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Reproductive biology of the Lessepsian migrant shrimp scad *Alepes djedaba* (Forsskal, 1775) from the Egyptian Mediterranean Sea coast.

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ABSTRACT

The shrimp sacd, Alepes djedaba is a dominant species of genus Alepes in the purse seine catch of Damietta and Port Said landing sites on the Egyptian Mediterranean Sea. It represents a commercial species in Mediterranean Sea fisheries. A total of 718 specimens were collected from January to December 2019. The sex ratio M: F was 1.0:1.24 as the total sample contained 321 males and 397 females. The P-value of the chi-square test showed a highly significant difference between both sexes. Macroscopic and histological examination of gonads and monthly gonado-somatic index distribution revealed that A. djedaba has a prolonged spawning season extending from April to November with a peak during May. The lengths at first maturity were 16.0 and 16.7 cm for males and females respectively. A. djedaba is distinguished by high fecundity as the absolute fecundity ranged from 31680 to 336675 with an average of 134940.6 eggs and the relative fecundity ranged from 980.8 to 2683.9 with an average of 2152.2 oocytes g^{-1}

INTRODUCTION

Family carangidae is one of the most commercial families in the Egyptian Mediterranean Sea sector. It comprises 140 species in 32 genera of worldwide distribution (Nelson, 2006). *Alepes djedaba* (Forsskål, 1775) is a pelagic member of the family Carangidae that has a widespread distribution in tropical and warm temperate areas between 40°N and 47°S, of the Indo-Pacific, Red Sea and East Africa, and immigrant to the eastern Mediterranean (Levant coast) (Golani, 1998). Other studies by Kuthalingam (1955) and Taskavak and Bilecenoglu (2001) showed that *A. djedaba* is one of the species involved in the Lessepsian migration and inhabits the east coast of the Mediterranean Sea around Lebanon and Egypt.

It is a very important species for commercial fisheries and some studies on its fisheries. The length-weight relationships were described by Raje (1993) from the Mediterranean coast of Turkey. Osman and Abdulhadi (2011) reported some aspects of

the reproductive biology of *A. djebaba* in the Arabian Gulf. Studies on the Length-Weight Relationship, Fecundity, Sex-ratio and Gonadal Maturation were reported by Shuaib and Ayub (2011) at the Karachi Fish Harbour, Karachi, Pakistan. Biological aspects and stock evaluation were conducted in the Arabian Gulf by Abd el Barr *et al.*, (2014). Biometrics studies were conducted on *Alepes djedaba* from Semarang waters, Indonesia by Siwat (2016).

In spite of the dominance and suitable price of *A. djedaba* in the Egyptian Mediterranean fisheries, the studies on its biology and fisheries are scarce. This study comes to throw light on the reproductive biology of the shrimp scad. The reproductive biology is a very important item for fishery management. Some reproductive aspects such as sex ratio, maturity stages development, histological examination of the gonds, the length at first maturity, gonado-somatic index and fecundity of *A. djedaba* were investigated for the first time in our Mediterranean sector from purse seine landings in Damietta and Port Said fishing harbor.

MATERIALS AND METHODS

A total of 718 specimens were collected from the Egyptian waters of the Mediterranean Sea from Damietta and Port Said landing sites. Specimens were monthly collected from January to December 2019. Samples were transported to the lab for measurements and dissection. For each specimen, total length to the nearest millimeter, total and gutted weight to the nearest 0.1 gram were measured. The sex were assigned and the maturity stages were recorded. Samples from gonads of all maturity stages were preserved for histological examination. The ripe ovary were preserved in 4% formalin for fecundity study.

Sex ratio was determined as the percentage of males to females (M: F).The variations in sex ratio were estimated monthly and for the different length groups. A chi-square test at 0.05 significance level was computed according to Snedecor (1956).

Maturity stages were determined macroscopically according to El Ganainy (1992) with some modifications reported by Osman (2020) as: I. stage of non-differentiated sex, II. Stage of immature as both tests and ovary have 1/2 abdominal cavity with white tests and reddish ovary, III. Maturing stage when the tests and ovary become 2/3 abdominal cavity with clearly recognizable eggs and sperms, IV. Ripe Stage as both gonads completely filled the abdominal cavity, the tests more compacted and ovary become orange compacted with ripe eggs, V. spawning stage soft and creamy white testes milt oozes out on pressing the gonad and ovary is reddish-yellow in color, opaque mature ova more numerous than maturing ova, and VI. Spent stage as both tests and ovaries become very loose with more blood capillaries.

