1 **Effects of temperature and salinity on four species of north-eastern Atlantic**

2 **scyphistomae (Cnidaria: Scyphozoa)**

3

4 **Running page head:** Effects of temperature and salinity on jellyfish reproduction

5

6 **Authors and addresses**

7 *Chad L. Widmer

8 University of St Andrews, Pelagic Ecology Research Group, Scottish Oceans Institute, St

9 Andrews, Scotland, UK

10 Present Address: Point Defiance Zoo and Aquarium, Tacoma, 98407, Washington, USA

11 T: 253 404 3676, email: chad.widmer@pdza.org

12

13 Clive J. Fox

14 Scottish Association for Marine Sciences, Dunstaffnage, Oban, Scotland, PA371QA UK

15

16 Andrew S. Brierley

17 University of St Andrews, Pelagic Ecology Research Group, Scottish Oceans Institute, St

18 Andrews, Scotland, KY16 8LB UK

19

20 **Abstract**

21 Laboratory incubation experiments were conducted to examine the effects of different

22 temperatures (4, 9, 14, 19, 23°C) and salinities (21, 27, and 34) on survival and asexual

23 reproduction of scyphistomae of *Cyanea capillata*, *Cyanea lamarckii*, *Chrysaora hysoscella*,

24 and *Aurelia aurita* in order to better understand how climate variability may affect the timing

25 and magnitude of jellyfish blooms. Significant mortality was only observed for *C. capillata*

26 and *Ch. hysoscella* at the highest and lowest temperatures respectively, but temperature and

27 salinity significantly affected the asexual reproductive output for all species. As temperature

28 increased production rates of podocysts increased and, if produced, progeny scyphistomae by

29 side budding also increased. However, strobilation rates, and therefore the mean number of

30 ephyrae produced, decreased when scyphistomae were exposed to elevated temperatures.

31 These results provide a mechanistic explanation for why ephyrae of these species tend to be

32 produced during colder periods of the year whilst summer and early autumn are probably

33 important periods for increasing the numbers of scyphistomae in natural populations.

34

35 **Key words:** jellyfish, scyphistoma, strobila, ephyra, temperature, salinity, life cycle

36

37 **Introduction**

38 In some locations jellyfish blooms appear to be occurring more often (Brotz et al., 2012;

39 Dong et al., 2010; Mills, 2001; Purcell et al., 2007; Richardson et al., 2009) while in others

40 decreases have been reported (Dawson et al., 2001; Mills, 2001). However, because of a

41 global lack of long-term monitoring (Nickell et al., 2010; Purcell et al., 2007) the question of

42 whether blooms are really increasing in frequency and intensity has been controversial

43 although it has been frequently stated that increasing global temperatures are likely to favor

44 jellyfish.

45

46 Analysis of available time-series suggests that the abundance of jellyfish medusae is often

47 linked with long-term climate cycles (Condon et al., 2013; Lynam et al., 2005, 2004) and

48 environmental conditions are undoubtedly important influences upon jellyfish populations.

49 For example, increases in numbers of *Chrysaora* spp. and *Aurelia* sp. in the Gulf of Mexico

50 have been linked with warm winters, cool dry springs, and warmer than average summers

51   (Robinson and Graham, 2013). In the North Sea, the abundance of scyphozoan medusae has

52   been linked with the North Atlantic Oscillation, although with differing patterns in the

53   northern and southern sub-regions (Lynam et al., 2010, 2005, 2004).   Several of the regions

54   and in particular to the west of Denmark, showed significant negative correlations between

55   medusa abundance of *A. aurita* and *C. lamarckii* and the NAO index of the previous winter.

56   This result seems surprising because positive NAO years are associated with warmer winters.

57   The finding of reduced medusae abundances during the following summers is thus the

58   opposite of the suggestions that warming with favor jellyfish.

59

60   The life cycles of most non-oceanic jellyfish include an asexually reproductive benthic stage

61   – the scyphistoma. Because planktonic medusae originate from the benthic scyphistomae

62   through the process of strobilation, factors affecting polyp growth and reproduction are likely

63   key controls on the abundance of medusae (Lucas et al., 2012). Benthic asexual reproduction

64   modes in scyphozoan scyphistomae have been grouped into nine categories (Adler and Jarms,

65   2009). These modes include production of lateral buds (two types); stolon buds, regeneration

66   from stolon fragments; production of podocysts; free-swimming buds; gastric cavity

67   regeneration; longitudinal fission and strobilation. Scyphistomae also release juvenile

68   medusae, known as ephyrae, through the process of strobilation and these ephyrae eventually

69   grow into the sexually reproductive pelagic medusae, (Adler and Jarms, 2009; Arai, 1997;

70   Lucas et al., 2012). It has been widely recognized that further studies into the effects of

71   environmental conditions on the asexual reproductive modes of the scyphistomae are

72   required (Boero et al., 2008; Lucas et al., 2012; Mills, 2001) since their success ultimately

73   determines whether or not medusae blooms will form (Lucas et al., 2012).

74

75 Apart from the widely occurring *Aurelia aurita*, the habitat preferences of the scyphistomae

76 of other species are largely unknown and information on the locations and timings of

77 strobilation remains based upon observations of the ephyrae in near-shore plankton samples

78 (Grondahl, 1988; Hernroth and Grondahl, 1985; Lucas and Williams, 1994; Verwey, 1942).

79 Until scyphistoma populations are found and studied *in situ* it will be necessary to rely on

80 laboratory experiments to learn more about how benthic life history stages may respond to

81 altered physical conditions.

82

83 Here we report on laboratory incubation experiments to investigate the effects of different

84 temperatures and salinities on the population growth and strobilation rates of scyphistomae of

85 four species of north-eastern Atlantic Scyphozoa. We sought to investigate the role that

86 changed environmental conditions may have on asexual reproduction of scyphistomae

87 because the numbers of scyphistomae and the rates of strobilation are likely key factors

88 controlling the numbers of medusae released into the plankton. Specific hypotheses tested

89 were that differences in both temperature and salinity would significantly affect (1) mortality,

90 and (2) asexual reproduction (3) timing of strobilation, and, (4) numbers of ephyrae released.

91

92 **Methods and materials**

93 *Founding stock cultures*

94 Experiments were conducted with scyphistomae of *Aurelia aurita*, *Cyanea capillata*, *Cyanea*

95 *lamarckii*, and *Chrysaora hysoscella*. Scyphistomae of *A. aurita* were sourced from the tests

96 of the ascidian, *Ascidia mentula*, growing at between 10 – 27 meters deep in Scapa Flow,

97 Scotland during summer 2010. The host ascidians were collected by divers and scyphistomae

98 carefully removed at the Scottish Oceans Institute (SOI) with fine tipped forceps.

99 Scyphistomae were placed inside plastic culture plates filled with 5µm-filtered North Sea

100 water, salinity 34. Ephyrae released from these scyphistomae were raised at SOI into mature

101 medusae to confirm that they were *Aurelia*. Specimens of scyphistomae collected from

102 Scapa Flow were also supplied to S. Piraino and G. Aglieri at the Universitia del Salento,

103 Leece, Italy, COI (cytochrome c oxidase subunit I) DNA barcoding confirmed them to be

104 *Aurelia.* During summer 2011 *C. capillata* medusae were collected near Oban, Scotland, and

105 *C. lamarckii* medusae near St. Andrews, Scotland. Stock cultures of scyphistomae of the

106 species were initiated using planulae collected from five female medusae of each species.

107 Planula larvae of *Ch. hysoscella* were harvested from 3 female medusae collected near

108 Dalefort, Wales, in August, 2011. Thestock cultures of scyphistomae were maintained at

109 salinity 34 at 10°C, in a dark temperature controlled roomin the SOI. They were fed one day

110 old *Artemia franciscana* (Kellog) nauplii once per week for at least 6 months prior to the start

111 of experiments in order to ensure that scyphistomae had time to fully develop.

112

113 *Incubation temperatures and salinities*

114 The locations of the benthic stages of most species of scyphozoa are cryptic. However, ones

115 that have been found are often located in water less than 30m deep so the temperatures

116 selected for the experiments were in the range reported for surface stations in the North Sea

117 (Beszczynska-Möller and Dye, 2013; Schulz, 2009) with the addition of a 23°C treatment

118 which is at the upper end of predictions for the southern North Sea by the 2080s (Mathis and

119 Pohlman, 2014). Offshore salinities in the North Sea are generally above 35 but lower

120 salinities are found closer inshore, particularly in the estuaries, coastal zone and German

121 Bight during late winter and early spring (Beszczynska-Möller and Dye, 2013). The

122 temperatures and salinities tested for each species (Table 1) were thus selected to cover a

123 plausible range which might be experienced by scyphistomae in the North-eastern Atlantic.

124

Equipment and acclimations

Experimental rearing was conducted inside temperature controlled incubators (Lucky Reptile

Herp Nursery II). The incubators were darkened to remove the potentially confounding

effects of light/dark period on asexual reproduction (Liu et al., 2009; Purcell et al., 2009), and

temperature in each incubator was continuously monitored using USB data loggers (Lascar

EL-USB-1). The salinity of North Sea water was adjusted by mixing with distilled water and

monitored using a calibrated hand held Bellingham and Stanley refractometer. One

scyphistoma from the stock cultures described above was placed in each well of 6-well

polycarbonate culture plates filled with 12 ml of the 5 µm-filtered North Sea water, and then

gradually acclimated to the target salinity at 10°C in a stepwise manner over 7 days. The

scyphistomae were then gradually acclimated to their target temperatures over an additional 7

days. All scyphistomae had attached to the bottoms of their replicate wells by the ends of the

acclimation period. During the experiments scyphistomae were fed one day old *A.*

franciscana nauplii to repletion once per week. Uneaten food was removed and water

changed the following day using a pipette, with the wells being refilled with 5µm-filtered

seawater of appropriate salinity and temperature.

Data recording

Scyphistomae were examined weekly under a dissecting stereomicroscope for the formation

of new podocysts or progeny scyphistomae, to check for strobilation, and to record any

mortality. Examinations were conducted as quickly as possible (~15 min observation^{-1}) at

room temperature (~15°C) to prevent large temperature fluctuations. Progeny scyphistomae

were removed from the wells as soon as they had separated from parent scyphistomae in

order to eliminate the effects of crowding on asexual reproduction. If at the end of the eight

week experiment a scyphistoma was observed to still be undergoing strobilation, incubations

150   were continued until the last ephyrae was released. At the ends of the incubations

151   scyphistomae were removed from their experimental wells with fine tipped forceps, and the

152   number of podocysts counted.

153

154   *Data analysis*

155   The response variables were: number of progeny scyphistomae and podocysts produced,

156   whether or not mortality or strobilation had occurred; time until strobilation began; duration

157   of strobilation events; and numbers of ephyrae produced per individual in each treatment

158   group. Since the response variables were either counts (e.g. number of podocysts produced),

159   or were binomial in nature (e.g. strobilated or did not) generalized linear models (GLMs)

160   were used to model the effects of temperature, salinity and their interaction. Best fitting

161   models were selected based on Akaike Information Criteria, followed by analysis of deviance

162   likelihood ratio tests. Model validation followed recommendations in (Ver-Hoef and

163   Boveng, 2007; Zuur et al., 2013). Relationships between temperature, salinity and response

164   variables were evaluated by calculation of Spearman's correlation coefficient. All analyses

165   were conducted using R version 2.15.1 (R Development Core Team 2012).

