

1 **Improving Pacific Oyster (*Crassostrea gigas*, Thunberg, 1793) Production in**
2 **Mediterranean Coastal Lagoons: Validation of the growth model “ShellSIM” on**
3 **traditional and novel farming methods**

4
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12
13 **Abstract**

14 Bivalve farming is a major European aquaculture activity, representing 48.5% of total biomass
15 produced. Italy is one of the largest consumers of oysters but local production does not meet the
16 market demand. Italy has approximately 384 000 ha of shallow lagoons in its coastal area, already
17 devoted to extensive aquaculture activities which could also represent potential locations for Pacific
18 oyster (*Crassostrea gigas*, Thunberg, 1793) farming.

19 The aim of this study is to enhance Pacific oyster farming in shallow coastal lagoons by testing
20 novel farming technologies and validating an existing bioenergetic growth model (ShellSIM).

21 Commercial performance of Pacific oysters and associated environmental parameters were
22 monitored in two Sardinian coastal lagoons (San Teodoro and Santa Gilla, Italy). Oyster growth and
23 survival were compared during a production cycle for two rearing systems: traditional systems
24 (floating bags or lanterns) and Ortac units. The latter has not been previously tested in coastal
25 lagoons. Measured performances were compared with ShellSIM predictions to evaluate the model's
26 ability to predict growth and the potential production in other coastal lagoons.

27 Results showed that at the end of a six months cycle the oysters mean weight and Condition Index
28 were significantly higher (p value < 0.05) in floating bags than in Ortac, (55.8 ± 0.9 g and $50.1 \pm$
29 1.3 g; 4.6 ± 0.1 and 3.9 ± 0.1 respectively). Also, the minimum commercial size (40 g) was reached
30 by 98 % and 68 % of the oyster farmed in floating bags and Ortac units respectively. On the other
31 hand, oysters reared in the Ortac showed a higher survival than in the floating bags (95.8 ± 0.9 %
32 and 82.1 ± 3.4 %, respectively).

33 ShellSIM growth predictions were highly correlated with the observed data in both lagoons.
34 However, high values for RMSD indicated that ShellSIM predictions were significantly validated
35 for San Teodoro lagoon but not for Santa Gilla suggesting further tailoring to some environmental
36 conditions to produce more realistic growth predictions.

37 Results of this study indicate that both floating bags and Ortac system should be employed during
38 the production cycle to maximise oysters' survival and growth performances. Furthermore, this
39 study provides a new validated tool to farmers and stakeholders to monitor oysters performances
40 and estimate productivity in local waters.

41

42 **Keywords**

43 Pacific oysters farming; Shellfish growth model; Farming technologies;

44

45 **Abbreviations**

46 POS 1, POS 2 and POS 3 are the three chosen experimental position

48 **1. Introduction**

49 Italy is one of the main seafood consumers in Europe and amongst the World's top 10 importers,
50 estimated at 5.6 million US dollars in 2016 (FAO, 2016). Different species of shellfish, crustaceans
51 and fish are farmed using both extensive and intensive methods.

52 In 2016, in Italy most aquaculture companies were farming shellfish contributing to over 64% of the
53 total aquaculture production being the largest producer of Manila clam (*Venerupis philippinarum*
54 (Adams and Reeve, 1850)) and the third producer of Mediterranean mussel (*Mytilus*
55 *galloprovincialis* (Lamarck, 1819)) in Europe. A smaller production includes grooved carpet shell
56 (*Ruditapes decussatus* (Linnaeus, 1758)) and Pacific oyster (*Crassostrea gigas* (Thunberg, 1793))
57 (Eurofish, 2016; FAO, 2016). Pacific oyster is native to Japan and coastal regions of Asia, and due
58 to its wide adaptation range at different environmental conditions, is the most widespread cultured
59 oyster species in the world (Shatkin *et al.*, 1997).

60 In 2016, Europe produced 23000 tonnes of Pacific oysters 145 of which were of Italian origin (30
61 tonnes by a single Sardinian company (FAO 2011 - 2018 Fishstat.J)). Italy is one of the largest
62 consumers of oysters in Europe importing 6500 tonnes per year primarily from France, major
63 reason why there is an opportunity to diversify Italian shellfish farming in the future
64 (Sardegnaagricoltura.it, 2016, FAO, 2016). Sardinia has approximately 10000 ha of shallow coastal
65 lagoons representing the 2.6% of the total lagoon area in Italy (Bazzoni *et al.*, 2013). Many of these
66 lagoons are used for extensive finfish farming, but are potential sites for Pacific oyster farming.

67 Currently, depending on environmental conditions such as water depth, tidal range, water exchange
68 rates and bottom substrates, three main oyster farming methods are used: off-bottom culture, on-
69 bottom culture and suspended culture (Buestel *et al.*, 2009). In Sardinian lagoons suspended culture
70 is the most commonly used due to their environmental conditions. More specifically, floating bags
71 are designed to keep the oyster growing at the water surface where most of the food is available.
72 These are manufactured in square and diamond mesh patterns (from 4 to 23 mm), suspended on the

73 surface thanks to two floaters which allow periodic exposure of the oysters to the air to reduce
74 biofouling and strengthen the adductor muscle.

75 Amongst suspended oyster culture methods, several new farming tools have been recently
76 developed, for example Ortac units (ABBLOX), OysterGro© (OysterGro) and Zapco Tumbler
77 (Zapco Aquaculture). These aim to improve oyster production by reducing manual labour,
78 increasing growth rates and improving oysters' quality (i.e. shell shape). The Ortac system has been
79 employed in this study. The Ortac system consists of baskets made of polypropylene plastic and
80 divided in two halves. These operate attached to a trestle and, due to their shape, an up-welling
81 water flow is passively generated by the surrounding water currents. Furthermore, thanks to the
82 constant movement under currents actions, this system has been designed to reduce fouling
83 therefore requiring less handling.

84 Aside from environmental conditions, also the use of different grow-out gears affects oyster
85 performances as suggested by the recent study from Rankin *et al.*, (2018).

86 The only independent trial for Ortac conducted to date to compare growth, survival and
87 physiological performances with traditional bag systems used *Ostrea edulis* in Scotland (Francouer,
88 2017). Results of this study indicated that there were no significant differences in growth between
89 *Ostrea edulis* reared in the two different systems (Ortac units and traditional bags) but higher
90 survival was observed within the Ortac units. The study presented here is the first investigation and
91 comparison of the performance of the Ortac system in warmer climates with a smaller tidal range.

92 Much effort has been dedicated to generate and validate growth models for bivalves (Pouvreau *et*
93 *al.*, 2006). Most of the energy budget models predicting growth are net production models, which
94 assume that energy is immediately available for the animal maintenance while the rest is used for
95 growth or deposited as a reserve. Others are based on a dynamic energy budget approach (DEB)
96 where energy is first stored as a reserve and then used for different metabolic processes at a
97 catabolic rate (Kooijman, 2000; Pouvreau *et al.*, 2006; Ren and Ross, 2001; Beadman *et al.*, 2002).

98 Most shellfish energy budget models are only able to simulate growth for locations where they have

99 been calibrated, therefore restricting their use in areas with different environmental conditions
100 (Hawkins *et al.*, 2013; Dowd, 1997). ShellSIM growth model has been calibrated for 16 shellfish
101 species in different locations throughout Europe, the U.S.A, China, New Zealand, Malaysia and
102 Australia. This includes *Mytilus edulis* and *Crassostrea gigas* (ShellSIM, 2011; Hawkins *et al.*,
103 2013).

