

ARTIFICIAL PROPAGATION OF SEABASS, *LATES CALCARIFER* (BLOCH, 1790) IN MYEIK ARCHIPELAGO

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Abstract

Artificial propagation of fish is an alternative to meet increasing demand for food fish. Seabass (*Lates calcarifer*) is one of the commercially important species in Myanmar. It is fast growing and euryhaline. This fact is seen as a valuable attribute for species enabling its adoption for ponds and cages culture under marine and brackish water environment. This study was conducted to assess the breeder management for artificial propagation of seabass. The study period was from September 2017 to December 2018 in Myeik Archipelago. The breeders were reared for two months before injection to be healthy fish. The male and female spawners were selected and injected hormone which was calculated to males and females, and their body weight. Induced breeding of study species was studied and number of fertilized eggs, per cent of mortality rate and survival rate were observed seasonally. The hatching rate and survival rate of study fish were highest in hot season. Larvae rearing and different diets on larvae were observed. The effects of weather and water parameters of the study area were collected.

Keywords : Artificial propagation, Seabass, *Lates calcarifer*, spawners, pond and cage culture, larvae rearing, embryonic developmental stages.

Introduction

The seabass (*Lates calcarifer*) is one of the species with a high potential for cultivation. It is fast growing and euryhaline. This fact is seen as a valuable attribute for a species enabling its adoption for pond and cage culture under marine, brackish, and freshwater environments. *Lates calcarifer* (Bloch, 1790) is one of the commercial important species in Myanmar. It is one from nine *Lates* species (the others mostly in fresh waters in Africa) of the Latidae (Parazo *et al.*, 1998). The fish comes under a diverse group of common names “Barramundi” in Australia, “Giant sea perch” in Papua New Guinea, “sea bass” in South-east Asia and “Ka-kadit” in juvenile and male phase and Ka-tha-baung in Myanmar (Tin Tin Aye, 2004).

Artificial propagation of sea bass was first achieved in Thailand during 1971 by stripping the ripe spawners collected from natural spawning grounds. Wongsomnuk and Maneewongsa (1974) successfully induced cultured broodstock to spawn in captivity by hormone stimulation. Captive broodstock of sea bass were successfully induced to spawn naturally using environmental stimulation (Kungvankij, 1981). Thailand is the most advanced country in the production of sea bass seed from spawners collected from the wild and induced to breed since 1973. Thailand is presently producing more than 100 million seeds annually. The popularity and demand for this species made it an obvious choice for the development of aquaculture technology. Techniques for the culture of barramundi were developed in Thailand in the early 1970s (Wongsomnuk and Manevonk, 1974) and considerable progress in aquaculture techniques for the species has been achieved since that time.

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The growth of barramundi was investigated by tagging and scale reading in northern Australia (Davis and Kirkwood 1984), by scale reading in the sexually precocious population of the northeast Gulf of Carpentaria (Davis 1984a). In present study, sea bass are cultured in cage and tested by tagging with biotag during the study period.

Lates calcarifer, known as sea bass in Asia and barramundi in Australia, is a euryhaline member of the family Centropomidae. It inhabits freshwater, brackish and marine habitats including streams, lakes, estuaries and coastal waters. The newly-hatched larvae are distributed along the coastline of brackishwater estuaries while 1cm size larvae can be found in freshwater bodies (Bhatia and Kungvankij, 1971). Under natural condition, sea bass grows in fresh water and migrates to more saline water for spawning. The sea bass (*Lates calcarifer*) is one of the species with a high potential for cultivation. Seabass spawn naturally in captivity (Toledo et al. 1991). Alternatively, they can be induced to spawn by hormonal or environmental manipulations (Kungvankij 1987, Garcia 1989a, b). This widely developed in South East Asia, sea bass (*Lates calcarifer*) are popular marine food fish of high market value and culture of sea bass has been successful in coastal regions in Myanmar.

Based on size and weight of larvae, the stocking density of 30 ind/l is the most appropriate in mass producing sea bass fry (Juario and Duray 1985). Since the seabass is cannibalistic, the larger ones eating up the smaller, it is essential to grade them from the fry stage onwards into different size groups. Stocking the same size of larvae and fry will reduce the rate of cannibalism, the survival rate will be increased and the growth rate of the fish could also be faster and more uniform in size (Chantarasri *et al.*, 1989). At about 2-3 months old stage they are ready for stocking in large meshed grow-out net cages and ponds for commercial culture.

