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Zaragoza : CIHEAM

Cahiers Options Méditerranéennes; n. 47

2000

pages 241-247

Article available on line / Article disponible en ligne à l'adresse :

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To cite this article / Pour citer cet article

Lazzari A., Fusari A., Boglione C., Marino G., Di Francesco M. **Recent advances in reproductional and rearing aspects of *Seriola dumerilii***. *Recent advances in Mediterranean aquaculture finfish species diversification*. Zaragoza : CIHEAM, 2000. p. 241-247 (Cahiers Options Méditerranéennes; n. 47)



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Recent advances in reproductional and rearing aspects of *Seriola dumerilii*

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SUMMARY – *Seriola dumerilii* is a very promising species for the Mediterranean aquaculture because of its high growth rate and the good performance in captivity. This study illustrates the results on the collection, transportation and captivity acclimation of wild *S. dumerilii* for building a broodstock. Preliminary results on hormone induced spawning, larval rearing and juveniles growth to date are also described. The study was carried out within a national research project started in 1996. Sexually mature fish (15-25 kg body weight) were collected in the Pelagie Islands (South Thyrrenian Sea) by purse seine vessels and stocked in a 200 m³ submersible cage at –18 meter depth. During the 1997 reproductive season several attempts were made to collect eggs released in the rearing cage. In 1998 the broodstock were transferred in circular concrete tanks and fertilised eggs were obtained after hormone treatment. Larval rearing was carried out using microalgae, rotifers and nauplii of *Artemia salina*. Biological and SEM observations were carried out to study the larval ontogenesis with particular regard to the feeding apparatus. The preliminary results validate some of the transfer and cage stocking techniques applied and point out the possibility of obtaining eggs from captive broodstock.

Key words: *Seriola dumerilii*, reproduction, broodstock, larval rearing, Mediterranean.

RESUME – "Progrès récents concernant les aspects de reproduction et d'élevage de *Seriola dumerilii*". La sériole, *Seriola dumerilii*, est une espèce très prometteuse pour l'aquaculture méditerranéenne ayant un taux élevé de croissance et réagissant très bien à la captivité. Cette étude présente les résultats sur la pêche, le transport et l'acclimation à la captivité des sérioles sauvages, pour pouvoir construire un stock de géniteurs. Des premiers résultats sur l'induction hormonale, sur l'élevage de larves et de jeunes poissons sont également présentés. Cette étude a été accomplie dans le cadre d'un programme national de recherche, débuté en 1996. Des poissons sexuellement matures (15-25 kg poids vif) ont été pêchés dans les Iles de Pélagie par des bateaux à, puis placés dans une cage submersible de 200 m³ à –18 mètres de profondeur. En 1998 les stocks de géniteurs ont été transférés dans des bassins circulaires et, après traitement hormonal, des œufs fertiles ont été obtenus. L'élevage de larves a été mené en utilisant des microalgues, des rotifères et des nauplii de *Artemia salina*. Les observations biologiques et SEM conduisent à l'étude de l'ontogenèse des larves, avec une attention particulière sur l'appareil alimentaire. Les premiers résultats ont validé certains des transferts effectués, ainsi que les techniques de stockage en cage appliquées et ont montré la possibilité d'obtenir des œufs dans un milieu de captivité.

Mots-clés : *Seriola dumerilii*, reproduction, géniteurs, élevage de larves, méditerranéenne.

Introduction

Greater amberjack has a very high growth both in natural environment (Andaloro *et al.*, 1992) and in captivity (Cavaliere *et al.*, 1989; Lazzari, 1991; García-Gómez, 1993). Juveniles, 60-70 g, caught at sea and reared in floating cages or in tanks reach a size of 1.2 kg after one year (Cavaliere *et al.*, 1989; Lazzari *et al.*, 1989; Mazzola *et al.*, 1996). Fish farmed on a fresh fish or semi-moist diet show the most remarkable growth (García-Gómez, 1993).

The need to diversify the scenery of the species farmed, together with good market value of the fish have focused on the *Seriola dumerilii* much of the attentions of the marine farmers.

The experimental trials performed in controlled environments, floating cages, land-based tanks, etc., point out the relevant importance that the *Seriola* could have in the Mediterranean context.

However, the good results obtained in the rearing of juveniles caught from the wild were not followed from relevant results on the controlled reproduction in the Mediterranean area until now.

The lack of relevant results in aquaculture in this area could be mainly due to the scarce experience grew up on the species together with the scarce availability of the fish in the sea, the restricted fishing areas and the difficulty of the fish catch.

