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Larval Width as Indicator of Growth Rate and Effect of Larval Classification on Final Body Composition and Flesh Quality in Cultured Gilthead Sea-bream (*Sparus aurata*, L.).

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Summary

The objective of this study was evaluating the effects of separating larvae according to larval width on growth rate and flesh quality in cultured gilthead sea-bream (*Sparus aurata*, L.).

Progeny from two broodstocks (A and B) were divided according to larval width into two groups, heads (being the biggest specimens) and tails. After 18 months, both groups were analyzed to evaluate growth, carcass traits and proximal composition.

The head specimens reached a bigger size and showed a greater level of well-being and degree of nourishment. The total body and fillet composition was found to be that generally expected for this species. Some differences were found in total body composition between head and tail specimens in both batches, and in fillet composition in batch B (higher fat and lower moisture content in head specimens).

Sensory analyses were carried out with untrained panelists. They were unable to distinguish between the head and tail samples in batch A, whereas in batch B differences were noticed. The panelists judged samples from the head group to be tastier and juicier, a consequence of their higher fat content.

Hence, fish separation according to larval width is an effective tool to separate progeny with slow and fast growing, whereas total body and fillet analyses and sensory test allow us to ensure that selection does not generate any negative effects on the product quality.

51 Introduction

52

53 The gilthead sea-bream (*Sparus aurata* L.) represents one of the most important
54 cultured species in Mediterranean Sea. Global production reached 151,346 tons in
55 2011, with Greece, Turkey and Spain being the main producers (APROMAR,
56 2012). To improve production, companies usually focus their efforts on nutrition,
57 stock management, disease prevention and facility location (Navarro et al.,
58 2009a). However, little attention is given to genetic aspects, due to high cost and
59 not immediate results obtained. Nevertheless, genetic selection and the rearing of
60 breeders with high genetic variability are important tools for improving progeny
61 quality and avoiding inbreeding; a bigger problem in fish farming than in
62 livestock (Borrell et al., 2007).
63 Selection programs focus on growth rate, food conversion efficiency, fecundity,
64 meat quality and resistance to stress and diseases, as enhancing these
65 characteristics can lead to shorter rearing times and lower mortality rates
66 (Gjedrem, 1983; Afonso et al., 1998; Thorland et al., 2007; Dupont-Nivet et al.,
67 2008; Antonello et al., 2009; Navarro et al., 2009a).
68 In selection programs, composition analyses are required to ensure that the
69 characteristics of the final product are not altered due to the selection process.
70 Body composition and flesh quality, especially skeletal muscle composition and
71 fat deposition, can influence consumer choice because they affect fish appearance,
72 smell and taste (Grigorakis, 2007).
73 Favourable and unfavourable traits can be positively correlated as in the case of
74 Condition Factor (CF) and visceral fat - desirable and not desirable, respectively -
75 in *S. aurata* (Grigorakis and Alexis, 2005; Navarro et al., 2009b) or in the case of

high harvest weight (favourable) and flesh fat content (unfavourable) in Atlantic salmon (*Salmo salar* L.) (Quinton et al., 2005).

In *S. aurata*, high hereditability for visceral fat has been found (Navarro et al., 2009b) and as it is an undesirable character, because it is not appreciated by consumers (Grigorakis, 2007), its indirect selection should be monitored and avoided.

However, in *S. aurata* a positive correlation between size and fat deposition (Grigorakis and Alexis, 2005) and between weight and length (Navarro et al., 2009b) has been found and in this case increase of both characters is desirable, as they increase size, fish appearance, taste and juiciness.

In the case of gilthead sea-bream, there is a lack of efficient breeding programs (Thorland et al., 2007). However, using samples from the progenies of the broodstocks used in the present study, it has been demonstrated that microsatellite analysis is a good molecular tool for assisting breeding programs, as it allows us to identify the breeders which primarily contributed to fast-growing progenies (Borrell et al., 2011).

Complementing these previous results, the main aim of this experiment was to test other not genetic tools (such as the larval classification, and the evaluation of flesh composition) which could be complementarily used as part of selection programs. Hence differences in growth rate, body parameters, proximate composition and flesh composition in progenies with different larval width obtained from two different broodstocks (A and B) were evaluated.