104 ShellSIM is based on principles of energy balance:

105

106
$$\text{Net energy balance} = \text{Energy ingested} - (\text{Energy egested} + \text{Energy excreted} + \text{Energy expended})$$

107

108 and was developed as a tool to be used by farmers, scientist and environmental regulators (Hawkins
109 *et al.*, 2013). Consequently, this growth model was considered to be appropriate to provide growth
110 forecasts in Sardinian coastal lagoons with suitable validation for local conditions.

111 The aim of this study is to validate this existing bioenergetic growth model in two ecologically
112 different Mediterranean coastal lagoons and for two different oyster farming systems: the Ortac and
113 the traditional floating bags and to compare the production efficiency between these two farming
114 systems.

115

116 **2. Materials and Methods**

117 2.1. Growth Trial: Ortac *vs* floating bags

118 This trial was performed between June 2017 and December 2017 in the lagoon of San Teodoro
119 (northeast Sardinia: 40°48' 38.08''N, 9°40'26.99''E). A total of 2400 Pacific oyster seeds (1.7 ± 0.1
120 g, 2.9 ± 0.2 cm), from a French hatchery located in the Loire region of France, were randomly
121 divided between 6 Ortac units and 6 Floating bags (200 individual per unit, mean total biomass per
122 unit was 260.7 ± 5.6 g). Thirty oysters from each unit were tagged with an underwater curing epoxy
123 resin (AquaScape) and biometric parameters were measured every two weeks (i.e. weight, length,
124 depth and width) using a portable scale (Steinberg SBS-LW-2000A, 0.01g) and callipers

125 (METRICA, 0.05 mm). At each sampling point, mortality was also recorded and 5 oysters per unit
126 were selected for dry weight measurements (Mo and Neilson, 1994) and Condition Index (CI)
127 calculations using the protocols described by Mo and Neilson (1994) for the dry weight and
128 Davenport and Chen (1987) and Walne and Mann (1975) for CI calculations where:

129

$$130 \quad CI = (\text{Dry weight meat (g)} / \text{Dry weight meat (g)} + \text{Dry weight shell (g)}) \times 100$$

131

132 The oyster culture systems were positioned in two rows of three Ortac units and three floating bags
133 (fig.1). Ortac units were mounted onto two trestles (3 units per trestle), floating bags were attached
134 to ropes as in the usual commercial setting of the Compagnia Ostricola Mediterranea, host of these
135 trials.

136 Oysters of both systems were cultured following the standard conditions of the company, with 24hrs
137 of air exposure every two weeks to prevent biofouling, changing of the floating bags mesh (4, 9, 14
138 and 19 mm) according to oysters' size, and based on the increasing Pacific Oysters biomass. Grading
139 was performed for both Ortac units and floating bags keeping similar biomass in both systems.

140

141 2.2. ShellSIM validation for floating Bag units (San Teodoro Lagoon)

142 A survey of the dominant currents in the area was conducted during the neap (minimum) and spring
143 (maximum) tides using GPS tracking drifters (drogues) (Cromey and Black, 2005). The drifters
144 were released at the same time at three different points, one hour before the tidal peak until one hour
145 after. During the survey the wind direction and speed was recorded by a fixed weather station (La
146 Crosse WS3650). These information was fed to ShellSIM model to account for the hydrodynamic
147 and ecological conditions within the lagoon.

148 Three floating bags per each area were stocked with *C. gigas* (838 ± 36.4 g, 811.5 ± 17.8 g and
149 709.8 ± 40.1 g total biomass) with a mean size in weight and length of 4.5 ± 0.3 g and 4.0 ± 0.2 cm

150 (POS 1), 4.5 ± 0.3 g and 3.9 ± 0.2 cm (POS 2), 3.9 ± 0.2 g and 3.9 ± 0.2 cm (POS 3). The oysters
151 were cultured following the standard procedures described above.

152 Sampling for oyster growth was performed monthly for 5 months. Each month 80 individuals unit⁻¹
153 were randomly measured for wet weight 30 of which also measured for length, depth and width.
154 Other 10 individuals unit⁻¹ were collected for dry weights measurements.

155 Environmental data: temperature (T, °C), salinity (Sal, ‰), dissolved oxygen (DO, mg L⁻¹), total
156 particulate matter (TPM, mg/L), particulate organic matter (POM, mg L⁻¹), particulate organic
157 carbon (POC, mg m⁻³) and Chlorophyll-a (Chl-a, µg L⁻¹) were collected in the immediate vicinity of
158 the farming gears. Temperature, salinity and dissolved oxygen were collected at a depth of 15 cm,
159 with a multiparametric probe (HACH HQ40d) and data loggers (HOBO: UTBI-001, U26-001 and
160 U24-002-C respectively for T, DO and Sal). Temperature data loggers were set-up to take
161 measurements every 30 minutes, while the Sal and DO probes measured values every 2 hrs.

162 Water for the TPM, POM, POC was collected using 1L pre-rinsed in sample water plastics bottles,
163 while 5L pre-rinsed in sample water plastic bottles were used to collect water for Chl-a analysis.

164 Laboratory analysis for TPM and POM were performed according to Hawkins *et al.* (2013), while
165 Chl-a analysis according to Lorenzen (1967). For POC measurements water was collected in 1 L
166 pre-rinsed plastic bottles then filtered in 47-mm diameter GF/F filters previously combusted at 450
167 °C. POC samples were analysed with a CEI Flash smart elemental analyser. The average values of
168 each environmental parameter were used to run the model, excluding September 2017, when no data
169 were collected due to farmers' activities and weather constraints.

170

171 2.3. ShellSIM validation for lantern systems (Santa Gilla Lagoon)

172 In order to validate the growth model in a different location, a growth trial was performed between
173 May 2017 and September 2017, in Santa Gilla lagoon (39°12'28.2''N 9°05'53.5''E). Three lanterns
174 with five compartments each and a mesh size of 3.5 x 5 mm, were stocked with 500 oysters per
175 compartment (mean weight = 4.4 ± 0.1 g; mean length = 3.6 ± 0.6 cm). The oysters were farmed

176 following the standard production protocols, grading and changing the mesh size according to
177 oysters' size and biomass. Growth was measured monthly when 70 individuals per lantern were
178 randomly sampled and weighted, 30 of which were also measured for shell length, depth and width.
179 Furthermore, 10 individuals per lantern were collected for dry weight measurements.
180 Environmental data sampling and analysis were conducted as described above. The monthly means
181 of all the environmental data were used to run the growth model.

182

183 2.4. ShellSIM Validation for Ortac units and floating Bags (San Teodoro Lagoon)

184 In order to validate the model for different gear types, a new experiment was set up in the lagoon of
185 San Teodoro (July 2017 – December 2017). In this occasion the model performance were also tested
186 on a different farming system i.e. the Ortac units.

187 Farming methods, growth measurements, sample collection and analysis of all environmental
188 parameters were conducted as described previously. A bi-weekly mean of all the environmental data
189 were used to run the ShellSIM, except for November and December, when data were collected only
190 one per month due to farmers' activities and weather constraints.

191

192 2.5. Statistical Analysis

193 Prior to analyses, data were tested for normality and homogeneity of variance. Weight gain,
194 biometrics measures differences, survival rate and Condition index differences, over time were
195 analysed by general linear model followed by a Tukey post-hoc test where significant differences
196 occurred.

197 End points of all biometrics measures, survival rate and condition index, were analysed by one-way
198 ANOVA followed by post-hoc Tukey's Multiple Comparison tests where significant differences
199 occurred.

200 To assess fitness between the prediction made by ShellSIM and observed data, Taylor diagrams and
201 skill scores (S) were used (Taylor, 2001). A Taylor diagram is a way to show graphically how well a

202 model prediction fits the observed data, using correlation, centred root mean square difference
203 (RMSD) and amplitude of their variation (standard deviations). The skill score proposed by Taylor
204 (2001) quantifies model performance against observed data.

