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SHORT COMMUNICATION

Origin of broodstock and effects on the deformities of gilthead sea bream (*Sparus aurata* L. 1758) in a Mediterranean commercial hatchery

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Abstract The use of broodstock of different origin as a method to improve fry production performance and consequently to minimize deformities was examined at industrial scale in a commercial gilthead sea bream hatchery. The outcome of fry production from three different broodstock groups (BA: broodfish (Mediterranean) with multiannual hatchery presence, BB: selected offspring originating from the BA group, and BC: broodfish of Atlantic origin) was investigated in the same rearing conditions and feeding protocol. Performance factors assessed were the survival and weaning of the larvae; the mortality rates from the "weaning until the end of the hatchery stage" of the larvae/fry; the percentage of fry without swim bladder; the percentage of fry with skeletal deformities and the feed conversion ratio. In all factors, no statistical differences among the experimental groups were detected. However, due to early rejection of the deformed individuals, benefits are expected from the decrease of the supplied amount of food and the reduced labor cost.

Keywords Broodstock · Gilthead sea bream · Hatchery · Larval mortality · Skeletal deformities

Introduction

One of the main stages of marine aquaculture is larval production. Several improvements in hatchery rearing technology and use of broodstock have been occurred in the last decades (Theodorou 2002; Pavlidou 2009). However, problems such as skeletal deformities still represent a major factor of quality degradation and fry losses for the hatcheries. Gilthead sea bream is one of the most important cultured species in the Mediterranean aquaculture. Several studies on fry deformities exist for this species, recording qualitative and quantitative data (Koumoundouros et al. 1995; Divanach et al. 1996; Koumoundouros et al. 1997a, b; Galeotti et al. 1999, 2000; Carrillo et al. 2001; Cahu et al. 2003; Boglione et al. 2013a). There are also studies focusing on the causes of these deformities, which are related to physicochemical, genetic and nutritional factors, as well as on improvement efforts (Johnson and Katavic 1984; Liao et al. 1993; Chatain 1994; Boglione et al. 2001; Cahu et al. 2003; Verhaegen et al. 2007; Boglione et al. 2013b; García-Celdrán et al. 2014, 2015). However, most of the studies include limited data in terms of the economic benefits from the reduction of fry deformity-

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caused losses. Furthermore, the existing literature on the reduction of fry deformities using broodstock of different origin is contradictory (Liao et al. 1993; Verhaegen et al. 2007).

Consequently, the objective of the present study was to evaluate the possible economic benefits for a large-scale commercial hatchery in Greece from the replacement of the existing broodstock. This broodstock (of Mediterranean origin), routinely operating for a long period, was replaced by other broodstock sources including broodstock of Atlantic origin, taking also into account other improvements during the fry production process. Since the aim of the study was to improve the gilthead sea bream fry production at industrial scale through the reduction of fry losses caused by skeletal deformities, the effort was concentrated on the analysis of the data obtained throughout the entire typical production cycle of the hatchery.

Materials and methods

Production data and hatchery operating profile

Production data includes the tanks' stocking conditions as well as the larvae feeding regime, the physico-chemical conditions in the rearing tanks, as well as data on broodstock characteristics. The experimental broodstock groups used were BA, BB and BC. Eggs from each broodstock group were stocked during 2008 at the same time in identical plastic circular tanks of 16 m^3 each, under the same culture conditions (physico-chemical, live food quality and quantities). Egg batches were stocked three times for each group (BA1, BA2, BA3; BB1, BB2, BB3 and BC1, BC2, BC3; see also Table 1) over the production period of the hatchery.

Each experimental tank was stocked with 1 kg of eggs, and the mean number of eggs (1,400,000) was estimated, by measuring the mean egg diameter, with a mean initial rearing density of 88 larvae l^{-1}. The eggs were collected from the broodstock tanks and hatching was completed after two days. The feeding scheme (from hatching until the first 120 days) is presented in Fig. 1. The amounts as well as the type of feed supplied, until D120, were kept the same for all experimental groups (Table 2).