Material and Methods

Rearing and fish samplings

From a total of 101 *S. aurata* adult individuals (80 females and 21 males) available in December of 2006 from a hatchery in Spain (Piscicultura Marina Mediterránea, S.L. Burriana, Castellón, Spain), two broodstocks (Broodstock A: 29 females, average weight 1998.2 ± 43.5 g and 11 males, average weight 1877.2 ± 83.4 g; Broodstock B: 28 females, average weight 1992.8 ± 50.1 g, and 10 males, average weight 1950.0 ± 76.3 g) were grouped using a combinatorial optimization approach that followed the procedures described in Borrell et al. (2007), ensuring the same genetic variability in both broodstocks.

In March 2007 breeders were allowed to spawn freely. Fertilized eggs coming from one single day were collected from every broodstocks and incubated under identical conditions in two tanks until hatching. Larvae were reared in large tanks and after 86 and 78 days post-hatching offsprings (batch A and B, respectively) were classified by body width, using different sieves, and a slow and a fast growing group, named tails and heads, respectively, were obtained from each batch. As measuring all the fish was not viable, selection size was determined according to the mean width after trying different sieves. In batch A, fish with a body width greater than 4.5 mm or lower than 3.5 mm were selected as heads and tails respectively; whereas in batch B fish with a body width greater than 3.5 mm or lower than 2.5 mm formed head and tail groups respectively. The difference between batch A and B is due to the different ages of the larvae. The mean weights are shown in table 1. Weights were obtained in the UPV facility where a precision balance was available and using absorbent tissue paper to minimize the effect of humidity.

Ultimately, four different fish groups were obtained: A-Heads (AH), A-Tails (AT), B-Heads (BH) and B-Tails (BT).

At the age of 165 and 157 days post-hatching (batches A and B, respectively) a sample of approximately 1,000 fish per group (mean fish weight of head groups: 6.5 g; mean fish weight of tail groups: 2.0 g) was moved to the facilities at the Universitat Politècnica de València (Valencia, Spain). The fish were distributed in 1,750 L fiberglass tanks where they were reared under intensive conditions. The tanks were kept in a re-circulating marine water system (30 m^3 capacity) with a rotary mechanical filter and a gravity biofilter. All tanks were equipped with aeration and the water temperature was maintained at 22 ± 3 ºC by a heat pump installed in the system, and oxygen at 6.25 ± 0.44 ml/L. Photoperiod was natural. Fish were fed by hand using commercial fish feed (Dibaq S.A., Segovia, Spain) twice a day to apparent satiety. Feed size was chosen according to fish size. Rearing conditions were made as much as possible similar to those present in fishfarms.

At an intermediate stage of growth (311 and 303 days post-hatching, batch A and B, respectively) 250 juveniles from each group (corresponding to 25% of the total) were chosen by weight, with the final aim of selecting the biggest fish for the head groups and the smallest fish for the tail groups. Weight ranges were established according to the estimated mean weight in each group. In batch A heads with a weight >41 g and tails with a weight <15 g were selected; while in batch B heads with a weight >31 g and tails with a weight <12 g were selected.

Finally, the head and tail groups were distributed into four 4,000 L tanks where they were reared under intensive conditions. At an intermediate stage of growth

(411 days post-hatching batch A and 403 days post-hatching batch B), 100 fish from each tank were weighed to calculate biomass and growth tendency.

At the end of the growth cycle (528 and 520 days post-hatching, batch A and B, respectively) all fish were weighed to establish the growth rate.

Fifteen animals from each group were randomly measured to calculate the body parameters and stored at -20 °C until carcass and fillet proximate analysis (moisture, fat, protein and ash content). In addition, ten animals from each group were gutted and filleted.

To perform the sensory test, the animals were filleted and the dorsal fillets were stored at -20 ºC until use.

Growth analysis

Growth was calculated by using weights recorded at 311, 411 and 528 days post-hatching (batch A) and 303, 403 and 520 days post-hatching (batch B).

Between 311 and 528 days post-hatching (batch A) and 303 and 520 days post-hatching (batch B) the following parameters were calculated to evaluate the differences between tail and head groups:

Daily growth index (DGI, %/d) = $100 \times (\text{final weight}^{1/3} - \text{initial weight}^{1/3}) / d$

Feed Intake (FI, %/d) = 100 × feed consumption (g) / average biomass (g) x d

Feed Conversion Ratio (FCR) = feed offered (g) / weight gain (g)

Thermal Growth Coefficient (TGC x 1000) = (Final weight $^{1/3}$ – Initial weight $^{1/3}$) / (∑ ºC effective x d)

Body analysis

Total length was measured and total body, liver, gonads, mesenteric fat, guts, head, skin, fins and bone were weighed in 15 animals from each group.