205

206 **3. Results**

207 3.1. Growth Trial: Ortac vs floating bags

208 At the end of the of the production cycle (October to December), the Pacific oysters farmed in the
209 floating bags had a significantly higher weight and shell depth (p value = 0.001, 0.001 respectively)
210 to those in the Ortac units (55.8 ± 0.9 g, 50.1 ± 1.3 g; 26.6 ± 0.2 g, 24.2 ± 0.3 g) (figs.2a, 2c).
211 Oysters farmed into the Ortac units showed instead a significant higher growth in shell length (86.9
212 ± 1 , 75.4 ± 0.6 , p value = 0.001 Ortac and floating bags respectively), and shell width (46.2 ± 0.5
213 mm, 44.6 ± 0.4 mm, p value = 0.017) (fig.2b, d).

214 Survival was significantly higher ($p = 0.001$) in the Ortac units compared to the floating bags (95.8
215 $\pm 0.9\%$, $82.1 \pm 3.4\%$) (fig.3). The highest mortality occurred between June and July ($3.8 \pm 1\%$,
216 $16.3 \pm 3.3\%$ Ortac units and floating bags respectively).

217 The condition index at the end of the production cycle was significantly higher (p value = 0.001) in
218 the floating bags compared to the Ortac units (4.6 ± 0.1 , 3.9 ± 0.1) (fig.4) and the smallest
219 commercial size (40 g) was reached by the 98 % and 69 % of the oyster farmed in the floating bags
220 and Ortac units respectively (fig.5).

221

222 3.2. ShellSIM validation in San Teodoro Lagoon

223 Three areas with different current speed (POS1 0.15 m s⁻¹, POS2 0.07 m s⁻¹; POS 3 0.04 m s⁻¹) were
224 identified in San Teodoro. A decreasing speed gradient from the sea mouth to the internal part of
225 the lagoon was identified. Consequently, these areas were used as experimental locations to monitor
226 the oysters' growth and the environmental parameters required by the growth model.

227 Environmental data are illustrated in Table 1. ShellSIM predicted, during a 6 months production
228 cycle, a final weight and length of 19.7 g, 48.4 g and 121.6 g; 6.0 cm, 8.3 cm and 11.5 cm,
229 respectively for POS 1, POS 2 and POS 3.

230 The measured weight and length at the end of this production cycle, was 16.4 ± 1.1 g, 46.9 ± 2.1 g,
231 48.9 ± 1.5 g and 5.4 ± 0.3 cm, 8.2 ± 0.3 cm, 9 ± 0.2 cm, respectively in POS 1, POS 2 and POS 3.

232 Figure 6 shows that measured growth in weight and length, fitted the predicted growth curve in
233 POS 2, while in POS 1 and POS 3 ShellSIM overestimate the final mean growth in weight and
234 length respectively 20.5, 12.1, 148.8 and 27.9 %. The calculated skill score for the three different
235 areas indicate the best fitting between observed and predicted measures of weight and length,
236 respectively in POS 2 (S=1; S=1), POS 1 (S=0.87; S=0.81) and POS 3 (S=0.42; S=0.79).

237 Standard deviation, Centred Root Mean square difference (RMSD), correlation and the overall skill
238 score of the performance of the predicted growth curve to fit the observed data in the lagoon of San
239 Teodoro are shown in Figure 7 and Table 2.

240

241 3.3. ShellSIM validation in Santa Gilla Lagoon

242 Environmental data collected in the lagoon of Santa Gilla and their seasonal variations are
243 illustrated in Table 3.

244 The measured growth in weight and length (79.5 ± 1.8 g and 9.1 ± 0.1 cm) did not fit the predicted
245 growth curve (data not shown), and the calculated skill score indicates a very poor fit between
246 observed and predicted measures of weight and length, respectively S=0.003 and S=0.17 (Table 2).

247 Standard deviation, Centred Root Mean square difference (RMSD) and correlation are shown in
248 Figure 8 and Table 2.

249

250 3.4 ShellSIM Validation on Ortac vs floating bags

251 Environmental data collected to run ShellSIM and their changes are shown in Table 4.

252 In this trial, ShellSIM was run in POS 2 for two different farming systems. It predicted a growth of
253 48.6 g and 8.3 cm in weight and length respectively for the Ortac system, and a growth of 49.1 g
254 and 8.2 cm for the floating bags over a 6 months production cycle. At the end of this production
255 cycle, the measured weight and length were 50.1 ± 1.3 g and 8.7 ± 0.1 cm for the Ortac and $55.8 \pm$
256 0.9 g and 7.5 ± 0.1 cm for the floating bags. Figure 9 shows that during the production cycle, the
257 measured mean weight and length in the Ortac units and floating bags were underestimated by
258 ShellSIM, except for the final length farmed in the floating bags which was accurate. Indeed, in
259 November there was a change in trend of the model prediction, from underestimation to
260 overestimation (the model overestimated the final mean length of 8.2 %), while the final weight was
261 still overestimated by 11.9%.

262 Moreover Figure 9 shows that ShellSIM at the end point of the production cycle of the oysters reared
263 inside the Ortac units, unlike the rest of the predictions, slightly underestimated growth in weight and length
264 by 3% and 4.4% respectively.

265 The calculated skill score indicates that the best fitting between observed and predicted measures of
266 weight and length, respectively in Ortac ($S=0.95$, $S=0.93$) compared to floating bags ($S=0.90$,
267 $S=0.89$). Standard deviation, Centred Root Mean square difference (RMSD), correlation and the
268 overall skill score of the performance of the predicted growth curve to fit the observed data in this
269 trial in the San Teodoro lagoon are shown in Figure 10 and Table 2.

270

271 **4. Discussion and conclusions**

272 The results of this study provide new information to improve *C. gigas* farming and a growth
273 prediction tool in shallow coastal lagoons. The higher survival rate in the Ortac units for the first two
274 months and the higher growth in weight and CI in the floating bags, suggest a potential mixed use of
275 the two systems during the production cycle. Specifically, the Ortac units may be employed when
276 Pacific oysters are more susceptible to stress and during the stressful period (e.g smaller size and
277 hottest periods) and floating bags thereafter. In this way it would be possible to reduce the costs of

278 the equipment, avoiding several floating bags with different meshes depending on the oysters' sizes,
279 and a minor loss of individuals due to mortality.

280 There was no statistically significant difference in growth between Ortac units and floating bags, but
281 at the end of the production cycle there was a significant higher mean weight in the floating bags
282 than in the Ortac units. Comparison of these results with previous studies are difficult due to
283 difference in culture techniques and effects of other experimental variables such as initial oyster size
284 and the production season.

285 In many studies it is reported that shell morphology in bivalves is influenced by population density,
286 predation responses, handling and grow-out methods (Telesca *et al.*, 2018; Seed, 1968; Brake *et al.*,
287 2003; Kube *et al.*, 2011; Griffiths and Buffenstein, 1981; Van Erkom Schurink and Griffiths, 1993;
288 Bayne, 2000; Sheridan *et al.*, 1996). We also observed a difference in shape between the animals
289 reared in floating bags or Ortac with the latter longer and wider compared to the former which were
290 instead thicker and with a higher C.I.

291 The morphological differences found between individuals farmed in the two different farming gears,
292 are probably due to the different interaction that these tools have with the currents. Under low
293 current speed typical of shallow lagoons, the shape of the Ortac units may have not promoted the
294 rocking motion required to generate enough rubbing between oysters and the farming gear, causing
295 less shell chipping, which is widely recognised as a factor promoting shell depth and a higher meat
296 content (Brake *et al.*, 2003; Holliday, 1991; O'Mealey, 1995; Robert *et al.*, 1993).