166

167   In order to visualize the predicted number of ephyrae under different temperature conditions

168   the best fitting models were used to predict 30 fitted values within the temperature ranges

169   reported to commonly occur during each month of the year during positive and negative

170   NAO years at stations in the North Sea (http://www.cefas.defra.gov.uk).

171

172   **Results**

173   During the present study scyphistomae to scyphistomae (StS) asexual reproduction of of *C.*

174   *capillata* and *Ch. hysoscella* were observed to be exclusively by the production of podocysts

175   while *C. lamarckii* produced both podocysts and typical lateral side buds. *A. aurita*

176   scyphistomae produced podocysts, lateral side buds and stolon budded progeny. However,

177   scyphistomae of all four speciesstrobilated during the experiments. A summary of the best

178   fitting GLMs for the effects of temperature, salinity and their interaction on asexual

179   reproductive output and survivorship of studied scyphistomae are given in Table 2. Tables of

180   descriptive statistics are given in the supplemental material, and a summary of results of

181   Spearman correlation tests is provided in Table 3.

182

183   *Surviving scyphistomae*

184   Temperature significantly affected the survival of *C. capillata* and *Ch. hysoscella*

185   scyphistomae, but did not significantly affect survival of *C. lamarckii* or *A. aurita*. At higher

186   temperatures survival of *C. capillata* scyphistomae was diminished, and all *C. capillata*

187   scyphistomae at perished within three weeks at 23°C (Fig. 1A).   In contrast, scyphistomae of

188   *Ch. hysoscella* survived at 23°C but died at 4°C by the end of the 7th week (Fig. 1C). Salinity

189   did not have significant effects on survival for any of the four species.

190

191   *Production of progeny scyphistomae*

192   *C. capillata* and *Ch. hysoscella* did not produce progeny scyphistomae during the any of the

193   incubations and asexual reproduction for these species was limited to the production of

194   podocysts, and ephyrae through strobilation. *C. lamarckii* and *A. aurita* produced progeny

195   scyphistomae by means of typical side budding in all treatments, but not in high numbers

196   (Fig. 2A). There were also no significant relationships between the number of progeny

197   produced by *C. lamarckii* and temperature or, salinity. *A. aurita* also produced progeny

198   scyphistomae (Fig. 2B) and there were significant relationships with temperature, but not

199   salinity. The interaction was however significant, therefore salinity was retained in the

200   model.

201

202   *Production of podocysts*

203   Podocysts were produced by scyphistomae of all four species during the study with the

204   general trend that the number of podocysts increased with temperature (Fig. 3). Podocyst

205   production was significantly and positively correlated with temperature for *C. capillata* ($r_s =$

206   0.354, p < 0.001), *C. lamarckii* ($r_s = 0.428$, p < 0.001) and *Ch. hysoscella* ($r_s = 0.659$, p <

207   0.001), but temperature was not significantly correlated with podocyst production in *A. aurita*

208   (Table 3). The mean number of podocysts produced by scyphistomae of *C. capillata* and *C.*

209   *lamarckii* was significantly linked with temperature in the GLMs. The greatest number of

210   podocysts, average 3.0 per scyphistoma, was produced at 23°C and salinity 27 by *Ch.*

211   *hysoscella*. The rates of *Ch. hysoscella* and *A. aurita* podocyst production was significantly

212   and positively correlated with salinity ($r_2 = 0.204$, p = 0.013), and significantly linked in the

213   GLM to temperature and salinity, and the interaction of these two factors was also significant

214   (Fig. 3C).

215

216   *Strobilation*

217   Scyphistomae of all four species strobilated during the study (Fig. 4). Strobilation was

218   significantly and negatively correlated with temperature for *C. capillata* ($r_s = -0.68$, p <

219   0.001), *C. lamarckii* ($r_s = -0.41$, p < 0.001) and *A. aurita* ($r_s = -0.61$, p < 0.001), but

220   temperature was not significantly correlated with strobilation in *Ch. hysoscella* (Table 3).

221   Salinity was not significantly correlated with strobilation for any of the species tested (Table

222   3.) No scyphistomae strobilated more than once during the eight week studies. Strobilation

223   of scyphistomae of *C. capillata*, *C. lamarckii* and *Ch. hysoscella* was significantly linked

224   with temperature alone in GLMs, and strobilation of *A. aurita* was significantly linked with

225   temperature and salinity, but not their interaction. None of the scyphistomae perished after

226   strobilating and appeared to be in good condition which was apparent by the regeneration of

227   mouths and feeding tentacles following liberation of the final ephyra.

228

229   *Onset of strobilation*

230   Scyphistomae of *C. capillata* and *C. lamarckii* maintained at warmer temperatures strobilated

231   sooner than scyphistomae incubated at cooler temperatures (Fig. 5A, B). However, the

232   number of scyphistomae that strobilated within two weeks was far fewer than those that

233   strobilated after more than two weeks (Fig 4, 5). For *C. capillata* there was a significant

234   negative relationship between the days taken to begin strobilation and temperature, but not

235   with salinity or their interaction. For *C. lamarckii* the mean time to onset was significantly

236   linked with temperature and salinity, but not their interaction. Neither temperature nor

237   salinity was significantly linked with onset of strobilation in *A. aurita* or *Ch. hysoscella* over

238   the ranges tested.

239

240   *Duration of strobilation*

241   Temperature was significantly linked with the duration of strobilation in all species except *A.*

242   *aurita*, with the general trend being that duration decreased as temperature increased (Fig. 6).

243   Salinity was only significantly linked with the duration of strobilation for *C. capillata* (Fig.

244   6A). Strobilation duration was significantly and negatively correlated with temperature for

245   *C. capillata* ($r_s = -0.49$, $p < 0.001$), *C. lamarckii* ($r_s = -0.69$, $p < 0.001$) and *Ch. hysoscella* (r_s

246    $= -0.66$, $p < 0.001$), but temperature was not significantly correlated with strobilation duration

247   in *A. aurita* since strobilation only occurred at 4°C (Table 3). Salinity was significantly

248 correlated with strobilation duration for *C. capillata* (r_s = -0.21, p = 0.03), and *A. aurita* (r_s = -

249 0.54, p = 0.001).

250

251 *Production of ephyrae*

252 Temperature and salinity were significantly linked with ephyra production with the general

253 trend being that mean ephyra production decreased as temperature increased for all species

254 tested (Fig. 7, Table 2). The interaction of temperature and salinity was also significant for

255 *C. lamarckii* and *Ch. hysoscella* in the GLMs. The greatest mean number of ephyrae, 20.3 per

256 scyphistoma, were produced by *A. aurita* at 4°C and salinity 27 (Fig. 7) The number of

257 ephyrae produced was significantly and negatively correlated with temperature for *C.*

258 *capillata* (r_s = -0.66, p < 0.001), *C. lamarckii* (r_s = -0.41, p < 0.001) and *A. aurita* (r_s = -0.6, p

259 < 0.001), but temperature was not significantly correlated with the number of ephyrae

260 produced in *Ch. hysoscella* (Table 3), and salinity was not significantly correlated with the

261 number of ephyrae produced for any of the four species examined. For *C. capillata* and *Ch.*

262 *hysoscella* the mean number of ephyrae produced increased to an optimum temperature but

263 then decreased as temperatures increased further. The temperature at which the maximum

264 number of ephyrae was produced was also slightly higher in *Ch. hysoscella* compared with

265 the other species. For *C. lamarckii* higher temperatures led to fewer ephyrae being released

266 per scyphistoma whilst strobilation did not occur at all in *A. aurita* when the scyphistomae

267 were held at temperatures above 4°C.

268

269 *The potential effect of high and low NAO scenarios on the production of ephyrae*

270 Our results suggest that when the NAO is in a positive phase warmer winter sea temperatures

271 may decrease the number of *A. aurita* and *C. lamarckii* scyphistomae that strobilate with the

272 effect being fewer ephyrae are added to the system (Fig. 8). Conversely, when the NAO is in

273   a negative phase cooler sea temperatures during winter may increase the number of

274   scyphistomae that strobilate, resulting in more ephyrae. By summer/autumn correlations of

275   sea temperature with previous NAO have largely disappeared (Lynam et al., 2005) so that we

276   would not expect much predictive power for the effect of NAO on the other asexual

277   reproductive modes e.g. via podocyst production.

278

279   **Discussion**

280   Results from the present study showed that asexual reproductive outputs of scyphistomae of

281   four species of north-eastern Atlantic jellyfish are significantly affected by temperature and

282   salinity. This provides a possible mechanistic explanation for previously reported correlative

283   interannual climate-related variability in jellyfish medusae abundance in the North Sea

284   (Lynam et al., 2005, 2004) as well as suggesting that a future warmer north-eastern Atlantic

285   may not be so jelly dominated as some hae suggested unless this is due to a strong increase in

286   the abundance of more lusitanian species.

287

288   Around the UK *C. capillata* medusae have been recorded more frequently along the north-

289   western coastline giving it a more northerly distribution. In contrast, *Ch. hysoscella* tends to

290   befound in more southerly waters although the medusae have occasionally been observed

291   along the northern Scottish coast (Doyle et al., 2007; Holst, 2012; NBN, 2016; Russell,

292   1970). Medusae of *C. lamarckii* have been recorded all around the UK including the

293   southern North Sea and English Channel (National Biodiversity Network Database, consulted

294   14 Feb. 2016) whilst *A. aur*ita is similarly broadly distributed. These broad geographical

295   patterns in medusae distribution seem to be broadly supported by the relationships between

296   scyphistomae survival and temperature seen in the experiments. *C. capillata* failed to survive

297   at 23°C whilst *Ch. hysoscella* scyphistomae suffered 100% mortality at the lowest

298   temperature tested. Temperature did not significantly affect survival of *C. lamarckii* or *A.*

299   *aurita* scyphistomae.

300

301   Across all the species studied, scyphistomae responded to warmer temperatures by increasing

302   benthic asexual reproductive output through the production of podocysts and/or progeny

303   scyphistomae. The greatest number of podocysts were produced in this study by

304   scyphistomae of *Ch. hysoscella* at a rate of about 0.375 podocysts per week at 23°C salinity

305   27 which was slower than the rates reported for *Chrysaora fuscescens* (1.65 podocysts w^{-1})

306   from the Northeast Pacific Ocean (Widmer, 2008a), and *Chrysaora quinquecirrha* (4.3

307   podocysts w^{-1}) in the Chesapeake Bay (Cargo and Schultz, 1967). In natural populations,

308   podocyst production is probably maximal during the summer to early autumn which would

309   be in agreement with findings for other *Cyanea* (Brewer and Feingold, 1991; Grondahl, 1988;

310   Thein et al., 2013) and *Chrysaora* spp. (Cargo and Rabenold, 1980; Cargo and Schultz, 1967;

311   Thein et al., 2013). Excystment of podocysts was not observed in the present study, but if

312   patterns are similar to some other species (Brewer and Feingold, 1991; Cargo and Rabenold,

313   1980; Cargo and Schultz, 1967; Grondahl, 1988; Thein et al., 2013) then podocysts may

314   excyst when sea temperatures drop during autumn. This behaviour could act as a mechanism

315   for timing the development of the emergent scyphistomae in time to strobilate during winter

316   or early spring.