For each fish, the hepatosomatic index [I_H (%) = 100 x liver weight (g) / total weight (g)], gonadosomatic index [I_G (%) = 100 x gonad weight (g) / total weight (g)], condition factor [C_F = 100 x total weight (g) / total length^3 (cm^3)], viscerosomatic index [I_V (%) = 100 x viscera weight (g) / total weight (g)], mesenteric fat index [I_M (%) = 100 x mesenteric fat weight (g) / total weight (g)], fillet yield [F_Y (%) = 100 x (total body weight – head – viscera – fins – bone – skin - gonad) (g) / total weight (g)] and head percentage [H_P (%) = 100 x head weight (g) / total weight (g)] were calculated.

Proximate analysis

Ten specimens from each group were analyzed for carcass composition following the AOAC procedure (1990). Moisture levels were determined by drying samples (2.5-3 g) in porcelain cups at 104 °C for 24 h. Ash levels were determined by incinerating the dried samples at 550 °C for 5 h. Protein content was determined in 0.5 g of lyophilized and minced fish using the Kjeldhal method. Fat content was determined in 0.5 g of lyophilized and minced fish using Soxhlet extraction.

In five specimens per group, the analyses previously described were carried out only on fish fillets with the aim to evaluate the composition of the edible part. All analyses were performed in triplicate. All these traits are expressed as a percentage of fresh weight.

199 Sensory analysis

200 A triangle test following ISO 1420 (1983) was performed to evaluate the sensory
201 differences between head and tail specimens. Dorsal fillets were thawed at room
202 temperature, vacuum-packed in plastic bags and cooked for 10 min using a water-
203 bath at 60 ºC. Each fillet was then cut into nine pieces, each weighing
204 approximately 3–4 g. The resulting equally sized pieces were coded, wrapped in
205 aluminum foil and kept in 40 °C thermo regulated boxes until the test. To ensure
206 that possible differences were not due to the effects of the fillet portions, samples
207 from a similar part of the fillet were compared in each test. At each session, three
208 triangle tests were presented to each panelist. A total of 120 triangles for 40
209 untrained panelists were prepared for each group and the panelists were asked to
210 identify the odd piece. 'No difference' was not accepted as a valid response,
211 according to Meilgaard et al. (1999). The minimum number of correct answers (x)
212 for a statistically significant result ($p<0.05$) was calculated according to $x = 0.4174 \times 1.64 \times (\sqrt{n}) + (2n + 3) / 6$ where n = number of responses (International
213
214 Organization for Standardization 1983).
215
216 Statistical analysis
217 After establishing data normality, an ANOVA (Newman-Keuls test) was carried
218 out to compare results. If normality failed (GSI and weights for growth evolution)
219 with transformed data, a Kruskal-Wallis test was carried out. All values are
220 expressed as mean ± standard error (SEM).
221 Growth curves were compared by regression analysis and regression coefficients
222 (slopes) were compared using analysis of variance. Differences were considered

223 significant at $p<0.05$. All statistical procedures were run using Statgraphics Plus®
224 5.1.
225
226 Results
227 Growth parameters at the end of the feeding period are shown in Table 2. During
228 the feeding period, the weight of the head specimens was significantly higher. The
229 tail specimens presented higher FI values, whereas no differences were recorded
230 in DGI, TGC and FCR. The comparison of the slopes (b) of the two regression
231 lines confirmed a higher growth rate in the head group specimens.
232 Body parameters at the end of the growth period are shown in Table 3. In batch A,
233 higher body weights, total length, C_F and I_G were recorded in head specimens
234 than in tails, demonstrating a significant difference in size. However, all
235 specimens were sexually immature. No differences were found in the rest of
236 parameters (I_H, I_V, I_M, F_Y and H_P). In batch B the head specimens showed higher
237 values in all the measured parameters, except head percentage.
238 With regards to body composition, analysis was carried out on the whole body in
239 both batch A and batch B (Table 4). No differences were found in either ash or
240 protein content between head and tail groups, however the head specimens
241 presented a higher fat content and a lower moisture percentage than the tail
242 specimens.
243 Concerning fillet composition (Table 5), no differences were recorded in batch A,
244 where both head and tail groups demonstrated the same moisture, ash, protein and
245 fat content. Nevertheless, in batch B, coinciding with the results observed in the
246 whole body analyses, no differences were found in ash or protein content, whereas