The objectives of the present study were as follows:

- to assess the breeder management for pre-spawning of seabass
- to conduct the induced breeding procedure for seabass
- to find out the seasonal variation of fertility rate, hatching rate and survival rate of seabass
- to identify the larvae rearing of seabass

Materials and Methods

Study period and study sites

The study period lasted from September 2017 to December 2018. The investigation on *Lates calcarifer* was conducted at Sarr kyun (Sarr insand) located in (12° 33' 1.98" N, 98° 28' 40.33"E), and Ye myit kyi village (12° 33' 27.0" N, 98° 19' 34.13"E) in Myeik Township, Taninthayi Region (Fig. 1)

Food and feeding the selected broodstock

The breeders fed twice daily with fresh fish and squid at the rate of 1 per cent of body weight for 1-2 time a week. Moreover, vitamin A, B, C and E were supplemented once daily alternately (by putting inside fresh fish). It is fed twice daily with 5 per cent of body weight for pellet diet to old breeding stock.

Selection of spawners

The breeders were stocked for two months before injection. It comes from both wild type and cultured fish. They were selected as broodstock size– female 3 kg up, male < 4 kg (between 2yr and 7yr). They must be healthy and active fish. Their fins and scales must be complete for sign of health. They were free from disease and parasite and also free from injury.

Tagging and preparation for injection

The breeders were marked tagging by biomass to calculate the dose for injection. Ten females and five males of broodstock were injected with single dose of 1 cc hormone per 1 kg fish. Suprefact and Dextrose ratio of (1:2) for female and ratio of (1:1) for male were injected. According to the spawning behavior of species, the breeders were injected at 11:00 in the morning, and then released injected-fishes back into spawning tank. Begin releasing mucus before spawning (after 30:30 hours of injection). Spawning was occurred at 32:30 hours after injection.

Procedures of seed production

After hormone-inducing, the brood fish was transferred into the spawning tank. The water overflowing method was used for collecting eggs from a spawning tank. The flowing water carries the eggs into the eggs collecting tank from which it transfers into the hatching tanks by using (200 μ) fine netting. The unfertilized eggs sinking to the bottom were discarded before placing in the larvae rearing tanks. The fertilized eggs were stocked in the incubation tanks until hatching about 12-15 hrs.

Egg collection and incubation

The fertilized eggs were collected by fine cloth and 50 μ hand net by using air lift method. It is used sampling method; density of egg, estimate egg number per litre for egg counting. The fertilized eggs were stock into hatching tanks that is concrete tank by using incubate equipments. The optimum water quality is monitored with five parameters. The eggs density was placed by about 1,000,000 in one tank (100 eggs/liter). The tanks were given gently aeration to prevent the eggs from settle on the bottom.

Data analysis

$$\text{Hatching rate} = \frac{\text{Total fertilized eggs} - \text{unfertilized eggs}}{\text{Total number of hatched eggs}} \times 100$$

$$\text{Survival rate} = \frac{\text{Total number of spawn}}{\text{Total number of spawn stocked}} \times 100$$



(Source: Department of Geography, Myeik University)