This study reports the preliminary results on broodstock formation, hormone induced spawning and rearing of larvae and juveniles.

Broodstock formation and management

The project has started in the summer 1996, in the Pelagie Islands, South of Sicily, with the localisation of fishing areas. Fifty amberjacks, ranged from 15 to 25 kg BW, have been caught in open sea with an appropriate purse seine vessels, 40 tonnes T.S.L. and transferred into a 80 m³ hand made floating cage. The cage was carried from a speedboat in a temporary sheltered site, and moored on a -10 sandy bottom depth. After 2 weeks from the catch, fish have been weaned first with live preys, *Spondyliosoma cantharus*, *Boops boops*, and later with fresh *Scomber scomber*.

When fish seem to be well adapted to the captivity conditions and conditioned to feed on the fresh fish given by farmer-hand, they were transferred into a 200 m³ cubic submersible cage, moored on a -18 sandy bottom depth. During the three year trial, water temperature and currents have been monitored by means of a *Hugrun Seamon mini* and an *Aandera RCM 7*, respectively.

Out the reproductive period, fish have been fed every other day at 1% of BW with fresh *Scomber scomber*, while in the pre-reproductive seasons diet has been integrated with fresh *Loligo vulgaris*.

Induced spawning

In May 1998, the brooders were moved, in-group of 2-3 animals, from the submergible open sea cage to the hatchery using a small transportation cage. Fish were stocked into 2 circular concrete tanks (60 m³) and into a 100 m³ raceway supplied with running seawater. Stocking density ranged from 3.7 to 4.7 kg/m³.

Fish were anaesthetised for gonads biopsy and hormone treatment. We observed that the amberjacks are very sensitive to handling and to anaesthesia and, depending on the dose and on the water temperature, during this operations a high mortality can occur. For this reason many efforts must be done to search molecules and dose for a safe handling of these large size fish.

Good results were obtained using the 2-phenoxyethanol, at 0.1 to 0.2 ml/l dosage, directly administered in the rearing tank. After anaesthesia, sample of gonad tissue was taken by means of a plastic catheter, 500 mm length and 3 mm in external diameter to assess the stage of gonad maturation. Few females showed vitellogenic oocytes (400-600 µm), whilst most of them were stopped in previtellogenic phase or early vitellogenesis. The maturing females had a group synchronous ovarian development, as already observed in fish caught from the wild environment (Marino *et al.*, 1995a,b).

No spontaneous gamete release was observed in the spawning tank. Therefore, first hormone induction trial was performed in May 1998, when the water temperature in the tank was the same measured at sea when some spawning events in wild fish were observed.

Two different hormone solutions were used to induce the final maturation and spawning: Des-Gly¹⁰ (D-Ala⁶) LHRHa (Sigma) at 2-10 µg/kg dosage and Decapeptyl (Triptorelin, Ipsen, Biotech) at 20 and 40 µg/kg.

Some of females submitted to gonad biopsy presented atretic oocytes and evident signs of gonad reabsorption, which indicate that we were in late with the hormone injections.

The first spawning occurred after two days from the hormone induction, at 21°C water temperature. Three spawning events were observed and all the eggs were collected and incubated (Table 1).

Table 1. Incubation of the eggs

Hatching tank	Eggs (g)	Hatched larvae (no.)	Temperature (°C)
ES2	700	350,000	20.5
ES5	850	650,000	20.5
ES6	700	450,000	20.5
Total	2250	1,450,000	

Fertilised eggs were collected with a 500 µ-mesh size surface collector placed at the exit of the tank.

Few hours after the seedling in the incubators, 20% of the eggs were not viable. Mean egg diameter was 1.15 mm and mean diameter of the oil drop was 0.3 mm. The eggs were incubated in 600-l incubators with weak aeration and no light. Larvae hatched after about 40 h at 20°C. Mean hatching rate was 62.2%. The total number of hatched larvae was 1.4×10^6 .

Rearing techniques and larval development

After hatching, larvae were transferred in a 60 m³ circular tank equipped with an air lift system. Salinity was 39‰, pH ranged from 8.1 to 8.3 and water temperature was maintained at 20-22°C (Table 2). During the first phase of the larval rearing, microalgae, *Chlorella* sp., *Nannochloropsis* sp. and *Isocricis* sp. were introduced in the tanks and the rotifer *Brachionus plicatilis* was used as first live preys. Nauplii of *Artemia salina* (450 µ) were utilised in the second rearing phase.

