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63

Larval rearing and weaning of Seabass, *Lates calcarifer* (Block), on experimental compounded diets.

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Abstract. — *Larval rearing trials of seabass larvae, either imported from Singapore or obtained from the captive broodstock of IFREMER/C.O.P. at Tahiti, were carried out since 1987, by testing different technologies originally used in South East Asia (green-water culture, sequential water changes) or developed by IFREMER for other species (clear water system, preys enrichment, continuous water renewal).*

The preliminary results reveal a high variation of survival and growth rates after the first month, which are not clearly related to one of the closen rearing technic. Best results are obtained at a starting density of 30 larvae/liter in 450 liter cylindro-conical tanks whith a survival rate superior to 50-60 %, for a final mean weight of 20 mg on day 25.

In other trials, strong mortalities are observed from day 12 to 14 with symptoms of depigmentation, over-inflation of the swimbladder and abnormal swimming behaviour followed by death. Histological observations studies reveal vacuols appearing in retinal, CNS and liver a few days before the first symptoms. Pathological researches are now under way to reduce these mortalities and explain these phenomenons. No clear relation with nutritional deficiency is demonstrated.

The weaning of seabass larvae on artificial pellet is feasible as early as the 25th day when the mean weight of fry ranges from 20 to 40 mg with alternative food sequences of live to frozen Artemia and dry pellet containing a high protein (56 %) and lipid (16 %) level. A grading sequence has been defined, which allows to reduce the cannibalism observed as soon as live preys are replaced by artificial diet. Survival rates (50 %) and growth results (mean weight between 1 and 2 g on day 80) are similar to those obtained by other research teams in Singapore and Australia, and confirm the good ability of seabass fry to accept dry pelleted feeds.

Preliminary experiments to replace live prey by artificial microparticulate diet (MD) as soon as the 15th day (mean weight inferior to 15 mg) have been conducted with encouraging results.

INTRODUCTION

Seabass, *Lates calcarifer* is an economically important foodfish in many Asian countries and in Australia. His production reached 18 099 tonnes in 1982 (FAO, 1984 in Grey, 1986), and aquaculture industry is in extension in Thailand (Sirikul et *al.*, 1988); Indonesia (Danakusuma et *al.*, 1986); Malaysia (Awang, 1986); Singapore (Cheong, 1987); Philippins (Duray et *al*, 1986) and Australia (Rimmer et *al*, 1989).

Fingerlings have been recently imported from Singapore to Tahiti to study their ability to be reared in the particular environmental conditions of French Polynesian lagoons (high temperature : 26 - 28°C; and salinity : 34 - 35 ppt), and their growth potential investigated, feeding them on a dry pelleted diet including 56 % CP and 12 % CL.

Encouraging results have already been obtained, indicating that seabass is easily adaptable to lagunal waters of Tahiti : 450 to 500g mean weight fish are obtained in one year from the eggs at a final density of 30 to 35 kg/m3, in cage with a food conversion ratio interior to 1.7 : 1 (Fuchs, 1986; Aquacop et *al.*, 1989).

The maintenance of the captive broodstock and the control of seed production are been investigated in a second step as the supply of a large number of fry will conditionne the futur development of this new aquaculture in the area.

Suitable rearing techniques have been developped several decades ago in Thailand and rapidly extended in Asia, after the successful artificial propagation achieved by Songkla Fisheries Research Station in 1971 (Maneewong, 1986).

Since that date, considerable improvements have been achieved and 50 Thai private farms have produced more than 100 millions fry in 1983-1984, mainly exported to other countries : Singapore, Hong-Kong, Indonesia and Taiwan (Ruangpanit, 1986).

Rearing techniques are based on green algae culture (*Chlorella sp*, *Platymonas sp*) in large rectangular concrete tanks (5 to 15 tons) where density of larvae decrease from 50 to less than 20 per litre when transferred on day 15 in nursing ponds (Ruangpanit, 1986; Kungvankij et *al.*, 1986).

The cycle of larval rearing lasts 20 to 30 days and includes the succession of following preys : green algae (D1 to D14), Rotifers (D2 to D15), *Artemia* (D10 to D20), moina or Dapnia (D20 to D30), (Maneewong, 1986; Lim et *al.*, 1986)

With this technique, survival can reach 50 to 60 % after one month, whith important variations depending on sanitary conditions, food quality and egg supply.

Weaning larvae on inert food generally starts on day 30 using minced trash fish. During this period, cannibalism is a major problem and frequent grading are practiced (every 5 to 7 days) with grading trays made of plastic basins with many holes (2.5 to 5mm diameter) to minimize mortality. With regular grading and accurate managment of the seed, survival rate is 40 to 60 % from fry to juvenile (Lim et *al.*, 1986).

More recently, Australian Department of Primary Industry, in Cairns (Q.L.) started a pilot-scale hatchery, adapting Thai research results to conditions of Australia (physical, chemical and economical). Mac Kinnon (1986) reported that using close system, clear water and prey enrichment, survival reached 20 to 30 % on day 20, unless strong mortalities were sometime observed on day 10 to 15. Weaning larvae with artificial salmon dry pellet from D25, at a length of approximately 13-15 mm, were tempted and survival and growth rate were encouraging : 90 mm (TL) and 10 g mean weight at 65 days after hatching. Recent informations reveals that a private hatchery, based south of Cairns, produced more than 200 000 fry weaned on artificial diet in 1988, half a million in 1989 (Rimmer pers. com).

Larval rearing trials were recently carried out in Tahiti with eggs imported from Singapore or locally produced by brooders maintained in cages since 1984 and 1985. Seabass green water rearing technique, originally used in South East Asian countries was compared in term of survival and growth potential to clear water rearing method developped by IFREMER on others species like European seabass and seabream (Chatain, 1986; Ronzani-Cerqueira, 1986)

A number of differents weaning trials were also conducted with 30 to 35 days old larvae held in tanks and adapted to high protein level pelleted diets. Several formulations and presentations successfully used for seabass, seabream and flatfish in France (Metailler et *al.*, 1983) were applied to seabass fry.