Figure 1 Map of study site



Spawning tank and egg collection tank



Hatching tanks



Larvae rearing tank

Plate 1. Hatchery facilities



Step1. Carrying with hand net



Step 2. Tagging with biomark



Step 3. Injection of hormone

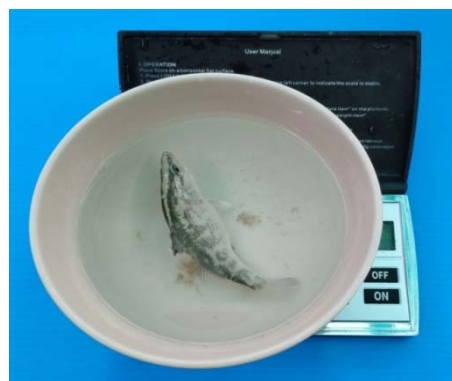


Step 4. Releasing injected fish into the spawning tank

Plate 2. Steps of inducing hormone to seabass



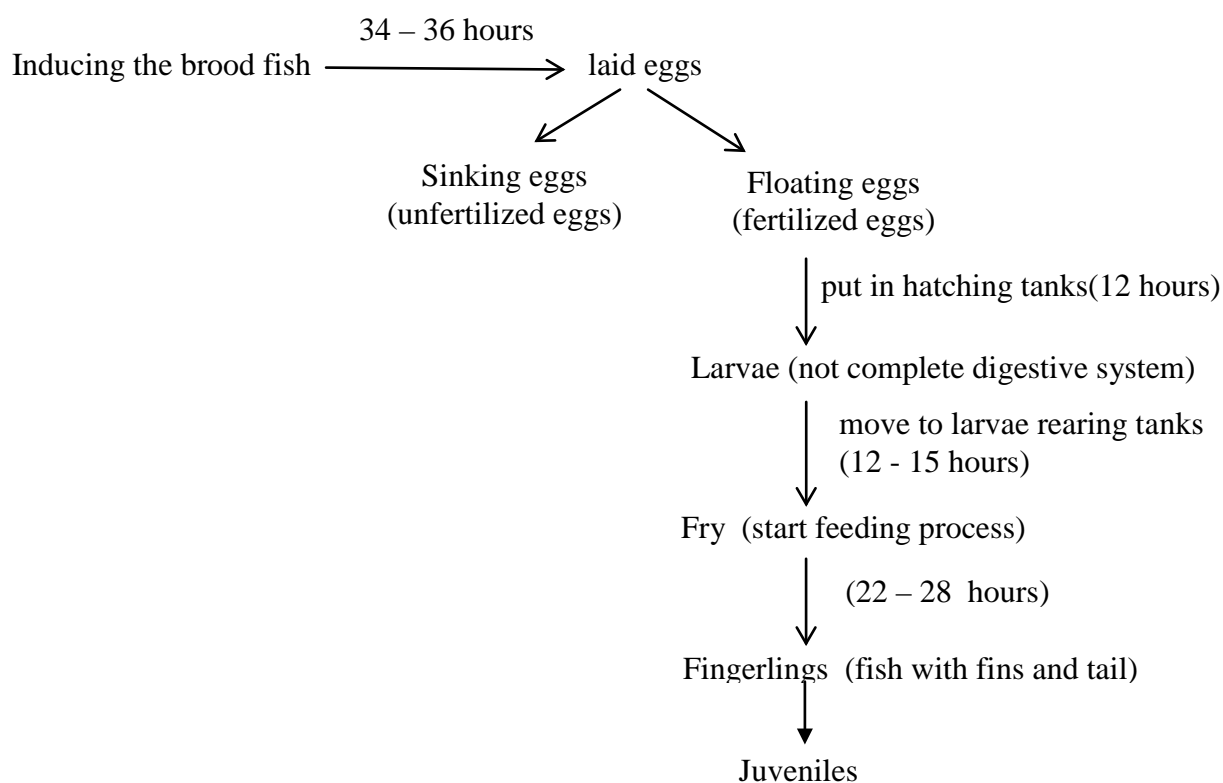
Length measuring



Weight measuring

Plate 3. Measuring size of seabass in larvae rearing tank

Larvae rearing procedure



Results

Systematic position of study species

The systematic positions of *Lates calcarifer* was followed after

Kingdom	-	Animalia
Phylum	-	Chordata
Class	-	Actinopterygii
Order	-	Perciformes
Family	-	Latidae
Genus	-	<i>Lates</i>
Species	-	<i>L.calcarifer</i> (Bloch, 1790)
Common name	-	Barramundi / Seabass
Local name	-	Ka kadit (Ka tha baung)



Plate 4. Male and female Seabass

Sex ratio and size of breeder

The sex ratio of male and female breeders were 1 : 2, some breeders were wild and some had been injected previous time. The size of female were 62.5 cm to 77.5 cm in length and 3.5 kg to 7 kg weight and male were 55 cm to 67.5 cm length and 2.8 kg to 3.5 kg weight (Table 1, 2 and 3).

Table 1 Measuring and tagging to Seabass for injection in hot season

No.	Date	Tag Number	Sex (M/F)	Length (cm)	Weight (kg)	Dose	Induced Time (pm)
1	29/4/18	wild stock	F	65	4.5	5 cc	10:18
2	29/4/18	2051374*	F	70	5.5	5 cc	10:20
3	29/4/18	wild stock	F	70	5.5	6 cc	10:22
4	29/4/18	wild stock	F	67.5	5	5.5 cc	10:25
5	29/4/18	wild stock	F	75	6	6.5 cc	10:28
6	29/4/18	wild stock	F	70	4	4.5 cc	10:30
7	29/4/18	wild stock	F	75	6	6.5 cc	10:31
8	29/4/18	wild stock	F	67.5	5	5.5 cc	10:33
9	29/4/18	wild stock	F	60	4	4.5 cc	10:24
10	29/4/18	1916542*	F	67.5	5.5	5 cc	10:39
11	29/4/18	wild stock	M	65	3	3.5 cc	10:27
12	29/4/18	wild stock	M	62.5	3	3.5 cc	10:35
13	29/4/18	wild stock	M	67.5	3.5	4 cc	10:37
14	29/4/18	wild stock	M	67.5	3	3.5 cc	10:42
15	29/4/18	wild stock	M	62.5	3	3.5 cc	10:45