317

318   During this study progeny scyphistomae were only produced by *C. lamarckii* and *A. aurita*,

319   and neither species produced them in great abundance. The results for *A. aurita* were

320   unexpected in light of other studies which have reported elevated scyphistoma production at

321   higher temperatures (Han and Uye, 2010; Schiariti et al., 2014). In our study the Orkney

322   population of *A. aurita* produced very few scyphistomae at 4°C and progeny were produced

323   at temperatures beyond that with a general trend toward fewer progeny being produced at

324   higher temperatures which was similar to results for scyphistomae of *Aurelia labiata* (Purcell,

325   2007). Furthermore several workers have suggested that *Aurelia* may be locally adapted

326   (Connelly et al., 2001; Edwards, 1965; Lucas et al., 2012; Pascual et al., 2014; Schroth et al.,

327   2002).

328

329   In the present experiments, scyphistomae exposed to cooler temperatures tended to decrease

330   their production of podocysts and progeny scyphistomae, and instead began strobilating. The

331   lowest numbers of ephyrae also tended to be produced at the highest temperatures but there

332   were some differences in the temperature at which the maximal number of strobilating

333   scyphistomae occurred. For *Ch. hysoscella* the maximal strobilation temperature was slightly

334   higher (9-14°C) compared with the other species. In the Gullmar Fjord, Sweden *C. capillata*

335   has been recorded as strobilating during the coldest months of the year (Grondahl, 1988) and

336   the same was observed for *C. capillata* from the Niantic River estuary, Connecticut (Brewer

337   and Feingold, 1991). In the southern North Sea *A. aurita* ephyrae are have been observed

338   from the end of January through to the middle of March (Lucas and Williams, 1994; Lucas,

339   2001). Observations on the timing of ephyrae release in natural populations of *Ch. hysoscella*

340   are lacking, but our experimental results suggest strobilation in this species is possible in

341   slightly higher temperatures, compared with the other species. Again this seems consistent

342   with the broad temperature preferences of the four species studied.

343

344   It has been suggested that there is a minimum temperature threshold required for

345   scyphistomae to strobilate (Russell 1970, and references therein). Numerous workers have

346   sought to uncover the internal mechanisms responsible for strobilation in scyphistomae (Arai,

347   1997; Lucas et al., 2012), and recent work has shown that the precursor hormone (CL390),

348   controlling strobilation in *A. aurita* is encoded in response to seasonal temperature change

349   (Fuchs et al., 2014). Strobilation has thus been associated with colder temperatures across a

350   range of Scyphozoa in temperate waters and is presumably a mechanism for maximising the

351   temporal match between the ephyrae and the later developing spring zooplankton bloom.

352   This hypothesis is supported by evidence of the remarkably long point-of-no return under

353   starvation demonstrated by *Aurelia* ephyrae (Fu et al., 2014) The findings here support the

354   hypothesis that the four species of scyphozoan studied must experience low sea temperatures

355   for appropriate durations in order for the majority of scyphistomae to strobilate but there were

356   inter-specific differences so that the precise minimum temperatures required are species, and

357   possibly population specific.

358

359   In the experiments reported here increasing temperature decreased strobilation durations.

360   This finding is in accordance with those reported elsewhere for a number of temperate (Holst,

361   2012; Purcell, 2007; Purcell et al., 1999) and tropical (Lotan and Fine, 1994; Suguira, 1965)

362   jellyfish species. In order to determine whether natural populations of scyphistomae are able

363   to strobilate more than once during an annual cycle it is important to know the amount of

364   time required for scyphistomae to initiate the process of strobilation (onset when

365   temperatures are below the critical threshold), the strobilation duration, and the amount of

366   time required for scyphistomae to recover and be ready to strobilate again. The complete

367   sequence of initiation, strobilation and recovery constitute a strobilation requirement timeline

368   (SRT). The recovery periods for scyphistomae were not the focus of the present study, so

369   those periods must be estimated, based on laboratory culturing experience, and probably have

370   durations of at least four weeks in well fed individuals (CW personal observation). For

371   example, the SRT for *A. aurita* scyphistomae in this study at 4°C and salinity 34, would be

372   about 19.4 weeks (data from Supplement 1: Table S4.) comprised of,

373

374   Onset (7 weeks) + Duration (8.4 weeks) + Estimated recovery (4 weeks) = 19.4 weeks.

375

376   Following this one can determine the "strobilation window," or length of time when annual

377   sea surface temperatures are likely to fall below the critical minimum temperature thresholds.

378   In Scapa Flow, *A. aurita* scyphistomae normally experience annual SSTs ranging from *ca.* 4

379   – 14°C (http://www.divesitedirectory.co.uk/uk_scotland_scapa.html) with salinities near 35

380   year round (Turrell et al., 1996). Since the SRT for *A. aurita* was 19.4 weeks one can

381   estimate that populations of this species probably do not strobilate more than once during an

382   annual season.

383

384   Once initiated, the process of strobilation can be inhibited by further changes in temperature

385   (Chen and Ding, 1983; Holst, 2012; Widmer, 2008b; You et al., 2008). Affected ephyrae

386   continue to develop and are released as normal, but no further ephyrae are produced

387   (Widmer, 2008b). Once sea temperatures begin to increase during spring the minimum

388   strobilation temperature thresholds cease to be met thus closing the strobilation window and

389   ending the process for the season. Asexual reproduction then shifts to the production of

390   podocysts and progeny scyphistomae.

391

392   Our findings for the numbers of ephyrae produced are similar to those for *A. aurita* from the

393   northwest Mediterranean Sea (Purcell et al., 2012), and from Taiwan (Liu et al., 2009).

394   Our results both concur and contradict with findings from previous similar studies on the

395   effects of temperature (Holst, 2012) and salinity (Holst and Jarms, 2010) on strobilation and

396   ephyra production of the same four species of scyphistomae. When scyphistomae from the

397   German Bight were maintained in simulated conditions reflective of warmer winter

398   temperatures (10°C versus 5°C) ephyra production was enhanced for *A. aurita*, *Ch.*

399   *hysoscella,* and *C. lamarckii* (Holst, 2012), and more ephyra were produced per strobila in *C.*

400   *capillata* and *C. lamarckii* (Holst, 2012). In the present study, scyphistomae of *Aurelia*

401   originating from Scappa Flow, Orkney only produced ephyrae at 4°C and the greatest

402   numbers of ephyrae were produced by *Aurelia*, *C. capillata* and *C. lamarckii* in the coldest

403   temperatures tested (4 – 9°C). However, our findings for strobilation and ephyra production

404   of *Ch. hysoscella* generally concur with those of Holst (2012).

405

406   Maximal numbers of *A. aurita*, *C. capillata* and *C. lamarckii* ephyrae from the German Bight

407   were produced at salinity 28 (Holst and Jarms, 2010). Our findings concur, in the present

408   study most ephyrae were produced at salinity 27. We found that there was a significant

409   interaction between temperature and salinity for the number of ephyrae produced by

410   scyphistomae of *C. capillata* and *C. lamarckii* meaning that for these species the synergistic

411   effects of temperature and salinity on ephyra production may be more prominent than either

412   factor acting alone. Assuming that scyphistomae are affected by sea surface conditions,

413   during years with abundant rainfall and low sea temperatures our findings suggest that more

414   ephyrae of these species are likely to be produced than in years with little rainfall and warm

415   sea temperatures.

416

417   Scyphistomae in the present study were cultivated singly in replicate wells and progeny were

418   removed as soon as they were produced in order to avoid the potentially confounding effects

419   of replicate mates. For example, it has been shown that scyphistomae of *Aurelia* from the

420   Gulf of Mexico release a water transportable substance, neck-inducing factor, that stimulates

421   nearby scyphistomae to strobilate (Loeb and Blanquet, 1974; Loeb, 1974). Additionally,

422   scyphistomae abundance has been shown to be density dependent with intraspecific

423   competition decreasing asexual reproduction rates until equilibrium is reached (Melica et al.,

424   2014; Willcox et al., 2007). Scyphistomae from the German Bight were cultivated for

425   extended periods with many scyphistomae in each replicate (Holst and Jarms, 2010; Holst,

426   2012) which may have been affected by water transportable substances or by scyphistoma

427   density, potentially affecting asexual reproduction rates. However, cosmopolitan species

428   such as *A. aurita*, may also actually comprise a species complex as revealed by recent

429   molecular studies (Dawson and Jacobs, 2001).   Contrasting results may be the result of local

430   adaptations suggesting that regionally focused studies will be required in order to predict

431   population responses under climate change (Connelly et al., 2001; Edwards, 1965; Lee et al.,

432   2013; Lucas et al., 2012; Pascual et al., 2014; Purcell, 2007).

433

434   Our acclimation periods to the experimentla conditions were relatively rapid but we did not

435   observe any mortality or obvious deleterious effects during our acclimation protocol.

436   Furthermore, many jellyfish medusae are able to quickly acclimate to new environmental

437   conditions. For example, pulsation rates of field collected medusae of *Chyrsaora*

438   *quinquecirrha* reached equilibrium in 3hr when transferred from 29 to 15°C (Gatz et al.,

439   1973). A number of hydromedusae from the Puget Sound osmoconform to salinites ranging

440   from 23 – 38 within a few hours, altering their densities and regaining equilibrium buoyancy

441   (Mills, 1984). Even though we used a rapid acclimation scheme relative to the natural

442   environment, our findings are in line with with the idea of minumum temperature thresholds

443   needing to be met in order for strobilation to occur (Russell 1970 and references therein) and

444   the timings of ephyrae release (Hernroth and Grondahl, 1985; Lucas and Williams, 1994;

445   Russell, 1970; Verwey, 1942). It would be useful for future work to determine how the rate

446   of change affects asexual reproductive output.

447   Our data support the hypothesis that temperature and salinity influence asexual reproductive

448   modes and rates of scyphistomae in north-eastern Atlantic waters.   Links between the NAO

449   and sea temperatures in the North Sea are strongest during the winter and early spring so

450   potentially affect the period when scyphistomae are strobilating (Lynam et al., 2004). A

451   hypothetical model derived from GLM predictions from our results shows the overall effect

452   of fewer ephyrae added to the system in positive phase NAO years (Fig. 8). Conversely,

453   when the NAO is in a negative phase cooler sea temperatures during winter may increase the

454   number of scyphistomae that strobilate, resulting in more ephyrae. Climate variability is

455   however likely linked with many other changes which may affect scyphistoma reproduction,

456   an obvious factor being changes in planktic food (Ottersen et al., 2001). Better nourished

457   strobilae produce more ephyrae than poorly nourished ones (Ishii and Watanabe, 2003;

458   Purcell et al., 1999; Spangenberg, 1967; Wiesenthal, 2012). Furthermore, enhanced survival

459   of ephyrae and young medusae could easily lead to changes observed in population

460   abundances later in the year, regardless of the numbers of ephyrae released

461

462   *Conclusions*

463   Plasticity in asexual reproductive modes of scyphistomae plays an important role in the long

464   term maintenance of jellyfish populations (Arai, 2009; Boero et al., 2008; Lucas et al., 2012).

465   In this study the general trend was that as temperature increased benthic asexual output

466   increased. Benthic asexual reproduction probably occurs throughout much of the year with

467   the majority occurring during summer when prey availability is high. For the species studied,

468   the present results suggest that the majority of strobilation probably takes place during the

469   colder months, which is in agreement with the presence of ephyrae in the north-eastern

470   Atlantic plankton samples (Hernroth and Grondahl, 1985; Lucas and Williams, 1994; Russell,

471   1970; Verwey, 1942) and other experimental data (Holst, 2012). During years when open

472   strobilation window durations are short (such as high NAO phases) it can be predicted that

473   fewer ephyrae will be produced by the scyphistomae, and they instead maximise benthic

474   asexual reproduction. During years with long open strobilation windows (such as low NAO

475   phases) benthic reproduction should be slowed, but more ephyrae are likely to be produced.