For histological preparation, ovarian and testicular sections were taken from the middle part of gonads. Sections were fixed in 10 % formalin for 24 hours, then washed with 70%

ethanol followed by several ascending series of ethyl alcohol, then kept in methyl benzoate overnight and Cleaned by two series of xylene (10-15 minutes for each). Samples were then embedded in paraffin wax and supported by paraffin block then sectioned (5-8 mm thickness) and stained using hematoxylin and aqueous solution of eosin 1%. Finally slides were cleared in xylene then examined and photographed using light microscope. Histological stages for ovarian sections were staged according to scale of (Arockiaragj *et al*., 2004) and testicular sections stages were staged according to (Schulz *et al.*, 2010) with some modification.

Gonado-Somatic Index was estimated according to Anderson and Gutreuter (1983):

G.S.I= [Gonad weight (G Wt) /Fish weight (Wt)] × 100

Length at first sexual maturity (L_m) which is the length at which 50% of males and females of each species attain maturity (L_{T50}) was calculated according to White (2007) By the logistic regression equation, where the proportion, P_L, of those shrimp scad that were mature at length L_T was calculated from the following equation:

$$P_L = \{1+e^{(-\ln(19)(Lt-Lt50)(Lt95-Lt50)-1}\}^{-1}$$

Lt50 and Lt95 are Parameters for the logistic regressions fitted to the total length (Lt) at maturity data.

Fecundity was estimated by counting the number of mature ova from a known weight of mature/ripe ovary according to Sujatha *et al.*, (2015).

$$\text{Absolute fecundity} = \frac{\text{(No. of ova in the subsample * total ovary weight)}}{\text{Weight of subsample}}$$

Relative fecundity was obtained by dividing absolute fecundity with the total weight of fish.

The relation of fecundity "F" with total length "TL", total weight "TW" and age was estimated according to the formulas given by Bagenal (1978)

$F = a\ TL^b$; $F = a\ TW^b$; $\text{Log } F = a + b\ T$

Where, a & b are constants, TL is the total length (mm), TW is body weight (g) and T is age of fish.

RESULTS

1.Sex Ratio:

The total collected sample of *Alepes djedaba* from the Mediterranean Sea revealed a sex ratio M/F of 1.0: 1.24, as the total 718 specimens contained 321 males and 397 females.

The chi-square value was 0.004564 (P<.05). This value showed significant difference between both sexes. The percentage of males and females was determined for different length groups and different months (Fig. 1 and 2) which revealed that males outnumbered females in small length groups 11 to13 cm and females is dominated in large length groups 14 to 23cm. but in the larger length groups more than 23 cm females only were represented in the samples.

According to the monthly variation in male and female percentage, females outnumbered males in all months except in April and June where males are dominant (Fig. 2) .