Table 2 Measuring and tagging to Seabass for injection in wet season

No.	Date	Tag Number	Sex (M/F)	Length (cm)	Weight (kg)	Dose	Induced Time (pm)
1	30/6/18	1299817*	F	67	5.5	5cc	10:00
2	30/6/18	wild stock	F	70	5.5	6cc	10:12
3	30/6/18	wild stock	F	67.5	5	5.5cc	10:04
4	30/6/18	1916542985*	F	75	6	5.5cc	10:16
5	30/6/18	2049387985*	F	67.7	6	5.5	10:22
6	30/6/18	2025977*	F	70	7.5	7cc	10:07
7	30/6/18	2043341*	F	65	5	4.5	10:20
8	30/6/18	2037659*	F	62.5	4.5	4	10:15
9	30/6/18	1802957*	F	65	5	4.5	10:32
10	30/6/18	1762213*	F	65	4.5	4cc	10:35
11	30/6/18	wild stock	M	67	3.5	4cc	10:10
12	30/6/18	wild stock	M	70	4	4.5cc	10:23
13	30/6/18	wild stock	M	62.5	3	3.5cc	10:25
14	30/6/18	wild stock	M	67.5	3.5	4cc	10:28
15	30/6/18	wild stock	M	67	3	3.5cc	10:38

Table 3 Measuring and tagging to Seabass for injection in cold season

No.	Date	Tag Number	Sex (M/F)	Length (cm)	Weight (kg)	Dose	Induced Time (pm)
1	30/12/18	02018978*	F	70	5.5	5 cc	11:00
2	30/12/18	wild stock	F	62.5	3.5	4 cc	11:13
3	30/12/18	wild stock	F	62.5	4	4.5 cc	11:18
4	30/12/18	wild stock	F	65	4.5	5 cc	11:21
5	30/12/18	wild stock	F	67.5	3.5	4 cc	11:24
6	30/12/18	wild stock	F	62.5	4	4.5 cc	11:25
7	30/12/18	wild stock	F	75	6	6.5 cc	11:27
8	30/12/18	01811216*	F	75	6	5.5 cc	11:28
9	30/12/18	wild stock	F	77.5	7	7.5 cc	11:30
10	30/12/18	02048854*	F	70	7	6.5 cc	11:34
11	30/12/18	wild stock	M	67.5	3	3.5 cc	11:02
12	30/12/18	wild stock	M	60	2.8	3 cc	11:04
13	30/12/18	wild stock	M	60	2.9	3 cc	11:08
14	30/12/18	wild stock	M	65	3	3.5cc	11:10
15	30/12/18	01934984*	M	55	3.5	3 cc	11:11

Seasonal fertility rate, hatching rate and survival rate of Seabass

Total number of eggs were collected differently in seasonally, during the hot season the total number of eggs are highest amount and lowest in wet season. The fertility rate was highest in hot season but lowest in wet season. The hatching rate was highest in hot season but lowest in cold season (Table 4). The survival rate of Seabass was different; highest survival rate (63 per cent) was found in hot season with salinity was 29 to 30 ppt and water temperature was 28 – 32 °C, and the lowest (30 per cent) was observed in cold season with salinity was 28 to 30 ppt and water temperature was 28 – 30 °C (Table 5 and Table 7).

