and late settlement periods as the repeated measure,

227   and site as a block) were used to compare the mean number of salmon per section at the

228   beginning and end of the early settlement periods. After we switched treatment in 2006, the

229   repeated measures analysis compared the numbers at the beginning (i.e. the number at the end

230   of the early settlement period) and end of the late settlement period.

231   For all analyses, only significant interactions between the effects of treatments, years,

232   sites, and time periods were reported, unless a non-significant interaction between the effects

233   of treatments, years, sites, and time periods might appear to be significant to the reader and

234   hence require an explanation. Results of all analyses are also presented in Table S1. To meet

235   the assumptions of parametric analyses, the number of YOY salmon counted during the day

236   and night and of parr salmon at the start and end of the treatment during early and late

237   settlement periods were $\log_{10} (X+1)$ transformed. For visual purposes, all data were back

238   transformed following the analyses and are shown with asymmetric S.E. bars.

239

240   **Results**

241   A total of 3682 fishes was counted in the two years, including six species: Atlantic

242   salmon, blacknose dace (*Rhinichthys atratulus*), lake chub (*Couesius plumbeus*), brook charr,

243   white sucker (*Catostomus commersonii*), and slimy sculpin (*Cottus cognatus*). YOY Atlantic

244   salmon were the most abundant (64.4 % of all observed fishes), followed by 1+ parr Atlantic

245   salmon (14.6 %), blacknose dace (11.4 %), 2+ parr Atlantic salmon (4.8 %), lake chub (2.7 %),

246   brook charr (1.6 %), white sucker (0.6 %), and slimy sculpin (one count).

247 **Early settlement**

248 As expected, the final number of YOY salmon in the 75 m^2 sites after the early

249 settlement period was higher during the day than at night (repeated measures one-way

250 ANOVA: $F_{1, 12} = 130.99$, $P < 0.001$; mean $\pm$ $SE = 31.18 \pm 4.92$ for day and 1.36 ± 0.62 for

251 night). Furthermore, the final number of YOY was higher in 2006 than in 2007 (repeated

252 measures one-way ANOVA: $F_{1, 12} = 20.60$, $P < 0.001$; mean $\pm$ $SE = 22.75 \pm 6.63$ for 2006

253 and 9.79 ± 2.89 for 2007). Hence, we analyzed the YOY data separately for day and night.

254 The final number of 1+ parr salmon in the 75 m^2 sites after the early settlement period

255 was higher in 2007 than in 2006 (repeated measures one-way ANOVA: $F_{1, 12} = 54.71$, $P <$

256 0.001; mean $\pm$ $SE = 2.21 \pm 1.50$ for 2006 and 12.43 ± 5.58 for 2007), but did not differ

257 significantly between day and night (repeated measures one-way ANOVA: $F_{1, 12} = 2.18$, $P =$

258 0.17; mean $\pm$ $SE = 8.64 \pm 2.00$ for day and 6.00 ± 1.45 for night). Hence, for the subsequent

259 analyses of parr, we used the average of the mean daytime and night-time densities.

260 As expected, the number of YOY salmon counted during the day increased over the

261 early settlement period (repeated measures two-way ANOVA, with site as block: $F_{1, 30} =$

262 40.04, $P < 0.001$; Fig. 2a). As predicted, there was a significant interaction between the effects

263 of treatments and the time periods on the number of YOY salmon counted during the day

264 (repeated measures two-way ANOVA, with site as block: $F_{2, 30} = 5.12$, $P = 0.012$; Fig. 2a).