476   The combination of SRTs and species specific minimum temperature strobilation thresholds

477   could explain the negative correlations between the NAO and medusa abundance in parts of

478   the North Sea (Lynam et al., 2005, 2004). However, these patterns are complicated by

479   differences at sub-regional scales which Lynam et al. (2005) suggested were linked to

480   complexities in the local oceanography. Furthermore, medusae of some species, such as *C.*

481   *capillata* may be able to overwinter (Hay et al., 1990), thus potentially masking the effects of

482   inter-annual temperature variability on their abundance (Lynam et al., 2004). Although the

483   scyphistomae of *C. capillata* appear able to continue strobilation over a wider range of

484   temperatures than *C. lamarckii* or *A. aurita* differences in the minimum temperature

485   strobilation thresholds suggest that *C. capillata* in particular, may become less common in

486   areas such as the North Sea under warming scenarios whilst *Ch. hysoscella* may be able to

487   increase its range (Mathis and Pohlman, 2014). Strobilation of *A. aurita* appeared to be

488   particularly sensitive to increased temperatures in our experiments but *Aurelia* is very widely

489   distributed and successful in coastal waters from the tropics to the sub-Arctic. One

490   explanation of the different results in the present study and Holst et al. (2012) is that we are

491   dealing with locally adapted sub-populations. If this is true then replacement of locally cold

492   adapted sub-populations by visibly similar *Aurelia* clades adapted to warmer waters may

493   occur (Dawson and Martin, 2001). Further experiments comparing the temperature responses

494   of *Aurelia* scyphistomae collected from different locations, ideally with accompanying

495   genetic taxonomy, are needed to test this.

496

497 In summary, scyphistomae responded to high temperatures by decreasing or ceasing

498 strobilation altogether, and by increasing rates of benthic asexual reproduction. The precise

499 minimum temperatures required to open strobilation windows are species and probably

500 population specific, and seem to explain the broad temperature preferences observed at the

501 medusa stages.

502

511

512 Bibliography

513 Adler, L., Jarms, G., 2009. New insights into reproductive traits of scyphozoans: special
514   methods of propagation in *Sanderia malayensis* GOETTE, 1886 (Pelagiidae,
515   Semaeostomeae) enable establishing a new classification of asexual reproduction in the
516   class Scyphozoa. Mar. Biol. 156, 1411–1420.

517 Arai, M.N., 2009. The potential importance of podocysts to the formation of scyphozoan
518   blooms: a review. Hydrobiologia 616, 241–246.

519 Arai, M.N., 1997. A functional biology of Scyphozoa. Chapman and Hall, London.

520 Beszczynska-Möller, A., Dye, S.R. (Eds.), 2013. ICES Report on ocean climate 2012, ICES
521   cooperative research report No. 321.

522 Boero, F., Bouillon, J., Gravili, C., Miglietta, M., Parsons, T., Piraino, S., 2008. Gelatinous
523   plankton: irregularities rule the world (sometimes). Mar. Ecol. Prog. Ser. 356, 299–310.

524 Brewer, R.H., Feingold, J.S., 1991. The effect of temperature on the benthic stages of *Cyanea*
525   (Cnidaria, Scyphozoa), and their seasonal distribution in the Niantic River estuary,
526   Connecticut. J. Exp. Mar. Bio. Ecol. 152, 49–60.

527 Brotz, L., Cheung, W.W.L., Kleisner, K., Pakhomov, E., Pauly, D., 2012. Increasing jellyfish
528   populations: trends in large marine ecosystems. Hydrobiologia 690, 3–20.

529 Cargo, D.G., Rabenold, G.E., 1980. Observations on the asexual reproductive activities of the
530   sessile stages of the sea nettle *Chrysaora quinquecirrha* (scyphozoan). Estuaries 3, 20–
531   27.

532 Cargo, D.G., Schultz, L.P., 1967. Further observations on the biology of the sea nettle and
533   jellyfishes in Chesapeake Bay. Chesap. Sci. 8, 209–220.

534 Chen, J.K., Ding, G.W., 1983. Effect of temperature on the strobilation of jellyfish
535   (*Rhopilema esculenta Kishinouye* – scyphozoa, rhizostomeae). Acta Zool. Sin. 29, 195–
536   206.

537 Condon, R.H., Duarte, C.M., Pitt, K.A., Robinson, K.L., Lucas, C.H., Sutherland, K.R.,
538   Mianzan, H.W., Bogeberg, M., Purcell, J.E., Decker, M.B., Uye, S., Madin, L.P.,
539   Brodeur, R.D., Haddock, S.H.D., Malej, A., Parry, G.D., Eriksen, E., Quiñones, J.,
540   Acha, M., Harvey, M., Arthur, J.M., Graham, W.M., 2013. Recurrent jellyfish blooms
541   are a consequence of global oscillations. Proc. Natl. Acad. Sci. U. S. A. 110, 1000–5.

542 Connelly, S.R., Menge, B.A., Roughgarden, J., 2001. A latitudinal gradient in recruitment of
543   intertidal invertebrates in the Northeast Pacific Ocean. Ecology 82, 1799–1813.

544 Dawson, M.N., Jacobs, D.K., 2001. Molecular evidence for cryptic species of *Aurelia aurita*
545   (Cnidaria, Scyphozoa). Biol. Bull. 200, 92–6.

546 Dawson, M.N., Martin, L.E., 2001. Geographic variation and ecological adaptation in *Aurelia*
547   (Scyphozoa , Semaeostomeae): some implications from molecular phylogenetics.
548   Hydrobiologia 451, 259–273.

549 Dawson, M.N., Martin, L.E., Penland, L.K., 2001. Jellyfish swarms, tourists, and the Christ-
550   child 131–144.

551 Dong, Z., Liu, D., Keesing, J.K., 2010. Jellyfish blooms in China: Dominant species, causes
552   and consequences. Mar. Pollut. Bull. 60, 954–63.

553 Doyle, T.K., Houghton, J.D.R., Buckley, S.M., Hays, G.C., Davenport, J., 2007. The broad-
554   scale distribution of five jellyfish species across a temperate coastal environment.
555   Hydrobiologia 579, 29–39.

556 Edwards, C., 1965. The hydroid and the medusa *Neoturris pileata*. J. Mar. Biol. Assoc.
557   United Kingdom 45, 443 – 468.

558 Fu, Z., Shibata, M., Makabe, R., Ikeda, H., Uye, S., 2014. Body size reduction under
559   starvation , and the point of no return , in ephyrae of the moon jellyfish *Aurelia aurita*.
560   Mar. Ecol. Prog. Ser. 510, 255–263.

561 Fuchs, B., Wang, W., Graspeuntner, S., Li, Y., Insua, S., Herbst, E.-M., Dirksen, P., Böhm,
562   A.-M., Hemmrich, G., Sommer, F., Domazet-Lošo, T., Klostermeier, U.C., Anton-
563   Erxleben, F., Rosenstiel, P., Bosch, T.C.G., Khalturin, K., 2014. Regulation of Polyp-to-
564   Jellyfish transition in *Aurelia aurita*. Curr. Biol. in press, 1–11.

565 Gatz, A.J., Kennedy, V.S., Mihursky, J.A., 1973. Effects of temperature on activity and
566   mortality of the scyphozoan medusa, *Chrysaora quinquecirrha*. Chesap. Sci. 14, 171–
567   180.

568 Grondahl, F., 1988. A comparative ecological study on the scyphozoans *Aurelia aurita*,
569   *Cyanea capillata* and *C. lamarckii* in the Gullmar Fjord, western Sweden, 1982 to 1986.
570   Mar. Biol. 97, 541–550.

571 Han, C.-H., Uye, S.-I., 2010. Combined effects of food supply and temperature on asexual
572   reproduction and somatic growth of polyps of the common jellyfish *Aurelia aurita* s.1.
573   Plankt. Benthos Res. 5, 98–105.

574 Hay, S.J., Hislop, J.R.G., Shanks, A.M., 1990. North Sea Scyphomedusae; summer
575   distribution, estimated biomass and significance particularly for 0-group Gadoid fish.
576   Netherlands J. Sea Res. 25, 113–130.

577 Hernroth, L., Grondahl, F., 1985. On the biology of *Aurelia aurita* (L.): 2. Major factors
578   regulating the occurrence of ephyrae and young medusae in the Gullmar Fjord, western
579   Sweden. Bull. Mar. Sci. 37, 567–576.

580 Holst, S., 2012. Effects of climate warming on strobilation and ephyra production of North
581   Sea scyphozoan jellyfish. Hydrobiologia 690, 127–140.

582 Holst, S., Jarms, G., 2010. Effects of low salinity on settlement and strobilation of scyphozoa
583   (Cnidaria): Is the lion's mane *Cyanea capillata* (L.) able to reproduce in the brackish
584   Baltic Sea? Hydrobiologia 645, 53–68.

585 Ishii, H., Watanabe, T., 2003. Experimental study of growth and asexual reproduction in
586   *Aurelia aurita* polyps. Sess. Org. 20, 69–73.

587 Lee, P.L.M., Dawson, M.N., Neill, S.P., Robins, P.E., Houghton, J.D.R., Doyle, T.K., Hays,
588   G.C., 2013. Identification of genetically and oceanographically distinct blooms of
589   jellyfish. J. R. Soc. Interface 10.

590 Liu, W., Lo, W., Purcell, J.E., Chang, H., 2009. Effects of temperature and light intensity on
591   asexual reproduction of the scyphozoan, *Aurelia aurita* (L.) in Taiwan. Hydrobiologia
592   616, 247–258. doi:10.1007/s10750-008-9597-4

593 Loeb, M.J., 1974. Strobilation in the Chesapeake bay sea nettle *Chrysaora quinquecirrha* II
594   Partial characterization of the neck inducing factor from strobilating polyps. Comp.
595   Biochem. Physiol. 47A, 291 – 301.

596 Loeb, M.J., Blanquet, R.S., 1974. Strobilation in the Chesapeake bay sea nettle *Chrysaora*
597   *quinquecirrha* III Dissociation of the neck-inducing factor from strobilating polyps.
598   Comp. Biochem. Physiol. 49A, 423 – 432.

599   Lotan, A., Fine, M., 1994. Synchronization of the life cycle and dispersal pattern of the
600   tropical invader scyphomedusan *Rhopilema nomadica* is temperature dependent 109,
601   59–65.

602   Lucas, C.H., 2001. Reproduction and life history strategies of the common jellyfish, *Aurelia*
603   *aurita*, in relation to its ambient environment. Hydrobiologia 451, 229–246.

604   Lucas, C.H., Graham, W.M., Widmer, C.L., 2012. Jellyfish life histories: role of polyps in
605   forming and maintaining scyphomedusa populations. Adv. Mar. Biol. 63, 133–96.

606   Lucas, C.H., Williams, J.A., 1994. Population dynamics of the scyphomedusa *Aurelia aurita*
607   in Southampton Water. J. Plankton Res. 16, 879–895.

608   Lynam, C.P., Attrill, M.J., Skogen, M.D., 2010. Climatic and oceanic influences on the
609   abundance of gelatinous zooplankton in the North Sea. J. Mar. Biol. Assoc. United
610   Kingdom 90, 1153–1159.