297 Moreover, the fact that the animals did not move enough inside the Ortac units probably induced
298 those in the innermost part to grow more in length and width in order to increase the filtering
299 surface. Nonetheless, results of this study are comparable with those obtained by Francouer *et al.*,
300 (2017) who reared *Ostrea edulis* using the Ortac system in Scotland.

301 Mortality may depend on the farming system (Pernet *et al.*, 2012). Improved survival in the Ortac
302 system could be due to the shading effect provided by a more solid structure, which would shelter
303 farmed individuals from direct sunlight and desiccation, particularly during the earlier part of the

304 growth cycle and during air exposure periods. (Potter and Hill, 1982; Spencer-Davies, 1970).
305 Moreover, different studies report that one of the stress factors associated to mortalities is
306 temperature, and sudden small changes may have a large effect on the survival of bivalves (Pernet *et*
307 *al.*, 2018; Pernet *et al.*, 2012; Petton *et al.*, 2015; Le Deuff *et al.*, 1994, Le Deuff *et al.*, 1996;
308 Sauvage *et al.*, 2009; Kennedy and Mihursky, 1971). Again the more solid structure of the Ortac
309 may have promoted more stable temperature and reduced stress.

310 Results of this study indicate that the predicted growth by ShellSIM, fitted well with field
311 measurements in the lagoon of San Teodoro. However, results from the growth trial in Santa Gilla
312 lagoon indicate that the model would require further tailoring to local conditions to produce realistic
313 growth projections. In particular, we tested the hypothesis that the assumption ShellSIM makes to
314 convert concentration of food into energy available to animals does not apply to Santa Gilla Lagoon.
315 To do this, we run the model reducing the amount of POC available to one quarter of the measured
316 POC and the model prediction was more accurate ($S = 0.97$ and $S = 0.95$ respectively for weight and
317 length). Indeed, POC can be considered as a very heterogeneous nutrient source composed by
318 different materials with large variations in digestible energy content. (Mazzola and Sarà, 2001;
319 Lawacz, 1977; Watanabe and Kuwae, 2015).

320 Further studies to identify the real digestible energy content of the Particulate Organic Carbon in
321 Santa Gilla area is required to modify the model assumption and improve its performances. Our data
322 also suggest that seasonality and farming system used can influence the accuracy of ShellSIM
323 providing scope for further tailoring of the model under the local conditions.

324 During the first-year trial in the lagoon of San Teodoro the measured growth closely fitted the
325 predicted growth in POS 2, while in POS 1 ShellSIM slightly overestimated and in POS 3
326 considerably overestimated the growth, both in weight and length. Similar results in POS 2 were
327 observed in the second-year validation trial. The growth in weight and length of the oyster was
328 different between the two farming tools, with a higher growth in weight recorded for oysters reared
329 in the floating bags and a higher growth in length for oysters reared in the Ortac units. In this trial,

330 ShellSIM underestimated the weight and length during the production cycle except at the end point
331 where it only slightly underestimated weight and length in the Ortac units providing a better
332 accuracy at harvest time. While in the floating bags the final mean weight was underestimated and
333 the length was overestimated.

334 These overestimation and underestimation can be potentially associated with a less than optimal
335 rearing method (the Ortac), combined with the potential different production capacity of the
336 different farming areas within the lagoon. Furthermore, as reported by several authors, the grow-out
337 methods employed could affect oyster growth (Bayne, 2000; Sheridan *et al.*, 1996). ShellSIM does
338 not consider different grow-out methods in its variables possibly generating the discrepancy between
339 observed and predicted growth measured in this study. Overall, ShellSIM predictions correspond
340 with the growth trends observed by the farmers over the years (POS 3 with higher growth rates and
341 POS 1 with lower growth rates) suggesting the good accuracy of the model with the general growth
342 dynamics in the different areas of San Teodoro lagoon. This is reflected in the calculated skill scores
343 (Taylor, 2001), for both validation trials in the fore mentioned lagoon.

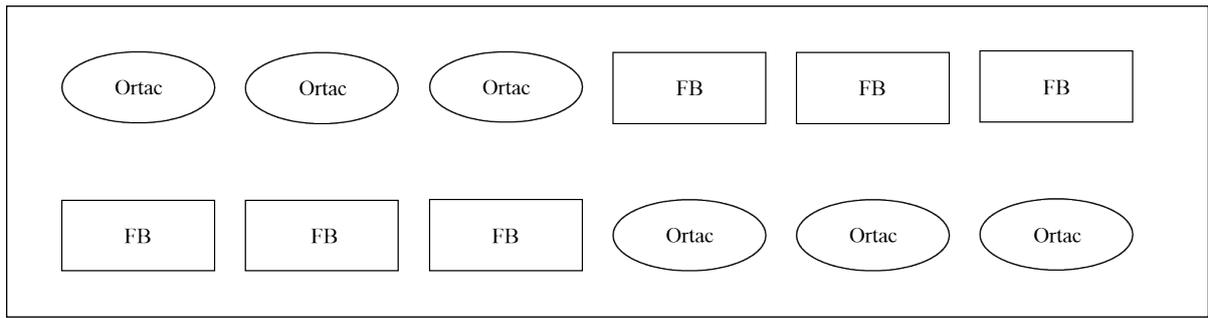
344 Taken together, the results of this study provide information to improve bivalve growth prediction
345 tools for Mediterranean lagoons. They could be applied to study the productivity of different sites to
346 potentiate the oyster's aquaculture industry and for coastal spatial planning. Moreover, the presented
347 results indicate that Ortac units improve the oyster's survival in the production early stage. The use
348 of Ortac units also reduces reliance on multiple mesh bags therefore simplifying production
349 protocols.

350

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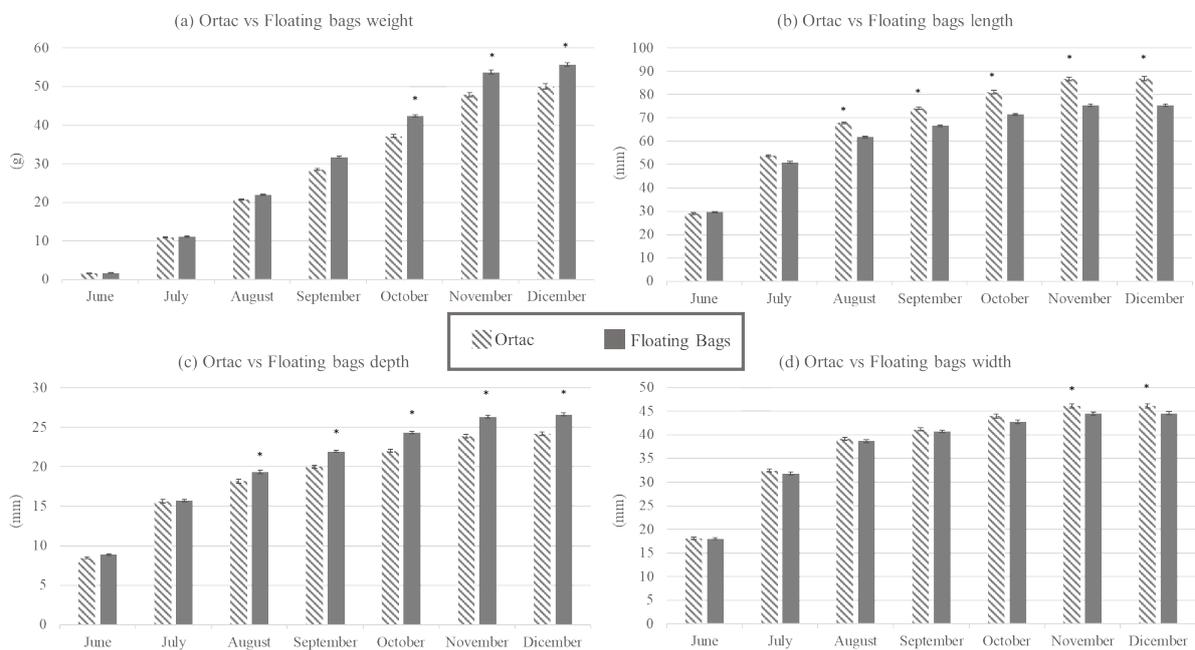


362

363 *Figure 1.* Diagram of the experimental layout of the Ortac and Floating Bags (FB) in the San Teodoro Lagoon.