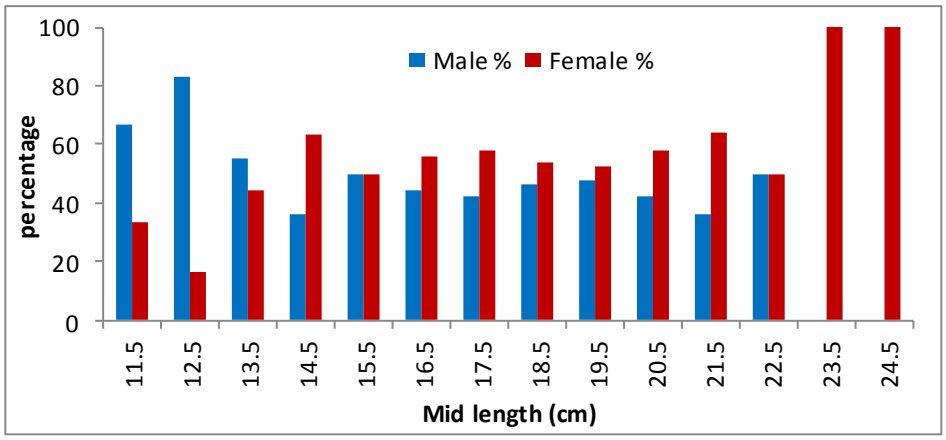

Fig. 1. Sex ratio of *A. djedaba* in relation to length in the Mediterranean Sea

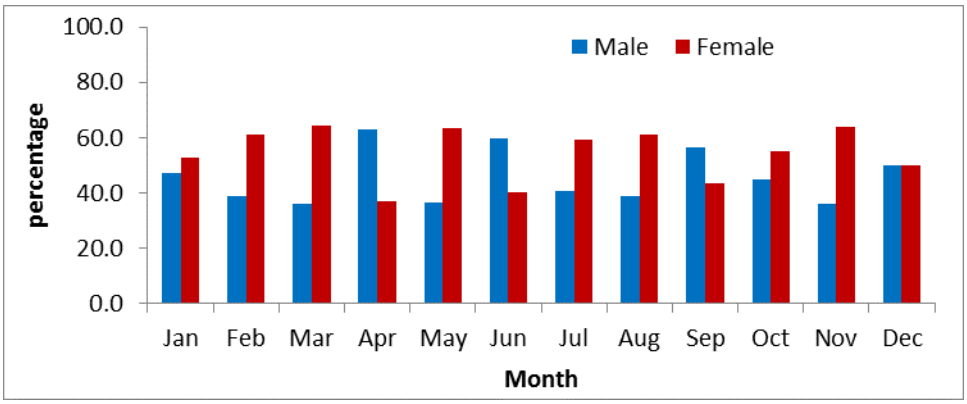

Fig. 2. Monthly variation in sex ratio of *A. djedaba* from the Mediterranean Sea

2.Maturity stages:

2.1.Macroscopic examination:

The results in (Fig. 3) showed that for males, the ripe stage started from April and extended to October with the highest percent of ripening stage during May. The running stage started from May to November. For females as shown in Fig. (4), the ripening started from April to October and the running stage was extended from May to November

with highest value during June. The spent stage were recorded from July to January and then the maturing appeared during December to March. From these results it can be noticed that the spawning season extends from April to November.

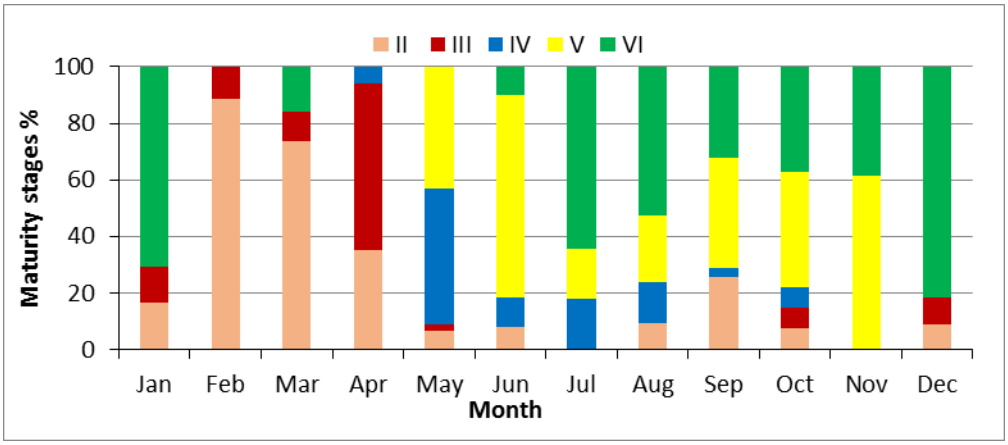

Fig. 3. Monthly distribution of maturity stages for male *A. djedaba*.