The physicochemical conditions were the same in all experimental tanks; Temperature until weaning was kept at 19–20 °C; afterwards and until the end of the "hatchery phase" it ranged between 18 and 22 °C. The level of oxygen saturation ranged between 85 and 105 % and pH was kept stable at 7.8. For the purpose of the study, several performance factors were evaluated as follows. The number of the weaned larvae per tank; the final survival expressed as the percentage of the initial stocking of the tank (based on the daily collection of the dead larvae/fry), as well as the percentages of: (1) the mortality (from the beginning of the weaning phase to fry sales); (2) the fry without swim bladder at a mean weight of 0.5–1.0 g which were rejected from the tanks and counted after buoyancy testing (i.e. these individuals sink to the bottom during a salinity increase from 35–36 to 58–60 ppt, after anaesthetized with 2-phenoxy-ethanol); (3) the fry with skeletal deformities which were also rejected by hand, following optical observation and counted during the previous buoyancy test; and (4) the total fry losses after weaning. The type of the skeletal deformities observed in this study were the lack of or the existence of atrophic operculum, snout/mouth dysplasia, lack of dorsal fin and hypoplasia of the caudal fin, with the last two of them being of significant commercial importance (Chatain and Corrao 1992; Morretti et al. 1999). Finally, feed conversion ratio (FCR = the amount of dry feed provided in kg/wet weight gain in kg) was also recorded for the supply period of dry feed.

The mean values of the measurements between broodstock groups were compared by applying one-way analysis of variance (ANOVA) at significance level of $p < 0.05$. All statistical tests were performed using the statistical program Statgraphics 2.1.

The hatchery used the BA broodstock group of Mediterranean origin since the early 2000s' (F_0 generation). The typical production cost per fry was estimated based on the financial statements of the hatchery's annual production of 22 million fry (Table 3). For the evaluations of the economic data the "Active Based Costing" analysis of the hatchery costs was used. The results are discussed as "production cost per fry" and "distribution of costs per fry".

A critical part of the decision-making process of the hatchery operating management is the planning for improved outcome of the next annual production cycle through innovations (Theodorou et al. 2015). In this case, the managing staff had to decide, if it is required to replace the given broodstock (BA) of Mediterranean origin with other broodfish groups such as BB broodstock (F_1 generation) from selected offspring originating

Table 1 Gilthead sea bream fry hatchery production data in three different broodstock groups and batches (standard deviations of means in parentheses)

	Experimental groups and batches of gilthead sea bream fry												p value
	BA1	BA2	BA3	BB1	BB2	BB3	BC1	BC2	BC3	Mean BA	Mean BB	Mean BC	
Final survival (%)[a]	13	15	12	15	22	15	19	28	13	13.3 (1.5)	17.3 (4.0)	20 (7.5)	0.32
Weaned fry ($\times 10^3$)	234.7	260.5	213.0	254.0	388.7	245.0	325.0	434.5	228.0	236.1 (23.8)	295.9 (80.5)	329.2 (103.5)	0.38
Mortality (%)[b]	6	6	6	5	6	8	4	8	15	6 (0.0)	6.3 (1.5)	9 (5.6)	0.52
Without swim bladder (%)	8	11	8	10	6	7	12	2	5	9 (1.7)	7.7 (2.1)	6.3 (5.1)	0.63
Deformed fry (%)	24	19	10	15	15	8	18	12	4	17.7 (7.1)	12.7 (4.0)	11.3 (7.0)	0.46
Total losses (%)[c]	37	36	22	30	27	23	34	23	24	31.7 (8.4)	26.7 (3.5)	27.0 (6.1)	0.58
Fry for sale (%)[d]	63	64	78	70	73	77	66	77	76	68.3 (8.4)	73.3 (3.5)	73.0 (6.1)	0.58
FCR[e]	1.53	1.09	0.97	1.11	1.22	0.99	1.16	1.13	1.04	1.2 (0.3)	1.1 (0.1)	1.1 (0.1)	0.77

