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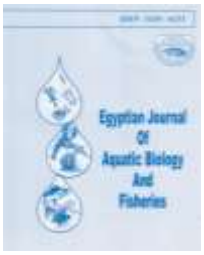

Reproductive biology of the yellow-striped goatfish *Mulloidichthys flavolineatus* (Lacepède, 1801) (Perciformes: Mullidae) in the Red Sea, Egypt.

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ABSTRACT

The Red Sea population of the yellow-striped goatfish *Mulloidichthys flavolineatus* (Lacepède, 1801) was studied to determine the spawning season and characteristics of its reproduction to support fisheries management. Adult fishes were caught with a gill net in the Egyptian Red Sea from January to December 2015. Specimens were dissected and sex ratio, gonado-somatic index, fecundity, maturity stages and egg diameter were determined. Larvae were collected with a plankton net at monthly intervals between January and December 2015. The gonado-somatic index was highest during summer (June-August) with that of females being significantly higher than that of males. Larvae were found between June and August with the highest densities in August. The spawning season of *Mulloidichthys flavolineatus* was recorded between June and August coinciding with increasing water temperatures. Spawning seasons of fish determined by fish larvae survey is in agreement with those determined by reproductive biology studies.

INTRODUCTION

Due to their economic value, goatfishes have been the subject of intense biological and taxonomical studies (Munro, 1976; Lee, 1983; Wahbeh, 1992). Goatfishes (Mullidae) are moderately sized (30-60 cm) bottom dwelling fishes often found in coral reefs. The various species of this family can be easily identified by the presence of a pair of barbles on the chin. These barbles are used to detect and uncover benthic invertebrates (Randall, 1986; Ormond & Edwards, 1987). Goatfishes are numerically small, with about 60 species in 6 genera. They are successfully distributed into shallow tropical and temperate regions (Thresher, 1984). Three genera of the Mullidae have been recorded in the Red Sea; *Parupeneus* spp., *Mulloidichthys* spp. and *Upeneus* spp. with 5, 2, 7 species respectively (Golani & Bogorodesky, 2010).

The yellow stripe goat fish *Mulloidichthys flavolineatus* (Lacepède, 1801) is widely distributed throughout the Indo-Pacific region (Holland *et al*., 1993) and also known from the Red Sea. It is one of the most important food fishes, and their juveniles are highly prized as bait. Despite their cultural and commercial importance and widespread distribution, very little is known about their distribution, movements, habitat utilization, and dispersal patterns.

Mulloidichthys flavolineatus is a migratory species migrating from deep water and coral reef areas to shallow soft bottom habitats to feed on invertebrate prey that is abundance when water temperatures rise.

This migration occurs between May and August and its onset coincides with full moon (Alkholy, 1964; Boraey, 1969; Ahmed, 2018).

The study of the ichthyoplankton is of great importance to fisheries management. Data about fish larva provide a basisfor studying population dynamics of major fishery species, such as *Mulloidichthys flavolineatus* (Smith & Richardson, 1977; Rutherford *et al*., 1997 and Butler *et al*., 2003; Abu El-Regal, 2017a). Knowledge about ichthyoplankton ecology comprises an important component of stock assessment and fishery management plans (Rutherford, 2002). Moreover, the estimation of egg abundance can be used to estimate the biomass of the spawning stock. Studies of larvae are also used to detect spawning grounds and spawning seasons (Smith & Richardson, 1977; Armstrong *et al*., 2001; Fuiman, 2002).

Published information on the comprehensive biology and ecology of *M. flavolineatus* is scarce despite its high economic value. Boraey, (1965) and Wahbeh (1992) studied the reproductive biology of the species in the Red Sea and there was an agreement between the two studies regarding the spawning seasons and sex ratio. The species was found to spawn in summer from June through August (Ahmed, 2018).

The present study, aimed at determining the spawning seasonof *M. flavolineatus* in the Egyptian Red Seaby studying the reproductive biology and larval presence and development of this species. These data provide a solid basis for the management and conservation of this species.

MATERIALS AND METHODS

Study area

The population of *Mulloidichthys flavolineatus* was studied in the coastal waters off Hurghada (Egypt) in the north western section of the Red Sea (Fig. 1).

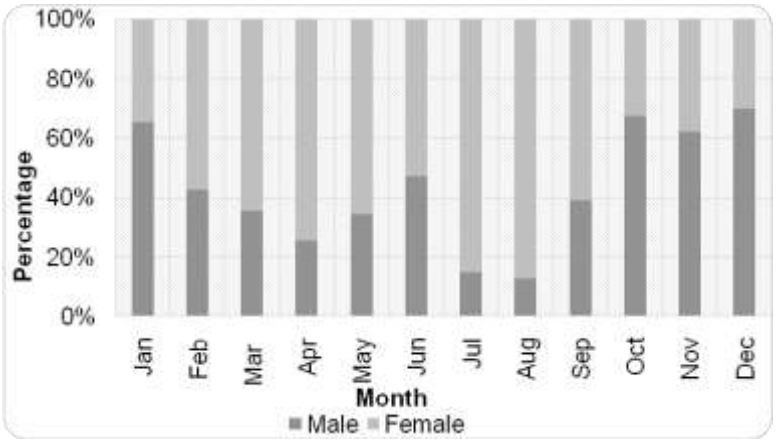

Fig. 1: The percentage contribution of females to males in the present collection

Sampling of adult fish

Adult specimens were fished monthly with a gill net of 3-5 cm mesh size from January to December 2015. All fish samples were collected early in the morning just after the sunrise. The collected fish were frozen till the examination in the laboratory.

Biometry and data analysis

The total length (TL in cm) and weight (W in g) were determined for each adult specimen.

Sex ratio

All specimens were dissected, sexed, and sex ratio was determined for each month. The Chi square test was used to assess sex ratio deviation from a 50 male:50 female ratio (Wootton, 1998).