364

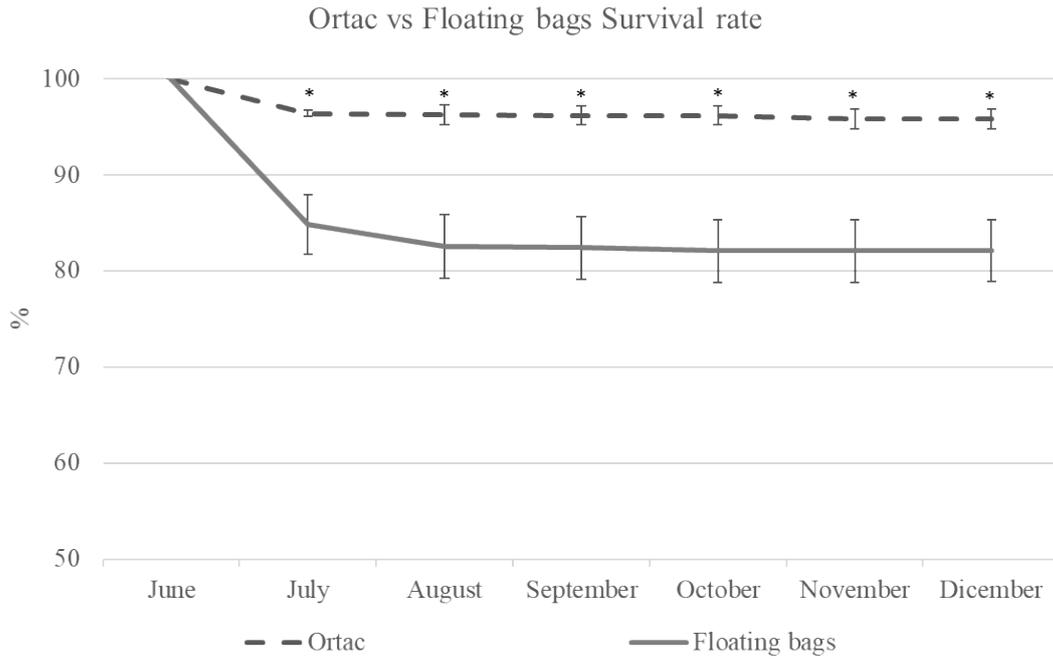
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366

367 *Figure 1.* (a) Difference growth in weight between *C. gigas* farmed in two different tools (Ortac units and Floating bags). (b)
 368 Difference growth in length between *C. gigas* farmed in two different tools (Ortac units and Floating bags). (c) Difference growth in
 369 width between *C. gigas* farmed in two different tools (Ortac units and Floating bags). (d) Difference growth in depth between *C.*
 370 *gigas* farmed in two different tools (Ortac units and Floating bags). Stars indicate where significant difference occurs. (p -value <
 371 0.05). Data are presented as mean \pm SE; n=6.

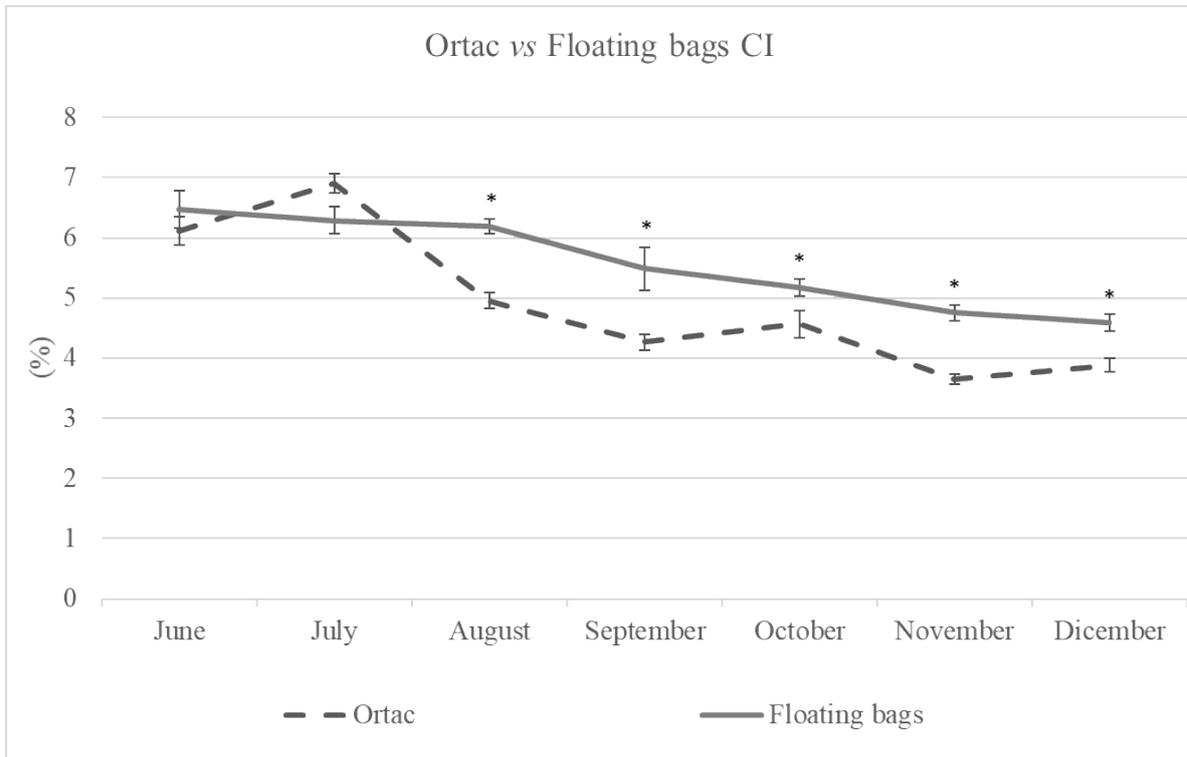
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373

374 *Figure 2.* Comparison of survival rate between *C. gigas* farmed into Ortac units and Floating bags. Stars indicate where significant
 375 difference occurs. (p-value < 0.05). Data are presented as mean ± SE; n=6.