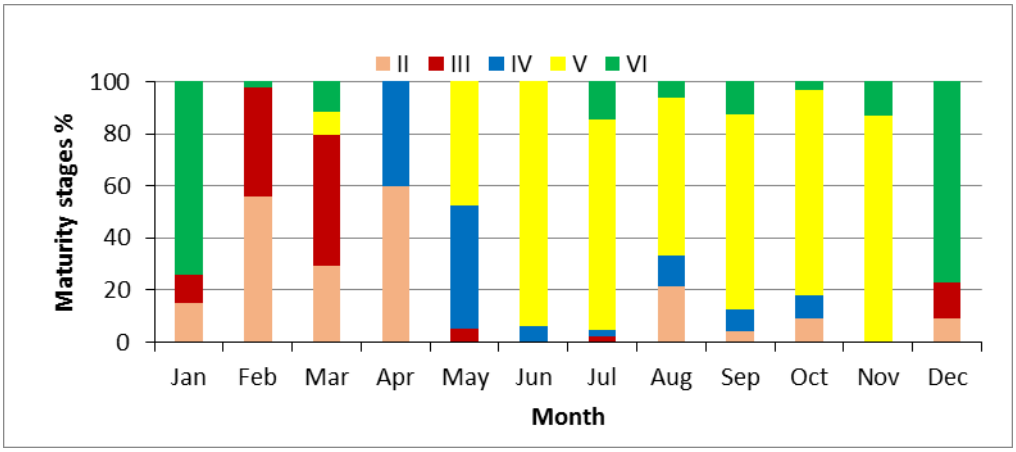

Fig. 4. Monthly distribution of maturity stages for female *A. djedaba*

2.2.Histological examination:

2.2.1.The results of microscopic examination of the ovarian sections showed that:

Ovaries are covered by tunica albuginea (dense connective tissue) and it have follicle which contain the oocytes. The size thickness and crowded of follicle and oocyte changed during different maturity stages (Fig. 5). The first non-differentiated sex sample, then theses stages were recorded

II. (Immature = previtellogenesis): It is a germ native cell and pre nucleus phase. The follicle is characterized by thin membrane and a polygonal oocytes. The oocytes include primary oocytes which is invaginated in the follicle membrane and secondary oocytes which is enlarged in size, leave the follicle membrane and detached to the center. This stage most dominant in the period from January to April.

III. (Maturing = vitellogenesis): It is the lipid deposition stage; the oocytes became more rounded than polygonal (opaque rounded). This stage is the cortical alveoli stage, it is characterized by the presence of small yolk vesicles in the Ooplasm. The cortical alveoli oocytes fill the follicle lumen. As this stage proceeds, the follicle increases in size and the follicle epithelium became more thickened with less space observed in the lumen. The nucleus enlarged and become irregular in shape. This stage is most dominant during April.

IV. (Ripe stage = post vitellogensis): This stage is distinguished by fully grown oocytes with unobserved nucleus and lipid granules (yolk) filled the oocyte ooplasm. The membrane dissolved and lipid and protein fused together to form homogenous appearance. Oocyte became clearly rounded and cytoplasm was hydrated which result in adhesion of yolk granules**.** The vitelline membrane started to disintegrate and the follicle become condensed and reached its maximum size. This stage is most dominant in late April and May.

V. (Spawning = Running stage): It is distinguished by less crowded follicle with spaces in the lumen because of releasing of mature oocytes. The follicle is characterized by informality and irregularity and the newly formed oogonia started to appear. This stage started to appear in the period from May and extended to November.

VI. (post spawning =Spent): This is atresia stage, it characterized by yolk re absorption and the vitelline envelope become disintegrate, fats and lipids were well fused in oocyte and the opening in the outer area of vitelline membrane is appeared. The follicle had a messy appearance. This stage started from July and its peak during December and January.

2.2.2.The results of microscopic examination of testicular sections showed that:

The tests of *A. dejedaba* covered by tunica albuginea (dense connective tissue) which extended inside the tests to form septum, in developing stage this tunica albuginea is covered by mesothelium. The testes composed of seminiferous tubules; it is consisting of germinate cysts which contain germinal cells. The stages of development can be classified as follow (Fig. 6).

II. Immature stage: This stage is characterized by nests of spermatogonia (**A;** which found solitary under tunica albuginea close to periphery of testicular membrane and **B;** which found deeper in the testicular tube). This stage characterized by faintly stained cytoplasm and nuclei of small spermatogonia as well as large spermatogonia (B) which is conspicuous boundaries and nuclear outline with dark stained nucleus. This stage is dominant in the period from January to April.