611   Lynam, C.P., Hay, S.J., Brierley, A.S., 2005. Jellyfish abundance and climatic variation:
612   contrasting responses in oceanographically distinct regions of the North Sea, and
613   possible implications for fisheries. J. Mar. Biol. Assoc. UK 85, 435–450.

614   Lynam, C.P., Hay, S.J., Brierley, A.S., 2004. Interannual variability in abundance of North
615   Sea jellyfish and links to the North Atlantic Oscillation. Limnol. Oceanogr. 49, 637–
616   643.

617   Mathis, M., Pohlman, T., 2014. Projection of physical conditions in the North Sea for the 21st
618   century. Clim. Res. 61, 1 – 17.

619   Melica, V., Invernizzi, S., Caristi, G., 2014. Logistic density-dependent growth of an *Aurelia*
620   *aurita* polyps population. Ecol. Modell. 291, 1–5.

621   Mills, C.E., 2001. Jellyfish blooms: are populations increasing globally in response to
622   changing ocean conditions? Hydrobiologia 451, 55–68.

623   Mills, C.E., 1984. Density is altered in hydromedusae and ctenophores in response to changes
624   in salinity. Biol. Bull. 166, 206–215.

625   NBN, 2016. National Biodiversity Network database [WWW Document]. URL
626   http://www.nbn.org.uk/

627   Nickell, T., Davidson, K., Fox, C., Miller, P., Hays, G., 2010. Developing the capacity to
628   monitor the spatial and temporal distribution of jellyfish in western Scottish waters.
629   London.

630   Ottersen, G., Planque, B., Belgrano, A., Post, E., Reid, P., Stenseth, N., 2001. Ecological
631   effects of the North Atlantic Oscillation. Oecologia 128, 1–14.

632   Pascual, M., Fuentes, V., Canepa, A., Atienza, D., Gili, J.-M., Purcell, J.E., 2014.
633   Temperature effects on asexual reproduction of the scyphozoan *Aurelia aurita* s .l.:

634   differences between exotic (Baltic and Red seas) and native (Mediterranean Sea)
635   populations. Mar. Ecol. 1 – 9.

636 Purcell, J., Hoover, R., Schwarck, N., 2009. Interannual variation of strobilation by the
637   scyphozoan *Aurelia labiata* in relation to polyp density, temperature, salinity, and light
638   conditions in situ. Mar. Ecol. Prog. Ser. 375, 139–149.

639 Purcell, J., Uye, S., Lo, W., 2007. Anthropogenic causes of jellyfish blooms and their direct
640   consequences for humans: a review. Mar. Ecol. Prog. Ser. 350, 153–174.

641 Purcell, J.E., 2007. Environmental effects on asexual reproduction rates of the scyphozoan
642   *Aurelia labiata*. Mar. Ecol. Prog. Ser. 348, 183–196.

643 Purcell, J.E., Atienza, D., Fuentes, V., Olariaga, A., Tilves, U., Colahan, C., Gili, J.-M., 2012.
644   Temperature effects on asexual reproduction rates of scyphozoan species from the
645   northwest Mediterranean Sea. Hydrobiologia 690, 169–180.

646 Purcell, J.E., White, J.R., Nemazie, D.A., Wright, D.A., 1999. Temperature, salinity and food
647   effects on asexual reproduction and abundance of the scyphozoan *Chrysaora*
648   *quinquecirrha*. Mar. Ecol. Prog. Ser. 180, 187–196.

649 Richardson, A.J., Bakun, A., Hays, G.C., Gibbons, M.J., 2009. The jellyfish joyride: causes,
650   consequences and management responses to a more gelatinous future. Trends Ecol.
651   Evol. 24, 312–22.

652 Robinson, K.L., Graham, W.M., 2013. Long-term change in the abundances of northern Gulf
653   of Mexico scyphomedusae *Chrysaora* sp. and *Aurelia* spp. with links to climate
654   variability. Limnol. Oceanogr. 58, 235–253.

655 Russell, F.S., 1970. The Medusae of the British Isles. II. Pelagic scyphozoa with a
656   supplement to the first volume on Hydromedusae. Cambridge University Press,
657   Cambridge.

658 Schiariti, A., Morandini, A., Jarms, G., von Glehn Paes, R., Franke, S., Mianzan, H., 2014.
659   Asexual reproduction strategies and blooming potential in Scyphozoa. Mar. Ecol. Prog.
660   Ser. 510, 241–253.

661 Schroth, W., Jarms, G., Streit, B., Schierwater, B., 2002. Speciation and phylogeography in
662   the cosmopolitan marine moon jelly, *Aurelia* sp. BioMed Cent. Evol. Biol. 10, 1–10.

663 Schulz, A., 2009. North Sea Atlas - Temperature, salinity, density and heat content - monthly
664   means for the period 1902 to 1954. Bundesamt für Seeschifffahrt und Hydrographie,
665   Hamburg und Rostock.

666 Spangenberg, D.B., 1967. Iodine induction of metamorphosis in *Aurelia*. J. Exp. Zool. 165,
667   441–449.

668 Suguira, Y., 1965. On the effects of temperature on the strobilation of *Mastigias papua*. Biol.
669   Bull. 128, 493–496.

670   Thein, H., Ikeda, H., Uye, S.-I., 2013. Ecophysiological characteristics of podocysts in
671   *Chrysaora pacifica* (Goette) and *Cyanea nozakii* Kishinouye (Cnidaria: Scyphozoa:
672   Semaeostomeae): Effects of environmental factors on their production, dormancy and
673   excystment. J. Exp. Mar. Bio. Ecol. 446, 151 – 158.

674   Turrell, W.R., Slesser, G., Payne, R., Adams, R.D., Gillibrand, P.A., 1996. Hydrography of
675   the East Shetland Basin in relation to decadal North Sea variability hydrography of the
676   East Shetland Basin. ICES J. Mar. Sci. 53, 899–916.

677   Ver-Hoef, J.M., Boveng, P.L., 2007. Quasi-Poisson vs. negative binomial regression: how
678   should we model overdispersed count data? Ecology 88, 2766–72.

679   Verwey, J., 1942. Die periodizität im auftreten und die aktiven und passiven bewegungen der
680   quallen. Arch. Neerl. Zool. 6, 363–468.

681   Widmer, C.L., 2008a. Life cycle of *Chrysaora fuscescens* (Cnidaria: Scyphozoa) and a key to
682   sympatric ephyrae. Pacific Sci. 62, 71–82.

683   Widmer, C.L., 2008b. How to keep jellyfish in aquariums: an introductory guide for
684   maintaining healthy jellies. Wheatmark, Tucson.

685   Wiesenthal, A.A., 2012. The effect of temperature and food availability on the asexual
686   reproduction of *Aurelia aurita* and *Cyanea capillata*. University of St. Andrews.

687   Willcox, S., Moltschaniwskyj, N.A., Crawford, C., 2007. Asexual reproduction in
688   scyphistomae of *Aurelia* sp.: Effects of temperature and salinity in an experimental
689   study. J. Exp. Mar. Bio. Ecol. 353, 107–114.

690   You, K., Ma, C., Gao, H., Li, F., Zhang, M., Wang, B., Wei, R., 2008. The Effects of
691   temperature decrease on the scyphistomae strobilation of jellyfish , *Rhopilema*
692   *esculentum* Kishinouye. J. world Aquac. Soc. 39, 706–711.

693   Zuur, A.F., Hilbe, J.M., Ieno, E.N., 2013. A Beginner's Guide to GLM and GLMM with R:
694   A frequentist and Bayesian perspective for ecologists, 1st ed. Highland Statistics Ltd.,
695   Newburgh.

696

697

698

699

700
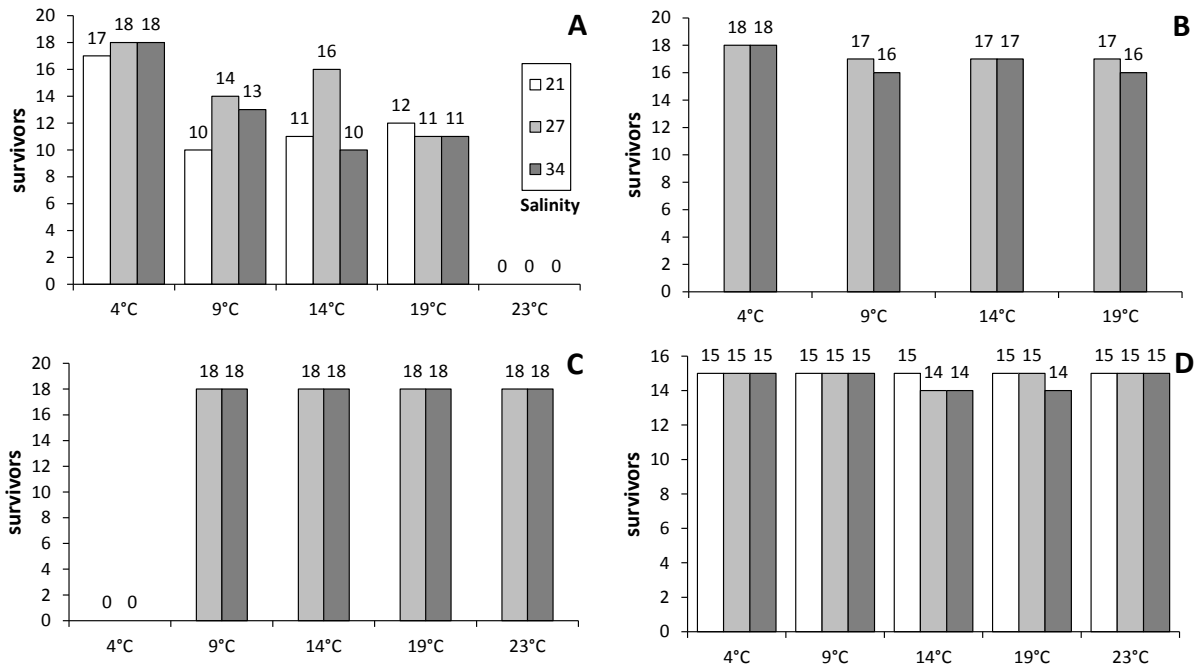
Figure 1. Total number of surviving scyphistomae at the terminus of the experiments. A. *C. capillata*. B *C.*

lamarckii. C *Ch. hysoscella*. D *A. aurita*. Note that the starting n was 15 for *A. aurita* and 18 for all other spp.

Note that the starting n was 15 for *A. aurita* and 18 for all other spp.

704

705

706

707

708
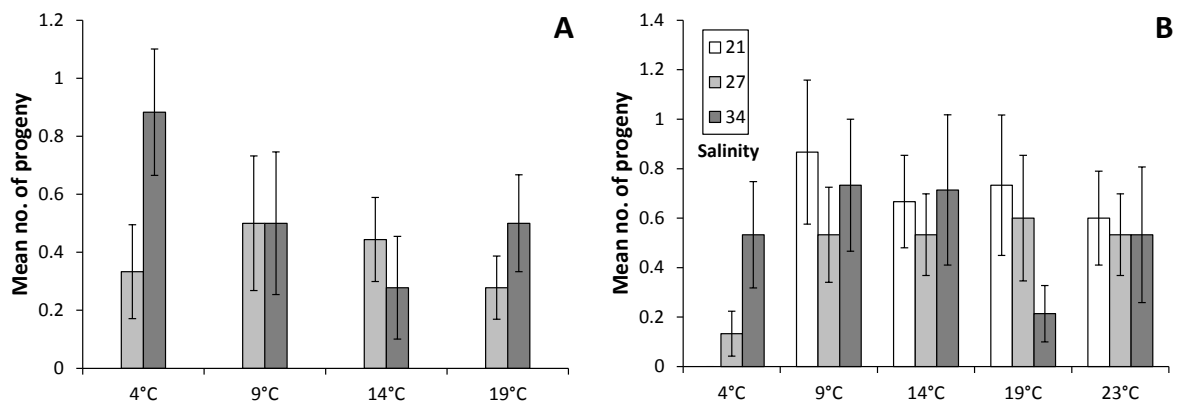
Figure 2. Mean numbers of progeny produced per scyphistoma. Error bars = standard error of the mean. A *C.*

lamarckii. B *A. aurita*.