MATERIAL AND METHODS

Origin of the larvae

Just hatched larvae or 15 days old larvae produced at the Primary Production Department of Singapore, following the technique described by Lim (1986), were air-shipped to Tahiti. Packaging was practiced in plastic bags at a mean density of 1000 to 1500 larvae per litre, each bag being fullfilled with one volume water for 3 volumes of oxygene. In these conditions, larval survival after 24 hours was rather high although water quality was deteriorated with pH inferior to 6.5 and ammonia level superior to 9 and 10 mg/litre. Quality of the larvae was fluctuating with each shipment which made difficult the interpretation of the preliminary larval rearing tests, conducted in 1987.

More recently, first maturation and spawning of local brooders were successfully carried out and a large supply of eggs was obtained.

Broodstock maintenance and managment

Two separate seabass broodstock batches were raised from 500g mean weight fish, imported in 1984 and 1985 at the stage of larvae. They were stocked in netcages mesuring 2 x 2 x 2m deep or 4 x 2 x 3m deep at a density of 4 to 8 pieces per m3. Feeding regime was mainly a dry pelleted diet, including 55 % protein, 12 % lipid offered at 1 to 1.5 % of the body weight per day. It was completed by a moist pelleted diet (50 % bonitos meal + 50 % fish meal) offered during the maturation season.

After 2 years, each fish was tagged and regular gonad conditions were checked. Maturation was monitored by canulation of gonads and date of sex-reversal of this hermaphrodite fish was identified.

Ripe male and female with ovocyte diameter superior to 400 microns were selected and transfered from cages to spawning tanks (10 m3 capacity) at a rate of 2 females and 4 males per tank.

Techniques of spawning, described by Lim et *al.* (1986) and Nacario (1986) were tested with one injection of 10 microgrammes of LHRH (a) or 500 UI of HCG per kg of female. After several unsuccess trials, it was observed that fertilization ratio was improved when brooders were maintained one or two months in the spawning tank before injecting. Injection was always practiced one or two day before the full moon, although no clear effect was noticed.

Natural spawning occured 36 to 40 hours after injection and the eggs were concentrated and their number and quality were investigated before transferring to the incubation unit. Each cylindro-conical incubator, 25 litres capacity, was filled with 100 to 150 000 eggs. Pourcentage of hatching was checked before transferring larvae in the rearing tanks.

Larval rearing

— *Rearing conditions*

Larval rearing trials were conducted in 80, 150 or 450 litres cylindro-conical tanks painted in grey. Each tank was supplied with mild central aeration and keep in shaded natural light condition (maximum intensity : 1000 Lux).

Seawater, pumped deep in the lagoon, was filtered through several pressure filters (maximum filtration 100 microns) and continously or partially distributed. A biological coral filter, corresponding to 10 % of the rearing volume was connected to the rearing tanks in trial 3.

Live food culture included microalgae : *Chlorella sp, Platymonas sp* or *Isochrysis,* mass cultured in 300l indoor or 2 m^3 outdoor fiberglass tanks at a final mean density of around 2-4 millions and 1-2 millions per ml respectively. Rotifers, *Brachionus plicatilis* were maintained in 1.5 m^3 tanks and feed on algae and baker yeast. Their density was increasing from 20 to 80-100 per ml in a 5-6 days cycle of production. Enrichment techniques, described by Gatesoupe et *al* (1983), were generally used.

Nauplii and two days old *Artemia* from different strains, were enriched with silco enrichment technique, proposed by Duray and *al.* (1986), before being fed to the larvae.

Green algae larval rearing technique, traditionnally developped in Asia (Lim, 1986; Maneewong, 1986) was compared to rearing method used for European seabass which include : clear water, continuous water flow from early stages, food quantity exactly ajusted to the consumption and Rotifers and 2 days old *Artemia* enriched with high fatty acids.

Physical parameters of water were identical with temperature increasing from 25 to 28-29°C and salinity stabilized around 30 ppt.- Desigh of experiments

The designs of 3 experiments conducted with larvae imported from Singapore are as follows :

Trial 1. 450 litres tanks. 4 treatments. No replicate. One clear water. 3 with green algae : *Chlorella* : 100 000 c/ml or *Isochrysis* : 30 000 c/ml. Initial density = 33 larvae per litre. Water exchange = 5 to 10 %/hour in continuous (Treatment 1) or 50 to 80 %/day (treatments 2-3-4). Duration : 25 days. Feeding regime : Rotifers D.5-15; Nauplii *Artemia* D.9-25; 2 days old *Artemia* D.15-25.

Trial 2. 80 litres tanks. 3 treatments. 2 replicates. One clear water. 2 with green algae : *Chlorella* : 100 000 c/ml or *Isochrysis* : 30 000 c/ml. initial density = 25 larvae per litre. Water exchange : 30 to 80 %/day in discontinuous. Duration : 25 days. Feeding regime : Rotifers D.3-15; Nauplii *Artemia* D.10-22; 2 days old *Artemia* D.18-25.

Trial 3. 450 litres tank. One treatment. 2 replicates. Clear water. Initial density : 33 larvae per litre. Water exchange : 5 to 15 %/hour. Close recirculated system. Feeding regime similar to trial 2.

Five other larval rearing trials were tempted with larvae locally produced in Tahiti : spawning n° 9 (March 1988); spawning n° 17 (November 1988); spawning n° 20 (january 1989). Clear water technique was used in 150 and 450 litres tanks as indicated in table 1.

Table 1. Experiment larval rearing trials conducted with larvae produced locally in Tahiti in 1988 - 1989, with continuous water exchange from early stage.

Trial	Spawning number	Total larvae reared	Tanks	Initial concentration /litre	Treatment	Water exchange
1	9	170 000	12 x 150 l 1 X 10 m^3	33 11	C W G A	C D
2	9	130 000	9 X 450 l	32	C W	C
3	17	135 000	9 X 450 l	33	C W	C
4	20	135 000	9 X 450 l	33	C W	C
5	20	60 000	12 X 150 l	33	C W	C

C W : clear water C : continuous
G A : green algae D : discontinuous

Weaning larvae on pelleted diet

After the first month of rearing, larvae were transferred from rearing tanks to circular tanks (1.5 m^3 capacity) and weaned on artificial diet. During transfer, larvae were graded and dispatched into 3 differents sizes (inferior to 1.5mm; superior to 1.5 — inferior to 2; superior to 2 mm) to minimize the rate of cannibalism appearing as soon as live preys were replaced by inert food. Graders used have been described by Vasselin (1984) for European seabass. Grading frequency was fixed every 10 to 15 days, depending on the behaviour of the fry.