Gonado-somatic index

Gonads were weighed to the nearest 0.01 g to calculate the monthly Gonado-Somatic Index (G.S.I.) using the following formula (Berhaut 1973; Siddeek et al., 1994; Valdes et al., 2004)

G.S.I =Wet weight of gonad (g)/ Wet weight of fish (g) X 100

Maturity stages

Maturity stage of each individual was determined according to the size of the gonad and oocytes categorizing them in five maturity stages: Immature, maturing, mature, ripe and spent.

Size at first maturity

The length at first sexual maturity (Lm); the length at which 50% of fish reach their sexual maturity was estimated by fitting the percentage maturity against mid lengths. L_m was estimated as the point on X-axis corresponding to 50% point on Y-axis.

Fecundity

The relationship between absolute fecundity (Fa) and total length was determined by regression analysis. Relative fecundity was calculated with weight, total length and ovary weight (Kostrzewa et al., 2003; Mousavi-Sabet et al., 2011a). Statistical tests were conducted with SPSS (version 22.0).

The relationship between absolute fecundity and the length or the weight of a fish were determined according to the equation:

$$F=axb$$

Where F is absolute fecundity, x is the length or weight of the fish and a, b are constants.

Egg diameter

Egg diameter of 50 specimens of *M. flavolineatus* was measured during the period from June to August. Egg diameter was measured to the nearest 1 μm using a micrometre scale in the ocular of a microscope at a magnification of 40X.

Sampling of larval stages

Fish larvae were collected from 12stations located at Hurghada area on the Egyptian coast of the Red Sea using a plankton net (100 cm diameter and 0.5 mm mesh size). Collection were performed monthly between January and December 2015 in the early morning just before sunrise to avoid the downward movement of larvae during the daylight. The net was towed for 10 minutes parallel to the coast and about from 10-100 m from the coast. The net was equipped with a flow meter to calculate the volume of water filtered allowing the calculation of larval fish densities. Samples were preserved in a buffered 5% formaldehyde seawater solution.

In the laboratory, larvae of *Mulloidichthys flavolineatus* were sorted, identified, counted and measured to the nearest 0.1 mm before final preservation in 70% ethanol.Identification guides (Leis & Rennis, 1983; Abu El-Regal, 1999; Carson-Ewart, 2004; Abu El-Regal, 2017a) were used to identify larvae of *Mulloidichthys flavolineatus*. A total of 324 hauls were taken where each site was hauled 3 times in month.

RESULTS

Length frequency distribution

The length of fish ranged from 11.5 cm to 31 cm with an average length of 21.5 cm. Females range in size from 14 cm and 31 cm with an average of 22 cm while males varied in size from 11.5 to 26 cm with an average of 18.4 cm. The largest size was observed in November with 31 cm while the smallest fish were recorded in January with 11.5 cm. Small-sized fish began to appear in November with 12 cm indicating a new recruitment.

Sex ratio

A total of 716 specimens were caught and used to determine the monthly sex ratio of *Mulloidichthys flavolineatus* (Table 1, Fig. 1). Throughout the entire year, 426 females (59.5%) and 290 males (40.5%) were caught. The overall female: male ratio of 1.5 was significantly different from expected 1:1 ratio ($F= 218.8$, $P<0.01$).

The sex ratio varied among months with the dominance of female fish most of the year. In the period from October to January, the population was dominated by male fish with the maximum number of male was recorded in December where male constituted 70% of the population. This situation is reversed in the period from February to September when female numbers were greatly higher than male numbers. Number of female started to increase in February forming about 57% of the population. Number of female goatfish reached its maximum in July and August with 85% and 87% respectively (Fig. 2).

Table 1: The numbers of both, sex ratio and percentage of male and female *M. flavolineatus* and Chi square values.

Month	Number of fishes			Sex ratio	Percentage		P-value	Chi square
	Total number	Male	Female		% Male	% Female		
Jan	72	47	25	1.9	65.3	34.7	0.01	6.7
Feb	40	17	23	0.7	42.5	57.5	0.3	0.9
Mar	17	6	11	0.5	37.5	68.8	0.2	1.5
Apr	51	13	38	0.3	25.5	74.5	0	12.3
May	29	10	19	0.5	34.5	65.5	0.09	2.79
Jun	95	40	55	0.7	42.1	57.9	0.12	2.36
Jul	80	12	68	0.2	15.0	85.0	0	39.2
Aug	110	14	96	0.1	12.7	87.3	0	51.8
Sep	59	23	36	0.6	39.0	61.0	0.09	2.86
Oct	40	27	13	2.1	67.5	32.5	0.2	1.28
Nov	63	39	24	1.6	61.9	38.1	0.05	3.57
Dec	60	42	18	2.3	70.0	30.0	0.02	9.6
Total	**716**	**290**	**426**					

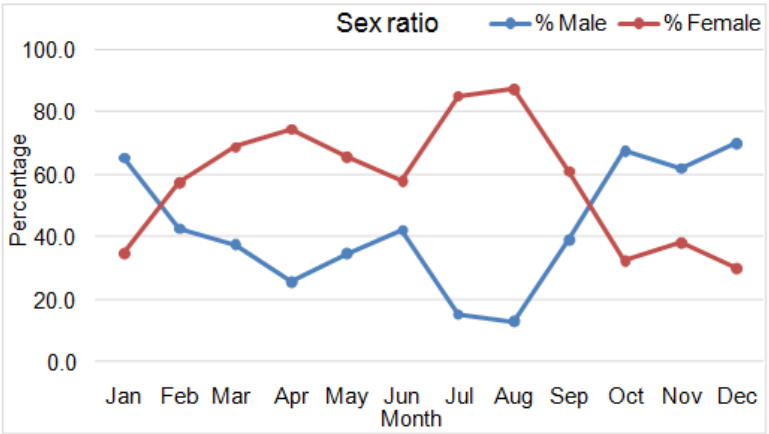

Fig. 2: Monthly variation in the sex ratio of male and female *M. flavolineatus* from the Red Sea.

