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# Influence of sole and plurispecific diets on larval consumption and development of Pacific oyster *Crassostrea gigas* (Thunberg)

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## 1. Introduction

Despite an indisputable know-how in mollusc hatchery some biological aspects are still unknown and beyond them mollusc feeding requirements are poorly understood. Because a mixed algal diet increases the chances of achieving a balanced feeding, microalgae are generally supplied in plurispecific rations for bivalves without a clear knowledge of their need in essential components. However significant larval production in hatchery relies on this empirical feeding method. The diet used for *Crassostrea gigas* larvae in the experimental hatchery of Argenton (North Britanny) was issued from that applied routinely for the scallop *Pecten maximus* (Robert et al., 1994). In *P. maximus* it was shown that such a diet led to good larval development and metamorphosis but lower larval growth or metamorphosis was recorded with single microalgae (Delaunay et al., 1993). Such information was unavailable for *C. gigas* in our larval rearing context and the litterature did not report complete data throughout the whole larval stage. The objective of the present work was to study effective consumption by veligers exposed to different monospecific bispecific and trispecific diets in relation with *C. gigas* larval development performances based on growth, survival and metamorphosis.

## 2. Materials and methods

Detailed information on microalgae used here have been already reported (Robert et al., 2004). Originated from the Culture Collection of Algae and Protozoa (GB) they showed similar size (40 to 45 $\mu m^3$) and dry individual weight (18 to 20 pg: Robert et al., 2004) and mixed feeding ration was accordingly based on a ratio 1/1.
Gametes were issued from conditionned broodstock at 19°C, in a flow through system, with a daily mixed diet of 6% *Isochrysis aff. galbana* (clone t. Iso), *Chaetoceros gracilis* and *Skeletonema costatum* per mg oyster (dry algal weight/dry meat weight). Two day old D larvae were then distributed in 30L cylindro-conical tanks ($5.ml^{-1}$ density, 1 µm filtered seawater, 25°C, 34‰, 0.5 $L. mn^{-1}$ aeration). Seawater renewal and tanks cleaning occured three times a week with no antibiotic addition. Larvae were daily fed on different sole or mixed diets, however at equal quantity and adjusted to larval increasing demand. Larval length and survival were achieved on days 2, 9 and 16 by use of image analysis technique.
The number of pediveligers ready to set was estimated prior to a selective grading on a 200 µm sieve mesh aperture. Then, the largest larvae were transferred in 30L tanks in presence of 15 cm diameter plastic disks, used as collectors. Six days latter, experiments ended and settlement was evaluated by counting precisely the number of remaining pediveligers.
For each nutritionnal condition, phytoplancton consumption was evaluated by means of an electronic particle counter ZM equipped with a 100 µm aperture tube. PTCp was the reference diet and when food limitation was detected in the control, daily rations were fitted accordingly and applied to the other nutritionnal conditions.
The influence of monospecific diets (P = *Pavlova lutheri*, T = t. Iso et Cp = *C. calcitrans* forma *pumilum*), and bispecific diets (PT, PCp et TCp) on larval growth, survival and metamorphosis were studied.

## 3. Results

TCp was the best diet leading to high survival (98% ; table 1) and growth ($13.2\ \mu m.d^{-1}$). Larvae fed exclusively *Chaetoceros calcitrans* forma *pumilum* exhibited a reasonable growth ($10\ \mu m.d^{-1}$) however with lower survival on day 16 (76%) and characterized by a high intertank variability