Food sequency included the overlapping from live Artemia (5 days) to frozen Artemia (10 days) and dry pelleted starter diet (56 % Protein, 16 % Lipid) offered from 5th day. (see exemple in figure 1). Live and frozen food were distributed 3 times per day, and artificial diet in 8 hours, thanks to automatic feeders. Quantity of dry pellet was calculated to be in excess (20 to 30 % of the body weight per day).

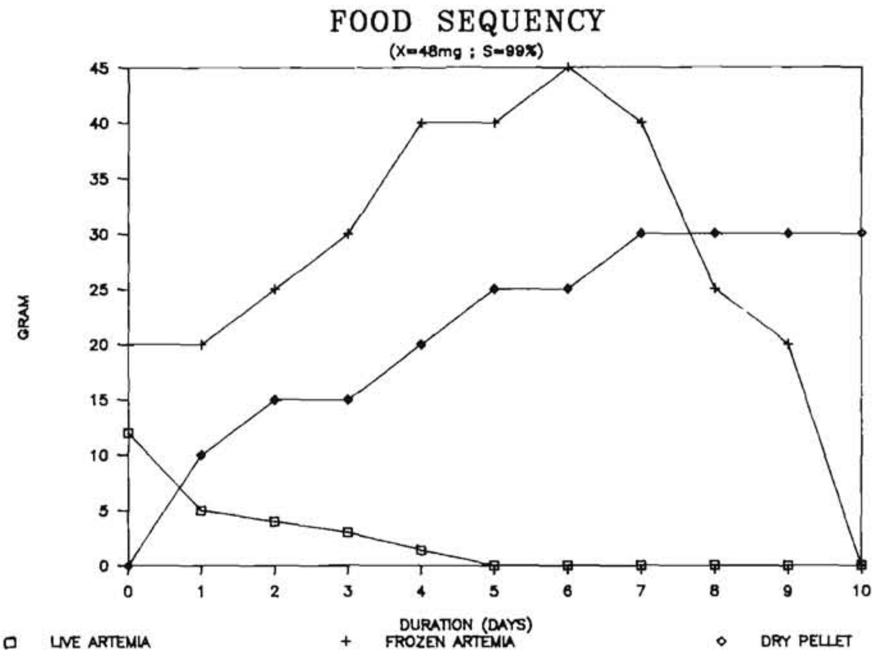


Fig. 1 — Exemple of Seabass larvae weaning with an overlap of 5 days on live Artemia and 10 days on frozen Artemia. (number : 2000 fry; initial mean weight : 48mg; age : 35 days).

Larval density was fixed between 0.5 to 1.5 larvae per litre and water quality was 27°C for temperature, 35 ppt for salinity. Water exchanges were increasing from 10 to 50 %/hour. Light intensity was low (inferior to 5000 lux) as seabass fry are sensitive to any environmental stress.

The whole population was weighed and regularly graded into different sizes varying from 1.5 to 5 mm. Grading was provided every 10 to 15 days to avoid cannibalistic behaviour and density was maintained between 0.5 to 1.5 larvae per litre.

Three tests were conducted in 1987 and 1988,following this design with 35 days old larvae issued from first larval imported from Singapore. Initial number of larvae was respectivelly 4800, 18 000 and 13 000 in trials 1-2 and 3 and duration varied from 20 to 60 days.

Weaning seabass larvae with attractive substances

Four different mixtures of chemical attractive substances, successfully tested on european seabass by Metailler et *al* (1983) were tentativelly incorpored in the weaning diet of seabass. The objective was to evaluate the effect of these substances on pellet acceptability and possible survival and growth improvement.

Rearing conditions were similar to the former. Density was fixed to 2 larvae per litre or 160 larvae, (50.55 ê 0.9 mg mean weight), in 70 litres rectangular tank.

Diet sequency included direct weaning on dry pellet, distributed 8 hours per day with a first stabulation of larvae during 6 days, feeding them with frozen Artemia.

Water quality was similar with a temperature of 27 ê 1 °C, 10 % water exchange per hour and a salinity of 35 ppt.

Six treatments and 2 replicates were tested

T.1 : control fish fed on frozen Artemia	
T.2 : weaning Diet without attractive substance :	Diet 1
T.3 : weaning Diet 1 plus 0.5 % glycine and betaine :	Diet 2
T.4 : Diet 2 plus 1.77 % neutral amino-acids :	Diet 3
T.5 : Diet 3 plus 0.37 % amino-acids :	Diet 4
T.6 : Diet 4 plus 0.054 % inosine and hypoxanthine :	Diet 5

Duration of the experiment was 24 days, without intermediar grading.

Early weaning of seabass larvae on microparticulate diets

Microbinded diets (MBD) similar to those described by Person-Le-Ruyet (1986-1989) and including fresh products, were tested on seabass larvae as early as the 20th day. Rearing conditions were differents to the formers with the use of 80 litres cylindro-conical tanks. Density was variable from 1.5 to 2.5 larvae per litre, and each treatment was replicated 2 times. Diet sequency included MBD distributed from the first day in continuous during 8 hours with automatic feeders. Diet quantity was ajusted to the needs (1 to 2 g per day).

From day 10, MBD was gradually replaced by former weaning diet in an overlap of 3 to 4 days. Size of particules were 125-250 M for MBD and 250-300 M for weaning diet. Live *Artemia* were given during the first 10 days of experiment; quantity was calculated to be exactly 10 % of the dose offered to the control.

Water temperature was fixed to 28°C and water exchange to 200 % per day.

Three experimental designs have been carried-out :

Trial 1 : 1 MBD formulation. 2 different binders : carragheen or alginate. 1 control with live *Artemia*. Initial larvae age : 20 days and 15.2 mg mean weight. Duration 15 days.

Trial 2 : 1 MBD formulation. 1 binder (carragheen). 1 principal ingredient (squid). 1 treatment with live *Artemia* : 10 % of control. 1

treatment with frozen *Artemia* : 10 % of control. 1 control with live *Artemia*. Initial larvae age : 21 days and 24.4 mg mean weight. Duration : 15 days.