Gonado-somatic index (GSI):

Gonado Somatic Indexofmales ranged from the lowest value in 0.02 in December to the highest value of 1.00 both in April and June. It started to increase in April with GSI value of 1.00 then decreased in May with a value of 0.59. Gonado-somatic index for females had two obvious peaks in June (3.14) and August (3.16), the value of GIS was also high in July (1.44) (Fig. 3).

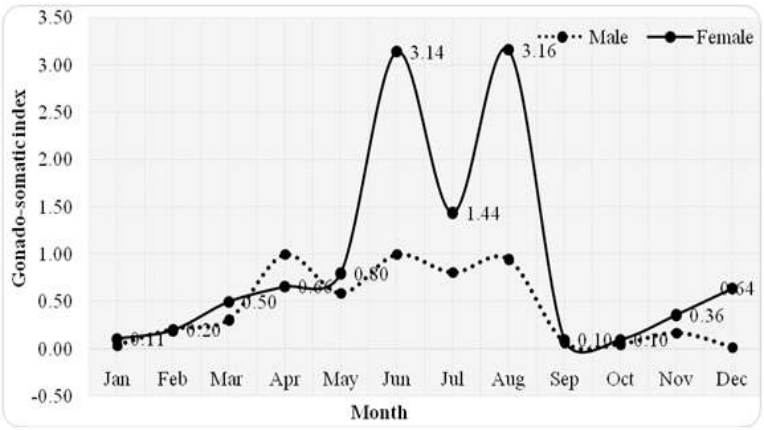

Fig. 3. Gonado-somatic index of male and female yellow-striped goatfish

Sexual maturity

Size at first sexual maturity

The smallest mature male had a TL of 11.5 cm and the smallest mature female had a TL of 12.5 cm. In general, there was a difference in the length at which 50% of the male and female of *M. flavolineatus* reach the sexual maturity. Male reach the first sexual maturity at 12.5 cm (Fig.4) whereas females reach sexual maturity at 14.5 cm (Fig.5).