265 The number of salmon in the alarm-cue sections decreased in 2006 and increased the least in

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266   2007 (Fig. 2a). Contrary to our expectations, however, the increase in number of YOY was

267   greatest in the buffer sections rather than control sections. The effect of treatments on the

268   number of YOY per section also appeared stronger in 2006 than in 2007; however, there was

269   no significant interaction among the effects of treatments, years, and time periods (repeated

270   measures two-way ANOVA, with site as block: $F_{2, 30} = 1.76$, $P = 0.19$; Fig. 2a). The number

271   of YOY counted during the day was greater in 2006 than in 2007 (repeated measures two-way

272   ANOVA, with site as block: $F_{1, 30} = 39.30$, $P < 0.001$; Fig. 2a).

273   Because YOY salmon were absent at night in 19 out of 21 sections in 2007, we only

274   tested the effect of treatments on the number of YOY at night during the early settlement

275   period in 2006. Moreover, because YOY salmon were absent at night in all 21 sections at the

276   start of early settlement period in 2006, we analyzed the data using a one-way ANOVA with a

277   randomized block design on the final number of YOY at night during early settlement. In

278   contrast to the patterns in the daytime data, the number of YOY at night appeared to increase

279   least in the buffer section and most in the alarm-cue and control sections (Fig. 2b), however,

280   this difference was not significant (one-way ANOVA, with site as block: $F_{2, 12} = 1.84$, $P =$

281   0.20; Fig. 2b). However, the contrasting patterns between the day and night data in 2006 led

282   to a significant interaction among the effects of treatments, time of the day, and time periods

283   (repeated measures two-way ANOVA, with site as block: $F_{2, 30} = 8.73$, $P = 0.001$). Hence, the

284   treatments had opposite effects on the number of YOY during the day and at night during the

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285   early settlement period in 2006.

286   In contrast to YOY salmon, there was no significant interaction between the effects of

287   treatments and the time periods on the number of 1+ parr (repeated measures two-way

288   ANOVA, with site as block: $F_{2, 30} = 2.31$, $P = 0.12$; Fig. 2c). However, there was a significant

289   interaction between the effects of time periods and years on the number of parr (repeated

290   measures two-way ANOVA, with site as block: $F_{1, 30} = 17.01$, $P < 0.001$); the number of parr

291   increased over the early settlement period in 2007 (repeated measures one-way ANOVA, with

292   site as block: $F_{1, 12} = 38.21$, $P < 0.001$; Fig. 2c), but remained the same in 2006 (repeated

293   measures one-way ANOVA, with site as block: $F_{1, 12} = 0.17$, $P = 0.68$; Fig. 2c). Furthermore,

294   the number of parr was greater in 2007 than in 2006 (repeated measures two-way ANOVA,

295   with site as block: $F_{1, 30} = 73.12$, $P < 0.001$; Fig. 2c).

296   **Late settlement**

297   As expected, the number of YOY during the day increased over the 13-day late-

298   settlement period (repeated measures one-way ANOVA, with site as block: $F_{1, 12} = 45.24$, $P < $

299   0.001; Fig. 3a), indicating that some fish were still selecting habitats in which to settle at this

300   time. The number of YOY salmon increased significantly more in control and buffer sections

301   than in risky sections (repeated measures one-way ANOVA, with site as block: $F_{2, 12} = 6.26$,

302     $P = 0.014$; Fig. 3a). Perhaps because of alarm cues dispersing downstream, the buffer section

303   had an intermediate final number of YOY (Fig. 3a).

304   At night, the number of YOY per section increased over the 13-day late-settlement

305   period (repeated measures one-way ANOVA, with site as block: $F_{1, 12} = 10.80$, $P = 0.007$; Fig.

306   3b), and this increase differed between treatments (interaction between treatments and time

307   periods: $F_{2, 12} = 5.84$, $P = 0.017$; Fig. 3b). Again, in contrast to the patterns during the day

308   time, the number of YOY salmon increased more in the risky sections than in the control or

309   buffer sections (Fig. 3a and 3b). Hence, the treatments again had opposite effects on the

310   number of YOY during the day and at night during the late settlement period in 2006.