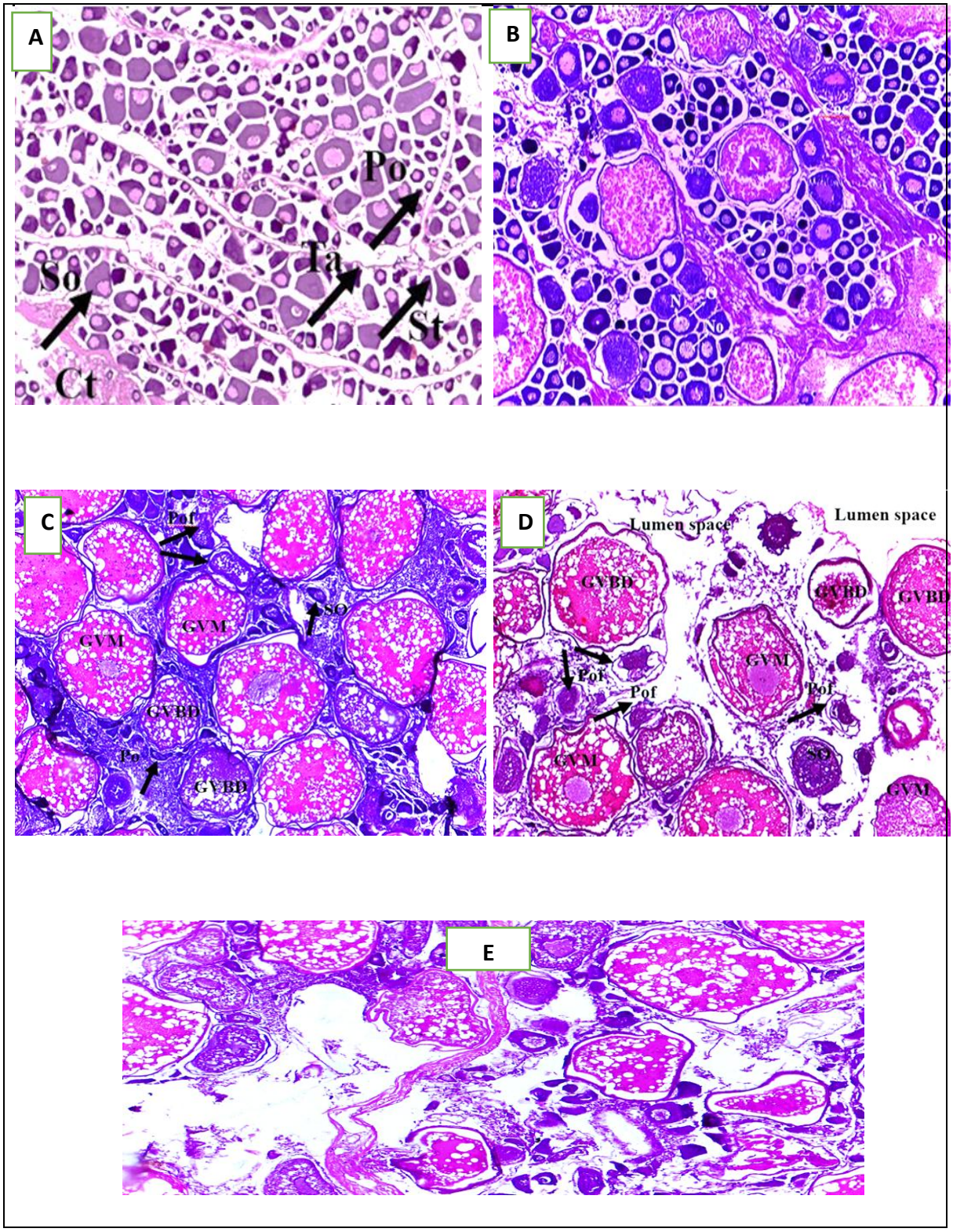

Fig. 5. A- immature, B -maturing, C- ripe, D -spawning, and E -spent female *of A. djedaba* as **Po**-primary oocyte, **GVBD**-germinal vesicle breakdown, **CA**-cortical alveolar oocyte, **N** –nucleus, **O**-ooplasm, **Ne**-nucleolus, **POF**- postovulatory follicles and **GVM**- germinal vesicle migration.

III. Maturing stage: The maturing stage characterized by nests of spermatogonia, spermatocytes and few spermatids. Spermatocytes classified into primary (smaller than spermatogonia with spherical nuclei and deeply stained chromatin) and secondary (highly smaller and characterized by large nuclei occupies most of cell, surrounded by thin cytoplasm). At the end of this stage nests of primary and secondary spermatocytes , spermatids as well as few cells of well-developed spermatozoa appeared. Spermatids were found in cysts and noticed as small cells with scantly cytoplasm and dense spherical nuclei. This stage is most dominant during February and March.

IV. Ripe = mature :The seminiferous tubules in this stage showed a fair quantity of spermatozoa and fewer amount of spermatids. The tubules is very crowded and the mature spermatozoa is elongated cell composed of head and tail. The lumen of follicle showed no spaces. This stage is dominated during April, May and June.