Table 2. Larval rearing experimental protocol

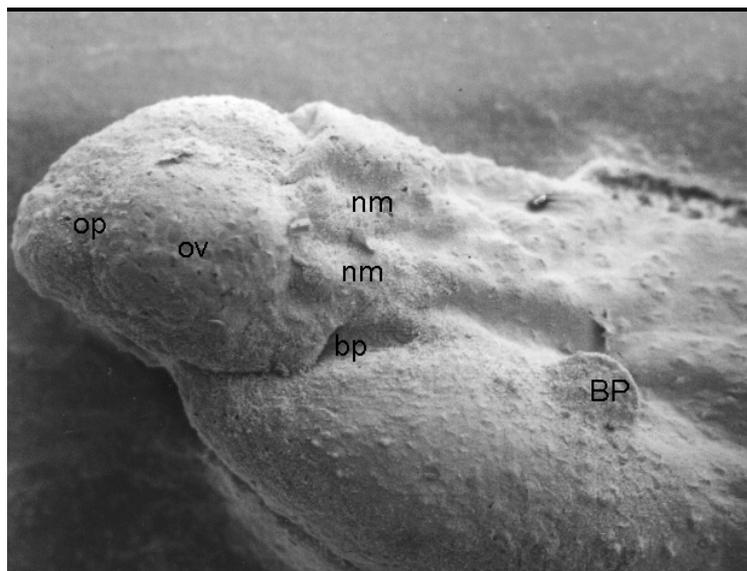
Age (days)	Temp. (°C)	No. x 10 ⁶	Larvae/l	Water renewal	Air bladder (%)	NH ₄	Fito M/cc	Rotifers M	Artemia (g)	L:D (h)
2-3	21	1.4	23	0.1	–	0	–	–		10-14
4	21	1.4	20	0.1	–	0	0.1	60	–	10-14
5	21.1	1.4	15	0.1	–	0	0.25	200	–	10-14
6	21.1	1.4	15	0.1	15	0	0.16	200	–	10-14
7	21.3	1.4	5	0.1	60	0.01	0.1	150	–	10-14
8	21.5	1.4	1	0.1	60	0	0.08	180	–	10-14
9	21.5	1.4	0.5	0.1	60	0.01	0.1	200	–	10-14
10	21.5	1.4	0.5	0.1	60	0.01	0.25	250	–	10-14
11	22	1.4	0.5	0.1	60	0	0.2	200	–	10-14
12	22	1.4	0.5	0.1	60	0.01	0.25	200	5	10-14
13	21.6	1.4	0.5	0.1	60	0.02	0.15	200	20	10-14
14	21.7	1.4	0.5	0.1	60	0.01	0.15	200	60	10-14
15	21.5	1.4	0.5	0.1	60	0	0.08	200	50	10-14
16	21.5	1.4	0.5	0.1	60	0.01	0.15	200	50	10-14
17	21.5	1.4	0.5	0.1	60	0.01	0.1	200	50	10-14
18	21.6	1.4	0.5	0.5	60	0.01	0.08	–	100	10-14
19	21.4	1.4	0.5	0.5	60	0.01	–	–	100	10-14
20	21.3	1.4	0.5	0.5	60	0.01	–	–	100	10-14

Mortality occurred in tank from the day 6th to the day 9th, probably due to lack of predation on rotifers. The weaning food was supplied up to the day 30th and from the day 40th they have been selected and transferred in raceways. At the day 60th the fingerling's BW was about 2 g, at the day 90th they reached 30 g and at the day 120th, BW was about 130 g.

Larvae and juveniles have been screened for quality and morphological observations have been carried out. Samples at different stages of growth have been fixed in glutaraldehyde 2.5% in monosodic cacodilate buffer 0.1 M, pH 7.2, for scanning electron microscopy (SEM) and in Bouin' fluid for optic microscopy.

Our investigation were aimed at widening ontogenesis knowledge on larvae and juveniles, with particular regard to *apparati* involved in trophic behaviour, such as digestive, sensorial and locomotory, in order to optimise larval diets and rearing protocols. Observations on digestive apparatus development are actually in progress; meanwhile some preliminary results are available on the development of some sense organs. The considered sensory organs have been vision, olfaction, hearing, mechano- and chemoreception.