[a] As a percentage of the initial stocking of the tank

[b] From the weaning phase to the sale of the fry

[c] These losses refer to the fry after the weaning phase

[d] The remaining fry for sale

[e] Feed conversion ratio was recorded throughout the dry feed supply period

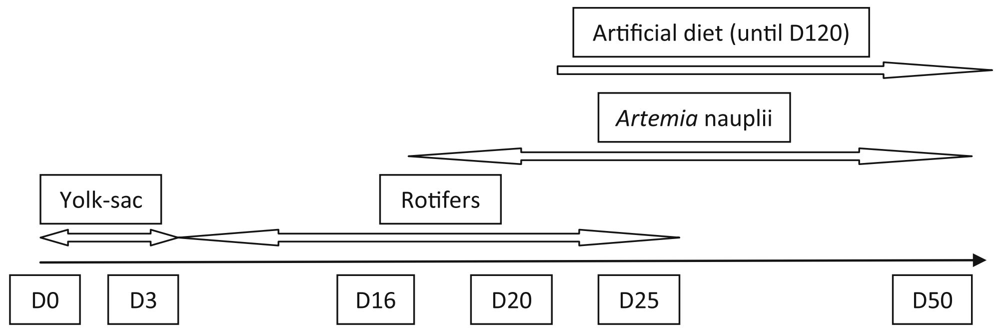

Fig. 1 Feeding scheme of larvae/fry during 120 days post-hatching

Table 2 Food supply per experimental culture tank of gilthead sea bream fry

Live food type	Quantity	Dry feed (μm)	Quantity (kg)
Algae	11.5 m^3	100–200	2
Rotifers	3200×10^6	200–300	20
Artemia nauplii	5300×10^6	300–500	35
		400–500	65
		500–800	190
		800–1200	350

Table 3 Typical cost structure of the gilthead sea bream hatchery with annual production of 22 millions fry per year

Type of hatchery costs	Total costs (%)	Values (€)
Staff-labor	40	580,000
Energy	6	85,000
Larvae-weaning feeds	15	215,000
Fry-feeds	7	95,000
Drugs-chemicals	1	18,700
Oxygen	3	41,500
General expenditures	6	102,000
Administrative costs	7	100,000
Depreciations	15	210,000
Total	100	1,447,200
Cost per fry		0.066

from the BA broodstock group. Eggs from this group were first used in the production cycle of the hatchery during 2007. The BC group consisted of Atlantic (French) broodfish, originally imported as fry of different geographic origin, and was compared to the previous two groups.

The BA broodstock group consisted of 43 fish of mean weight of 4.9 kg, BB group of 45 fish with mean weight of 2.4 kg and BC group of 40 fish with mean weight of 2.6 kg. In all groups males were less than 10 %. The exact number of males and the effective breeding numbers are not available. Sex ratio in sea bream broodstock tanks is not stable given that the species is a protandrous hermaphrodite. Moreover, hatchery managers avoid disturbing the broodfish with manual handling (i.e., stripping) in spawning tanks in order to reduce the stress levels. The eggs in all groups used in the current experiment were produced in a 7–10 day period (from 4 to 15 of May 2009), under increasing artificial photoperiodic regime, which is suitable for spawning synchronization. The protocol was LD 10:14 during the middle of the spawning period (May), starting from LD 8.5:15.5 at the beginning (15th of March) to LD 12:12 close to the end (30th of June). Hatching took place more or less simultaneously. The typical broodstock selection process was based on growth rate and the desired phenotype (i.e., body shape). The first selection stage was performed during the pre-ongrowing phase, where the 2 % of the fastest growing fry was transferred to floating cages in the sea. Then, at a body weight >800 g, the best 500 fish were transferred to the hatchery in quarantine and finally the best 50 broodfish were selected at most to become the broodstock group.

Results

Production results

The production data in the three broodstock groups are presented in Table 1. Fry without swim bladder ranged between 2 and 12 %, the percentage of deform fry varied at 4–24 %, while ready-to-sale fry rates ranged between 63 and 78 %. Although no statistical differences were detected ($p > 0.05$ in all performance factors), the BA group presented the higher percentages of fry without swim-bladder and fry with deformities. Accordingly, total losses were increased and conversely the available fry for sale decreased compared to rest of broodstock groups. The most common deformities detected in all groups were those on the head region, while specimens with opercular deformities represented 2 % of the overall deformities. No significant differences were also evident in the final survival rate, fry rejections due to skeletal deformities and fry without swim bladder as well as in the FCR.

Economic results

The recording of all economic data for the typical production procedure of the hatchery revealed that production cost was €0.066 per fry when the hatchery was fully operating year-round for a production capacity of 22 million fry, as shown in Table 3.

The distribution of the annual costs in terms of both feeds and personnel costs is presented in Figs. 2 and 3, respectively. *Artemia* cysts together with the enriched food represented 40 % of the total food costs, while starter feeds during the weaning phase represented 22 % of the costs (Fig. 2). The personnel costs represented approximately 50 % of the entire costs of the hatchery (Fig. 3).

Given that there was no statistical differences in the production outcome, the elimination of the deformities between the existing operating broodstock group (BA) and the tested alternative sources (BB and BC groups) is not expected to affect dramatically the economic performance and accordingly the annual returns.

