

SEA BASS, GROUPERS, AND SNAPPERS

Marietta N. Duray

Aquaculture Department
Southeast Asian Fisheries Development Center
Tigbauan, Iloilo 5021, Philippines

ABSTRACT

Studies on sea bass (*Lates calcarifer*) broodstock were directed at techniques to maximize egg production. Now known are the: optimum luteinizing hormone releasing hormone analogue (LHRHa) dose range to induce spawning, optimum egg size responsive to LHRHa induction, appropriate time for induction, proper storage conditions for LHRHa, and induction of spermiation in males. Gonadal maturation and spawning are successfully induced by LHRHa and/or 17 alpha-methyltestosterone. An experiment on photoperiodic induction of sexual maturation is being conducted to produce seed year round. Increased information on larval morphology and physiology of sea bass led to improvements in feeding strategies and transport techniques. Studies on nutrient requirements and practical diets are currently being undertaken for different stages/sizes of sea bass. An economic assessment found an integrated sea bass production system viable.

Studies on groupers (*Epinephelus* spp.) have been geared towards broodstock development including induction of sex inversion by hormonal control, intraspecific interaction, and sex control using synthetic anabolic steroids. Spontaneous maturation and successive spawnings of captive *Epinephelus suillus* were achieved in 1990. Larval rearing techniques used for other marine fish species were tried but with limited success. Culture techniques in ponds and floating cages using SEAFDEC-formulated diets or commercial pellets are being developed.

Studies on snappers (*Lutjanus* spp.) have been started with the identification of species common in Panay Island.

INTRODUCTION

Fishfarmers are currently searching for alternative high-value aquaculture species following the decline of the export market value of shrimp. The popularity and high market demand of tropical sea bass, groupers, and snappers make them obvious choices for culture. However, culture of these species is constrained by insufficient seed supply, high production costs, and lack of cost-effective feeds.

Cognizant of the need to eliminate these constraints, the Aquaculture Department of the Southeast Asian Fisheries Development Center (SEAFDEC/AQD) conducted research in the following general areas: 1) sea bass breeding, larval rearing, grow-out culture, nutrition, and farm socioeconomics, (2) grouper species identification, breeding, larval rearing, grow-out culture, and disease control, 3) snapper species identification and market survey.

This paper reviews the results of studies conducted at SEAFDEC/AQD over the past four years. However, many of these results are yet to be published.

SEA BASS

Breeding

Sequential and multiple spawnings of sea bass were first achieved by injection, pellet implantation, or by osmotic-pump implant of mammalian and salmon gonadotropin hormone-releasing hormone analogue (GnRH_a) (Almendras et al. 1988). Pellet implantation is most effective. Stimulation of multiple spawnings is almost similar if mammalian or salmon GnRH_a is used or if the stimulation is applied at the first quarter or new moon phase.

Hormone-induced sequential spawning is dose-related. Pelleted luteinizing hormone-releasing hormone analogue (LHRH_a) is optimum at doses of 38-75 microgram per kilogram body weight of fish (Garcia 1989a). Mean egg production is highest on the first day and gradually declines on subsequent days. Despite the advantages of pellet implantation, injection is still the most convenient and popular mode of LHRH_a administration. The median effective dose of injected LHRH_a is about 25 µg/kg among mature female sea bass having a minimum egg diameter of 0.40 millimeter (Garcia 1989b). Fish spawn 30-36 hours after a single injection. An ovarian biopsy technique developed for grey mullet can also be used to determine the stage of ovarian maturity in sea bass (Garcia 1989c).

Sea bass usually spawns in the evening (Garcia 1988). If 20 µg/kg LHRH_a is injected at 1100 H or 1700 H, sea bass spawns at dawn. However, those given a single injection at 2300 H or 0500 H spawn at daytime (Garcia 1990b). Unlike spawnings at daytime, spawning at dawn of LHRH_a-injected fish results in more eggs. Administration of LHRH_a at certain times of the day could provide

a convenient means of timing the onset of spawning of mature female sea bass so that a maximum number of eggs may be collected.