247 a higher fat content and a lower moisture percentage were recorded in head
248 specimens.
249 Sensory test results are reported in the Table 6. One hundred and twenty triangle
250 tests were performed, but in both batches four ‘no differences’ replies were
251 rejected, thus only 116 tests were considered.
252 For 116 triangle tests, the number of correct answers necessary for a $p<0.05$ is 47.
253 For batch A the number of correct answers was 42, demonstrating that the
254 panelists were unable to distinguish between head and tail specimens. However,
255 for batch B the number of correct answers was 48, showing that the panelists
256 could distinguish between heads and tails. In terms of the differences noticed, the
257 panellists described the head samples as tastier and juicier than the tail samples, in
258 both batches.
259
260 Discussion
261 At the end of the growth period (528 days post-hatching for batch A and 520 days
262 for batch B), higher length, weight and growth rate values were seen in fish from
263 the head groups. The tail groups presented a higher feed intake (FI) but this did
264 not signify a more rapid increase in size as demonstrated by the lack of
265 differences in DGI, TGC and FCR between heads and tails.
266 Growth results show that separating larvae according to body width is an effective
267 tool for individualizing and obtaining fish with a higher growth rate. Apparently,
268 this could be a good complementary tool to be included together with genetic
269 analyses as part of selection programs. Due to the difficulties in comparing
270 growth parameters between fish of different sizes, as was the case in this study,
271 fish from this experiment were compared with fish of similar size and culture

conditions. For comparisons with fish from ingredient replacement experiments control group results were considered. TGC and FCR were recalculated on the basis of the results, if not shown in the articles.

Head specimen results were compared with those shown by Martínez-Llorens et al. (2008), who obtained a TGC = 1.41, a lower value than the one obtained in selected heads during the present experiment (TGC = 1.49). Moreover, a lower FCR (1.40) was recorded in the head specimens in the present study than that obtained by these authors (FCR = 1.93).

However, the tail group specimens selected during the experiment showed a lower TGC (TGC = 1.67) than that obtained by Sitjà-Bobadilla et al. (2005) (TGC = 2.04). Moreover, tail specimens from the present experiment showed a higher FCR (FCR = 1.35) than that obtained by Sitjà-Bobadilla et al. (2005) and Benedito-Palos et al. (2008) (1.18 and 0.99, respectively).

Therefore, size classification allows the possibility of keeping animals with faster growth and discarding those with slow growth.

Differences in C_F indicate that the head specimens have a higher level of well-being and degree of nourishment than the tail specimens. C_F can vary according to sexual maturation, age and sex, but in this case it is due only to weight and length differences as the specimens were the same age and had not reached sexual maturation. In fact, I_G for mature *S. aurata* is higher than 1% for females and 0.70-1% for males (Zohar et al., 1984), whereas the maximum I_G recorded at the end of the experiment was 0.2%. Sexual maturity was not reached, despite the animal age, probably because of the water temperature, which was maintained at 22 ºC, while *S. aurata* spawns during winter, with an optimal temperature range

296 of 15-17 ºC (Moretti et al., 1999). In any case, C_F results are consistent with
297 literature (Navarro et al., 2009a,b; Sánchez-Lozano et al., 2009).
298 In batch A, body parameters differences were limited at length, weight, C_F and I_G.
299 However, in batch B, the head specimens presented differences in all the
300 considered parameters, except head percentage, probably as consequence of the
301 greater difference between head and tail specimens in this batch.
302 Results here reported for I_M and I_H are similar to those shown by Grigorakis et al.
303 (2002) for specimens reared at the same temperature. In the same way, I_V
304 obtained in this experiment is consistent with the dressing percentage reported by
305 Navarro et al. (2009a), as these parameters are inversely proportional.
306 The differences in the lipid content between head and tail specimens, recorded in
307 the whole body analysis in both batches A and B, are due to their difference in
308 weight. In fact, it has been demonstrated that muscle, perivisceral and peritoneal
309 fat increase with size (Grigorakis and Alexis, 2005) as animals first use energy for
310 their muscles and then to deposit fat (Gjedrem, 1997). In accordance with Navarro
311 et al. (2009b) our experiments also demonstrated that the percentage of fat is
312 inversely proportional to the percentage of moisture (Tables 4 and 5). When
313 analyzing only the fillets, no differences were found between the head and tail
314 groups in batch A, whereas in batch B fillets from the head specimens presented a
315 higher fat content and a lower moisture percentage than the tail specimens.
316 The differences between batch A and B with regard to body parameters and body
317 and fillet composition are due to the greater difference in weight between the head
318 and tail specimens in batch B as opposed to batch A.
319 Protein and ash content maintained constant values in both batches (A and B) both
320 in whole body and fillet composition. In general, ash and protein are considered