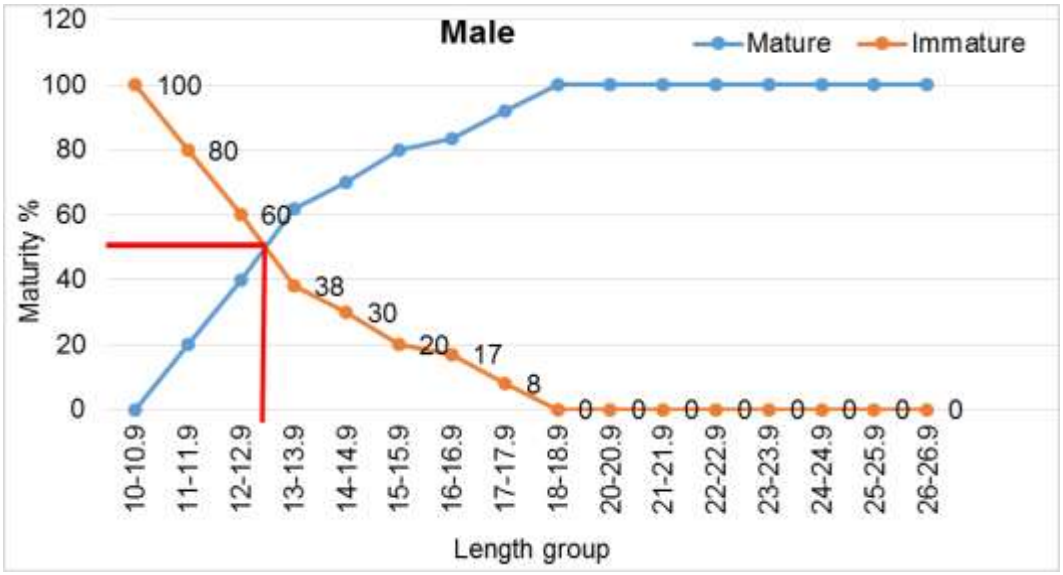

Fig.4: Size at first maturity of male *M. flavolineatus*

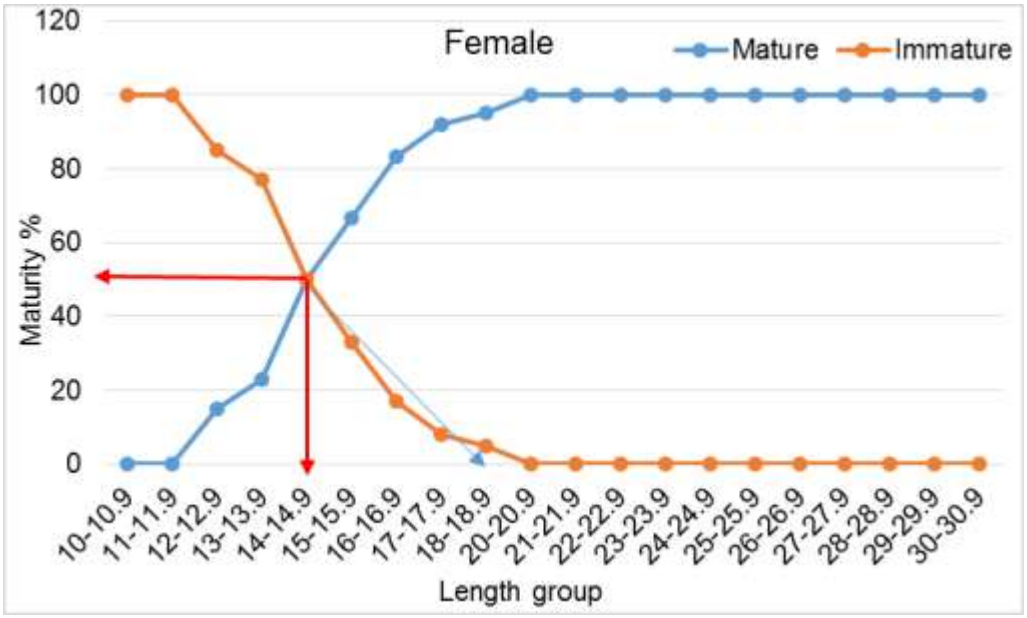

Fig.5: Size at first maturity of female *M. flavolineatus*

Maturation cycle of *M. flavolineatus*

Both male and female specimens could be divided into five groups, immature, maturing, mature, and ripe and spent according to maturation of gonads. Maturing male occurred in April and May with 20% and 40% respectively. Males collected in June were either mature or ripe whereas most of fish in July were mature forming about 70% of all fish collected in July. The ripe male dominated in August constituting about 80% of all fish (Fig.6).

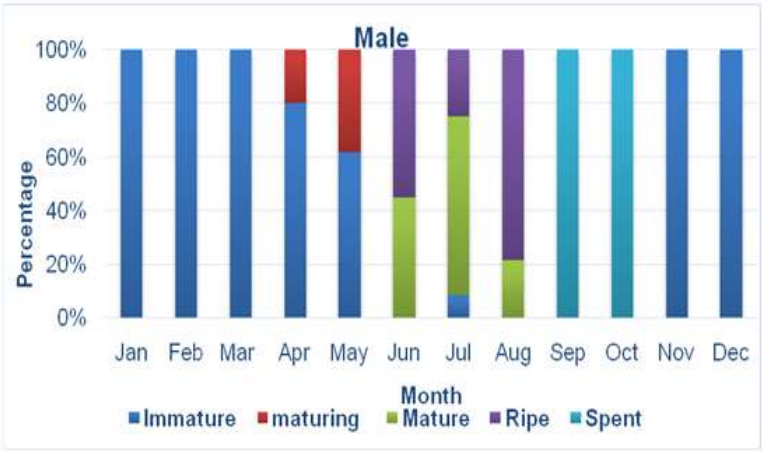

Fig. 6: Percentage contribution of different maturity stages of male *M. flavolineatus*

The maturing female was found in May with the ovary small, and tube like occupying a very small portion of the abdomen. Eggs were not invisible at all by the naked eye. Maturing male fish started to appear in and May where they formed 48 % and 28% of all fish respectively. Maturing male fish started to appear in and May where they formed 48 % and 28% of all fish respectively. The mature and ripe stages were recorded in June, July and August (Fig. 7). Eggs are very large and can be distinguished by naked eyes. Ripe fish started to appear in June where they formed

75% of all fish collected in this month. About 60% of fish collected in August were ripe. About 75% of fish collected in July were mature whereas, ripe fish were very few (1%). The spent stage begins from September where the gonads were shrunk. In the period from October to March, gonads cannot be distinguished at all by naked eyes. It can be concluded that *M. flavolineatus* spawns from June to August (Fig.7).

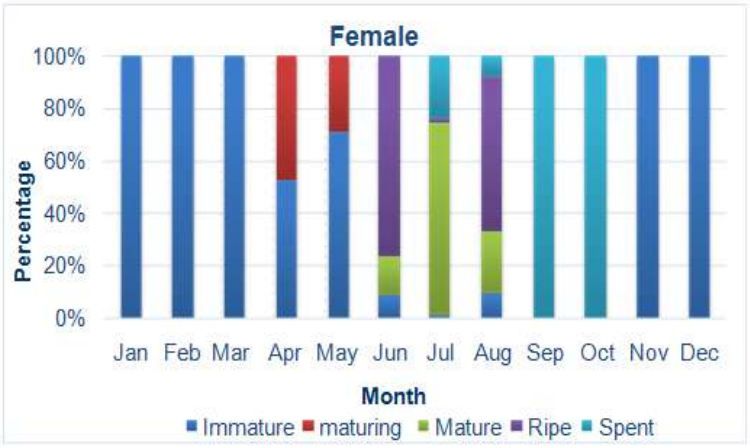

Fig.7: Percentage contribution of different maturity stages of female *M. flavolineatus*

Ripe fish were collected in June, July and August with a peaks in June and August (Figs.8 & 9).

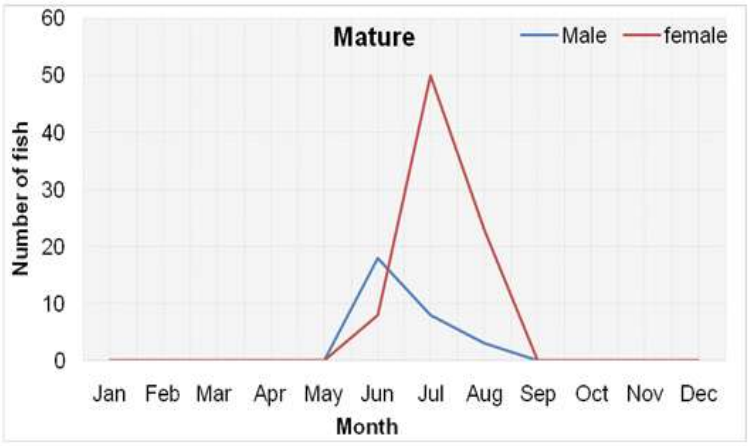

Fig. 8: Monthly variation in the numbers of mature fish over the whole year.