Trial 3 : 2 MBD formulations. 1 binder (carragheen). 2 principal ingredients in the diet : mussel or squid. 1 control with live *Artemia*. Initial larvae age : 31 days and 39.9 mg mean weight. Duration : 11 days.

No grading were provided during the experiments.

RESULTS

Maturation in Captivity

Although, the complete cycle of reproduction of this imported species is still under study, first observations indicate that seabass seems to mature during the rainy season, like the local species as grouper (Debas et *al.*, 1989). Peak of maturation is observed when light duration and temperature are increasing (November to April) and sexual rest is noticed from june to October (Guiguen, pers. com.).

Seabass males reach sexual maturity at the age of 2 to 2.5 years and are generally ripe 5 to 6 months per year. First sexual reversal changes were noticed in November 1987 and February 1988 for the 2 broodstocks respectivelly imported from Singapore in 1984 and 1988. (The proportion of reversal rapidly increased in few months and a significant number of female was obtained in 1988).

More accurate observations, realized on 3 batches of 30 brooders, isolated from the second broodstock in June 1988 indicated that a minimum duration of 3.5 years was necessary to observe the first reversal and 4 years for 40 % of the population to reverse from male to female (Nedelec, pers. com.).

Spawning and hatching

Spawning trials, were conducted in November, December 1987 with the first broodstock and from February 1988 to May 1989 with the second broodstock. Males and females were always issued from the same original batch (imported in 1984 or 1985). Synthesis of results are summarized in table 2.

The preliminary trials conducted in November, December 1987 with females issued of the first broodstock (1984) were not successfull and only one female spawned 6 000 unfertilized eggs.

Between February and May 1988, 7 females spawned and 12 depositions were compted. A total of 6.39 millions eggs fertilized at a rate of 27 %, were collected. and 1.7 millions were incubated in conical incubators.

During the last spawning season, from October 1988 to March 1989, 6 females naturally spawned more than 7.8 millions eggs fertilized at 65 %.

Table 2. Results of Seabass spawning between November 1987 and May 1989.

	November December 1987	February to May 1988	October 1988 to March 1989	Total
Origine of female	Broodstock n°1	Broodstock n°2	broodstock n°2	
Number of female treated	2	9	9	20
Number of female spawning	1	7	6	14
(%)	50	78	67	70
Total number of deposition	2	12	12	26
Number of deposition with fertilized eggs	1	3	10	14
(%)	50	25	83	54
Total number of eggs (millions)	0.006	6.39	7.86	14.246
Number of fertilized eggs (millions)	0	1.72	5.13	6.82
% fertilization	0	26.9	65	48
Mean egg diameter (mm)	0.88	0.77	0.817	0.822

10 of the 12 deposition were normally fertilized and 5.1 millions eggs were transfered in incubators.

Figure 2 illustrate this regular increase in the pourcentage of fertilized eggs between 1987 and 1989.

The regular improvment in the management of broodstock can partially explained thes increasing results. The pourcentage of fertilization was very low when brooders were injected 2 to 3 days after transfer from cage to spawning tank due to stress behaviour. A minimum of 1 month of stabulation in the spawning tank is highly recommended to improve the pourcentage,as previously noticed by Lim et *al.* (1986) in Singapore.

Origin of the brooder and the choose of large females (mean weight superior to 4-6 kg) are also important factors to consider.

Finally, determination of the natural cycle of reproduction and sex-reversal period of this imported species in Tahiti, would certainly help to choose the better period of the year to successfully induce the brooders.

Larval rearing

Trial n°1

The first larval rearing trial, conducted in 1987 with larvae imported from Singapore gave encouraging results as shown in table 3.

Traditionnal S-East Asian rearing techniques using green algae and a discontinuous water exchange was successfully transfered to the local conditions of Tahiti. Survival after 25 days was superior to 50 % with a maximum of 82 % in Tank n°3, where larvae received Rotifers and *Isochrysis sp* algae. Mean weight at the end of the rearing period reached 25 to 30 mg and food intake for each produced larvae was estimated to 5-6000 Rotifers, 3000 nauplii of *Artemia* and 600 of 2 days old *Artemia.*

Table 3. Larval rearing trial n° 1 conducted in 1987 in 450 litres tanks with larvae imported from Singapore. Traditionnal S.E.Asian larval rearing techniques was compared to clear water rearing method where Rotifers and Artemia are enriched.

Tank	Treatment	Initial number	Survival (%)	Final density /litre	Final mean weight (mg)	Food intake/larvae produced		
						Rotifer	Na Artemia	2 days old Artemia
1	Clear water	11 600	52	13	46	4 800	3 000	1 200
2	Green water (Chlorella)	11 600	56	14	29	5 100	3 700	680
3	Green water (Isochrysis)	11 600	82	21	24	6 000	2 300	410
4	Green water (Chlorella)	11 600	78	20	26	6 300	2 700	650