311   Contrary to the results for YOY, the number of 1+ parr per section did not differ

312   significantly over the 13-day late-settlement period (repeated measures one-way ANOVA,

313   with site as block: $F_{1, 12} = 3.82$, $P = 0.074$; Fig. 3c). Furthermore, there was no significant

314   interaction between the effects of the treatments and the time periods on the number of parr

315   per section ($F_{2, 12} = 1.70$, $P = 0.22$; Fig. 3c).

316

317   **Discussion**

318   Our results suggest that YOY Atlantic salmon can use chemical information to

319   directly assess the relative activity of predators in different habitats and then avoid

320   particularly dangerous habitats. During the early settlement period, the number of YOY

321   during the day in risky sections decreased or increased less than in the buffer and control

322   sections. The magnitude of increase in density in the buffer sections was not expected and

323   may have been caused by fish moving upstream out of the risky sections. During the late

324   settlement period, the number of YOY salmon increased most in the control and buffer

325   sections and least in the risky sections, where the perceived predator activity increased the

326   most.

327   It is also possible that YOY salmon remained within risky sections but altered their

328   behaviour, by spending more time hidden in the substrate (Leduc et al. 2007), and were not

329   counted during surveys. We think this explanation of our results is unlikely for three reasons.

330   First, the delay between the release of the stimulus and the subsequent density survey (i.e. >

331   one hour) was much longer than the latency to resume feeding in response to the same

332   stimulus (i.e. < 50 sec; Kim et al. 2009). Second, YOY salmon are most active during the day

333   (Breau et al. 2007), so it is unlikely that the decrease in daytime density could be entirely

334   accounted for by more YOY salmon hiding in refuges. Third, if YOY were indeed hiding in

335   the substrate during the day, then we would have presumably detected them during night

336   surveys. While the increase in night densities was greatest in the risky sections, the magnitude

337   of this increase could not account for differences observed during the day. Nevertheless, we

338   cannot rule out the alternate explanation that YOY salmon responded to the alarm cues by

339   hiding in the substrate during the day and night. If true, this result would suggest that YOY

340   salmon hide in the substrate much longer during the summer than previously thought (Breau

341   et al. 2007). Taken together, these results suggest that YOY salmon detect and avoid the sites

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342   with alarm cues. Furthermore, our data suggest that alarm cues have longer-lasting effects on

343   the behaviour of YOY salmon at larger spatial scales than is typically measured during short-

344   term anti-predator experiments (Leduc et al. 2006; Kim et al. 2009).

345   Following the switch of treatments in 2006, we found no significant decline in

346   numbers in the alarm-cue sections (formerly the buffer sections), suggesting that once YOY

347   have settled on a foraging territory within a habitat, an increase in predator activity is not

348   sufficient to cause them to relocate to 'safer' habitats. Because the quality of alternative

349   locations is uncertain, the increased energy expenditure and decreased energy intake during a

350   relocation may decrease the net benefits of leaving risky sections (McDougall and Kramer

351   2007). In addition, if most YOY salmon have established their territories, then the prior

352   residency effect (Cutts et al. 1999) may make the costs of relocation outweigh the costs of

353   staying in risky habitats. Alternatively, YOY salmon can defend smaller territories to

354   compensate for the increased risk (Kim 2009; Kim et al. in press). However, the increase was

355   less in the alarm cue than in the other treatments, suggesting that new settlers either avoided

356   risky sites or hid in the substrate.

357   Interestingly, the patterns of YOY abundance at night during early and late settlement

358   periods were significantly different from and opposite to those observed in the day; the

359   increase in density at night was highest in the alarm-cue sections and lowest in the buffer

360   sections. The relatively few YOY found in risky habitats may become more night-active to

19

361   avoid the higher perceived activity of predators during the day. Furthermore, the few fish in

362   risky habitats may benefit from the reduced competition for food and space, which often leads

363   to an increased growth rate (Imre et al. 2005).