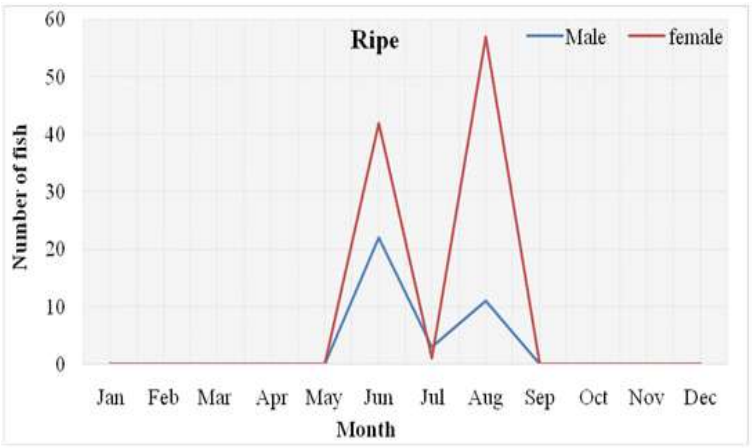

Fig. 9: Monthly variation in numbers of ripe fish over the whole year.

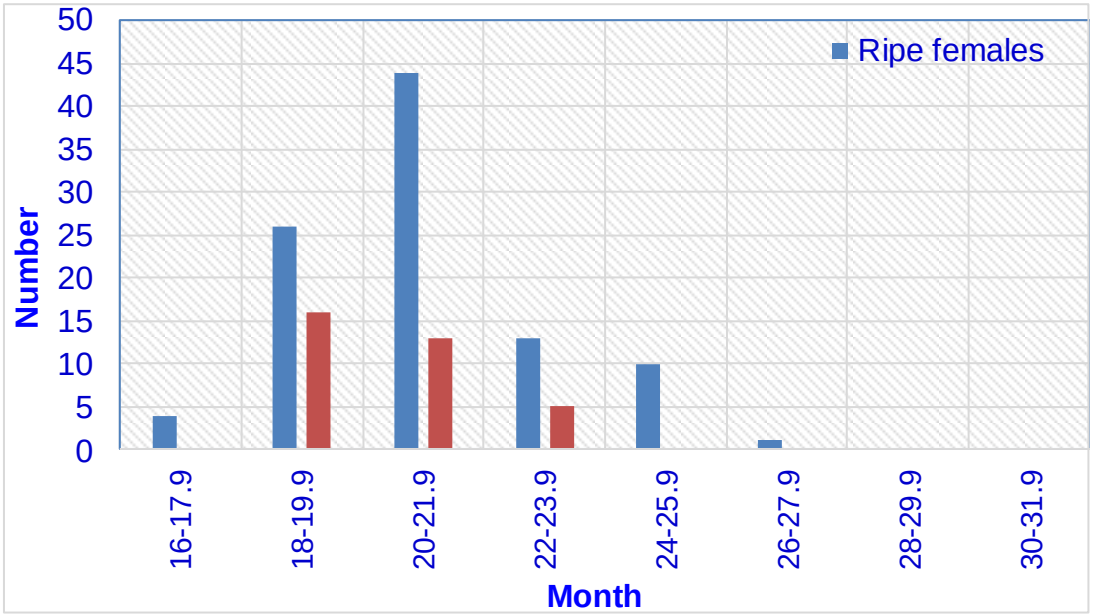

Fig. 10: Frequency distribution of the ripe females of *M. flavolineatus* in the Red Sea

Fecundity:

Fecundity *of M. flavolineatus* was determined for ripe female collected in the period from June to August. The absolute fecundity ranged from 60,000 eggs to 112,000 eggs.

The mean absolute fecundity ranged from 41,333 eggs at a TL of 16 cm to 128,000 eggs at a TL of 26 cm (Fig. 11). This relationship was represented by the equation

$$\mathbf{y= 23747x\ R^2=0.96.}$$

The absolute fecundity increased with the length of the females. The relative fecundity of *Mulloidichthys flavolineatus* varied from 2560 eggs/female for TL range from 16 to 18 cm to 7720 eggs/female for the TL range of 22-24 cm.

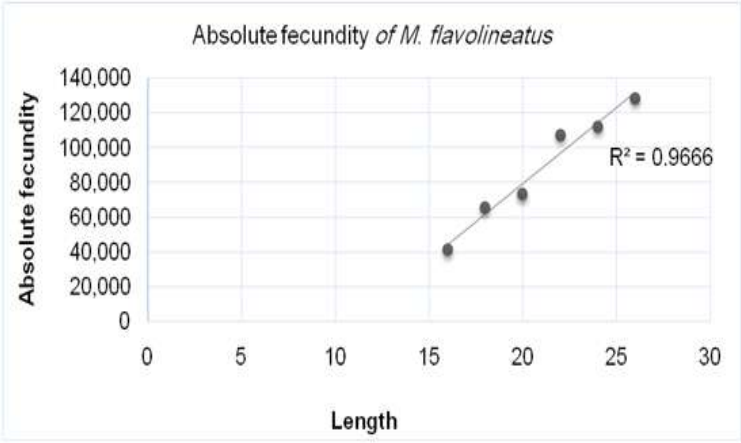

Fig. 11: Relation between fish length and fecundity of *M. flavolineatus* in the Red Sea

Egg diameter

Eggs of *Mulloidichthys flavolineatus*are spherical and small ranging in size from 0.6 mm to 0.9 mm with an average egg diameter of 0.8 mm. The maximum egg

diameter of 0.9 mm was observed in June and August. Most of the eggs vary in size from 0.8 to 0.99 constituting about 70% of all eggs investigated (Fig.12).