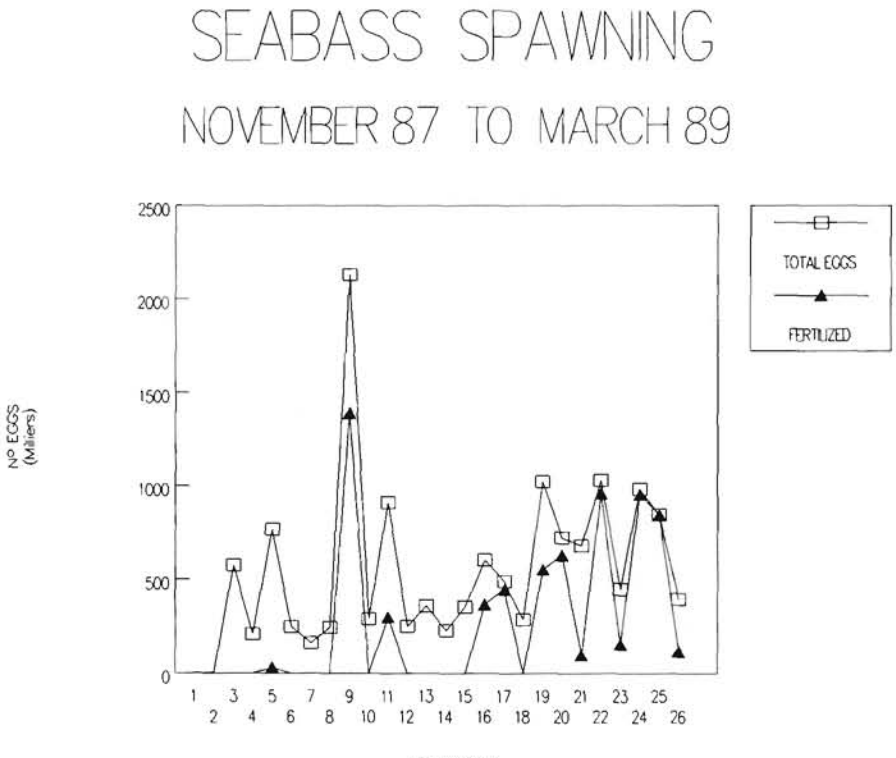


Fig. 2. — Total number of eggs and fertilized eggs obtained by spawning of induced female of Seabass (*Lates calcarifer*) in Tahiti from November 1987 to March 1989.

In comparison, the first batch, reared in clear water and fed on enriched Rotifers gave lower survival rate (52 %), but the final mean weight (46 mg on day 25) was significantly higher than in the 3 others batches. Food intake was similar but 2 days old *Artemia* were offered in larger quantity due to a higher growth speed.

Trial n°2

First experimental design was replicated in 80 litres tanks, with 2 tanks per treatments (Table 4).

When larvae were fed on Rotifers and algae (*Chlorella* or *Isochrysis*) : tank 1 to 4, survival rate was superior to 50 % (maximum 71 %) except in tank 1 where an anormal strong mortality was observed on day 15 although a high initial survival rate. Food intake was similar to trial 1 with a total of 8000 Rotifers, 3000 Nauplii of *Artemia* and 450 2 days old *Artemia* offered per larvae produced on day 25. Final mean weight reached 18 to 25 mg without clear difference when larvae received *Chlorella* or *Isochrysis* algae strains with Rotifers.

Table 4. Larval rearing trial n° 2 conducted in 1987 in 80 litres tanks with larvae imported from Singapore. Traditionnal rearing techniques are compared to clear water rearing method where Rotifers and Artemia are enriched.

Tank	Treatment	Initial number	Survival (%)	Final density /litre	Final mean weight (mg)	Food intake/larvae produced		
						Rotifer	Na Artemia	2 days old Artemia
1	Green water (Isochrysis)	2 000	14.5*	4	12.5	36 000	1 100	1 400
2	«	2 000	51.9	14	17.5	10 000	3 500	470
3	Green water (Chlorella)	2 000	63.7	16	22.8	7 800	3 000	380
4	«	2 000	70.7	18	13.1	6 900	2 700	340
5	Clear water	2 000	31	8	32.9	8 700	5 400	1 000
6	«	2 000	31	8	27.6	5 600	4 700	700

* Mortality on day 15.

In comparison, larvae reared in clear water from early stage had the same lower survival rate in the 2 replicates (31 %). Important mortality was noticed during the first 5 days of larval rearing (first feeding). Total amount of live prey was 7000 Rotifers, 5000 Nauplii of *Artemia* and 350 of 2 days old *Artemia* per larvae produced on day 25. Final mean weight was also significantly higher with a mean value of 30 mg when Rotifers were enriched.

Trial n° 3

Althoug the difficulty encountered to compare these results due to the lack of control, it has been observed that seabass larvae where able to survive when water was totally recirculated on a biological filter as previously mentionned in Australia by Mac Kinnon (1986). (Table 5).

Survival rate was 30 % after 20 days with high mortality, observed as in the former trial, from day 1 to 5. Food quantity intake was similar with around 7000 Rotifers, 3500 Nauplii of *Artemia* and 600 of 2 days old *Artemia* offered to each day 20 old larvae. Growth speed was satisfying and final mean weight varied from 14 to 17 mg.

Table 5. Larval rearing trial n°3 conducted in 450 litres tanks. Water is recirculated on a biological filter and larvae are fed on enriched Rotifers and Artemia.

Tank	Treatment	Initial number	Survival (%)	Final density /litre	Final mean weight (mg)	Food intake/larvae produced Rotifer	Na Artemia	2 days old Artemia
1	Clear water and	12 000	26	7	17.2	8 300	3 700	710
2	Close system	12 000	34	9	14.4	6 400	3 300	550

Larval reared trials conducted with eggs produced in Tahiti

Five trials have been conducted in 1988-1989 with eggs produced by brooders maintained in captivity since 1985. Clear water and enriched Rotifers wzere offered in each treatment. Unsuccessfull results have been obtained with extremely high mortalities occuring from day 10 to 15 (Table 6). Clinical symptoms were similar to those described by Rimmer (1989) in Australia with

+ stop feeding
+ depigmentation of the larvae
+ overinflation of the swimbladder
+ anormal swimming behaviour

Mortality generally occured 2 to 3 days after the aparition of the first symptoms and rapidly extended to the whole population. Histological studies shown clear vacuolisation in the retina and CNS. Final survival was so poor that larval rearing were stopped after 3 to 4 days.

Table 6. Larval rearing trials conducted with eggs locally produced. Rearing techniques include clear water or green algae.

Date	Number of larvae reared	Number of tanks/litre	density	Treatment mortality	Peak of survival	Final
1th Ap. 88	170 000	12x150 l	33	Clear Water	8	0 %
		1x 10 000 l	11	Green Algae	12-13	0 %
	130 000	9x450 l	32	Clear water	5-6	0 %
24th Nov. 88	135 000	9x450 l	33	Clear water	11-13	0 %
24th Jan. 89	135 000	9x450 l	33	Clear water	12-13	0 %
24th Jan. 89	60 000	12x150 l	33	Clear water + Green water	15-17	5 %

Weaning seabass on a dry pelleted diet

Weaning seabass larvae on a dry pelleted diet after an overlap of 10 days on live and frozen *Artemia* has demonstrated to be feasible. Results of the first two trials, conducted in 1987, (table 7), confirme the preliminary observations carried-out in Tahiti in 1985 (Fuchs, 1986).