364   Contrary to our results for YOY, 1+ parr did not respond to the experimental

365   treatments. 1+ parr clearly detect and respond to alarm cues in the short term by decreasing

366   their foraging rates and increasing their latency to foraging (Kim et al. 2009). Because 1+ parr

367   already forage primarily at night (Imre and Boisclair 2004; Breau et al. 2007), they may be

368   less influenced by the increased perceived predator activity of alarm-cue sections during the

369   day. Alternatively, 1+ parr may have reached a size that makes them less vulnerable to gape-

370   limited fish predators such as brook charr and Atlantic salmon (Sogard 1997).

371   Overall, our data suggest that YOY Atlantic salmon can use chemical information to

372   assess the quality of habitat in terms of predator activity. Our study suggests that if habitats

373   differ markedly in ambient predator activity, YOY will avoid settling in particularly risky

374   habitats, or hide in the substrate for long periods of time. Whether the differences in perceived

375   predator activity between the treatments in our study represent the degree of spatial variability

376   in actual predation risk in salmon streams is an open question, especially so given that the

377   treatment effect was not strong enough to over-ride the differences in initial density. While

378   our study tested whether the differences in perceived predator activity in each section

379   influence settling decisions, it would be important to examine how the intrinsic riskiness of

380   habitat (Lima and Dill 1990; Verdolin 2006) influence the population density of a given

381   habitat, potentially affecting the relative predation risk of an individual. It would also be

382   interesting to examine whether and how predators redistribute themselves in response to the

383   altered distribution of prey. Moreover, investigating the potential interaction between the

384   effects of predation risk and population density on habitat selection would lead to the better

385   understanding of population consequences at broader spatial and temporal scales (Werner and

386   Peacor 2003; Blanchet et al. 2008). Further research is also needed to determine if YOY

387   salmon use alarm cues to assess the spatial variation in predation risk in an unmanipulated

388   system. Furthermore, given the well known difficulties in studying long-term predation risk in

389   natural populations (Lima and Steury 2005), our current study suggests that chemical alarm

390   cues may provide a valuable tool for future studies examining the ecologically relevant effects

391   of predation.

392

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570   **Figure legends**

571   Figure 1. Experimental design in one of seven stream sites during the (a) early and (b) late

572   settlement periods of YOY salmon where a risky section received conspecific alarm

573   cues; a control section received stream water; and, a buffer section received nothing,

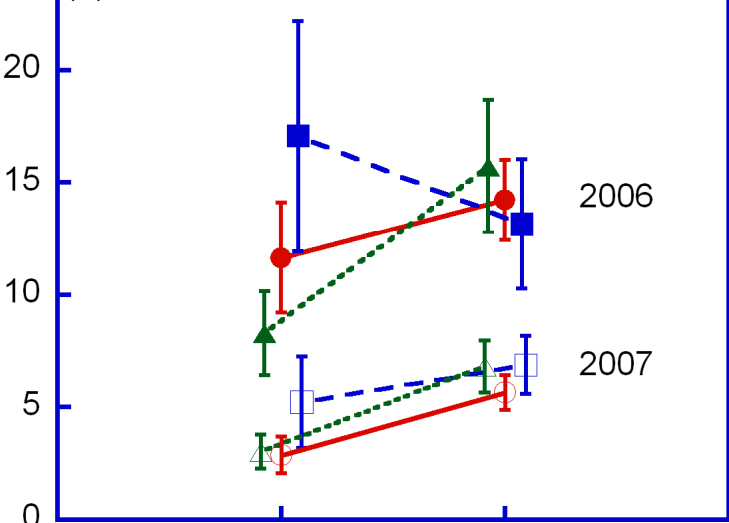574   twice a day for 13-17 days in 2006 and 2007.