Gonadal maturation and spawning can be advanced two months before the peak of the breeding season by long term exposure to pelleted LHRHa or 17 alpha-methyltestosterone (MT) (Garcia 1990a). A single implant in March of 200 µg/kg LHRHa alone or in combination with 20 µg/kg MT induces greater percentage of ovarian maturation (43-71%) 45 days after administration compared to the control group. A lower dose (100 µg/kg) of these hormones also stimulates ovarian maturation (78-80%) in May following three previous monthly implantations. Two to three monthly implantations of LHRHa alone or in combination with MT or MT alone likewise induces testicular maturation in April (90%) and May (100%). Lower rate of maturation (83%) is exhibited by the control group from April to June.

Males mature earlier than females during the spawning season. Hence, milt consistency becomes a problem in the latter part of the season leading to low fertilization and hatching rates. Injection of 20-80 µg/kg LHRHa or 100 µg/kg MT enhances milt production and induces thinning of milt and spontaneous milt release in captive sea bass at the beginning and peak of the breeding season (Garcia 1992). The fertilization rate of hormone-induced sea bass varies and declines towards the end of the breeding season.

LHRHa is now routinely used to spawn mature sea bass broodstock at SEAFDEC/AQD. Unlike a pelleted hormone preparation, solubilized LHRHa diminishes in biological potency after storage for more than 90 days in a refrigerator (4-10°C) or 30 days at room temperature (28-30°C) (Garcia, unpublished results). In these studies, an opercular tag facilitates identification and monitoring of the individual response of fish to hormonal treatments (Garcia and Gapasin 1988).

Nursery

Rotifer consumption of sea bass increases exponentially with growth (Kohno and Duray 1990). Although the mouth width of larvae at initial feeding ranges from 200 to 240 millimeters the larvae ingested rotifers 80-160 µm in size (Duray and Kohno 1990)

Growth and survival of sea bass larvae are promoted when they are fed 6 *Artemia* ml/ five times daily starting on the 14th day of rearing (Duray 1990a). High survival is also attained by supplementing *Artemia* with *Moina* (water flea), suggesting the possibility of using *Moina* in rearing sea bass (Fermin 1991). A feeding scheme for sea bass larviculture has now been developed to optimize *Artemia* feeding (Dhert et al. 1990a). Newly hatched *Artemia nauplii* of San Francisco Bay type is offered from Day 8 to 10 and Great Salt Lake *nauplii* from Days 10 to 15 and 24 h enriched *Artemia* instar II thereafter. Feeding highly unsaturated fatty acid (HUFA)-enriched *Artemia* to sea bass larvae (17 mm TL) accelerates the onset of metamorphosis and improves their resistance to salinity stress (Dhert et al. 1990b). "However, fry reared on rice bran-fed pre-adult *Artemia* grew and survived better than those fry fed partially enriched *Artemia* (Bombeo, personal communication).

Simulated transport studies of sea bass fry at loading densities of 200 and 1600 fry per liter showed similar survival rates after 2-8 hours at 20°C and 20 parts per thousand salinity (Parazo, unpublished results).

Sea bass can be weaned over to an appropriate formulated diet as early as Day 10, but results are not satisfactory (Juario et al. 1991). The best method is to wean larvae gradually starting Day 20. Survival is high and cannibalism is minimized when larvae are reared until Day 60 using this method. Day 21 larvae abruptly weaned over to commercial pellets show lower survival rates (39-49%) compared with *Artemia*-fed larvae (74%) (Duray 1990b). Studies on the morphological and histochemical development of the digestive tract of sea bass supports the present weaning strategy (Minjoyo 1990). Sea bass juveniles swallow their prey whole and maximum prey size depends on predator mouth width, prey body depth, and total length. The size of prey ingested increases with increasing predator size but not to exceed 60-67% of predator total length (Parazo et al. 1991).

Based on the seed production studies conducted at SEAFDEC/AQD, a manual on sea bass hatchery operations and results of hatchery runs was published (Parazo et al. 1990, Parazo et al. 1991).

Although sea bass fry are already commercially produced in three private hatcheries in the Philippines, inconsistent survival has been reported. Thus, aside from food and feeding, other parameters like stocking density, light, etc. were investigated. Sea bass larvae survived and grew better when reared under natural lighting conditions than those under continuous light (Duray 1990c). Routine sea bass larviculture used an initial stocking rate of 30 newly-hatched larvae per liter. The stock is reduced halfway every week starting on Day 14. Cannibalism is minimized by culling the "shooters" and transferring them to a separate container (Parazo et al. 1991). In a related study, high growth rates were obtained when fry were stocked in an illuminated cage at 300 fry per cubic meter with or without supplemental feeding; poor growth was obtained at stocking rates of 150 or 600 fry per cubic meter (Fermin, unpublished results). Larvae acclimated to 16, 24, and 32 ppt salinities and subjected to eight test salinities ranging from 0 to 56 ppt showed that Day 23 larvae was most resistant (Javier, unpublished results).