Mortality (up to 30-40 %) was concentrated during the first 15 days as shown in figure 3, with a strong cannibalistic behaviour when live preys were replaced by artificial pellets. After 20 days, fry were already weaned and the number of dead larvae dicreased, and final survival, after 2 months was around 50 % in trial 2.

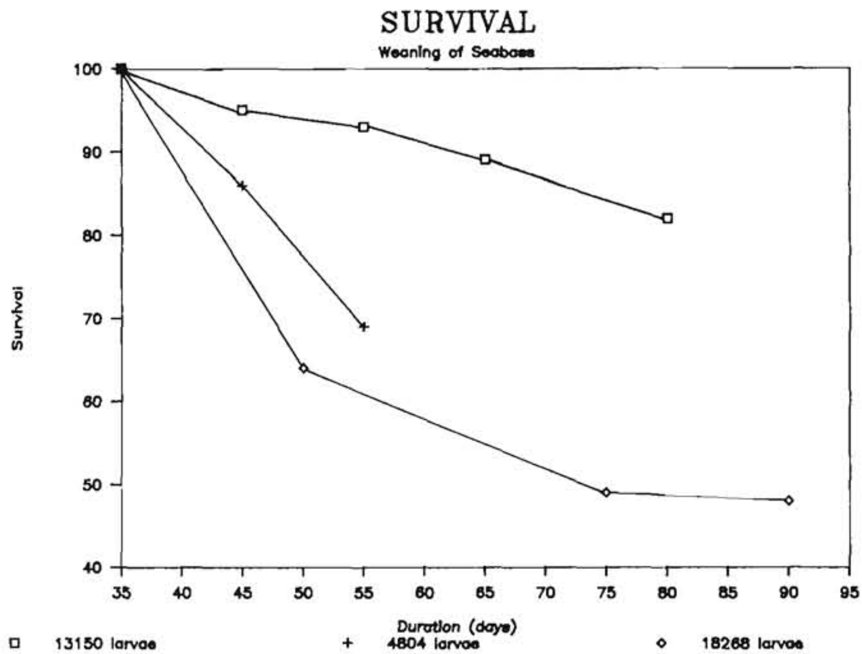
Growth speed was slow during the first period of adaptation to pellet but drastically increased to reach a final mean weight of 2 grammes at the end of the weaning period in trial 2 (see figure 3). Food efficiency was regularly increasing and final conversion ratio was 2.3 : 1 in trial 2, 4.05 : 1 in trial 1 (Table 7).

Size dispersion after 2 months was large with 67 % of the whole population distributed in grading size : 3.5 to 4 mm, 23 % between 4-5 mm and 10 % between 5 and 6 mm.

Table 7. Seabass weaning on dry pelleted diet after an overlap of 10 days on live and frozen Artemia (trial 1-2, 1987 ; trial 3, 1988).

Trial	Age (d)	Duration (d)	Number	Survival (%)	Density /litre	Rearing volume m^3	Mean weight mg	Conversion ratio
No 1	35	0	4 800	100	0.8	6	44	—
	45	10	4 110	86	0.5	9	119	5.2 :1
1987	55	20	3 300	69	0.4	9	332	3.5 :1
	Total	20	3 300	69	0.4	9	332	4.1 :1
No 2	35	0	18 300	100	1.8	10	84.2	—
	50	15	11 610	64	1.0	12	220	4.4 :1
1987	75	30	8 927	49	0.75	12	1 010	2.2 :1
	90	55	8 821	48	0.7	12	2 100	1.1 :1
	Total	55	8 821	48	0.7	12	2 100	2.3 :1
No 3	35	0	13 200	100	1.8	5	47	—
	45	10	12 535	95	1.7	6	122	2.3 :1
1988	55	20	12 206	93	1.4	6	340	1.7 :1
	65	30	11 807	90	1.3	6	720	1.1 :1
	80	45	10 829	82	1.2	6	2 400	0.7 :1
	Total	45	10 829	82	1.2	6	2 400	0.7 :1

The better knowledges of seabass larvae behaviour and the regular improvment of weaning technique and tank management from 1987 to 1988 allowed to increase the performances as shown by results obtained in 1988 (Table 7). Survival at day 80 range 82 %, average mean weight increased from 47 mg to 2.4 g in 45 days and final conversion ratio reached 0.8 : 1 in as fed. Cannibalism of fingerlings was controlled by a frequent grading, every 10 days during the first month, then every 15 days (Figure 3). Size distribution was more homogeneous with 53 % of the population distributed in the range 4-5 mm, 23 % inferior to 4 mm and 24 % superior to 5 mm.



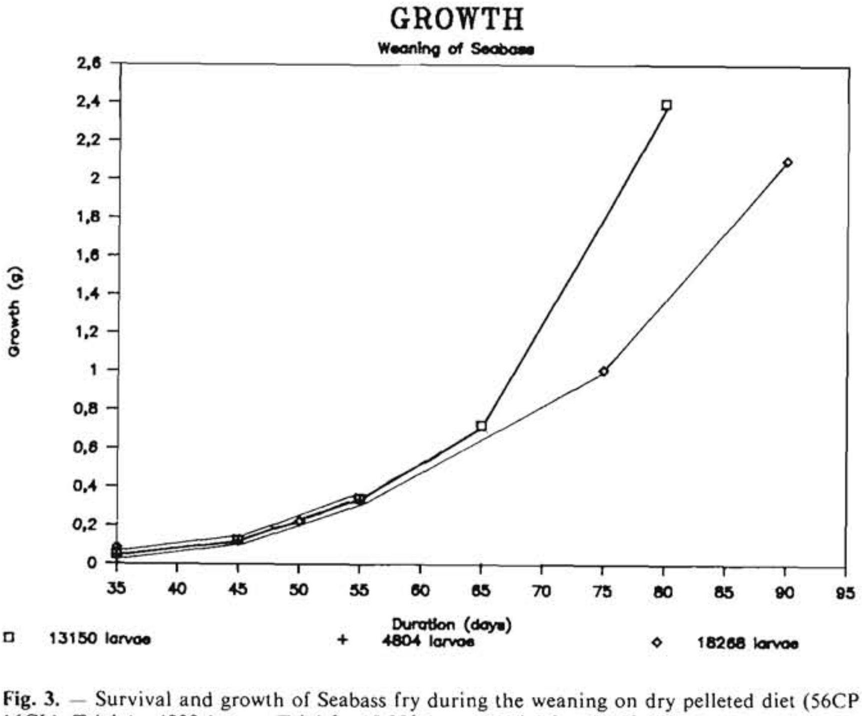


Fig. 3. — Survival and growth of Seabass fry during the weaning on dry pelleted diet (56CP 16CL). Trial 1 : 4800 larvae, Trial 2 : 18 300 larvae, Trial 3 : 13 150 larvae.

