The effect of PC/PI ratio on malformations in Gilthead seabream

Amos Tandler & Bill Koven

Introduction

Malformation in commercially raised fish, such as cranial, skeletal and gill cover deformities is a major factor reducing their market value (Koumoundourous et al., 1997). Although these deformities are most apparent in the juvenile and adult stages they may originate from suboptimal nutrition during the critical larval rearing stage. Previous research has hypothesised that dietary phosphatidylinostol (PI) was more effective in reducing deformities than the main membrane phospholipid phosphatidylcholine (PC) (Geurden et al., 1997, 1998). Consequently, the aim of this study was to test the effect of different dietary ratios of PC and PI fed to the gilthead sea bream (Sparus aurata) larvae, on developmental performances in juvenile fish in terms of survival, growth and malformation rate.

Materials and Methods

Four microdiet (MD) treatments, that differed in their PC/PI ratio and replaced 75% of the normal Artemia ration (wt/wt), were fed to 20-34dph (days post hatching) sea bream larvae. In addition to the high PC/PI or low PI containing MD control, a commercial reference treatment (100% Artemia ration) was given. The dietary PC/PI ratio in the experimental treatments is outlined in Table 1. *The composition of the microdiets is given at the end of this section.*

Treatment	A	B	C	D	Art
(g/100 g dry diet)	25% Artemia +75% MD[1]	25% Artemia +75% MD[1]	25% Artemia +75% MD[1]	25% Artemia +75% MD[1]	100% Artemia
Phospholipids					
Phosphatidylcholine (PC)	5.7	4.54	4.42	3.88	4.42
Phosphatidylinositol (PI)	1.86	1.95	2.76	3.04	1.78
PC/PI in total diet	**3.07**	**2.32**	**1.6**	**1.28**	**2.48**

Table 1: The dietary PC/PI ratio in the experimental treatments

At 40 dph, the fish were graded in all treatments into small (<1.3mg dw larva^{-1}) and large (>2.9mg dw larva^{-1}) larvae, in order to test if growth rate influenced treatment effect throughout development to 141 dph.

The experiments were carried out in four replicates and results analyzed by a one-way ANOVA followed by the Tukey-Kramer Honestly Significant Difference (HSD) multiple range test (P<0.05).

Results

There was no marked (P>0.05) treatment effect on growth rate in 40 dph larvae although larvae fed the MDs were significantly (P<0.05) smaller then the commercial reference treatment (Art) larvae.

On the other hand, in later juvenile development (67 dph), decreasing dietary PC/PI ratio contributed to a significantly (P<0.05) better growth and non-significant (P>0.05) higher survival (Figure 1).

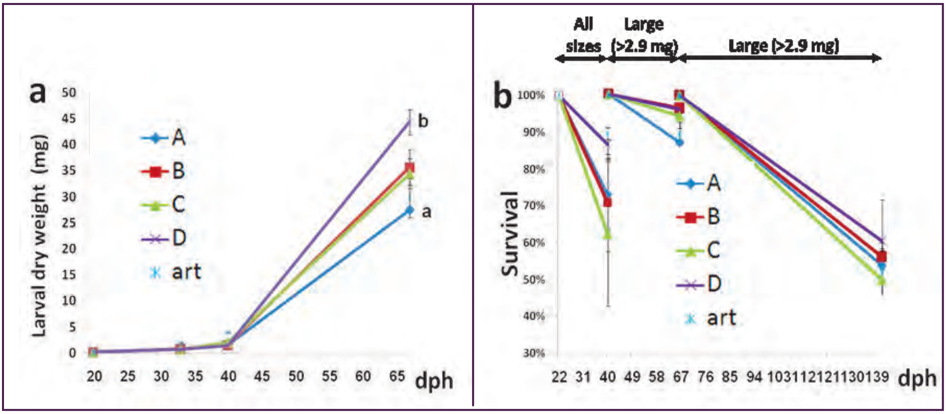

Figure 1: The effect of dietary PC/PI ratio on large (>2.9mg dw larva^{-1}) larvae (a) dry weight (20-67dph) and (b) survival (22-141dph).

Moreover, reducing dietary PI markedly (P<0.05) increased jaw (cranial) deformity in both size groups at 67dph, which may have adversely affected their feeding once weaned completely on to a dry hard starter feed. This is suggested since fish fed the high PC/PI ratio (low PI) diet demonstrated poorer growth at 67dph.

Conversely, increasing dietary PI (reducing PC/PI ratio) showed a non-significant trend of increased skeletal deformity which was markedly (P<0.05) higher in faster growing larvae in all MD treatments (Fig. 2). Interestingly, both Cranial and skeletal malformations increased throughout development (67-141dph) in most of the treatments ,suggesting that they were not deleterious.