Grow-out Culture

Polyculture of sea bass using tilapia as forage fish is often practiced. Growth and survival were higher among sea bass reared with tilapia in the same compartment than among those reared in ponds with net partition to separate the larger prey from predator (Triño, unpublished). Under laboratory conditions, sea bass actively select tilapia over sibling prey within the first 2 hours of exposure to prey (Avila et al. 1990b). When fed commercial pellets at 1.2 g/kg body weight (BW), the rate of food intake is higher in fishes held in groups than those kept individually. The specific growth rates of sea bass reared on commercial pellets were similar at salinities of 0, 15-16, and 34 ppt (Avila et al. 1990a).

Sea bass grow-out culture is still dependent on trash fish as food. A nutritious cost-effective artificial feed is now being developed to replace trash fish. There are several ongoing studies on the nutritional requirements of sea bass at various life stages. Dietary protein at 43% and dietary lipid at 12% are optimum for growth of sea bass fry (Alava 1990). High survival rates were obtained among juveniles given a high-protein high-energy feed (Catacutan, unpublished results). The quantitative amino acid requirements of sea bass were also studied. Methionine requirement of juveniles was estimated at 2.4% (Coloso, personal communication).

To reduce feed costs, cheaper sources of dietary protein and lipids must be used in formulations. As alternative to fish meal, protein sources such as shrimp head meal, meat and bone meal, soybean meal, and yeasts were evaluated. Sea bass fry and juveniles show better weight gain when fish meal is partially replaced (20%) by shrimp head meal in a 43 or 45% protein diet (Alava 1990). Similarly, cod liver oil, soybean oil, and coconut oil alone or in combination were tried on sea bass fry. Sea bass fed diet with cod liver oil and soybean oil in a 1:1 ratio show higher survival and growth than those fed diet with cod liver oil alone or soybean oil alone (Borlongan and Parazo 1991). Growth and survival are poor in the coconut oil diet and poorest in the diet without lipid supplement. Feed cost can be reduced if fish oil is partially substituted with soybean oil.

Since sea bass is commercially cultured under a wide range of salinities, its physiological response to osmotic stress was studied. Freshwater-adapted fingerlings and sub-adults transferred to seawater are able to regulate plasma osmolality and chloride ion back to normal levels in 2 days (Almendras 1991a). Seawater-adapted fingerlings and sub-adults can do the same in 1 day and 4 days, respectively. Ammonia excretion rate were also studied and found to be higher in freshwater-adapted fry than in seawater-adapted fry (Almendras 1991b). Ammonia excretion rates among freshwater and seawater-adapted sub-adults increased after feeding and returned to pre-feeding level after 10 hours.

Socioeconomics of Sea Bass Production

Species diversification is one factor that will help stabilize the aquaculture industry and sea bass culture offers such an opportunity. The profitability of sea bass aquaculture was assessed to entice fish farmers to go into this venture. A 10-year discounted cash flow projection indicates that a floating cage sea bass broodstock farm could be an economically viable enterprise (Agbayani, personal communication).

GROUPERS

Species Identification and Market Survey

Guides to grouper classification in the Philippines (Kohno 1986, 1987a) and in Southeast Asia (Kohno et al. 1990) had been published. Forty-six species from 7 genera were listed. Species identification was based on color patterns.

A fry collection survey (Solis, unpublished results) showed that grouper fry were most abundant in October with *Epinephelus suillus* and *E. megachir* as the dominant species. *E. suillus* fry were usually caught along channels of inshore waters while *E. megachir* on rocks close to open waters.

To identify the species available for aquaculture, species composition and abundance in grouper catch landed in Iloilo central market was monitored from April to October 1987 (Kohno and Duray 1989). Of the many grouper species found, *E. suillus* followed by *E. megachir* are the dominant species (Kohno and Duray 1989).