Weaning seabass larvae with attractive substances

Survival and growth results of the 6 treatments are summarized in Table 8.

When attractive chemical substances were incorporated in the weaning diet by coating, no significant difference was noticed, between treatments, after 25 days. Survival was superior to 75 % in each trial and the mean weight was increasing from 50 to 380 mg in 25 days, corresponding to a daily weight increase of 7.7 %.

In comparison, the control fish, fed on frozen *Artemia* shown a better survival rate (up to 87 %) but their growth speed was significantly lower with a final mean weight less than half of the weight obtained when larvae were fed with pellets. These results suggest that frozen Artemia would not cover the requirements of fingerlings during this phase, although survival rates are satisfying.

Table 8. Weaning of Seabass larvae on dry pelleted diet including different attractive chemical substances during a 25 days experiment (initial mean weight : 50.55 mg).

Treatment	% Survival		Final mean weight (g)		D.W.I. (%)	
	Per replicate	Mean s.	Per replicate	Mean w.	Per replicate	Mean D.W.I.
Control	91.8		176.3 ± 30.6		5.3	
frozen Artemia	82.5	87.1	186.1 ± 28.8	181	5.2	5.25
Diet 1	79.9		422.5 ± 71.3		8.5	
without Artemia	66.9	73.4	407.7 ± 75.2	415.1	8.3	8.4
Diet 2	79.6		368.8 ± 80.7		7.9	
	70.5	75.1	443.3 ± 106.8	406.1	8.7	8.3
Diet 3	71.1		442.4 ± 104.4		8.7	
	84.7	77.9	32[illegible].9 ± 46.6	382.1	7.4	8.05
Diet 4	79.0		391.9 ± 88.1		8.2	
	73.0	76	339.7 ± 61.5	365.8	7.6	7.9
Diet 5	88.6		386.0 ± 54.1		8.1	
	74.2	81.1	322.5 ± 55.8	354.2	7.4	7.8
Control		87.1		181		5.25
Artificial diet		76.7		384.7		8.09

Early weaning of seabass fry on microbinded diets

Results of preliminary early weaning tests (Table 9) clearly shown that seabass larvae were able to accept microbinded diets as early as 20 th day when offered with a few amount of live *Artemia* (10 % of control dose) during 10 days. In trial 1 and 2, survival rate, at day 35, end of the weaning period, ranged from 22 to 56 % with best results obtained when microdiets were binded with carragheen. Microdiet, incorporated alginate binder was not accepted and induced a strong mortality after 10 th day. Replacement of live *Artemia* by frozen ones (trial 2) did not affected survival as shown in table 9.

Table 9. Early weaning of Seabass larvae with microbinded diet incorporating different ingredients. 10 % of live Artemia offered to the Control are distributed during the first 10 days.

Diet	Number Initial	Number Final	Survival (%)	Mean weight (mg) Initial	Mean weight (mg) Final	D.W.I. %/Day	Conversion Ratio
Trial 1. Day 20 to 35							
MBD Carragheen	200	111	56	15.2	142	15	1.5 :1
MBD Alginate	200	44	22	15.2	76.5	11	7.3 :1
CONTROL	200	163	82	15.2	142	15	1.3 :1
Trial 2. Day 21 to 36							
MBD Live Artemia	200	84	42	24.4	163	13	6.6 :1
MBD Frozen Artemia	200	89	44	24.4	178	13	5.5 :1
CONTROL	200	102	51	24.4	166	13	2.3 :1
Trial 3. Day 31 to 42							
MBD Mussel	110	95	87	39.9	95.3	8	3.1 :1
MBD Squid	110	95	87	39.9	142	12	1.7 :1
CONTROL	110	104	94	39.9	214	15	0.7 :1

Similar differences were noticed on growth performances and average mean weight increased from 15-24 mg to 150-160 mg after 15 days using caragenane against 75 mg when alginate was incorporated as binder in the diet.

Growth potential were similar when live or frozen *Artemia* where comparatively offered as complement to the MBD.

Conversion ratio also fluctuated with the differents tested diets (range 2 to 7.1 : 1) confirming the good potential of MBD incorporating squid.

In comparison, control larvae, exclusively fed on live Artemia had a higher survival rate (up to 82 % in trial 1) but final mean weights were no significantly higher than when larvae were fed on a well balanced MBD.

Results of trial 3 (Table 9), conducted with larger larvae, (31 days old, initial mean weight : 39.9 mg) indicated that the MBD formulation has a major importance. Significant better growth was obtained when squid was incorporated in large amount in the MBD, compared to mussel (140 mg again 95 mg after 15 days). Survival was very high in both case with values superior to 85 %. In this trial, results of survival and growth were improved when live *Artemia* were offered as unique food with respectively 94 % survival and final mean weight : 214 mg after 15 days.

In this trial, conversion ratio were respectively 0.7, 1.7 and 3.1 : 1 respectivelly when larvae were fed with live *Artemia*, MBD quid or MBD mussel.

DISCUSSION

Seabass, *Lates calcarifer* has been selected as one of the most promising species to develop a new aquaculture industry in French Polynesia. Successfully nursing and grow-out trials have been carried-out since 1985 with fry imported from Singapore and no particular problem has been encountered when fry were exclusively fed on a compounded diet, pelletized in Tahiti (Aquacop et *al*, 1989).

The major contraint to increase the production and really initiate a development programme was the seed production which has to be manage with appropriate feasible production techniques. The climatic conditions of French Polynesian oceanic waters (high salinity) and the difficulties encountered to supply large amount of natural fresh food to larvae and juveniles were identify as possible limiting factors to overcome.