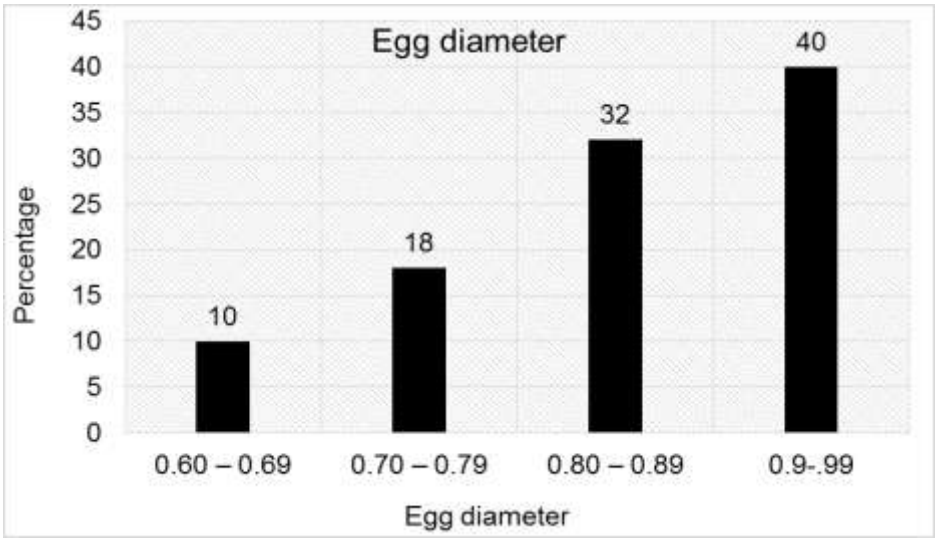

Fig. 12: Percentage of different egg diameters of *M. flavolineatus*.

Abundance of Fish larvae

A total of 3970fish larvae were collected from all sites over the year of which 750 larvae ofconstituting19% of all larvaebelong to the yellowstripegoatfish *Mulloidichthys flavolineatus* (Fig. 13).

The overall density of fish larvae recorded was 191 larvae/1000 m^3 water and the average density was 184 larvae/1000 m^3.

Mullid larvae were taken from 16% of all hauls (69 of 432 hauls). It occurred in all sites with the maximum densityof 156larvae/1000 m^3forming 19% inArabia station 1 followed by Marine Biological Station where 110 larvae forming 14.6% of all mullid larvae were collected……..(Fig.14). The lowest abundance of fish larvae of *M. flavolineatus* was recorded in MGW2 where 19 larvae were collected. In general larvae were abundant in Arabia and Marine Biological Station and were less abundant in Magawish (Fig. 14).

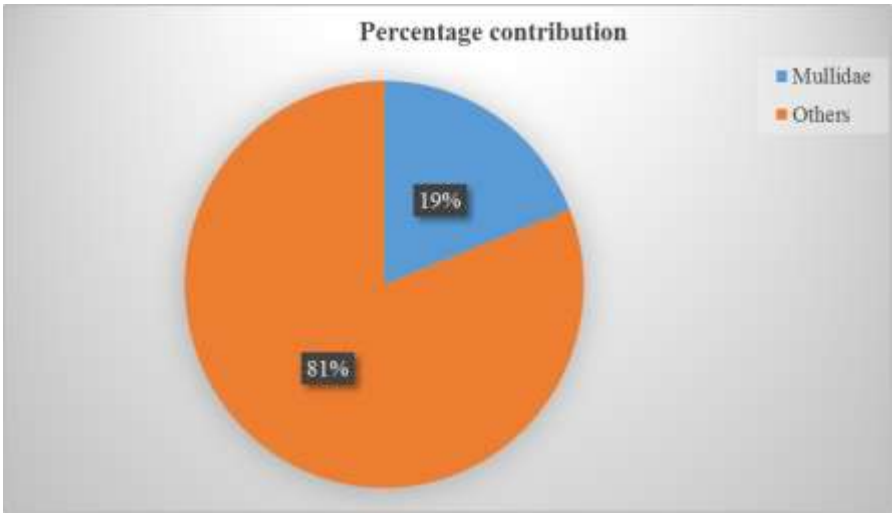

Fig. 13: Percentage contribution of mullid larvae in the fish larvae community

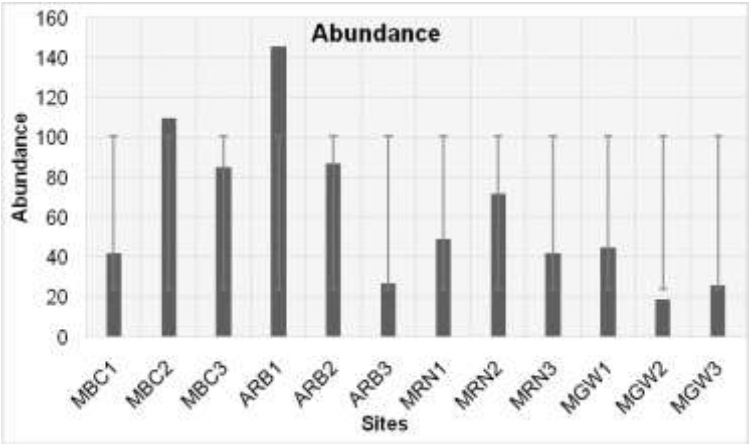

Fig. 14: Abundance of mullid larvae at all stations

Larvae were restricted to the warmer months of the year, June, July and August. The highest density was recorded in August where 156 larvae/1000m^3 (Fig. 15).