Discussion

Deformities such as the opercular absence affected until recently almost up to 80 % of the farmed gilthead sea bream (Chatain 1994; Andrades et al. 1996; Verhaegen et al. 2007). Increased number of fry without swim

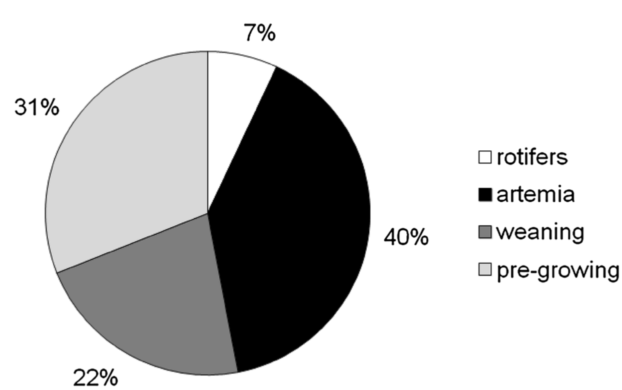

Fig. 2 Hatchery's typical distribution of food costs for gilthead sea bream fry production

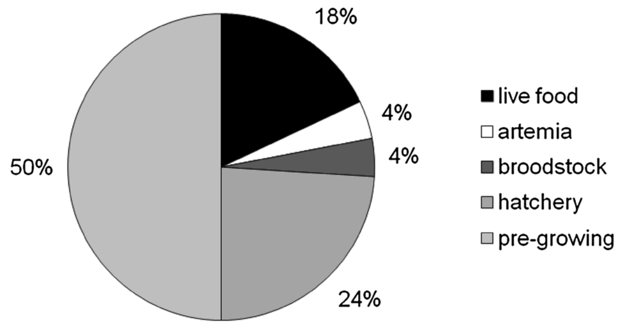

Fig. 3 Hatchery's typical distribution of personnel costs for gilthead sea bream fry production

bladder was observed in the second batch of BA broodstock group and in the two first batches of BB and BC groups. It was attributed to technical problems in the air supply system that occurred in the hatchery, causing problems to the oil skimmers. This confirmed the importance of the skimmers in inhibiting lipid formation on water surface (Chatain and Ounais-Guschemann 1990).

In the present study, the three different broodstock groups used in the hatchery showed no significant differences on fry deformities and total losses. In addition, all physicochemical conditions and the feeding protocol applied were identical for all groups and no such effects as environmental or nutritional could be detected. Existing literature on the genetic influence on deform fry is contradictory, while interactions between genetic, environmental factors and rearing procedures (e.g., water velocity, temperature and artificial diets are probably the most critical factors) (Sfakianakis et al. 2006; Vatsos and Angelidis 2010) have also been suggested to explain the origin as well as the incidence of fry deformities (Kause et al. 2007; Bardon et al. 2009). Opercular deformities are considered non inheritable in *Tilapia nilotica* (Tave and Handwerker 1994) as well as in gilthead sea bream (Castro et al. 2008). However, inbreeding increased opercular deformities in *Cichlasoma nigrofasciatum* (Winemiller and Taylor 1982). It was also reported that critical performance factors such as fecundity and egg quality vary among different genetic origin of broodstock (Brooks et al. 1997).

The cost analysis revealed qualitative as well as quantitative typical data of the hatchery (Table 3; Figs. 2, 3). Significant economic losses have been attributed to high mortalities during the early developmental stages, which are common in marine aquaculture; thus any effort towards the reduction of the production cost even based only on the reduction of the deformed fry is expected to have significant impact on hatchery economics. The early rejection of deformed fry which took place in the present study reduced both feeding and personnel costs required to prepare live food and to short the fry. The reduction of the personnel occupied either in fry rearing or in live food production could result to a corresponding labour reduction in the pre-growing stage. The decrease of feeding costs became also important, since no differences were apparent in FCR in the present study.

It is concluded that, although no statistical differences were evident between the performance of the different broodstock and fry sources (BA, BB and BC), there could be economic benefits even by the slightly better values of BB and BC groups in critical factors such as the percentage of total losses and eventually the percentage of fry for sale. At any case, the scope for fry improvement through improved broodstock is open-wide; therefore, new trials should be carried out to introduce in the hatchery new possible broodstock candidates of different origins and by genetic selection programmes aiming to pedigree certification.

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