The first observation of ripe males (in 1987) after 2.5 years of stocking in floating cage raised our dubiousness of getting maturations of seabass in Tahiti meanwhile his catadromous characteristic (Grey, 1986). One year later, sex-reversal phenomeous was identified and the first females appeared. Since that date, many induced spawning have been successfully tempted, confirming the validity of the spawning techniques, described by Nacario et al (1986), Lim et al (1986) or Duray et al (1986). Fecondity ratio of female regulary increased from very few eggs up to 600 000 per kg, fertilized at a high rate.

These encouraging results seem to indicate that maturation and egg supply are not limiting factors but many improvements are actually purchassed to propose an appropriate feasible technique.

In this objective, several axes of research are actually investigated :

- Determination of the natural cycle of reproduction of *Lates calcarifer* in the particular conditions of Tahiti and determination of the period of sex-reversal.
- Improvement of the quality of the gametes in relation with the food quality and the optimal environmental conditions (temperature, salinity...).
- Extension of the spawning season by environmental manipulations (temperature-photoperiod).
- Cryopreservation of sperm, testing existing technologies.

Larval rearing trials, carried-out with larvae imported from Singapore, demonstrated that seabass could easily be produced in Tahiti using the simple technique communly developped in many Asian countries (Maneewong et *al*, 1986 in Thaïland; Duray et *al*, 1986 in the Philippines). Interesting preliminary results have already been obtained in stagnant water with a feeding regime including algae, Rotifers and Artemia. Several contraints have nevertheless been noticed, limiting our interest to extend this method in Tahiti : survival and growth results are highly fluctuating with the quality of algae and Rotifers offered and water exchanges and tank management are time consumming.

In comparison, the technique developped in Europe to produce European Seabass and Seabream larvae (clear water, prey enrichment and continuous water exchange), was tentativelly adapted to tropical seabass larvae with interesting preliminary results. It has been observed that the growth potentail of larvae fed on enriched preys, as recommanded by several authors (Watanabe et al, 1983; Gatesoupe et al, 1983)was significantly increased compared to larvae fed on Rotifers grown on a mixture of algae and yeast. In the preliminary larval rearing tests, survival rate was nevertheless lower than with the former technique (30 to 50 % on Day 25) due to the high mortality observed during the first feeding stage (day 5-6). The strict adjustment of the quantity of preys offered to the consumption of the larvae which is recommanded with this technique, could partially explain these mortalities. The concentration of Rotifers was not exceeding 1 per ml due to the small consumption of preys at that age and the quantity available seem to be insufficent to feed all the larvae. In comparison, Duray et *al* (1986) maintain a concentration of 10 to 15 Rotifers/ml from day 2 to 15 and Rimmer et *al* (1989) supplies Rotifers at a concentration of 20 Rotifers/ml, remove every day.

Meanwhile these difficulties, the clear water larval rearing technique presents several advantages in simplifying the tank managment, reducing the rearing volumes and increasing the growth speed of the larvae and is recommanded for the futur. Many attemps have nevertheless to be carried-out before providing an appropriate feasible seed production technique, adapted to the local conditions.

The strong mortalities observed from day 12 to 14 when the larvae were issued of local brooders were difficults to explain. The symptoms of depigmentation and over-inflation of the swimbladder were very similar those previously described by Mac Kinnon (1986) and Rimmer et *al* (1989) in Australia but, in our conditions, no clear relation to the former techniques or to nutritionnal deficiencies of preys was noticed.

It has been verified that the larvae, imported from singapore and reared exactly in the same conditions in clear and green water did'nt show any symptoms of mortality (survival > 50 to 60 % on day 25) in both case which suggest that the sanitary quality and the origin of the larvae in relation to the manadgment of brooders has a major importance.

Research efforts are actually focused on :

- Sanitary conditions of brooders and eggs because some authors (Owens, pers. comm.) suspect a viral disease to be responsible for seabass larvae mortalities in Australia.
- Managment of broodstock in relation with food quality and optimisation of environmental conditions.
- Quality of live preys with the use of efficient enriched products like Frippak booster, recommanded by Rimmer et *al* (1989) to limit mortalities during the *Artemia* feeding stage.
- General managment of larval rearing environment with the use of biofilter in close system which allows to stabilize the chemical and bacterial containt of rearing water.

Weaning Seabass larvae from early stage with microparticulated diets or starter diet (56 CP, 16 CL) after an overlap of several days on live or frozen *Artemia* has been demonstrated to be feasible. Encouraging results have already been obtained in adapting to *Lates calcarifer*, the knowledges on European Seabass and Seabream (Chatain, 1986; Rozani-Cerqueira, 1986; Person-Le-Ruyet, 1986-1989).

Seabass larvae are able to accepte dry pelleted particules after a few days and survival and growth after 15 to 20 days are not drastically decreased, compared to the control fish, fed on lived or frozen *Artemia* if a certain number of conditions are respected :

- In order to homogenize the population and limit the size dispersion,, a grading of larvae is recommanded before starting the weaning.
- Selection of savety larvae issued of successfull larval rearing give a higher survival and growth potential and a lower dispersion.
- An overlap from live to frozen *Artemia* in 10 days is recommanded when larvae are weaned on starter diet to facilitate the adaptation of fry to the new rearing conditions and to the feeding regime. Preliminary trials conducted with high quality and attractive microparticulate diets suggest that this overlap could probably be reduced with the improvement of the diets.
- Continuous distribution of particules with automatic feeder is also an important factor in providing diet during the all days. Traditional feeding regime, proposed by Maneewong et *al* (1986), with fine trash fish meat, distributed 2 to 3 times per day induce strong dispersion and poor final survival (20 to 30 % in general).
- Frequent grading, provided every 10 to 15 days is also recommanded although constant improvement of diet and water management would probably reduce dispersion and cannibalistic behaviour.

In conclusion, the futur of seed production of *Lates calcarifer* seems to be encouraging but the problems encountered in these very preliminary trials suggest that the technique need several adjustments and improvements to be feasible and to allow the development of a new aquaculture activity of this very promising species in French Polynesia.

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