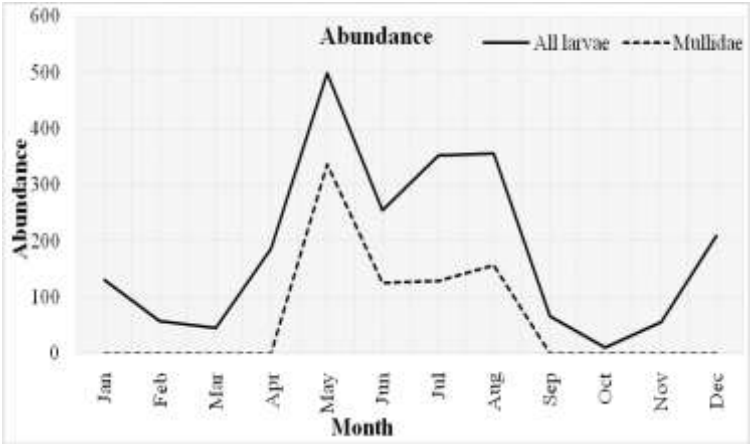

Fig. 15: Monthly variations of abundance of mullid larvae

Length frequency distribution

The total length of the larvae ranged from 1.8 to 8 mm. Of the 750 larvae taken during the current study 683 larvae were small and preflexion constituting about 85% of all larvae. Whereasthe postflexion larvae (larger than 4.5mm) formed ١٥% of all larvae. Along with size groups II and III, the first group formed the bulk of the preflexion larvae. In June, the all larvaecollected were in preflexion of which 223 larvae (97% of mullid larvae in June) are smaller than 2mm. larvae were divided into 8 size classes. Larvae in June belong to size classes I and III ranging in size from 1.8mm to 3 mm. The newly hatched larvae (group I) formed 97% of larvae collected in June.

In July, 233 larvae were collected most of which were smaller than 4 mm and in the preflexion stage forming 90% of all larvae collected in this month (Fig.16). Four size groups (I, II, III, IV) were represented in July of which the group II (2-2.9mm) peaked constituting 52% of all larvae taken in July. Postflexion larvae started to appear in July where 23 larvae were found constituting 10% of all larvae taken in July. The size ranged from 1.8mm to 4.9mm.

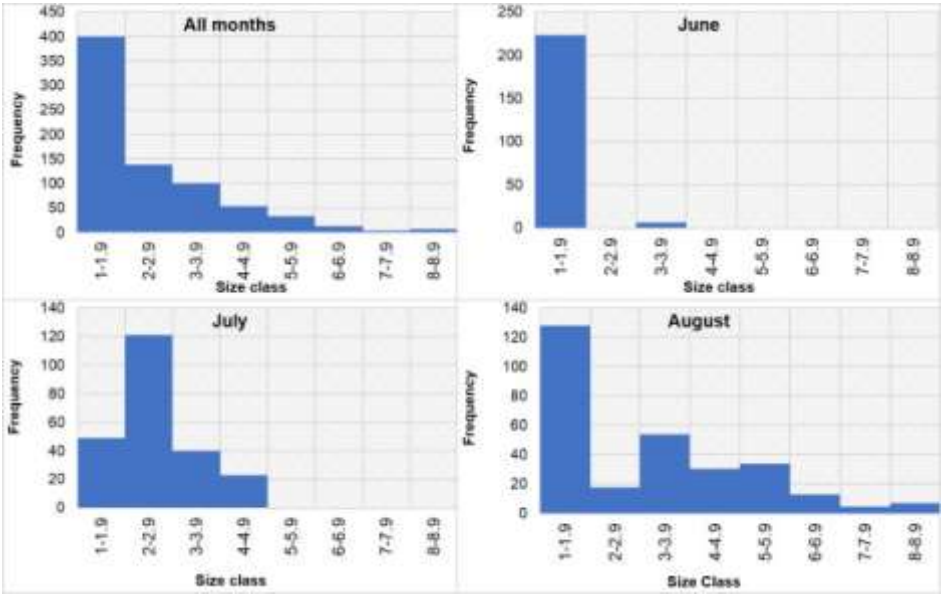

Fig. 16. Length frequency distribution of larvae of *M. flavolineatus*

In August, all size groups were represented in the collection. However, the first size group (1-1.9mm) dominated constituting44% of all larvae collected in this month. Number of prefelxion larvae decreased in August compared to those of June and July where 199 preflexion larvae constituting 70% were taken. Most of the larvae taken in August were new hatching and small preflexion larvae where they formed 70% of larvae taken in this month. The larger postflexion (> 4mm) larvae formed about 30% of all larvae in August

The large number of newly hatched larvae (1.8mm) in July and August indicated the spawning season at this time.The size of the collected larval fishes ranged between 1.8mm and 8mm. The smallest larvae (1.8mm) were preflexion with elongate body and elongate gut forming 44% of the total body length. They lack all fins but have finfolds and fin bud instead. Larvae have a very characteristic pigmentation pattern with pigment on the dorsal surface of the gut and small melanophores along the ventral midline of the tail. The brain has three melanophores

in a triangular pattern on the midbrain. The collected larvae ranged in size from 1.8mm to 8mm. About 72% of the collected larvae were small and preflexion ranging in size from 1.8 to 4mm (Fig.16).

DISCUSSION

The yellow-striped goatfish*Mulloidichthys flavolineatus* is widely distributed throughout the Indo-Pacific region. In the Red Sea, the species is one of several mullid species that are important food fishes and its juvenile isused as bait. Despite their commercial importance and widespread distribution, very little is known about their early life history in the area. The species attracted the attention of scientist since earlier times (Alkholy, 1964; Boraey, 1969). Larval development of the family Mullidae was studied in Sharm El-Sheikh (Abu El-Regal, 1999) and Hurghada (Abu El-Regal, 2008).

In general, females *Mulloidichthys flavolineatus* dominated the catch during the sampling period as the overall female's ratio was calculated to be 0.6. These results indicate that females were more vulnerable to fishermen than males. During the spawning seasons from June to August, the number of female fish increases to 6 times of male's number. This high value of female's ratio may refer to the reproduction strategy of the species (Thresher, 1984).