At hatching (Fig. 1) larvae showed several mucous cells scattered on the whole body, two olfactory placodes with sensorial ciliated cells, optic vesicles, otic capsules. One-two neuromasts are dorsally placed to both the branchial pores, and one on each side of the trunk. Some larva showed already pigmented eye.



op: olfactory placode
 ov: otic vesicle
 nm: free neuromasts of the lateral line
 bp: branchial pore
 BP: pectoral fin bud

Fig. 1. Anterior region of a just hatched amberjack. The bud of larval pectoral fin is present dorsally to the yolk sac.

From the 4th day, each observed larva has showed an open mouth, a quite completely reabsorbed yolk sac, and developed larval pectoral fins. Taste buds have been observed starting from the 5th day in the ventral mucosa of pharynx, and on the lip by the 33rd day. Neuromasts placed near the branchial slit progressively augmented in number, and migrate towards the cephalic region (lined in supra- and sub-ocular rows), converging near the olfactory region (20th day, Fig. 2). Others migrate dorsally, reaching the medial region of the trunk to form the trunk lateral line. Retina is completely differentiated by the 7th day. At the 35th day, amberjack has upward turned the notochord and shows completely differentiated impair fins. Teeth are present on buccal rim, palate, pharynx, and on branchial arches, both dorsally and laterally. Nares are forming and taste buds are scattered on lips, oral cavity, pharynx and lateral region of branchial arches.

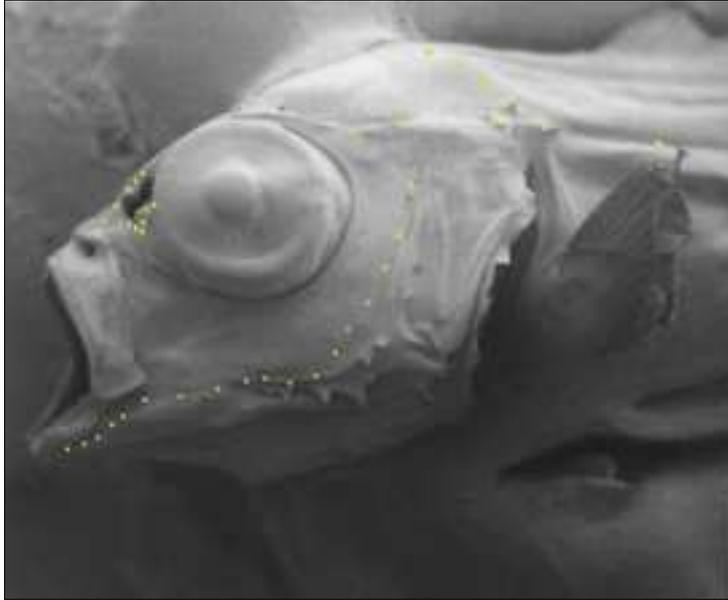


Fig. 2. Cephalic region in a 34th days-old amberjack: circles indicate the position of lateral line free neuromasts.

Data we obtained indicated that, at the beginning of heterotrophic phase, the amberjack larvae probably locate the prey by sight and mechano-reception at first, also by olfaction later. Starting from the 5th day post hatching, larvae have, furthermore, the capacity to decide to swallow or reject captured prey, basically on organoleptic characteristics. This organoleptic selectivity is destined to progressively increase along with the development. From the day 35th from hatching, amberjack may be considered as a juvenile, nourishing by branchial filtering and tasting each ingested particle with chemoreceptors. This type of "ram" feeding still persists at 152 day.

Conclusions

Most of the large pelagic species are threatened in the Mediterranean sea for their reproductive behaviour in a multiple pressure context (fishery, pollution, etc.). One of the duties of aquaculture is to widen the number of species farmed and to provide to the farmers fast growth valuable species for quick return charges. The amberjack, *Seriola dumerilii*, hold all the qualifications to become in the next years a candidate for a large scale Mediterranean aquaculture.

The main conclusion which can be drawn from the on course project regard the:

(i) *Catch of the broodstock*, which is challenging but made simpler than to the reproductive behaviour of the species.

(ii) *Acclimation*, easily reached in floating cage, which should be preferred to other structures before the transfer in land-based tanks.

(iii) *Handling*, is one of the most delicate steps in the management of *Seriola dumerilii* broodstock, mainly because to the lack of experience in the handling of large size fish.

(iv) *Reproductive condition of broodstock*, gonad maturation is achieved under captivity conditions and induced spawning can be induced with appropriate hormone treatment.

(v) *Controlled reproduction and larval rearing*, are preliminary but promising achievements and open new perspectives for *Seriola dumerilii* farming in Mediterranean area.

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