Table 4 Seasonal variation of fertility rate of Seabass

Season	Total no. of eggs	No. of fertilized eggs	No. of unfertilized eggs	Fertility rate (%)	No. of hatched eggs	Hatching rate (%)
Hot	16,000,000	14,000,000	2,000,000	87.5	13,600,000	97.1
Wet	9,000,000	6,250,000	2,750,000	69.4	5,000,000	80
Cold	15,000,000	12,000,000	3,000,000	80	9,500,000	79.2

Table 5: Seasonal variation of survival rate of Seabass

Rearing (Hot Season)	Larval age	Survival rate %	Number	Salinity	Temp (C)
	Day 1	100	13,600,000	29-30	28-32
	Day 30	91	12,404,000	29-30	28-32
	Day 60	76	9,358,000	29-30	28-32
	Day 90	65	6,080,000	29-30	28-32
	Day 100	63	3,850,000	29-30	28-32
Rearing (Wet Season)	Larval age	Survival rate %	Number	Salinity	Temp (C)
	Day 1	100	5,000,000	23-25	25-28
	Day 30	83	4,130,000	23-25	25-28
	Day 60	24	975,000	23-25	25-28
	Day 90	61	590,000	23-25	25-28
	Day 100	43	253,000	29-30	28-32
Rearing (Cold Season)	Larval age	Survival rate %	Number	Salinity	Temp (C)
	Day 1	100	9,500,000	28-30	28-30
	Day 30	84	7,950,000	28-30	28-30
	Day 60	74	5,875,000	28-30	28-30
	Day 90	61	3,560,000	28-30	28-30
	Day 100	30	1,050,000	28-30	28-30

Age and amount of supplementary food for Seabass larvae

The larvae were supplied live food (such as rotifer and artemia) until they reached the age of 14 days and then supplied the artificial pellet with respective size and weight (Table 6).

Growth rate of Seabass larvae

Larvae were metamorphosed from fry to fingerlings after one month with the weight of 8.3 gm and length 9 cm, in two month larvae were not much increase growth rate of 8.76 gm to 9 cm. After two and half month, the larvae reached 11 cm to 14. 78 gm which stage was fingerling (Fig 2, Table 8).

Table 6 Age and amount of food supply to Seabass larvae

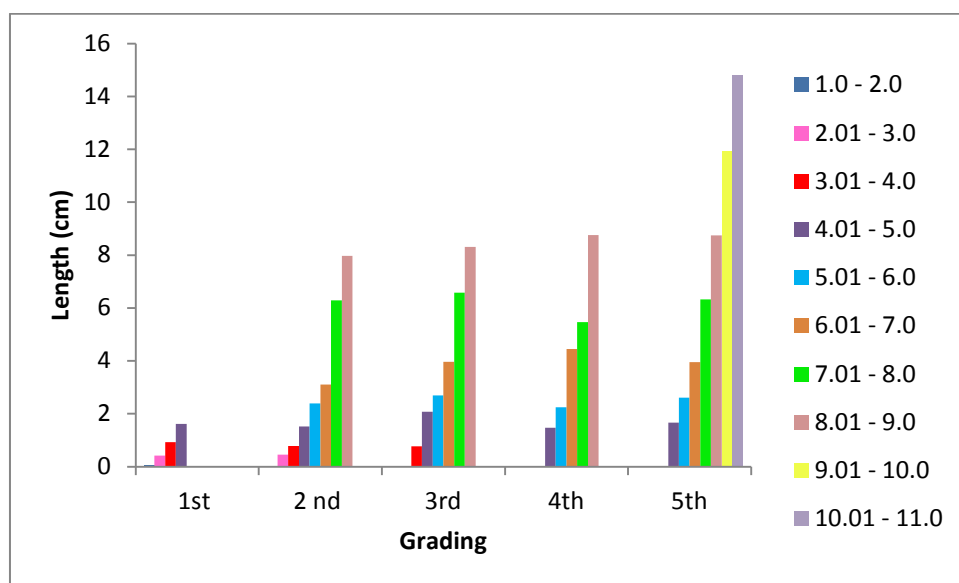
Age	Food supply	Amount of food gm / thousand
2 - 4 days	Rotifers	12
5 - 9 days	Rotifers	12
10 - 13 days	Rotifers	24
	Artemia	0.4
14 - 16 days	Artemia	40
	Inve $\frac{1}{2}$ + $\frac{2}{3}$	40
17 - 22	Artemia	67
	Pellet (size - Inve $\frac{3}{8}$ + $\frac{5}{8}$)	67
23-31 days	Pellet (size - Inve $\frac{3}{5}$ + $\frac{5}{8}$)	67g
1½ months	Pellet (size - Inve $\frac{5}{8}$ + G8)	16g
2 months	Pellet (size - Inve G8 + G12)	16g
2½ months	Pellet (size - Inve G12 + 1042)	125g