575   Figure 2. Mean number ($\pm$ SE, $n = 7$) of Atlantic salmon exposed to three treatments: stream

576   water (●, solid line), alarm cue (■, dashed line), and a buffer (▲, dotted line) during

577   the early settlement period in 2006 (closed symbols) and 2007 (open symbols) for

578   young-of-the-year (a) during the day, (b) at night, and (c) for parr during the day and

579   night (Note that the results for YOY at night is reported for 2006 only)

580   Figure 3. Mean number ($\pm$ SE, $n = 7$) of Atlantic salmon exposed to three treatments: stream

581   water (●, solid line), alarm cue (■, dashed line), and a buffer (▲, dotted line) during

582   the late settlement period in 2006 for young-of-the-year (a) during the day, (b) at

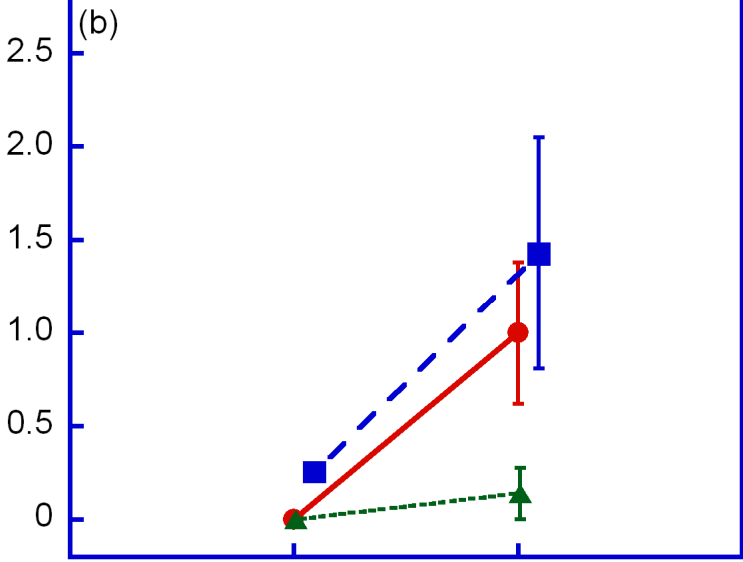583   night, and (c) for parr during the day and night
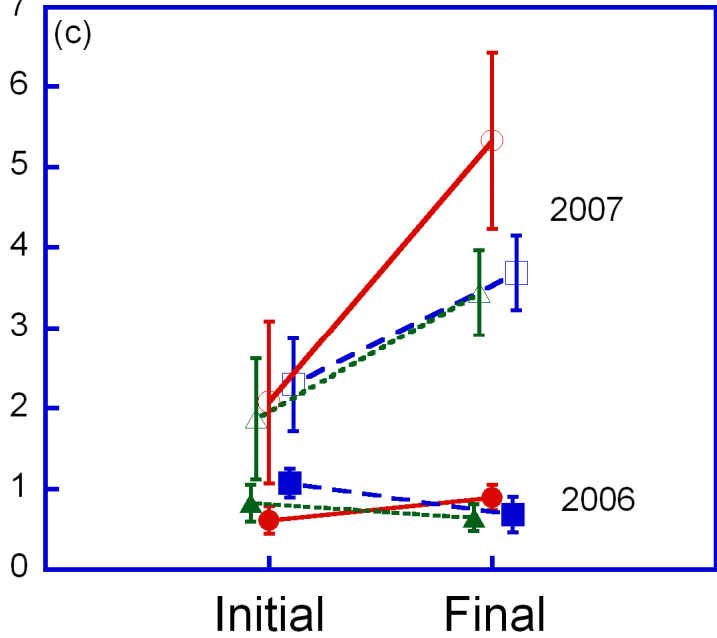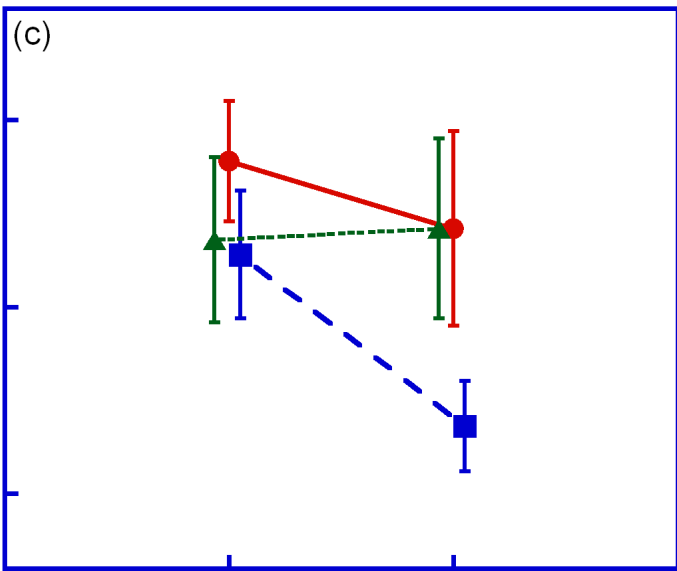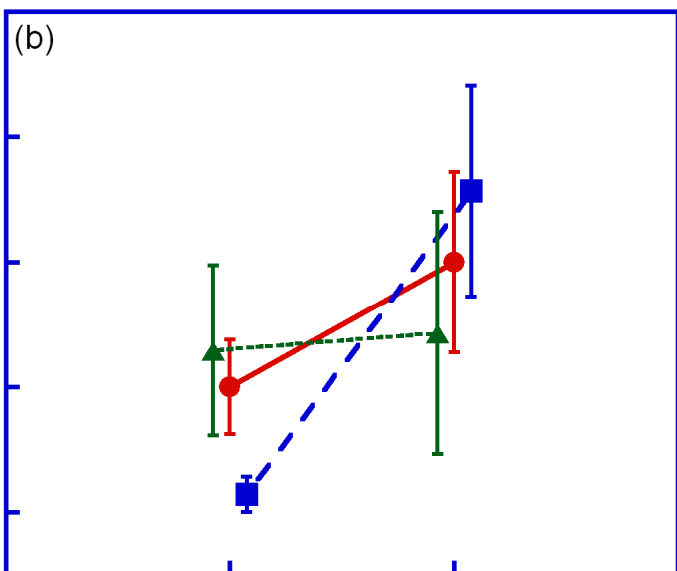
(a)