(CV = 26%). In contrast, feeding larvae with *Pavlova lutheri* led to a poor development, low survival (29%) and low growth (1.7 µm.d$^{-1}$) closed to starved larvae performances (Table 1) while t. Iso led to intermediate results (86 % survival and 6 µm.d$^{-1}$ growth).
While the number of pediveligers competent to metamorphosis was highest on day 16 for TCp diet (70%) metamorphosis, recorded one week later, was similar to the other mixed diets (72-77%) except PT that never led to competent larvae (Table 1). In contrast, larvae solely fed *C. calcitrans* f. *pumilum* exhibited lower larval competence and metamorphosis on day 16. The larvae fed the other diets did not showed any competence on day 16 and despite a prolongation of their rearing a similar situation occurred on day 22.
Grazing was determined for all diets, throughout larval development from days 6 to 15, and during metamorphosis up to day 22 for three of them (PCp, TCp and PTCp). Such a study, performed for each condition in triplicate and expressed as the number of cells eaten per day per larva, showed a preferential uptake of microalgae with avec P< PT<< T<<Cp <<TCp = PCp = PTCp. When only fed *P. lutheri* a *C. gigas* larva daily consumed 1 000 to 3 000 from days 6 to 15 (Fig. 1). Associated with t. Iso, uptake of such haptophytes mixture (PT) was weakly improved while a better ingestion of t. Iso (1 000 to 15 000 cells) was paradoxically observed when larvae was fed on that single diet. In the meanwhile 7 000 to 65 000 cellules of *C. calcitrans* f. *pumilum* were removed on a similar period. Associated to T or P a higher consumption was recorded as well as an excellent pattern reproductivity. Indeed, from days 6 to 15, larvae fed bispecifics diets daily grazed 8 000 to 35 000 microalgae to reach 85 000 on day 21 while an uptake food decrease occurred during metamorphosis (Fig. 1).

## 4. Discussion

This study clearly shows than a bispecific diet based on the Haptophyte *Isochrysis aff. galbana* and the diatom *Chaetoceros calcitrans* forma *pumilum* in a proportion of one to one fits well to *Crassostrea gigas* larval demand and allow good development from 2 days old D larvae to one week old postlarvae. The addition of *Pavlova lutheri* (P) to TCp did not support any advantages in term of larval performances and metamorphosis. *C. gigas* larvae and postlarvae poor development has been reported when fed *P. lutheri* used as fresh or preserved single diets (Ponis et al., 2003). Moreover, similar results have been recently obtained on other Pavlophytes (Ponis et al., unpublished data) and this trend seems to be a general rule for the genus *Pavlova* which is of low interest for *C. gigas* production. However, it might be maintained in hatchery as an alternative to *Isochrysis aff. galbana* in case of culture collapse. Indeed, its association with *C. calcitrans* f. *pumilum* led to good larval performances, not very different from that obtained with TCp. In this case, others considerations must be taken in account such as microalgal productivity and in Argenton hatchery, t. Iso is more efficient and reliable on a long term period than *P. lutheri*. Its utilization for *C. gigas* larvae is accordingly preferential. The role of *C. calcitrans f. pumilum* in the growth of *C. gigas* is clearly showed in the present work because such a single diet provided 75% of larval growth. This is the only single diet which led to mature larvae and to metamorphosis. The role of that diatom is accordingly essential for oyster larval development. Its association with an Haptophyte induced higher larval survival and metamorphosis. Moreover, larval performances were clearly related to feeding behaviour, species specific, with a low ingestion of *P. lutheri*, a high ingestion of *C. calcitrans* f.*pumilum,* and a moderate ingestion of t. Iso*.* In presence of bispecific diets, except PT with almost a nil uptake, grazing largely increased with a high reproductivity of the pattern in all tanks.

## Litterature


Delaunay, F., Marty, Y., Moal, J., Samain, J.F., 1993. The effet of monospecific algal diets on growth an fatty acid composition of *Pecten maximus* (L.) larvae. Journal Experimental Marine Biology Ecology 173, 163-179.

Ponis, E., Robert, R., Parisi, G., Tredici, M., 2003. Assessment of the performance of Pacific oyster (*Crassostrea gigas*) larvae fed with fresh and preserved *Pavlova lutheri* concentrates. Aquaculture International 11, 69-79.

Robert, R., Miner, P., Mazuret, M., Connan, J.P., 1994. L'écloserie expérimental de mollusques d'Argenton, bilan et perspective. Équinoxe 49, 20-33.