V. Runing = spawning : This stage is characterized by discharge of a considerable number of sperms. This accompanied by decrease in size of seminiferous tubules and increases in spaces among follicles. This stage was dominated and extended in the period from May to November.

VI. Spent stage : The tubular lumen contain a residual sperms after spawning and vacuoles appeared in the sertoli cell. Phagocytic cells appeared in close contact of basement membrane of sertoli cells. The lyding cells were less and smaller in size. The lobules were distorted and vacuolated, they contain the remains of non-discharged spermatozoa. At the end of this stage the follicle showed messy appearance and new generation of spermatogonia were formed. This stage started to appear in July and most dominated during December and January.

3.Gonado- Somatic Index (GSI):

The weight of gonad in comparison to total weight was used to obtain the gonad-somatic index. By studying the GSI for shrimp scad from the Mediterranean Sea, there were noticed differences between the values of GSI for males and females and this is because the weight of testes is less than the weight of ovaries. The highest values of GSI were recorded during May and June for females and males but the lowest values was during January and February for females and during January to March for males (Fig. 7). These results indicated that the spawning season of *A. djedaba* in the Mediterranean Sea is a prolonged spawning season extends from April to November with two beaks in May and October.

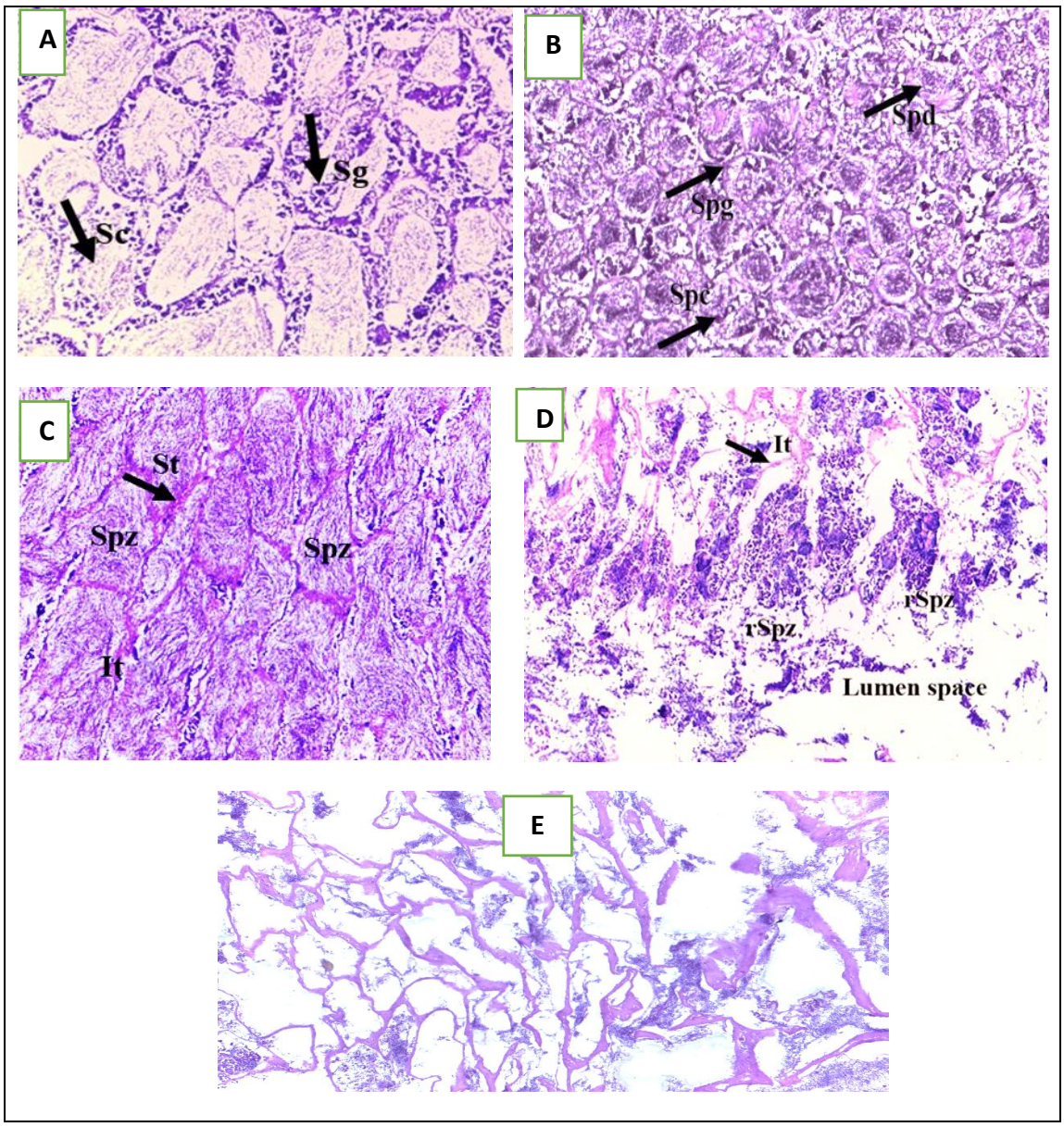

Fig. 6. a- immature, b-maturing, c-ripe, d-spawning and e-spent male of *A. djedaba*. As SPg- Spermatogonia, Spc- spermatocytes, Spz- spermatozoa, St- testicular membrane and It- inertial tissue.

4. Length at first sexual maturity:

Length at first maturity of *A. djedaba* which is the length at which 50% of the fish become mature was studied by grouping both males and females specimens in different length group into mature and immature individuals as shown in (Figs. 8A, and B.), length at first sexual maturity for both *A. djedaba* female and male was estimated at 16.7 and 16.0 cm respectively. These results indicated that all individuals of *A. djedaba* in the Mediterranean Sea over one year of age are sexually mature.

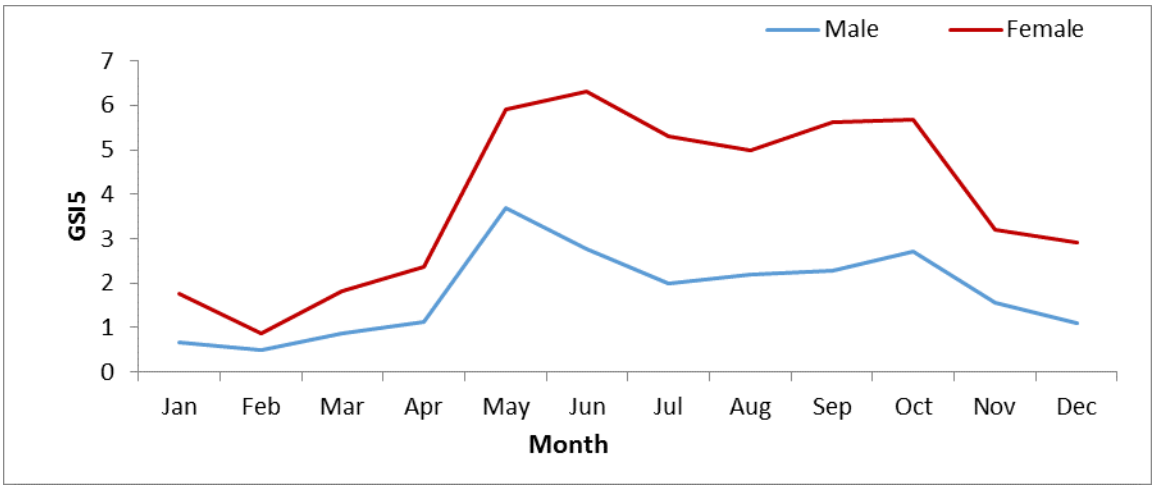

Fig.7. gonad-somatic index for males and females *A. djedaba* from the Mediterranean Sea in the period from January to December 2019.