Table 7 Water parameter in larvae rearing tank

Parameters	Range of value (standard) by FAO	Range of value (experiment)
Water temperature	26 – 32 °C	23 - 32 °C
Salinity	10 – 30 ppt	29 - 32 ppt
pH	7.5 – 8.5	6.8 – 8
Dissolved oxygen	4 – 9 mg/ l	6 – 9 mg / l
Ammonia(NH ₃)	Less than 1 ppm	0.3 – 0.5 ppm

Table 8 Grading of fish by length in larvae rearing tank (n = 100)

Weight(gm) Length(cm)	1 st Grading	2 nd Grading	3 rd Grading	4 th Grading	5 th Grading
1.0 - 2.0	0.07				
2.01 - 3.0	0.42	0.45			
3.01 - 4.0	0.93	0.78	0.77		
4.01 - 5.0	1.62	1.52	2.08	1.47	1.67
5.01 - 6.0		2.39	2.69	2.24	2.61
6.01 - 7.0		3.1	3.96	4.45	3.95
7.01 - 8.0		6.29	6.58	5.46	6.32
8.01 - 9.0		7.97	8.31	8.76	8.75
9.01 - 10.0					11.91
10.01 - 11.0					14.78

**Figure 2** Larvae size grading of Seabass

Discussion

Grace (1985) described that *Lates calcarifer* was one of the important cultured fish as a commercial and subsistence food fish and its relatively high market value, it had become an attractive commodity of both large and small-scale aquaculture enterprises. Sea bass is farmed with high production in Southeast Asia, generally from small coastal cage farms and large-scale sea bass farms in Australia. In Myanmar, sea bass are successful in induced breeding and also cultured in cage for its commercial value.

Adult seabass (3 – 5 kg) migrated towards the mouth of the river from inland water into the sea where the salinity ranges between 30 – 32 ppt for gonadal maturation and subsequent spawning (Grace, 1985). Sources of adult seabass spawners could be collected from wild-caught adults and from cages or ponds about 2 – 6 years old species averaging in weight from 2 – 8 kg (Ramon Y. Tangon, 2016). In present study, 2 – 7 kg breeders were selected for induced breeding and optimum salinity range is between 28 – 32 ppt, the previous study is in agreement with previous authors Grace Mathew, 1985 and Ramon Y. Tangon, 2016). Captive broodstock of sea bass were also successfully induced to spawn naturally using environmental stimulation (Kungvankij 1981).

Good broodstock management and proper nutrition are essential for full maturity to ensure sustainable supply of quality eggs and fry for stock enhancement (Reyes, 2014). In present study, breeders were well feed and care for two months before the induced breeding, the present study is in agreement with Reyes, 2014.

The hatching were occurred approximately 14hrs after fertilization at 28°C and 32-33 ppt according to Parazo *et al.*, 1998). MacKinnon *et al.* found barramindi spawned in a northern Gulf of Carpentaria estuary when water temperatures ranged from 27°C to 33°C and salinities from 28 to 34 ppt. Salinities in the range necessary for barramindi egg fertilization in Thailand 28-32 ppt (MacKinnon *et al.*, 1984). Newly hatched *nauplii* incubate at a density of (1g cysts/l) in plastic hatching vessels filled with clean filtered seawater and provided with vigorous aeration. The cysts will hatch normally within 24 h (Parazo *et al.*, 1998).

In one month, the spawn metamorphose into the fry and fingerling stages was completed in one month, which has the appearance very similar to the parent fish. The length measured 1.5-2.0 cm (Tookwinas, 1987) which was coincide with the present finding of 28 days and 0.07 – 1.58 cm. Ergun Buke (2002) found that the hatching rates were 80% to 90% and the survival rates were 40% to 65% in Turkey, Europe. In the present study, 97% of the hatching rate and 63% of the survival rate was found in hot season, 80 % of the hatching rate and 43 % of survival rate and then 79.2 % of hatching rate and 80 % of survival rate were observed, so the present study was agreement with above authors.

As the grading result, sea bass larvae were better growth rate in larvae rearing tank during the hot season. The present finding was agreement with Charles *et al.*, (2003), he said that the effects of temperature and salinity are significantly different in weight of juvenile black sea bass.

Summary

The seabass breeders were stocked for two months at Sarr kyun (salt island) before injection and 1: 2 ratio of male and female breeders were injected for artificial propagation at Ye Myint Kyi village in Myeik Township, Taninthayi region deported from September 2017 to

December 2018. The fertilized eggs, hatched eggs and survival fry were observed with estimate counting method. The hatching rate and survival rate were highest in hot season and lowest in cold season.

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