Robert, R., Chrétiennot-Dinet, M.J., Kaas, R., Martin-Jézéquel, V., Moal, J., Le Coz, J.R., Nicolas, J.L., Bernard, E., Connan, J.P., Le Déan, G., Le Gourrierec, B., Leroy, B., Quéré C., 2004. Amélioration des productions phytoplanctoniques en écloserie de mollusques : caractérisation des microalgues fourrage. Rapport de la Direction des Ressources Vivantes, DRV/RST/RA/LPI/2004-05, 126 pp.


## Tables

**Table 1.** Mean shell length (SD), survival (SD) and harvest yield (SD) of *Crassostrea gigas* larvae fed on different mono or pluri specific diets, with P = *Pavlova lutheri*, T = *Isochrysis affinis galbana* and Cp = *Chaetoceros calcitrans* forma *pumilum*, on days 9 and 16. Values with same letters are no significant at *P > 0.05*. Initial larval shell length on D2 = 78.37 µm (4.05).

| Diet | Length (D16) | Survival (D16) | Morphological competence (D16) | Metamorphosis (D22/D16) |
|---|---|---|---|---|
| TCp | 263.51 (49.56) | 98.33 (1.32) [a] | 69.00 (12.12) | 72.02 (0.89) [a] |
| PCp | 244.71 (44.78) [a] | 98.84 (0.13) [a] | 47.66 (8.50) | 76.49 (1.10) [a] |
| PTCp | 239.91 (52.77) [a] | 95.40 (2.09) | 58.50 (13.43) | 77.36 (1.58) [a] |
| Cp | 219.00 (33.43) | 76.19 (20.16) [b] | 18.36 (11.53) | 54.13 (10.05) |
| PT | 181.52 (53.10) | 81.81 (7.70) [b] | * | * |
| T | 162.98 (41.40) | 85.98 (7.88) [b] | * | * |
| P | 101.88 (18.57) | 29.23 (7.02) [c] | * | * |
| Unfed | 81.86 (7.53) | 33.67 (3.78) [c] | * | * |

* no data.

## Figures

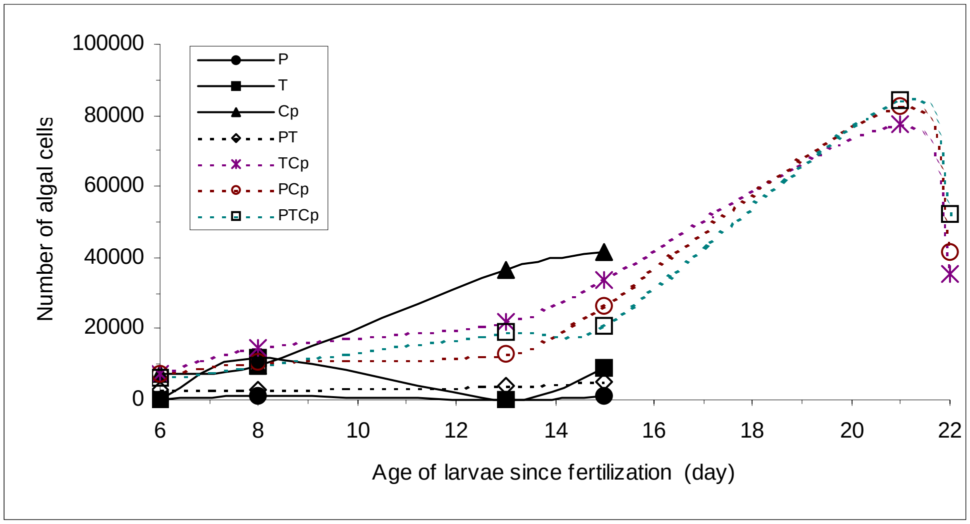


**Figure 1.** Evolution of grazing per day per larvae of *Crassostrea gigas* fed on different mono or pluri specific diets: from left to right *P. lutheri* (P); *P. lutheri* + t. Iso (PT); t. Iso (T); *C. calcitrans* f. *pumilum* (Cp); *P. lutheri* + *C. calcitrans* f. *pumilum* (PCp); t. Iso + *C. calcitrans* f. *pumilum* (TCp). Each condition was run in triplicate. From Day 6 to 15 this consumption was exclusively related to larvae while during metamorphosis it also concerned new settled postlarvae.