stable components of fish body and do not seem to be dependent on weight (Navarro et al., 2009b). In salmonids, the mechanism for protein homeostasis seems to be more effective than the one for lipid homeostasis, and body protein percentage shows low phenotypic and genetic variations (Kause et al., 2009). In *Salmo salar* carcass protein value stabilizes when fish reach 100 g in weight (Shearer et al., 1994). As highlighted by Grigorakis et al. (2002), no studies have been carried out to determine the relationship between body weight and protein percentage in Mediterranean fish.

In cultured *S. aurata* values for fillet yield and moisture, ash, fat and protein in fillet are, 31.3-48%, 68-76%, 1.2-1.6%, 2.5-11% and 18-23 %, respectively (Grigorakis, 2007). Hence, the values obtained from the present experiment (Tables 3 and 5) are those expected for this species.

During the sensory test, the panelists were unable to distinguish between the head and tail fish from batch A. When sampling fish from batch B, the panelists distinguished head and tail samples. In general, head samples were appreciated more because they were considered tastier and juicier. Taste and juiciness depend on the percentage of fat and the opinion of the panelists is consistent with proximate analyses. The ability to distinguish head and tail samples in batch B and not in batch A is consistent with the differences in fillet composition. These results show that consumers can detect the differences in fish with different levels of fat content, confirming the importance of to carry out composition and quality analyses during selection programs, as selection itself should not jeopardize the final product quality.

Due to the limitations of the experimental design, in particular related with the lack of true replicates, results here obtained have to be considered as preliminary.

However conditions under which experiments were carried on are very similar to the ones fish farms have to deal with, especially the limited number of tanks. Moreover, the study was aimed to identify a valid, effective and immediate tool which could be easily used in fish farms to optimize the productive cycle obtaining fish with a fast growth.

Also, the study aimed to study not the growth performance of an experimental group but the individual growth performance of some subsamples. Analyzing fish as single allowed establishing the parental contribution of breeders to the progeny, as shown in the parallel article by Borrell at al. (2011). The evidence that some breeders contribute more to fast growth progeny reinforces the idea that progeny is an important element to select breeders.

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References

Afonso, J.M.; Montero, D.; Robaina, L.; Fernández, H.; Izquierdo, M.; Ginés, R.; 1998: Selection programmes for stress tollerance in fish. CIHEAM – Options Mediterraneennes **34**, 235-245.