711

712
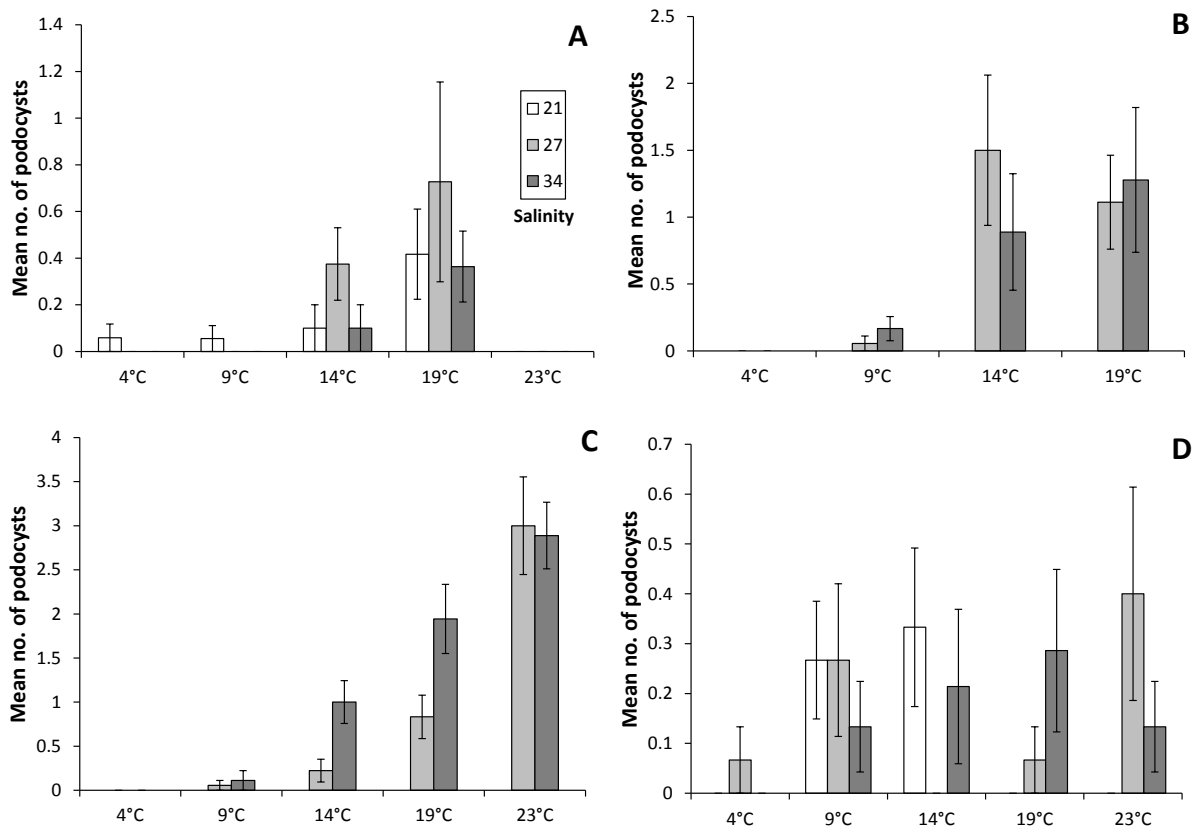
713

714   Figure 3.   Mean number of podocysts produced per scyphistoma.   A. *C. capillata*. B *C. lamarckii*. C *Ch.*

715   *hysoscella*.   D *A. aurita*.   Error bars = standard error of the mean.

716
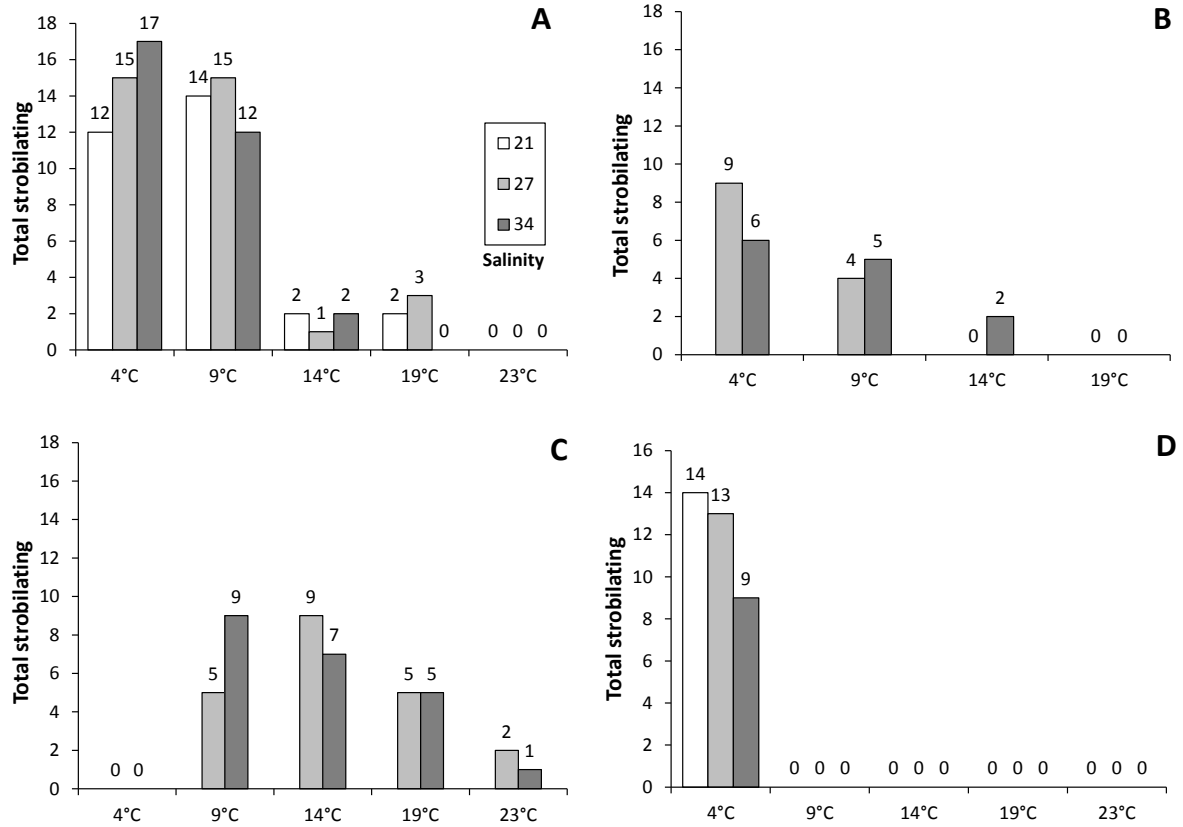
717

718   Figure 4. Total number of scyphistomae that strobilated during the experiments. A. *C. capillata*. B *C.*
719   *lamarckii*. C *Ch. hysoscella*. D *A. aurita*.

720

721

722

723

724
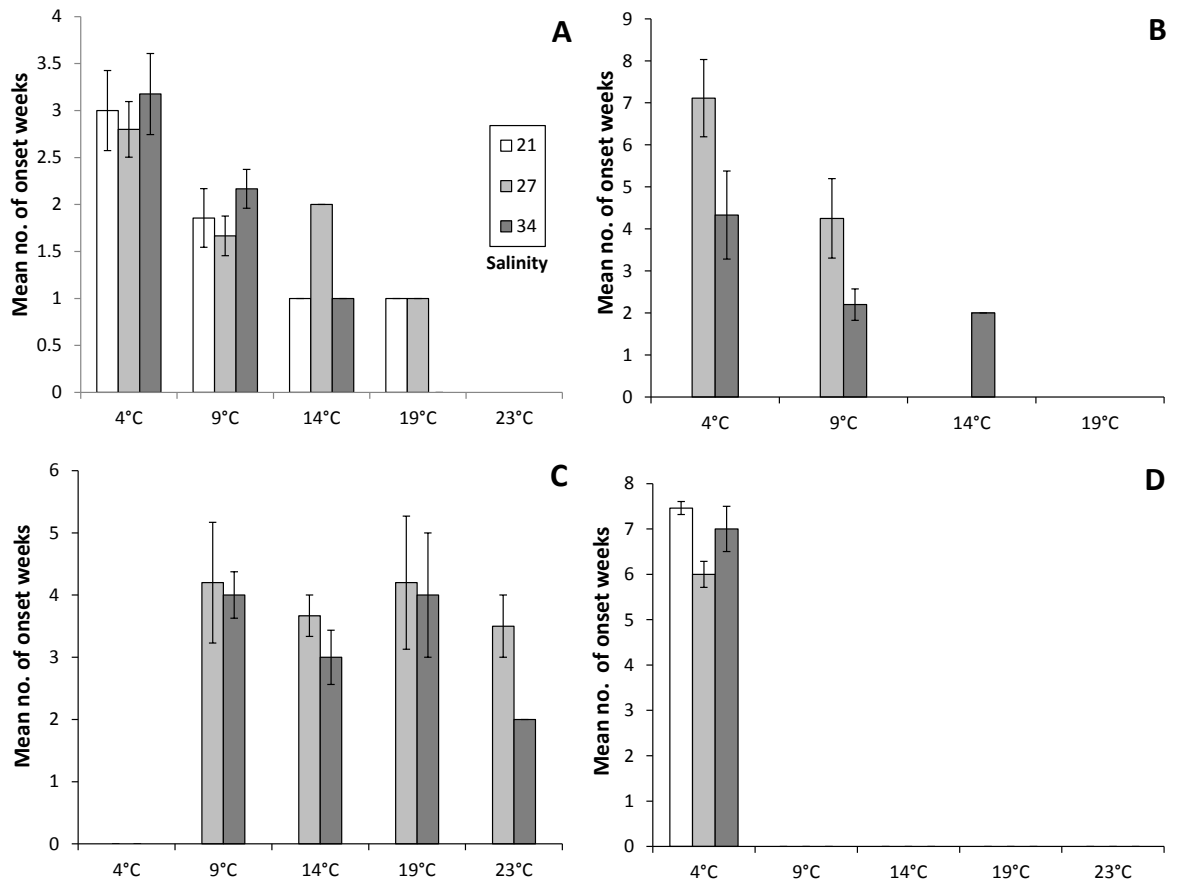
Figure 5. Mean number of weeks before the onset of strobilation. Error bars = standard error of the mean.