5 m — Stream water

5 m — Undisturbed buffer

5 m — Alarm cue

5 m

Stream flow

(b)

5 m — Stream water

5 m — Alarm cue

5 m — Undisturbed buffer

5 m

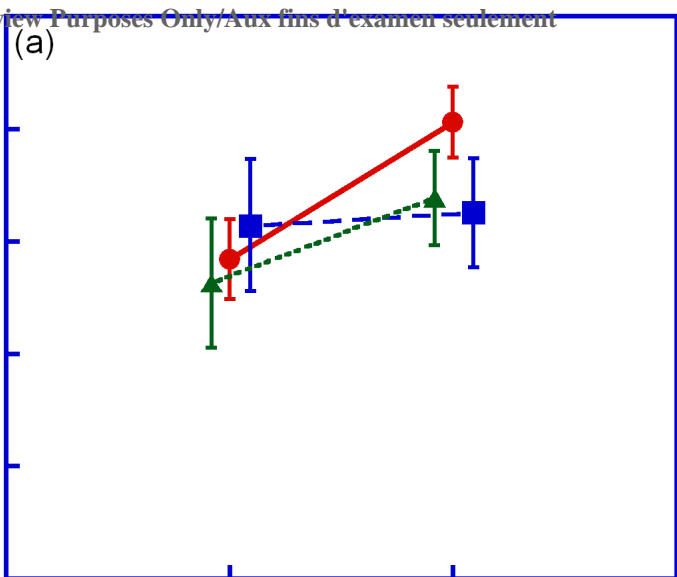**Number of salmon per section**

(a)

(b)

(c)

Initial   Final