371 Antonello, J.; Massault, C.; Franch, R.; Haley, C.; Pellizzari, C.; Bovo, G.;
372 Patarnello, T.; de Koning, D. J., Bargelloni, L., 2009: Estimates of heritability and
373 genetic correlation for body length and resistance to fish pasteurellosis in the
374 gilthead sea bream (*Sparus aurata* L.). Aquaculture **298**, 29–35.
375 AOAC, 1990: 15th ed. Official Methods of Analysis, vols I and II. Association of
376 Official Analytical Chemists, Arlington, VA, USA, 1298 pp.
377 APROMAR, 2012: La acuicultura marina en España. http://www.apromar.es. In
378 Spanish.
379 Benedito-Palos, L.; Navarro, J. C.; Sitjà-Bobadilla, A.; Bell, J. C.; Kaushik, S.;
380 Pérez-Sánchez, J., 2008: High levels of vegetable oils in plant protein-rich diets
381 fed to gilthead sea bream (*Sparus aurata* L.): growth performance, muscle fatty
382 acid profiles and histological alterations of target tissues. Br. J. Nutr. **100**, 992-
383 1003.
384 Borrell, Y.J.; Carleos, C. E.; Asturiano, J.F.; Bernardo, D.; Vázquez, E.; Corral,
385 N.; Sánchez, J.A.; Blanco, G., 2007: Use of microsatellites and a combinatorial
386 optimization approach in the acquisition of gilthead sea-bream (*Sparus aurata*, L.)
387 broodstocks for hatcheries. Aquaculture **269**, 200-210.
388 Borrell, Y.J.; Gallego, V.; García-Fernández C.; Mazzeo, I.; Pérez, L.; Asturiano,
389 J.F.; Carleos, C.E.; Vázquez, E.; Sánchez, J.A.; Blanco, G., 2011: Assessment of
390 parental contributions to fast and slow growth progenies in the sea bream *Sparus*
391 *aurata* L. using a new multiplex PCR. Aquaculture **314**, 58-65.
392 Dupont-Nivet, M.; Vandeputte, M.; Vergnet, A.; Merdy, O.; Haffray, P.;
393 Chavanne, H., Chatain, B. 2008: Heritabilities and GxE interactions for growth in
394 the European sea bass (*Dicentrarchus labrax* L.) using a marker-based pedigree.
395 Aquaculture **275**, 81–87.

396 Gjedrem, T., 1983: Genetic variation in quantitative traits and selective breeding
397 in fish and shellfish. Aquaculture **33**, 51-72.
398 Gjedrem, T., 1997: Flesh quality improvement in fish through breeding. Aquacult.
399 Int. **5**, 197-206.
400 Grigorakis, K., 2007: Compositional and organoleptic quality of farmed and wild
401 gilthead sea bream (*Sparus aurata*, L.) and sea bass (*Dicentrarchus labrax*) and
402 factors affecting it: A review. Aquaculture **272**, 55-75.
403 Grigorakis, K.; Alexis, M. N., 2005: Effects of fasting on the meat quality and fat
404 deposition of commercial-size farmed gilthead sea bream (*Sparus aurata* L.) fed
405 different dietary regimes. Aquacult. Nutr. **11**, 341-344.
406 Grigorakis, K.; Alexis, M.N.; Taylor, K.D.A.; Hole, M., 2002: Comparison of
407 wild and cultured gilthead sea bream (*Sparus aurata*); composition, appearance
408 and seasonal variations. Int. J. Food Sci. Technol. **37**, 477-484.
409 International Organization for Standardisation, ISO 4120, 1983: Sensory
410 Analysis. Triangle Test. International Organisation for Standardisation, Geneva,
411 Switzerland.
412 Kause, A.; Quinton, C.D.; Ruohonen, K.; Koskela, J., 2009: Genetic potential for
413 the regulation of variability in lipid and protein content of European whitefish
414 (*Coregonus lavaretus*). Br. J. Nutr. **101**, 1444-1451.
415 Martínez-Llorens, S.; Tomás Vidal, A.; Moñino, A.V.; Gómez Ader, J.; Pla
416 Torres, M.; Jover Cerdá, M., 2008: Blood and haemoglobin meal as protein
417 sources in diets for gilthead sea bream (*Sparus aurata*): effects on growth,
418 nutritive efficiency and fillet sensory differences. Aquacult. Res. **39**, 1028-1037.
419 Meilgaard, M.; Civille, G.V.; Carr, B.T., 1999: Sensory Evaluation Techniques
420 (3th edition), CRC Press, Boca Raton, Florida, USA. 387 pp.