726   A. *C. capillata*. B *C. lamarckii*. C *Ch. hysoscella*. D *A. aurita*.

727

728

729

730

731
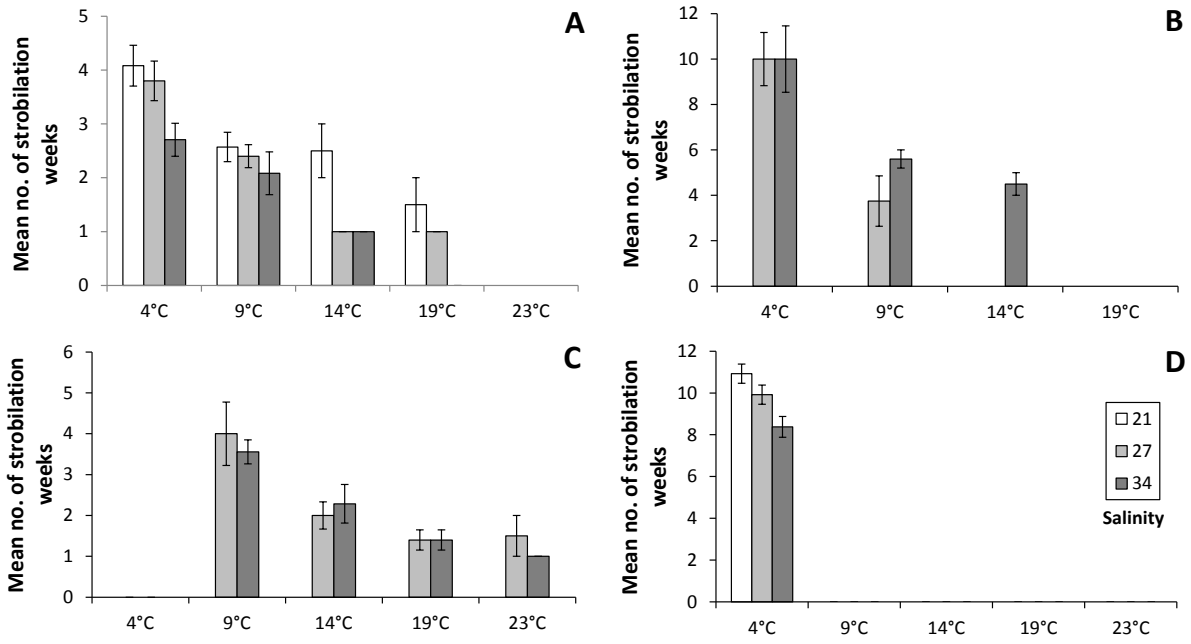
Figure 6. Mean number of weeks to complete the process of strobilation. Error bars = standard error of the mean. A. *C. capillata*. B *C. lamarckii*. C *Ch. hysoscella*. D *A. aurita*.

734

735
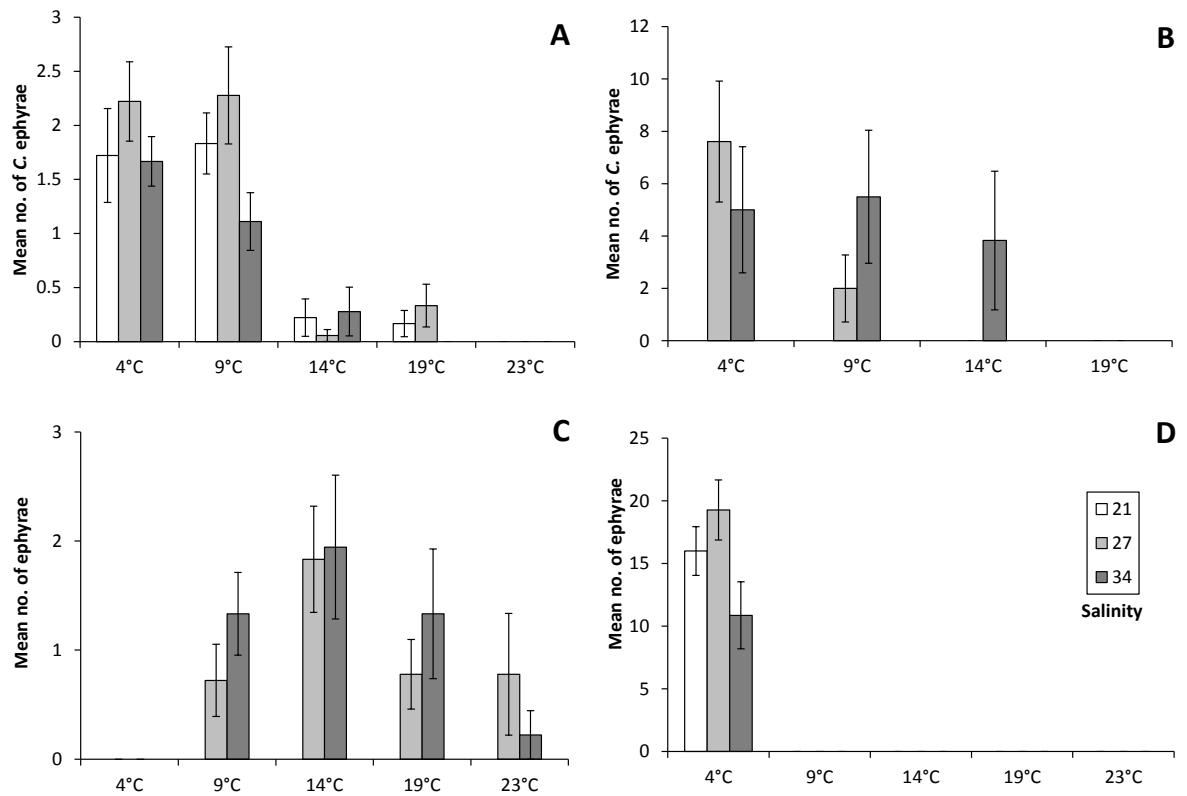
736

737   Figure 7. Mean number of ephyrae produced per scyphistoma. Error bars = standard error of the mean. A. *C.*
738   *capillata*. B *C. lamarckii*. C *Ch. hysoscella*. D *A. aurita*.
739

740

741
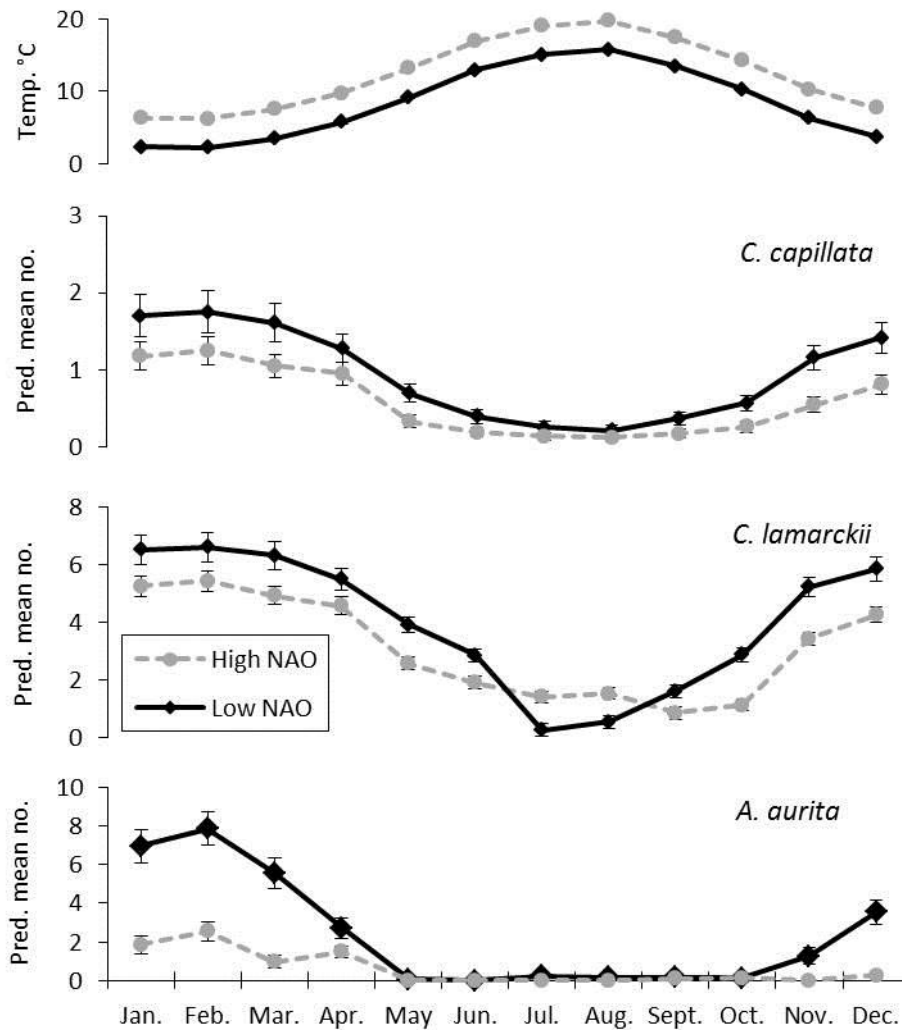
742

743   Figure 8. Hypothetical model of the mean number of ephyrae produced per scyphistoma of *C. capillata*, *C.*
744   *lamarckii* and *A. aurita* under high and low NAO sea surface temperature conditions. GLM predictions were
745   derived from present experimental results made at salinity 34, confidence limits = ± SE.
746

Table 1. Summary of temperatures and salinities tested for each species, n refers to the total
number of scyphistomae incubated per temperature and salinity combination.

Species	Temperature ±1 °C	Salinity	n
Cyanea capillata	4, 9, 14, 19, 23	21, 27, 34	18
Cyanea lamarckii	4, 9, 14, 19	27, 34	18
Chrysaora hysoscella	4, 9, 14, 19, 23	27, 34	18
Aurelia aurita	4, 9, 14, 19, 23	21, 27, 34	15

747

Table 2. Summary of best fitting generalized linear models for the results of experiments testing the effects of temperature (T) and salinity (S) on asexual reproductive output, strobilation and mortality of scyphistomae. The full model was: Response Variable ~ T + S + T x S + ε.

Species	Response variable	Significant predictor variables	Family	Link	Explained deviance (%)
Cyanea capillata	Surviving scyphistomae	~ T	Binomial	Logit	44.8
	Progeny scyphistomae	None produced			
	Podocysts produced	~ T	Poisson	Log	43.1
	Strobilating scyphistomae	~ T	Binomial	Logit	49.2
	Onset of strobilation	~ T	Poisson	Log	27.7
	Strobilation duration	~ T + S	Poisson	Log	35.6
	Ephyrae produced	~ T + S	Poisson	Log	53.5
Cyanea lamarcki	Surviving scyphistomae	None	Binomial	Logit	NA
	Progeny scyphistomae	None	Poisson	Log	NA
	Podocysts produced	~ T	Poisson	Log	31.0
	Strobilating scyphistomae	~ T	Binomial	Logit	23.0
	Onset of strobilation	~ T + S	Poisson	Log	53.0
	Strobilation duration	~ T	Poisson	Log	48.0
	Ephyrae produced	~ T + S + T x S	Poisson	Log	29.0
Chrysaora hysoscella	Surviving scyphistomae	~ T	Binomial	Logit	42.0
	Progeny scyphistomae	None produced			
	Podocysts produced	~ T + S + T x S	Poisson	Log	60.2
	Strobilating scyphistomae	~ T	Binomial	Logit	18.8
	Onset of strobilation	None	Poisson	Log	NA
	Strobilation duration	~ T	Poisson	Log	50.9
	Ephyrae produced	~ T + S + T x S	Poisson	Log	23.1
Aurelia aurita	Surviving scyphistomae	None	Binomial	Logit	NA
	Progeny scyphistomae	~ T + S + T x S	Poisson	Log	12.0
	Podocysts produced	~ T + S + T x S	Poisson	Log	24.0
	Strobilating scyphistomae	~ T + S	Binomial	Logit	80.0
	Onset of strobilation	None	Poisson	Log	NA
	Strobilation duration	None	Poisson	Log	NA
	Ephyrae produced	~ T + S	Poisson	Log	86.0

Table 3. Summary of spearman correlation results for experiments testing the effects of temperature and salinity on asexual reproductive output, strobilation and mortality of scyphistomae; n= 18 for *C. capillata*, *Ch. hysoscella* and *A. lamarckii*, and n = 15 for *A. aurita*. Significant correlations are highlighted in bold.

