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LIPID CLASSES OF *PAGRUS PAGRUS* EGGS AND STARVED LARVAE FROM ADULTS FED ON THREE DIETS

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Introduction

In fish culture, the highest mortality rates always occurs during embryo and larval development, and they are widely influenced by the rearing conditions and broodstock quality. When females are capable of spawning several times, the broodstock diet has a greater effect on egg composition than species with one spawn that requires a longer gonad maturation time. The success of *Sparus aurata* culture has led the way for the culture of similar species like *Pagrus pagrus*.

Many authors stress the importance of lipids during embryo and larval development, due to their role as an energy source and in cell construction. They also suggest the use of a condition index based on triacylglycerol (TG) and polar lipid (PL) function (Lochmann et al., 1995). It is well known that the important lipids remain as long as possible during a starvation period (Rainuzzo, 1993), and this knowledge aids in choosing the starter food or the right enrichment. It is therefore necessary to follow the evolution of lipids from the egg to starvation death.

Materials and methods

Adults of *P. pagrus* were caught in summer of 1998 and kept in floating open-ocean cages until November 1998, when they were separated between six 8 000-l tanks and fed on three different diets: (A) natural food composed of minced fish and mussels; (B) a commercial diet for gonad maturation (Ewos #9); and (C) a mixture of A and B. The spawns were spontaneous and started in January, continuing until the end of June 1999, but only spawns of February are included in this work. The newly spawned eggs were transferred to 100-l tanks with a continuous water flow and natural photoperiod. The temperature varied from 19-21°C and no food was added.

Fertilization samples were taken every six hours until hatch, which took place at 48 hours. For larvae, the samples were taken every twelve hours until 72 hours (three-day-old larvae) and every 24 hours after that until 144 hours (seven days), belonging to the last starvation larvae, although in the present paper, only newly spawned, hatched, and last starved data are offered.

Eggs and larvae were lyophilized before biochemical analysis. The total lipids were obtained in triplicate according to Folch et al. (1957), and neutral (NL) and polar (PL) lipids were separated by adsorption chromatography, passing the samples through a sepak of silica with chloroform and chloroform:methanol 49:1 for NL and methanol alone for PL.

The lipid classes were obtained by TLC using an Iatroscan with a flame ionization detector. NL were developed in hexano:diethylether:formic acid 85:15:0.04 v:v solution, and in chloroform:methanol:distilled water 75:35:3.5 for PL

In this paper, just the major classes in each case are given, and the data are given in percentage of lipid class in relation to 100% neutral or polar lipids (Table I).

Table I. Lipid classes percentage at spawn (sp) hatch (ht), and 72- and 144-hour larvae. TL percentage is related to dry weight and the rest are related to TL.

	Diet A				Diet B				Diet C			
	sp	ht	72h	144h	sp	ht	72h	144 h	sp	ht	72h	144h
SE	18.5	22.96	10.53	5.59	17.81	18.8	7.6	3.19	22.38	22.73	12.44	2.36
TG	25.34	29.31	14.15	0.86	28.45	25.3	15.52	0.23	25.24	19.19	3.75	0.18
Cho	2.75	5.1	13.2	18.25	6.13	5.63	13.27	16.53	3.58	6.55	14.29	18.78
Other NL	TR	0.62	1.08	3.55	0.34	4.85	3.61	7.19	1.96	7.87	12.82	3.45
NL	46.59	57.99	38.96	28.25	52.05	54.58	40.01	27.14	53.9	56.34	43.3	24.77
PC	40.79	34.66	43.89	42.57	38.74	36.53	34.34	39.34	37.55	36.42	40.35	38.86
PE	5.68	2.35	10.82	20.03	3.86	3.76	7.52	16.38	4.16	3.55	8.61	22.97
Other PL	6.94	5.01	6.33	9.16	5.26	5.13	18.14	17.14	4.39	3.69	7.74	13.4
PC/PE	7.18	14.75	4.06	2.13	10.04	9.72	4.57	2.4	9.03	10.26	4.69	1.69
PL	53.41	42.01	61.04	71.75	47.95	45.42	59.99	72.86	46.1	43.66	56.7	75.23
TI	19.67	24.25	20.04	13.94	19.97	23.55	18.6	10.92	20.05	22.7	19.66	11.36

Results and discussion

Embryo. There was a decrease in phosphatidylcholine (PC) and phosphatidylethanolamine (PE) percentage before hatch, but an increase in NL, probably because the embryo is preparing to utilize more 'fast' energy (free fatty acids, triacylglycerols, steryl esters) for swimming. TG decreased in treatments B and C (mixed and commercial diets), but its catabolism released the FFA and

monoglycerids (MG) as an energy source. At hatch, the natural food treatment showed more TG content and SE synthesis.

Newly hatched larvae to start feeding. During this stage, larvae endure exponential growth, increasing the PL to synthesize cell membranes. For diets with natural food (A and C), PC and PE increased until the onset of starvation, but a bit earlier for diet B. Cholesterol became an important class along this period duplicating its percentage in all diets. Brockerhoff (1974) and Rainuzzo et al. (1992) suggest cholesterol and PL or PC form a complex to build the biomembranes.

Starvation. Without a food supply, the more energetic lipids were withered and CHO was the only important NL class. At seven days old, larvae lost almost all the 'fast' energetic classes. Even the FFA, DG, and MG resulted from catabolism of others, and PC could be an alternative metabolic energy source during this condition. The PE is not easily explained simply by PC catabolism in these cases, and probably other classes could release the n-3 HUFA that PE synthesis requires.

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