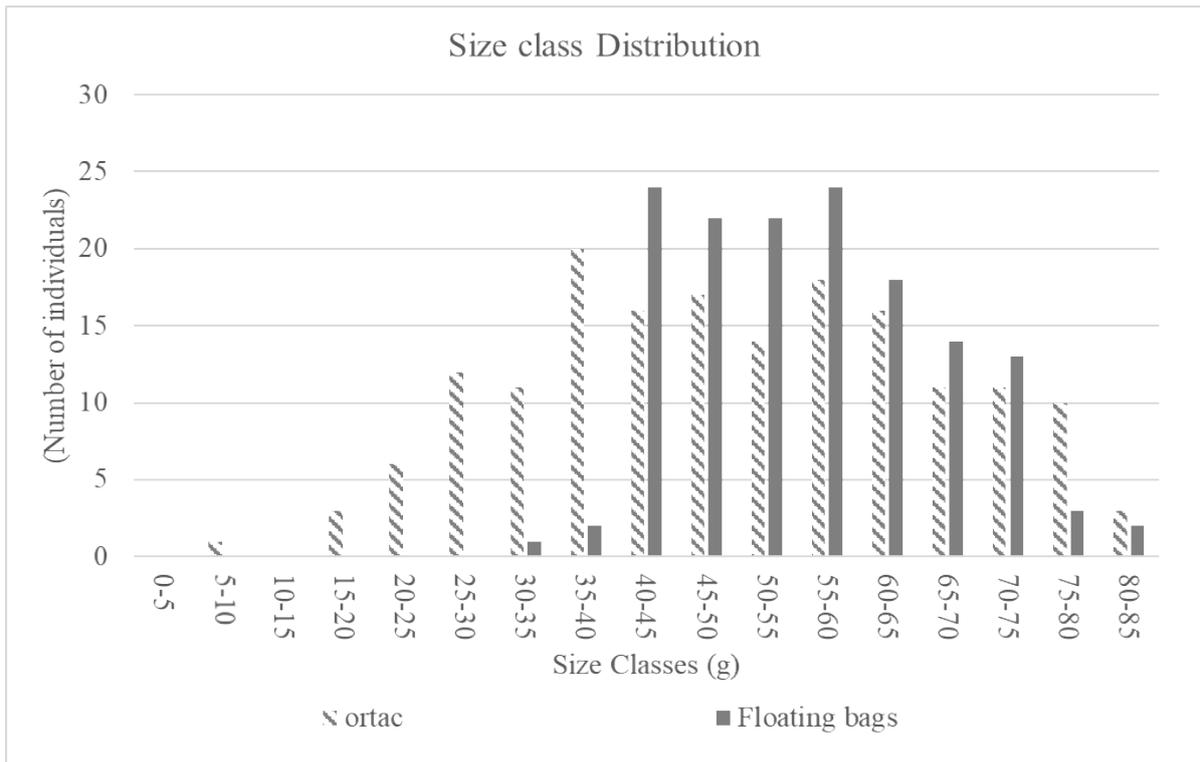
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378 *Figure 3.* Comparison of condition index (CI) calculated as (Dry weight Meat (g)/ Dry weight Meat + Dry weight shell) *100,
 379 between *C. gigas* farmed into Ortac units and Floating bags. Stars indicate where significant difference occurs. (p-value < 0.05). Data
 380 are presented as mean ± SE; n=6.

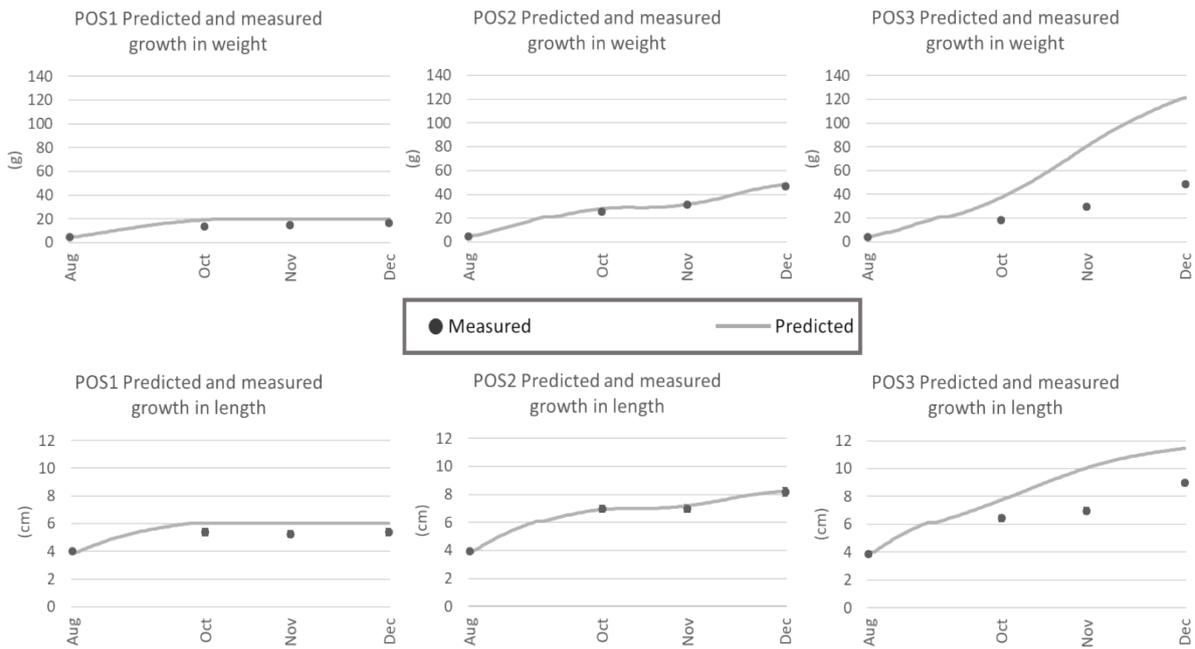
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383 Figure 4. Comparison of size class distribution between *C. gigas* farmed into Ortac units and Floating bags.

384

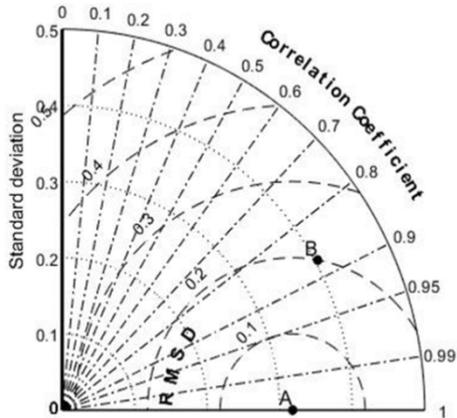


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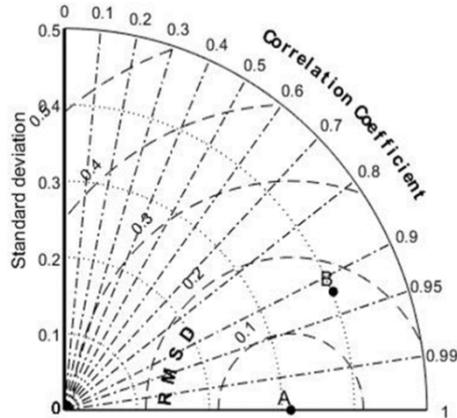
386 Figure 5. ShellsIM growth prediction compared to the measured oyster growth in weight and length, during a production cycles
 387 performed in to three different areas (POS1, POS2 and POS3) of the San Teodoro lagoon. Measured growth data are presented as
 388 mean ± SE; n=3.

389

SHELLSIM VALIDATION IN SAN TEODORO LAGOON (g)



SHELLSIM VALIDATION IN SAN TEODORO LAGOON (cm)

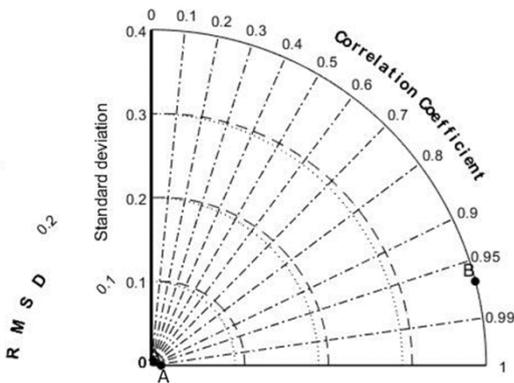


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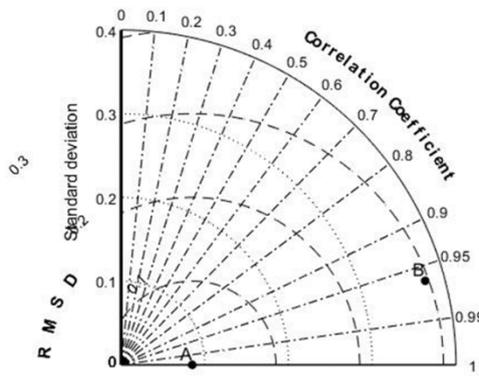
391 *Figure 6.* Taylor diagrams representing how closely model performance (B) match the observed data (A). The similarity between
 392 model prediction and observed data is quantified in terms of their correlation, the amplitude of their variation (normalised standard
 393 deviation) and their root mean square difference (RMSD) (dashed circular arcs). The left panel contain the results for the ShellSIM
 394 validation in the san Teodoro lagoon in terms of predicting the growth in weight of the *C.gigas*. The right panel contain the results
 395 for the ShellSIM validation in the san Teodoro lagoon in terms of predicting the growth in length of the *C.gigas*.