421 Moretti, A.; Pedini Fernandez-Criado, M.; Cittolin, G.; Guidastri, R., 1999:
422 Manual on hatchery production of Seabass and Gilthead sea-bream, FAO, vol 1
423 pp 194.
424 Navarro, A.; Zamorano, M.J.; Hildebrandt, S.; Ginés, R.; Aguilera, C.; Afonso,
425 J.M., 2009a: Estimates of heritabilities and genetic correlations for growth and
426 carcass traits in gilthead seabream (*Sparus aurata* L.), under industrial conditions.
427 Aquaculture **289**, 225-230.
428 Navarro, A.; Zamorano, M.J.; Hildebrandt, S.; Ginés, R.; Aguilera, C.; Afonso,
429 J.M., 2009b: Estimates of heritabilities and genetic correlations for body
430 composition traits and G x E interactions, in gilthead seabream (*Sparus aurata*
431 L.). Aquaculture **295,** 183-187.
432 Quinton, C.D.; McMillan, I.; Glebe, B.D., 2005: Development of an Atlantic
433 salmon (*Salmo salar*) genetic improvement program: Genetic parameters of
434 harvest body weight and carcass quality traits estimated with animal models.
435 Aquaculture **247**, 211-217.
436 Sánchez-Lozano, N.B.; Martínez-Llorens, S.; Tomás-Vidal, A.; Jover Cerdá, M.,
437 2009: Effect of high-level fish meal replacement by pea and rice concentrate
438 protein on growth, nutrient utilization and fillet quality in gilthead seabream
439 (*Sparus aurata*, L.). Aquaculture **298**, 83-89.
440 Shearer, K.D.; Åsgård, T.; Andorsdöttir, G.; Aas, G.H., 1994: Whole body
441 elemental and proximate composition of Atlantic salmon (*Salmo salar*) during the
442 life cycle. J. Fish Biol. **44**, 785-797.
443 Sitjà-Bobadilla, A.; Peña-Llopis, T.S.; Gómez-Requeni, P.; Médale, F.; Kaushik,
444 S.; Pérez-Sánchez, J., 2005: Effect of fish meal replacement by plant protein
445 sources on non-specific defence, mechanisms and oxidative stress in gilthead sea

446 bream (*Sparus aurata*). Aquaculture **249**, 387-400.

447 Thorland, I.; Papaioannou, N.; Kottaras, L.; Refstie, T.; Papasolomontos, S.; Rye,
448 M., 2007: Family based selection for production traits in gilthead seabream
449 (*Sparus aurata*) and European sea bass (*Dicentrarchus labrax*) in Greece.
450 Aquaculture **272S1**, 314.

451 Zohar, Y.; Billard, R.; Weil, C., 1984: La reproduction de la daurade (*Sparus*
452 *aurata*) et du bar (*Dicentrarchus labrax*) connaissance du cycle sexuel et controle
453 de la gametogenese et de la ponte. In: L'Aquaculture Du Bar et Des Sparides. Eds:
454 G. Barnabé; R. Billard, Paris, France, INRA. pp. 3–24.

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Tables

Table 1. Weights and longitude at the moment of the first larval selection

Batch	Size	Heads	Size	Tails
A	>4.5 mm	0.74 ± 0.20 g	<3.5 mm	0.18 ± 0.24 g
		3.91 ± 0.30 cm		2.52 ± 0.24 cm
B	>3.5 mm	0.31 ± 0.10 g	<2.5 mm	0.06 ± 0.02 g
		3.91 ± 0.30 cm		1.89 ± 0.19 cm

Table 2. Growth parameters of cultured gilthead sea-bream at the end of the feeding period. Values are reported as mean ± SEM. AH=A Heads, AT=A Tails, BH=B Heads, BT=B Tails. $n_{AH,\ AT,\ BH,\ BT\ Initial\ weight} = 250$; $n_{AH\ final\ weight} = 206$; $n_{AT\ final\ weight} = 153$; $n_{BH\ final\ weight} = 155$; $n_{BT\ final\ weight} = 136$.

	AH	AT	BH	BT
Initial weight (g)	47.8 ± 8.6 b	10.6 ± 2.3 a	41.5 ± 8.5 b	8.9 ± 2.2 a
Final weight (g)	301.9 ± 51.7 b	195.9 ± 28.9 a	320.7 ± 52.0 b	183.9 ± 32.7 a
	Heads		Tails	
FI (g 100 / g fish x d)	0.93 ± 0.02 a		1.09 ± 0.02 b	
DGI (% / d)	1.49 ± 0.45		1.67 ± 0.45	
TGC (x1000)	1.49 ± 0.05		1.67 ± 0.05	
FCR	1.40 ± 0.05		1.35 ± 0.05	
Growth slope (b)	1.24 ± 0.05 b		0.84 ± 0.05 a	

FI (Feed Intake, g 100 / g fish x d) = 100×feed consumption (g) / average biomass (g) x d; DGI (Daily Growth Index, %/d) = 100 × (final weight$^{1/3}$- initial weight$^{1/3}$) / d; TGC (Thermal Growth Coefficient, TGC x 1000 = (Final weight $^{1/3}$ – Initial weight $^{1/3}$) / (∑ ºC effective x d); FCR (Feed Conversion Ratio = feed offered (g) / weight gain (g).