Species	Response variable	Predictor variable	$r_s=$	$p =$
C. capillata	Surviving scyphistomae	Temp	**-0.633**	**< 0.001**
		Sal	-0.056	0.357
	Podocysts produced	Temp	**0.354**	**< 0.001**
		Sal	-0.029	0.702
	Strobilating scyphistomae	Temp	**-0.68**	**< 0.001**
		Sal	0.009	0.876
	Onset of strobilation	Temp	**-0.494**	**< 0.001**
		Sal	0.132	0.203
	Strobilation duration	Temp	**-0.498**	**< 0.001**
		Sal	-0.218	**0.033**
	Ephyrae produced	Temp	**-0.667**	**< 0.001**
		Sal	-0.021	0.728
C. lamarckii	Surviving scyphistomae	Temp	-0.108	0.195
		Sal	-0.061	0.470
	Podocysts produced	Temp	**0.428**	**< 0.001**
		Sal	-0.013	0.874
	Progeny scyphistomae	Temp	-0.064	0.44
		Sal	0.071	0.394
	Strobilating scyphistomae	Temp	**-0.419**	**< 0.001**
		Sal	0.0	1.0
	Onset of strobilation	Temp	**-0.604**	**0.001**
		Sal	**-0.586**	**0.001**
	Strobilation duration	Temp	**-0.699**	**< 0.001**
		Sal	-0.103	0.615
	Ephyrae produced	Temp	**-0.409**	**< 0.001**
		Sal	0.015	0.851
Ch. hysoscella	Surviving scyphistomae	Temp	**0.707**	**< 0.001**
		Sal	0.0	1.0
	Podocysts produced	Temp	**0.659**	**< 0.001**
		Sal	**0.204**	**0.013**
	Strobilating scyphistomae	Temp	0.018	0.806
		Sal	0.013	0.862
	Onset of strobilation	Temp	-0.187	0.229
		Sal	-0.099	0.526
	Strobilation duration	Temp	**-0.668**	**< 0.001**
		Sal	0.116	0.456
	Ephyrae produced	Temp	0.035	0.631
		Sal	0.019	0.794
A. aurita	Surviving scyphistomae	Temp	-0.027	0.682
		Sal	-0.094	0.155
	Podocysts produced	Temp	0.052	0.433
		Sal	0.022	0.736
	Progeny scyphistomae	Temp	0.107	0.108
		Sal	-0.036	0.590
	Strobilating scyphistomae	Temp	**-0.6172**	**< 0.001**
		Sal	-0.074	0.267
	Onset of strobilation	Temp	NA	NA
		Sal	-0.222	0.206
	Strobilation duration	Temp	NA	NA
		Sal	**-0.542**	**0.001**
	Ephyrae produced	Temp	**-0.604**	**< 0.001**
		Sal	-0.058	0.386

Table S1. *Cyanea capillata*: descriptive statistics for results of an 8 week experiment testing the effects of 15 different combinations of temperature and salinity on asexual reproductive output of *C. capillata* scyphistomae.

Salinity	Temperature °C				
	4	9	14	19	23
Total surviving scyphistomae					
21	17	10	11	12	0
27	18	14	16	11	0
34	18	13	10	11	0
Mean no. of podocysts produced scyphistoma^{-1} (SE)					
21	0.06 (0.06)	0.06 (0.06)	0.10 (0.1)	0.42 (0.19)	0
27	0	0	0.30 (0.2)	0.73 (0.43)	0
34	0	0	0.10 (0.1)	0.35 (0.15)	0
Total strobilating scyphistomae					
21	12	14	2	2	0
27	15	15	1	3	0
34	17	12	2	0	0
Mean number of weeks before strobilation initiated					
21	3.0 (0.43)	1.9 (0.31)	1.0 (0.0)	1.0 (0.0)	NA
27	2.8 (0.29)	1.7 (0.21)	2.0 (0.0)	1.0 (0.0)	NA
34	3.2 (0.43)	2.2 (0.21)	1.0 (0.0)	NA	NA
Mean strobilation duration in weeks (SE)					
21	4.1 (0.38)	2.6 (0.27)	2.5 (0.5)	1.5 (0.5)	NA
27	3.8 (0.37)	2.4 (0.21)	1.0 (NA)	1.0 (0.0)	NA
34	2.7 (0.31)	2.1 (0.39)	1.0 (0.0)	NA	NA
Mean number of ephyrae scyphistoma^{-1} (SE)					
21	1.7 (0.43)	1.8 (0.28)	0.2 (0.17)	0.2 (0.12)	0
27	2.2 (0.37)	2.3 (0.45)	0.1 (0.06)	0.3 (0.19)	0
34	1.7 (0.23)	1.1 (0.27)	0.3 (0.23)	0.0 (0.0)	0
Total number of ephyrae produced treatment group^{-1}					
21	31	33	4	3	0
27	40	41	5	6	0
34	30	20	1	0	0

*The format of this table is modelled after Purcell 2007.

760
761
762
763
764
765
766
767
768
769
770
771
772

Table S2. *Cyanea lamarckii*: descriptive statistics for results of an 8 week experiment testing the effects of 15 different combinations of temperature and salinity on asexual reproductive output of *C. lamarckii* scyphistomae.

Salinity	Temperature °C			
	4	9	14	19
Total surviving scyphistomae				
27	18	17	17	17
34	18	16	17	16
Mean no. of progeny scyphistomae produced parent scyphistoma^{-1} (SE)				
27	0.33 (0.16)	0.5 (0.23)	0.44 (0.15)	0.28 (0.11)
34	0.83 (0.22)	0.5 (0.25)	0.28 (0.18)	0.50 (0.18)
Mean no. of podocysts produced scyphistoma^{-1} (SE)				
27	0	0.06 (0.06)	1.50 (0.56)	1.11 (0.35)
34	0	0.17 (0.09)	0.89 (0.44)	1.28 (0.54)
Total strobilating scyphistomae				
27	9	4	0	0
34	6	5	2	0
Mean number of weeks before strobilation initiated				
27	7.11 (0.92)	4.25 (0.95)	NA	NA
34	4.33 (1.05)	2.2 (0.38)	2.0 (0.0)	NA
Mean strobilation duration in weeks (SE)				
27	10.0 (1.17)	3.75 (1.11)	NA	NA
34	10.0 (1.46)	5.60 (0.4)	4.50 (0.5)	NA
Mean number of ephyrae scyphistoma^{-1} (SE)				
27	7.6 (2.31)	2.0 (1.28)	0	0
34	5.0 (2.24)	5.5 (2.54)	3.83 (2.65)	0
Total number of ephyrae produced treatment group^{-1}				
27	137	36	0	0
34	99	99	69	0

773
774
775
776
777
778
779
780
781
782
783
784
785
786
787
788
789
790
791
792

Table S3. *Chrysaora hysoscella*: descriptive statistics for results of an 8 week experiment testing the effects of 15 different combinations of temperature and salinity on asexual reproductive output of *C. lamarckii* scyphistomae.

Salinity	Temperature °C				
	4	9	14	19	23
Total surviving scyphistomae					
27	0	18	18	18	18
34	0	18	18	18	18
Mean no. of podocysts produced scyphistoma^{-1} (SE)					
27	0	0.06 (0.06)	0.22 (0.13)	0.83 (0.25)	3.0 (0.55)
34	0	0.11 (0.11)	1.0 (0.24)	1.94 (0.39)	2.9 (0.38)
Total strobilating scyphistomae					
27	0	5	9	5	2
34	0	9	7	5	1
Mean number of weeks before strobilation initiated					
27	NA	4.2 (0.97)	3.6 (0.33)	4.2 (1.07)	3.5 (0.5)
34	NA	4.0 (0.37)	3.0 (0.44)	4.0 (1.0)	2.0 (NA)
Mean strobilation duration in weeks (SE)					
27	NA	4.0 (0.78)	2.0 (0.33)	1.4 (0.25)	1.5 (0.5)
34	NA	3.5 (0.29)	2.2 (0.47)	1.4 (0.25)	1.0 (NA)
Mean number of ephyrae scyphistoma^{-1} (SE)					
27	0	0.72 (0.33)	1.83 (0.49)	0.78 (0.32)	0.79 (0.56)
34	0	1.33 (0.38)	1.94 (0.66)	1.33 (0.59)	0.22 (0.22)
Total number of ephyrae produced treatment group^{-1}					
27	0	13	33	14	14
34	0	24	35	24	4

793
794
795
796
797
798
799
800
801
802
803
804
805
806
807
808
809
810
811
812
813
814
815

Table S4. *Aurelia aurita*: descriptive statistics for results of an 8 week experiment testing the effects of 15 different combinations of temperature and salinity on asexual reproductive output.

Salinity	Temperature °C				
	4	9	14	19	23
Total surviving scyphistomae					
21	15	15	15	15	15
27	15	15	14	15	15
34	15	15	14	14	15
Mean no. of progeny scyphistoma produced scyphistoma^{-1} (SE)					
21	0 (0.0)	0.8 (0.29)	0.6 (0.18)	0.7 (0.28)	0.6 (0.19)
27	0.1 (0.09)	0.5 (0.19)	0.5 (0.16)	0.6 (0.25)	0.5 (0.16)
34	0.5 (0.22)	0.7 (0.26)	0.7 (0.30)	0.2 (0.11)	0.5 (0.27)
Mean no. of podocysts produced scyphistoma^{-1} (SE)					
21	0.0	0.27 (0.12)	0.33 (0.16)	0.0	0.0
27	0.07 (0.07)	0.27 (0.15)	0.0	0.07 (0.07)	0.4 (0.21)
34	0.0	0.13 (0.09)	0.21 (0.16)	0.29 (0.16)	0.13 (0.09)
Total strobilating scyphistomae					
21	14	0	0	0	0
27	13	0	0	0	0
34	9	0	0	0	0
Mean number of weeks before strobilation initiated					
21	7.5 (0.14)	NA	NA	NA	NA
27	6.0 (0.3)	NA	NA	NA	NA
34	7.0 (0.5)	NA	NA	NA	NA
Mean strobilation duration in weeks (SE)					
21	10.9 (0.46)	NA	NA	NA	NA
27	9.9 (0.45)	NA	NA	NA	NA
34	8.4 (0.49)	NA	NA	NA	NA
Mean number of ephyrae scyphistoma^{-1} (SE)					
21	16.0 (1.9)	0	0	0	0
27	19.27 (2.4)	0	0	0	0
34	10.87 (2.6)	0	0	0	0
Total number of ephyrae produced treatment group^{-1}					
21	240	0	0	0	0
27	289	0	0	0	0
34	163	0	0	0	0

816
817

818

819

820