396

SHELLSIM VALIDATION IN SANTA GILLA LAGOON (g)

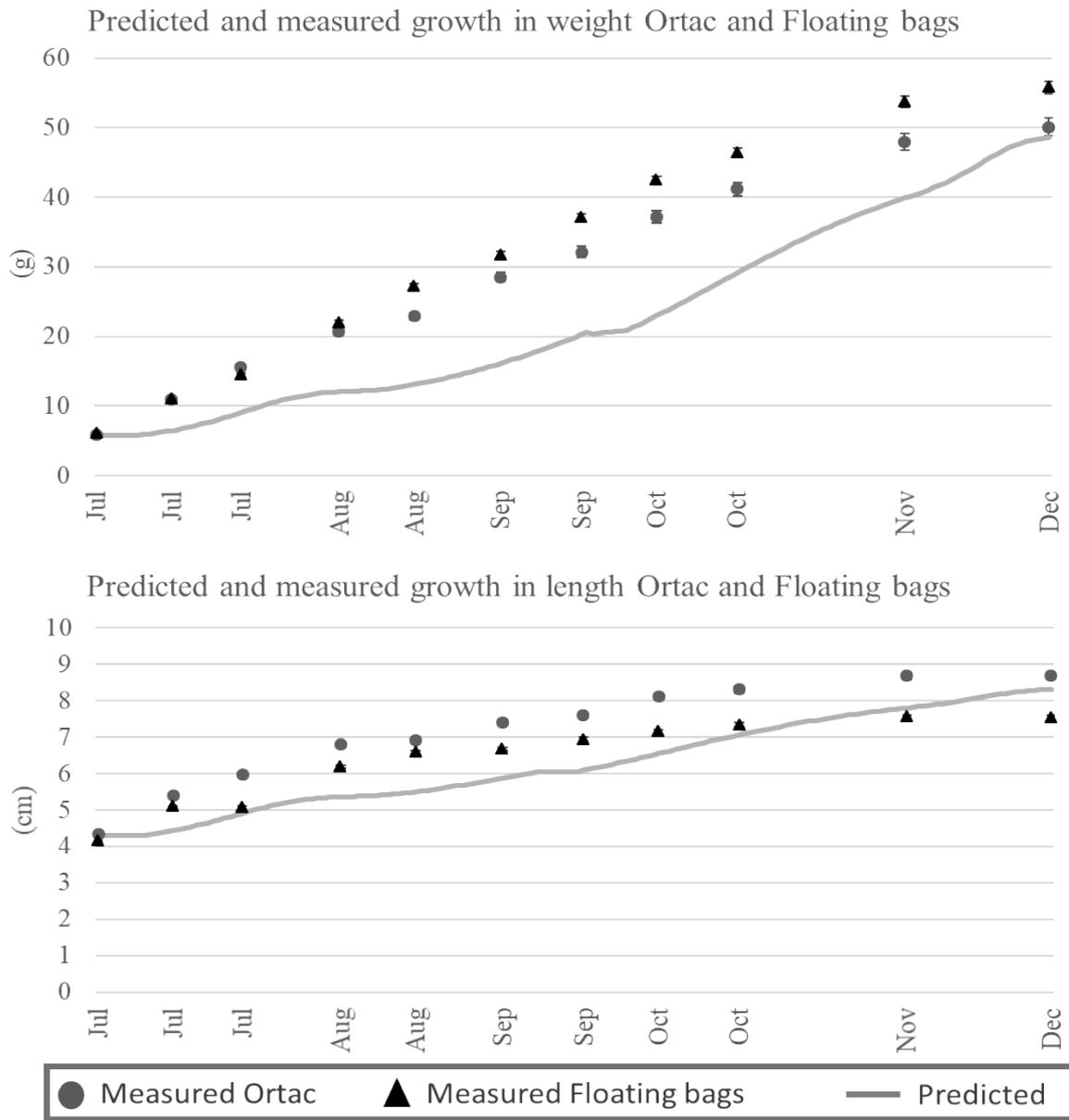


SHELLSIM VALIDATION IN SANTA GILLA LAGOON (cm)



397

398 *Figure 8.* Taylor diagrams representing how closely model performance (B) match the observed data (A). The similarity between
 399 model prediction and observed data is quantified in terms of their correlation, the amplitude of their variation (normalised standard
 400 deviation) and their root mean square difference (RMSD) (dashed circular arcs). The left panel contain the results for the ShellSIM
 401 validation in the Santa Gilla lagoon in terms of predicting the growth in weight of the *C.gigas*. The right panel contain the results for
 402 the ShellSIM validation in the Santa Gilla lagoon in terms of predicting the growth in length of the *C.gigas*.

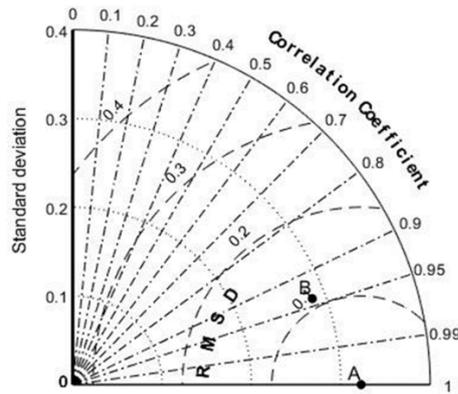


404

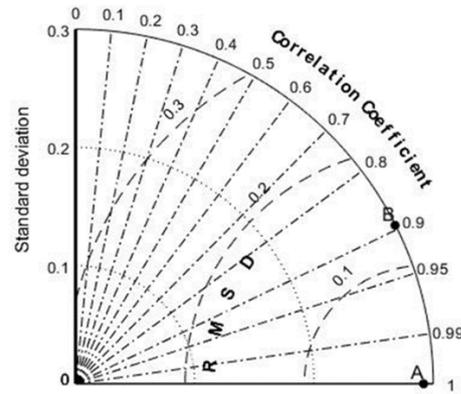
405 *Figure 9.* ShellSIM growth prediction compared to the measured oyster growth in weight and length, during a production cycles
 406 performed in to two different farming systems (Ortac units and Floating bags) in the San Teodoro lagoon (July 2017 – December
 407 2017). Measured growth data are presented as mean ± SE; n=6.

408

SHELLSIM VALIDATION ORTAC AND FLOATING BAGS (g)



SHELLSIM VALIDATION ORTAC AND FLOATING BAGS (cm)



409

410 *Figure 10.* Taylor diagrams representing how closely modelled performances (B) matched the observed data (A). The similarity
 411 between model prediction and observed data is quantified in terms of their correlation, the amplitude of their variation (normalised
 412 standard deviation) and their root mean square difference (RMSD) (dashed circular arcs). The left panel contain the results for the
 413 ShellSIM validation in the San Teodoro lagoon on Ortac and Floating bags in terms of predicting the growth in weight of the
 414 *C.gigas*. The right panel contain the results for the ShellSIM validation in the San Teodoro lagoon on Ortac and Floating bags in
 415 terms of predicting the growth in length of the *C.gigas*.