Breeding

The orange-spotted rockcod (*E. suillus*) is a protogynous hermaphrodite; sex inversion to male may occur at the age of seven years. The induction of sex reversal through hormone therapy is, therefore, vital to grouper propagation and culture. Regardless of the dose applied, only females weighing at least 1.2 kg undergo spermatogenesis after 3 months of biweekly injections of MT (Tan-Fermin et al. 1990). However, milt is obtained from both vehicle- and MT-treated fish six months after treatment. Seven monthly implantations of MT in silastic capsule did not induce sex inversion in 3-9 kg fish (Castillo, unpublished results). Allowing intraspecific interaction of *E. suillus* of different sizes in communal tanks for 11 weeks resulted in 4% of juvenile female grouper developing ovotestes (Tan-Fermin, unpublished results).

A single injection or pellet implantation of LHRHa plus MT did not induce *E. suillus* to spawn (Castillo, unpublished results). However, *E. suillus* spontaneously spawned in concrete tanks and in cages starting July 1990 (Toledo et al. 1990). Sequential spawnings occurred 5-10 times monthly. Spawnings were recorded late in the afternoon (1600 to 1800H) within four days before or after the last quarter moon phase.

Nursery

Larval rearing trials carried out for some grouper species over the past decade have had limited success. For *E. suillus*, similar results have been obtained at SEAFDEC/AQD by Nagai (1990) and Nagai et al. (1990). Preliminary data indicated an optimum stocking density of 10-20 larvae per liter. Oyster eggs, artificial plankton, or a combination of these with rotifers have been tried for larval rearing, but with little success (Nagai 1990). A practical diet containing 30% soybean meal, 25% squid meal, and 25% squid liver meal gave optimum

survival and growth rates in grouper fry (3.8-5.1 cm total length) (Nagai et al. 1990). High survival (92-97%) was attained when grouper fry (1.0 g BW) were fed minced fresh fish alone or in combination with fermented soybean meal. A 39% survival resulted when grouper fry (0.3 g BW) were weaned to formulated diet using fermented soybean meal as attractant; lower survival rates were obtained using either shrimp meal paste, squid meal, or squid liver meal as attractants (Nagai 1990).

Grow-out Culture

Kohno et al. (1988) reviewed the grouper culture practices in the Philippines. The existing culture method is usually based on the fishfarmers' own experience. Grouper culture is done in ponds or in cages. The most popular and desirable cultured species is *E. malabaricus* (= *E. suillus*). Increased production is constrained by insufficient fingerlings supply and lack of reliable culture technique. Kohno et al. (1989) investigated the effect of feeding rations and feeding frequencies on *E. suillus* in ponds. From an initial weight of 110-130 g, *E. suillus* can attain the marketable size (500 g) in 12 weeks, when fed trash fish at 5% BW once daily.

Disease Control

Cage and tank-held broodstock and juveniles of *E. suillus* were infected by *Vibrio*. Injured fish died when continuously exposed for 96 h to the bacteria. Intramuscular injection of oxytetracycline-HCl at 25 mg/kg BW for 5 consecutive days controlled the infection (Lavilla-Pitogo et al. in press).

SNAPPERS

Species Identification

A survey of snappers common in Iloilo markets revealed 13 species, the most abundant of which are: *Lutjanus decussatus*, *L. fulviflamma*, *L. vitta*, *L. carponotatus*, *L. malabaricus*, *L. gibbus*, and *L. fulvus* (Cheong, unpublished results).

SUMMARY

Spawning and egg production of sea bass have been maximized as a result of additional information on optimum dose, minimum egg size responsive to induction, timing of hormonal application, advancement of sexual maturation, and the shelf-life of LHRHa. Based on the results from recent studies, the stocking density, feeding and water management schemes for sea bass larval rearing have been modified to reduce cannibalism and improve survival. Sea bass nursery techniques under various conditions (tanks, ponds, and cages) are presently being investigated.

The spontaneous maturation and natural spawning of grouper (*E. suillus*) have paved the way for the development of reliable larval rearing techniques for this species.

Notwithstanding recent advances, many more aspects of sea bass, grouper, and snapper biology and culture require study: a) diseases of sea bass and grouper in captive broodstock and in the hatchery, b) cost-effective feeds from larval to grow-out stages, c) development of male spawners for sea bass and grouper, and c) broodstock development techniques for snapper.