506 Table 3. Body parameters at the end of the growing period. Values are reported as mean ± SEM. Different letters mean significant differences
507 between head and tail specimens of a same batch. AH=A Heads, AT=A Tails, BH=B Heads, BT=B Tails. $n_{AH, AT, BH, BT} = 15$.

	AH	AT	BH	BT
Body weight (g)	290.5 ± 11.6 b	186.0 ± 11.6 a	319.3 ± 12.0 b	176.3 ± 12.0 a
Total length (cm)	24.3 ± 0.4 b	21.7 ± 0.4 a	24.7 ± 0.3 b	20.9 ± 0.4 a
C_F	1.97 ± 0.04 b	1.81 ± 0.04 a	2.07 ± 0.04 b	1.93 ± 0.04 a
I_G (%)	0.11 ± 0.02 b	0.00 ± 0.02 a	0.20 ± 0.03 b	0.02 ± 0.03 a
I_H (%)	1.67 ± 0.11	1.68 ± 0.11	2.04 ± 0.14 b	1.63 ± 0.14 a
I_V (%)	7.01 ± 0.32	7.69 ± 0.32	8.26 ± 0.22 b	7.62 ± 0.22 a
I_M (%)	1.51 ± 0.20	1.26 ± 0.20	2.19 ± 0.22 b	1.46 ± 0.22 a
F_Y (%)	36.91 ± 1.19	35.25 ± 1.19	38.85 ± 1.61 b	32.72 ± 1.61 a

H_P (%)	23.40 ± 0.68	23.40 ± 0.68	22.26 ± 0.49	22.49 ± 0.51

C_F (Condition Factor) = 100 x total weight (g) / total length^3 (cm); I_G (Gonadosomatic index, %) = 100 x gonad weight (g) / total weight (g); I_H (Hepatosomatic index, %) = 100 x liver weight (g) / total weight (g); I_V (viscerosomatic index, %) = 100 x viscera weight (g) / total weight (g); I_M (mesenteric fat index, %) = 100 x mesenteric fat weight (g) / total weight (g); F_Y (fillet yield, %) = 100 x (total body weight – head – viscera – fins – bone – skin - gonad) (g) / total weight (g); H_P (head percentage, %) = 100 x head weight (g) / total weight (g).

535 Table 4. Whole body composition at the end of growing period. Values are reported as mean ± SEM. Different letters mean significant
536 differences between heads and tails of a same batch. AH=A Heads, AT=A Tails, BH=B Heads, BT=B Tails. $n_{AH, AT, BH, BT} = 10$.
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	AH	AT	BH	BT
Moisture (%)	60.91 ± 0.45 a	64.44 ± 0.45 b	60.95 ± 0.69 a	65.79 ± 0.69 b
Ash (%)	2.29 ± 0.12	2.13 ± 0.12	2.19 ± 0.12	2.12 ± 0.12
Protein (%)	15.70 ± 0.14	15.98 ± 0.14	15.73 ± 0.10	15.44 ± 0.10
Fat (%)	20.77 ± 0.53 b	17.06 ± 0.53 a	20.14 ± 0.69 b	16.02 ± 0.69 a

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Table 5. Fillet composition at the end of growing period. Values are reported as mean ± SEM. Different letters mean significant differences between heads and tails of a same batch. AH=A Heads, AT=A Tails, BH=B Heads, BT=B Tails. $n_{AH, AT, BH, BT} = 5$.

	AH	AT	BH	BT
Moisture (%)	73.50 ± 0.41	74.56 ± 0.41	71.82 ± 0.39 a	73.35 ± 0.39 b
Ash (%)	1.37 ± 0.03	1.43 ± 0.03	1.43 ± 0.02	1.43 ± 0.02
Protein (%)	20.20 ± 0.28	20.56 ± 0.28	20.08 ± 0.27	20.70 ± 0.27
Fat (%)	4.58 ± 0.20	4.23 ± 0.22	6.52 ± 0.38 b	4.61 ± 0.38 a

Table 6. Results for sensory test. AH=A Heads, AT=A Tails, BH=B Heads, BT=B Tails.

Set	AH vs AT	BH vs BT
Number of triangles	116	116
Number of correct responses	42	48
Significance	> 0.05	< 0.05