416

417

418

419 *Table 1.* Summary of the environmental data collected to run ShellSIM. These data were collected during the production cycles
 420 started in August 2016, in three different areas (POS1, POS2 and POS3) of the San Teodoro lagoon. Data are presented as mean \pm
 421 SE.

	<i>T</i> °C	<i>Sal</i> ‰	<i>DO</i> mg/L	<i>TPM</i> mg/L	<i>POM</i> mg/L	<i>POC</i> mg/m ³	<i>Chl-a</i> µg/L
August 2016							
POS 1	27.1 \pm 0.1	39.3 \pm 0.1	7.2 \pm 0.2	31.5 \pm 12.4	5.3 \pm 1.9	848.2 \pm 18.6	2.1 \pm 0.2
POS 2	27.2 \pm 0.1	39.7 \pm 0.1	7.5 \pm 0.1	15.5 \pm 1.6	3.2 \pm 0.3	1213.1 \pm 67.9	4.3 \pm 0.7
POS 3	27.2 \pm 0.2	39.5 \pm 0.1	6.8 \pm 0.1	19.4 \pm 1.1	3.6 \pm 0.2	1421.9 \pm 68.5	4.1 \pm 0.1
October 2016							
POS 1	23.5 \pm 0.1	39.2 \pm 0.1	8.7 \pm 0.1	5.2 \pm 0.2	1 \pm 0.1	206.9 \pm 6	0.3 \pm 0.1
POS 2	24.1 \pm 0.1	38.8 \pm 0.1	8.5 \pm 0.1	5 \pm 0.2	1 \pm 0.1	211.3 \pm 18.1	0.3 \pm 0.1
POS 3	23.1 \pm 0.3	38.8 \pm 0.1	8.9 \pm 0.3	21.1 \pm 2.4	3.9 \pm 0.2	1192.5 \pm 55.8	3.7 \pm 0.3

November 2016

POS 1	17.1 ± 0.1	39.2 ± 0.1	9.3 ± 0.1	0.6 ± 0.03	0.5 ± 0.1	167.3 ± 9.1	0.4 ± 0.02
POS 2	15.8 ± 0.5	37.7 ± 0.5	8.8 ± 0.2	2.3 ± 0.1	1 ± 0.1	485.2 ± 33.9	2.8 ± 0.2
POS 3	14.5 ± 0.2	38.8 ± 0.1	9.6 ± 0.1	3.0 ± 0.04	1.1 ± 0.1	473.4 ± 20.4	2.9 ± 0.1

December 2016

POS 1	16 ± 0.1	36.4 ± 0.2	10 ± 0.2	1.7 ± 0.1	0.6 ± 0.1	232.8 ± 21	0.7 ± 0.1
POS 2	17.5 ± 0.3	37 ± 0.3	10.7 ± 0.4	1.0 ± 0.1	0.6 ± 0.1	199.1 ± 28.4	0.4 ± 0.1
POS 3	15.1 ± 0.3	36.2 ± 0.2	9.2 ± 0.6	4.8 ± 0.2	1.2 ± 0.1	408.4 ± 24	0.9 ± 0.03

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Table 2. Summary of how well observed data match predicted data by ShellSIM in terms of their correlation, their root-mean square difference (RMSD), the ratio of their variances and skill score (Taylor, 2001).

SHELLSIM VALIDATION IN SAN TEODORO LAGOON (FLOATING BAGS)

	St.dev Obs.	St.dev Pred.	RMSD	Correlation	Skill score
POS 1 (g)	0.3	0.43	0.14	0.98	0.87
POS 1 (cm)	0.27	0.43	0.16	1	0.81
POS 2 (g)	0.35	0.36	0.02	1	1
POS 2 (cm)	0.36	0.37	0.02	1	1
POS 3 (g)	0.14	0.38	0.24	0.99	0.42
POS 3 (cm)	0.24	0.38	0.16	0.97	0.79
OVERALL (g)	0.32	0.4	0.2	0.87	0.83
OVERALL (cm)	0.31	0.4	0.17	0.92	0.87

SHELLSIM VALIDATION ON ORTAC AND FLOATING BAGS IN SAN TEODORO LAGOON

ORTAC (g)	0.32	0.30	0.1	0.95	0.95
ORTAC (cm)	0.31	0.28	0.11	0.93	0.93
FLOATING BAGS (g)	0.33	0.27	0.12	0.94	0.9
FLOATING BAGS (cm)	0.27	0.31	0.13	0.9	0.89
OVERALL (g)	0.32	0.29	0.11	0.94	0.93
OVERALL (cm)	0.29	0.30	0.14	0.89	0.9
SHELLSIM VALIDATION IN SANTA GILLA LAGOON					
SANTA GILLA (g)	0.01	0.4	0.39	0.97	0
SANTA GILLA (cm)	0.08	0.38	0.3	0.96	0.18

426

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428

429 *Table 3. Summary of the environmental data used to run ShellSIM. These data were collected during the production cycles started in*
430 *June 2017, in the Santa Gilla lagoon. Data are presented as mean ± SE.*

	<i>T °C</i>	<i>Sal %</i>	<i>DO mg/L</i>	<i>TPM mg/L</i>	<i>POM mg/L</i>	<i>POC mg/m³</i>	<i>Chl-a µg/L</i>
<i>June 2017</i>	24.7 ± 0.02	37.6 ± 0.03	7 ± 0.03	2.3 ± 0.2	0.9 ± 0.1	413.1 ± 8.2	0.7 ± 0.1
<i>July 2017</i>	24.8 ± 0.02	43.3 ± 0.1	6.1 ± 0.04	4.6 ± 1.1	1 ± 0.1	349.5 ± 3.3	0.7 ± 0.1
<i>August 2017</i>	27 ± 0.01	35.4 ± 0.03	5.2 ± 0.04	6.6 ± 0.9	1.4 ± 0.1	451.4 ± 12.6	1.8 ± 0.1
<i>September 2017</i>	24.4 ± 0.02	36 ± 0.03	6 ± 0.03	8.1 ± 0.7	2.1 ± 0.3	561.6 ± 19.9	1.9 ± 0.1

431

432

433 *Table 4. Summary of the environmental data used to run ShellSIM. These data were collected during the production cycles started in*
434 *July 2017, in the San Teodoro lagoon. Data are presented as mean ± SE.*

	<i>T °C</i>	<i>Sal %</i>	<i>DO mg/L</i>	<i>TPM mg/L</i>	<i>POM mg/L</i>	<i>POC mg/m³</i>	<i>Chl-a µg/L</i>
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July 2017	27.5 ± 0.2	38.9 ± 0.4	9.6 ± 0.3	6.1 ± 4.0	1.7 ± 1.2	358.2 ± 62.7	1.7 ± 0.7
August 2017	28.4 ± 0.3	41.1 ± 0.4	7.8 ± 0.3	3.9 ± 0.8	1.5 ± 0.2	557.6 ± 94.2	3.1 ± 1.1
September 2017	21.5 ± 0.1	40.2 ± 1.1	9.2 ± 0.1	5.4 ± 0.3	1.5 ± 0.2	491.1 ± 30.1	1.7 ± 0.6
October 2017	18 ± 2.1	40.7 ± 0.1	8.3 ± 0.3	5.2 ± 0.3	1.4 ± 0.1	769.2 ± 99	2.8 ± 0.6
November 2017	18.3 ± 0.3	38.4 ± 0.3	9.4 ± 0.1	23.2 ± 3.3	3 ± 0.4	151.3 ± 23.9	0.5 ± 0.03
December 2017	14.7 ± 0.1	36.7 ± 0.5	10 ± 0.1	12.3 ± 9.5	1.9 ± 1.3	222.4 ± 67.4	0.8 ± 0.3

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