A possible explanation for the jaw-skeletal contradiction is by the Osteocalcin (BGP) mRNA levels. High level of PI contributed to higher BGP levels, which reduced significantly (P<0.05) the jaw deformity levels while simultaneously elevated the skeletal deformities due to over mineralization.

Although there was no clear affect of PC/PI ratio on gill cover deformity rate, there was a size dependent susceptibility to this deformity where smaller larvae showed the highest incidence of this malformation.

Moreover, throughout development (67-141dph), gill cover deformity generally decreased, suggesting the possibility of operculum regeneration or that the exposure of the gills was deleterious with age.

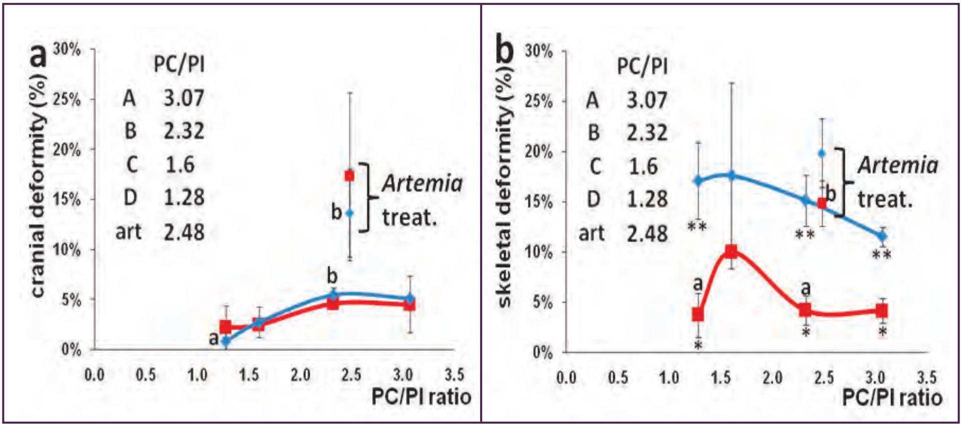

Figure 2: The effect of dietary PC/PI ratio on (a) cranial and (b) skeletal deformities level (%) in small (<1.3mg dw larva^{-1}) and large (>2.9mg dw larva^{-1}) juveniles at 67 dph.

This work has demonstrated an effective dietary PC/PI ratio of 1.28 for the sea bream larvae, effecting positively on jaw (cranial) deformity rate, growth and survival in juvenile fish.

Conclusions

Decreasing the dietary PC/PI ratio (5% total phospholipid) during larval rearing significantly increased juvenile growth. This may have resulted from a reduced incidence of jaw deformity which leads to more effective pellet feeding. More-over, the decreased rate of cranial deformity may be due to PI enhancing osteocalcin synthesis and normal jaw development. The results conclude that a PC/PI ratio of 1.28 or a PI level of 3.04g.100g^{-1} DW diet gave the best larval and fry performance.

Recommendations (seabream)

1. **For best larval growth performance, when microdiets are used, never exceed the ratio of 5 between PC/PI in the phospholipid fraction.**
2. **The best PC/PI ratio is 1.28 and a PI dietary level of 3.04g 100g^{-1} diet (DW)**

Composition of microdiets

Microdiets	A	B	C	D
Formulation (g/kg^{-1}) **Basal diet** [2]	950	950	950	950
De-oiled Soybean Lecithin [3]	0	20	40	50
Phospholipon 80 H [4]	50	30	10	0

[1] Produced at KARMAT Coating Industries Ltd., Israel.

[2] Per kg diet: 559.5 g fish meal; 44 g gelatin; 6 g acacia; 63 g fish oil (Matmor Feeds, Israel); 27 g EPAX 1050 (EPAX AS, Aalesund, Norway); Coating amino acids: 20 g glycine ; 20 g arginine; 20 g alanine and 20 g betaine (Sigma, St. Louis, USA); 0.5 g vitamin C (Stay C, Hoffman LaRoche, Switzerland); 40 g mono calcium phosphate (Fluka, Buchs, Switzerland); 10 g vitamin and mineral premix; 10 g choline chloride (Sigma, St. Louis, USA); Supplemented amino acids: 10 g valine; 10 g isoleocine; 10g tryptophan; 20 g phenylalanine(Sigma, St. Louis, USA).

[3] Deoiled soybean lecithin (Enzymotec, Migdal HaEmeq, Israel): 70.7% total PL; 23.9% phosphatidylcholine; 20.2% phosphatidylinositol; 14.5% phosphatidylethanolamine; 6.2% phosphatidic acid; 5.2%

[4] Phospholipon 80 H (Phospholipid GmbH, Köln, Germany): hydrogenated PC; 60%, Lyso PC: 10%.

References

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