REFERENCES

- Alava VR. 1990. Alternative sources of protein for fish meal in *Lates calcarifer* diets. Terminal report submitted to SEAFDEC/AQD. 41 p.
- Almendras JME. 1991a. Plasma osmolality and chloride regulation in the sea bass, *Lates calcarifer*. Terminal report submitted to SEAFDEC/AQD. 16 p.
- Almendras JME. 1991b. Ammonia excretion rates of the sea bass, *Lates calcarifer*, in fresh and seawater. Terminal report submitted to SEAFDEC/AQD. 13 p.
- Almendras JM., Dueñas C, Nacario J, Crim LW. 1988. Sustained hormone release: III. Use of gonadotropin-releasing hormone analogues to induce multiple spawnings in sea bass, *Lates calcarifer*. *Aquaculture* 74: 97-111.
- Avila EM, Salde R, Espada LA. 1990a. The specific growth rate of sea bass (*Lates calcarifer* Bloch) fed minimum pelleted rations at different rearing salinities. Terminal report submitted to SEAFDEC/AQD. 10 P.
- Avila EM, Ticar RB, Espada LA. 1990b. Selective predation on tilapia and sea bass by sea bass (*Lates calcarifer* Bloch) predators. Terminal report submitted to SEAFDEC/AQD. 18 p.
- Borlongan I, Parazo M. 1991. Effect of dietary lipid sources on growth, survival and fatty acid composition of sea bass (*Lates calcarifer* Bloch) fry. *The Israeli J. Aquaculture - Bamidgeh* 43: 95-102.
- Dhert P, Duray M, Lavens P, Sorgeloos P. 1990a. Optimized feeding strategies in the larviculture of the Asian sea bass *Lates calcarifer*. p. 319-323. In: *The Second Asian Fisheries Forum*. Hirano R, Hanyu I (Eds.). Asian Fisheries Society, Manila, Philippines. 991 p.
- Dhert P, Duray M, Sorgeloos P. 1990b. Improved larval survival at metamorphosis of Asian sea bass (*Lates calcarifer*) using ω 3-HUFA-enriched live food. *Aquaculture* 90: 63-74.
- Duray MN. 1990a. Level and frequency of feeding of sea bass larvae with *Artemia*. *Asian Aquaculture* 12(1):8-9.
- Duray MN. 1990b. Shifting sea bass larvae from live *Artemia* to artificial feeds. *Asian Aquaculture* 12(1): 7-8.
- Duray MN. 1990c. Growth and survival of milkfish and sea bass larvae in continuous light. *Asian Aquaculture* 12(3): 2-4.
- Duray MN, Kohno H. 1990. The relationship between mouth width and larval length to prey size in three marine fish species. *Phil. J. Sri.* 119: 237-245.
- Fermin AC. 1991. Freshwater cladoceran *Moina macrocopa* (Strauss) as an alternative live food for rearing sea bass *Lates calcarifer* (Bloch) fry. *J. Appl. Ichthyol.* 7: 8-14.
- Garcia LMB. 1988. Induced breeding in sea bass. *Asian Aquaculture* 10(3): 1-14.