The variation in the gonado-somatic index indicated that *M. flavolineatus* spawn in the warmer months of the year from June to August. These results were confirmed by the ichthyoplankton survey in the Red Sea. During his survey on the ichthyoplankton in the Gulf of Aqaba, Abu El-Regal (1999) recorded large number of larval stages of goatfishes. Unfortunately, larvae could not be identified below the family level. However, two peaks of spawning of the goatfishes were recognized; the first period was from June to August and the second peak was found from December through February. The spatial and temporal distribution of the family was studied at Hurghada and was found to be abundant in the in winter and summer (Abu El-Regal et al., 2014a, b; Maaty, 2015; Abu El-Regal, 2017b). Larvae of dominated the larval community in the period from May to August whereas larvae of another mullid species; *Parupeneus rubescens* was collected in large numbers in winter(Abu El-Regal et al, 2017). The current study focused on the yellow-striped goatfish, *Mulloidichthys flavolineatus*as it constituted the main part of the catch of this family in the Red Sea. It forms a considerable part of the commercial catch in Hurghada where they formed more than 10% of the total catch in 2015 (GAFRD, 2016).

Mulloidichthys flavolineatus seems to have one peak of spawning from June to August as indicated by the gonado-somatic index analysis (Fig.7) and abundance of fish larvae (Fig.9). Previous studies on the developmental stages of *M. flavolineatus* larvae showed that larvae smaller than 4 mm were in the preflexion stage (Abu El-Regal, 1999; Abu El-Regal, 2008).

Data about the spawning season of *M. flavolineatus* is in agreement with the very few reports about the species either globally (Munro *et al*., 1973; Houde *et al*., 1986) or locally in the Red Sea (Boraey, 1969). In the Arabian Gulf, mullid larvae were abundant from late spring to autumn and were absent in winter (Houde *et al*., 1986). In the Caribbean Sea, ripe females were caught in March and April (Munro *et al*., 1973). Boraey (1969) studied the reproductive and feeding biology of the species in Hurghada and stated that the species started the spawning season early in May till August.

The smallest size of *M. flavolineatus* recorded during the current study was in January indicating the addition of new recruitment at this time.

There was a dramatic decrease in goatfish catch in Hurghada from 9.4% of the total catch to in 1990 to 0.2% in 2000 because fishing activities are concentrated in the period from May through August at the time of goat fish migrations. This is the time of spawning for *M. flavolineatus.* One purpose of this study is the determination of the spawning season of the species to help manage fisheries of the species in the Red Sea.

Schools of *M. flavolineatus* occur in consistent locations at daytime and disperse at night (Al-Kholy, 1964; Boraey, 1969; Hobson, 1974; Holland, *et al*., 1993) to forage polychaetes and small crustaceans that predominate in its diet (Sorden, 1982). Many fish species, both coastal (Hobson, 1965; Gruber *et al*, 1988) and pelagic (Yuen, 1970; Holland *et aI.*, 1990), have spatially separated daytime and nighttime ranges.

Mulloidichthys flavolineatus migrates from the deep water and coral reef areas to shallow water along the shore at the time the water temperature rises and invertebrate food organisms are abundant (Boraey, 1965). This migration occurs in May through August to specific places that fishermen keep. It has been noted that the migration follows the lunar cycle and that they generally start at the full moon.

Catch of the yellow-striped goatfish is suffering from drastic decline in the last twenty years due to the concentration of fishing activities during the spawning aggregation.

Bayoumi (1972) mentioned that *Mulloidichthys flavolineatus* is a seasonal fish that spends most of the year hidden among corals and frequent the shallow coastal waters during summer months from May-August. *Mullus surmuletus* spawns in the spring in the Mediterranean Sea (Amin et al. 2016).

A tag-recapture experiment carried out in the in Kaneohe Bay, Oahu, Hawaii pointed to a high degree of site-fidelity *M. flavolineatus*. More than 90% of tagged-recaptured fish occurred at the release site, with times at liberty of up to 531 day indicates very little emigration from the home range, which was probably the recruitment site of the population as small juveniles.

This strong site fidelity bodes well for using marine conservation zones to protect populations of reproductive size adults. Their principal contribution would be in supplying eggs and larvae to adjacent reef areas because there appears to be very little post-recruitment movement of the adults from their home range. The possible exception to this would be during spawning migrations. Eggs and larvae of this species were completely absent from ichthyoplankton samples from the coastal areas despite the extensive sampling program. Instead, all mulled larvae were collected from the outer reefs from Giftun Islands.

These results showed the importance of the ichthyoplankton survey in the determination of the precise spawning season of the commercially important fishes (Smith and Richardson, 1977; Kendall *et al*., 1984; Armstrong *et al*., 2001; Fuiman, 2002).

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ARABIC SUMMARY

بيولوجيا التكاثر لسمكة البربونى ***Mulloidichthys flavolineatus*** بالبحر الاحمر، مصر.

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تم دراسة مجتمع الأسماك من نوع البربونى ذى الخط الأصفرلتحديد موسم التبويض بمنطقة الغردقة بالبحر الأحمر بمصر بهدف تنظيم مصايد هذا النوع. وقد تم جمع الأسماك البالغة من منطقة الغردقة باستخدام شباك خيشومية خلال الفترة من يناير حتى ديسمبر ٢٠١٥. تم تشريح العينات وتحديد نسبة الذكور للإناث والخصوبة ومراحل النضج وحجم البويضات وكذلك معامل وزن المناسل/الجسم. تم جمع يرقات الأسماك باستخدام شباك بلانكتون بصورة شهرية خلال الفترة من يناير حتى ديسمبر ٢٠١٥. وجد أن أعلى معامل للمناسل/الجسم سجلت فى الفترة من يونيو حتى أغسطس كما وجد أن أعلى كثافة ليرقات هذا النوع قد وجد أيضاً فى شهور يونيو ويوليو وأغسطس وهذه النتائج تتفق مع النتائج التى تم تسجيلها باستخدام معاملات بيولوجيا التكاثر. وهذا يدل على امكانية استخدام يرقات الأسماك فى تحديد مواسم التبويض.