- Garcia LMB. 1989a. Dose-dependent spawning response of mature female sea bass, *Lates calcarifer* (Bloch), to pelleted luteinizing hormone-releasing hormone analogue (LHRHa). *Aquaculture* 77:85-96.
- Garcia LMB. 1989b. Spawning response of mature female sea bass, *Lates calcarifer* (Bloch), to a single injection of luteinizing hormone-releasing hormone analogue: effect of dose and initial oocyte size. *J. Appl. Ichthyol.* 5:177-184.
- Garcia LMB. 1989c. Development of an ovarian biopsy technique in sea bass, *Lates calcarifer* (Bloch). *Aquaculture*. 77:97-102.
- Garcia LMB. 1990a. Advancement of sexual maturation and spawning of sea bass, *Lates calcarifer* (Bloch), using pelleted luteinizing hormone-releasing hormone analogue (LHRHa) and 17alpha-methyltestosterone. *Aquaculture* 86: 333-345.
- Garcia LMB. 1990b. Spawning response latency and egg production capacity of LHRHa-injected mature female sea bass, *Lates calcarifer* Bloch. *J. Appl. Ichthyol.* 6:167-172.
- Garcia LMB, Gapasin RSJ. 1988. An inexpensive tag for short term studies in milkfish (*Chanos chanos* Forsskal) and sea bass (*Lates calcarifer*). *J. Appl. Ichthyol.* 4:104-110.
- Juario JV, Duray MN, Fuchs J. 1991. Weaning of sea bass, *Lates calcarifer* B., to artificial diet. p. 183. In: Larvi'91-Fish and Crustacean Larviculture Symposium. Lavens P, Sorgeloos P, Ollevier F (Eds.). European Aquacult. Society Special Publ. 15, Gent, Belgium. 427 p.
- Kohno H. 1986. An introduction to lapu-lapu (*Epinephelus*) of the Philippines. *Asian Aquaculture* 8(6): 8-11.
- Kohno H. 1987a. An introduction to lapu-lapu (*Epinephelus*) of the Philippines. *Asian Aquaculture* 9(1): 5 & 10.
- Kohno H. 1987b. An introduction to lapu-lapu (*Epinephelus*) of the Philippines. *Asian Aquaculture* 9(2): 5-8.
- Kohno H. 1987c. An introduction to lapu-lapu (*Epinephelus*) of the Philippines. *Asian Aquaculture* 9(3): 5-8.
- Kohno H, Duray M. 1989. Species composition of groupers in Iloilo market. *Asian Aquaculture* 11(3): 6-8.
- Kohno H, Duray M, Juario J. 1988. State of grouper (*lapu-lapu*) culture in the Philippines. *Asian Aquaculture* 10(2): 4-8.
- Kohno H, Duray M. 1990. Daily amount of rotifers taken by a sea bass *Lates calcarifer* larva. *Phil. J. Sci.* 119:247-255.
- Kohno H, Duray M, Sunyoto P. 1990. A field guide to groupers of Southeast Asia. Pusat Penelitian dan Pengembang PHP/KAN/PT No. 14/1990. Jakarta. 26 p.
- Kohno H, Triño A, Gerochi D, Duray M. 1989. Effects of feeding frequency and amount of feeding on the growth of grouper, *Epinephelus malabaricus*. *Phil. J. Sci.* 118:89-93.
- Lavilla-Pitogo CR, Castillo AR, de la Cruz MC. Occurrence of *Vibrio* sp. infection in grouper, *Epinephelus suillus*. *J. Appl. Ichthyol.* (In press).
- Minjoyo H. 1990. Histochemical studies on the early stages of development of the digestive tract of sea bass *Lates calcarifer* (Bloch). M.Sc. Thesis. UP-MSI, Diliman, Quezon City, Philippines. 61 p.
- Nagai A. 1990. Natural spawning and larval rearing of grouper, *Epinephelus malabaricus*. Terminal report submitted to SEAFDEC/AQD. 40 p.
- Nagai A, Kume T, Javellana D. 1990. The effect of new formulated dry diet on the growth and survival of small grouper fry (*E. malabaricus* Bloch and Schneider). Terminal report submitted to SEAFDEC/AQD. 26 p.
- Parazo MM, Avila EM, Reyes DM Jr. 1991. Size and weight-dependent cannibalism in hatchery-bred sea bass (*Lates calcarifer* Bloch). *J. Appl. Ichthyol.* 7:1-7.

- Parazo MM, Garcia LMB, Ayson FG, Fermin AC, Almendras JME, Reyes DM Jr., Avila EM. 1990. Sea bass hatchery operations. Aquaculture Extension Manual No. 18. Aquaculture Dept., SEAFDEC. 38 p.
- Parazo MM, Reyes DM Jr., Avila EM. 1991. Hatchery rearing of sea bass, *Lates calcarifer* Bloch. Philipp. Sci. 28:65-76.
- Tan-Fermin JD, Garcia LMB, Castillo AR. 1990. Induction of sex inversion in juvenile grouper, *Epinephelus malabaricus* (Bloch and Schneider) by biweekly injections of 17alpha-methyltestosterone. p. 19-20 In: Hormones and the Environment. Chan DKO (Ed.). Proceedings of an international symposium held at the University of Hongkong, 18-20 December 1989. Society for the Study of Endocrinology, Metabolism and Reproduction, H.K. 168 p.
- Toledo JD, Nagai A, Javellana D, Tan-Fermin JD. 1990. Successive spawning of grouper, *Epinephelus malabaricus*, in tank and floating net cage. Asian Aquaculture 12(4): 8-10.