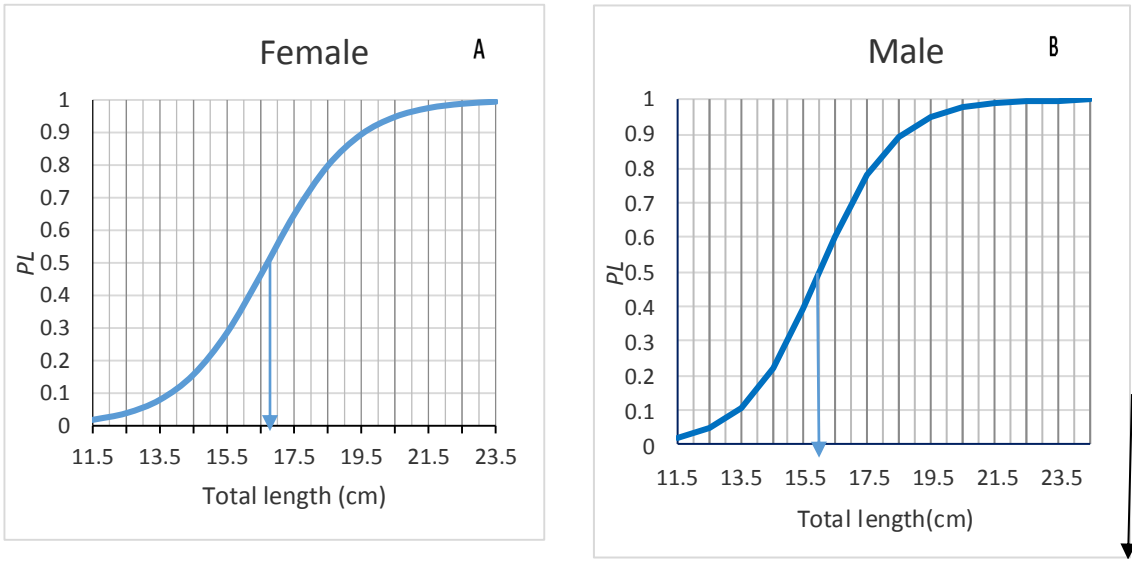

Fig. (8, A, B). Length at first sexual maturity for male and females of *A. djedaba* from the Mediterranean Sea.

5. Fecundity

5.1.Absolute and relative fecundity:

In *A. djedaba*, the absolute fecundity ranged from 31680 egg in 16.5 cm total length ripe individual to 336675 egg in 23.0 cm total length ripe sample. The average of absolute fecundity was 134940.6 egg.

The relative fecundity ranged from 980.8 to 2683.9 oocytes g^{-1}. With an average of 2152.1 oocytes g^{-1}.

5.2. Absolute fecundity-total length relationship

There were a positive relationship between absolute fecundity and total fish length of *A. djedaba.* The absolute fecundity ranged from 31680 egg to 336675 egg with length interval from 16.5 to 23 cm. The regression equation for the absolute fecundity and fish length as in (Fig. 9) was:

$$Log\ F_{abs}=-3.0413+6.396\ log\ L\ (R^2=0.89).$$

5.3. Absolute fecundity total weight relationship

The relation between absolute fecundity and gutted weight is also positive as the range of absolute fecundity increase by increasing growth in weight (Fig. 10). The regression equation can be expressed as: $(Log)F_{abs}=2.321+1.569\ log\ (W)$ ($R^2=0.83$).

5.4. Absolute fecundity and age relationship:

The relation between absolute fecundity and age of fish is also positive as the absolute fecundity increases with increasing the age (Fig. 11). The regression equation can be expressed as: $(Log)F_{abs}=4.6088+1.5466\ log\ (A)$ ($R^2=0.99$).

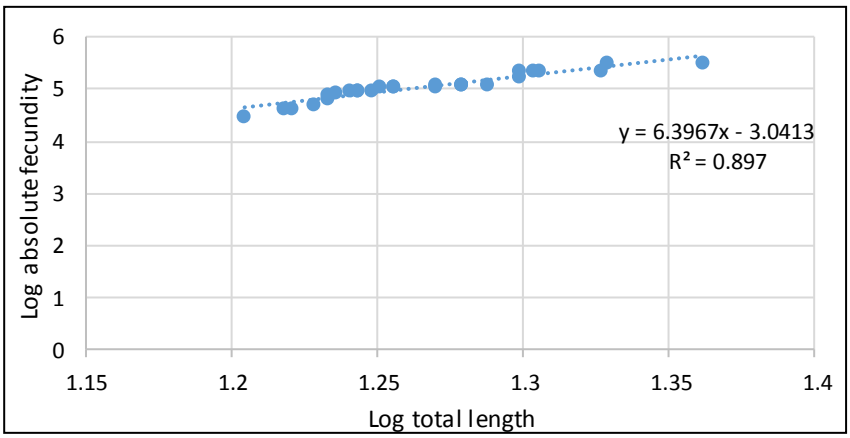

Fig. 9. logarithmic Relation between absolute fecundity and total length of *A. djedaba* from the Mediterranean Sea.

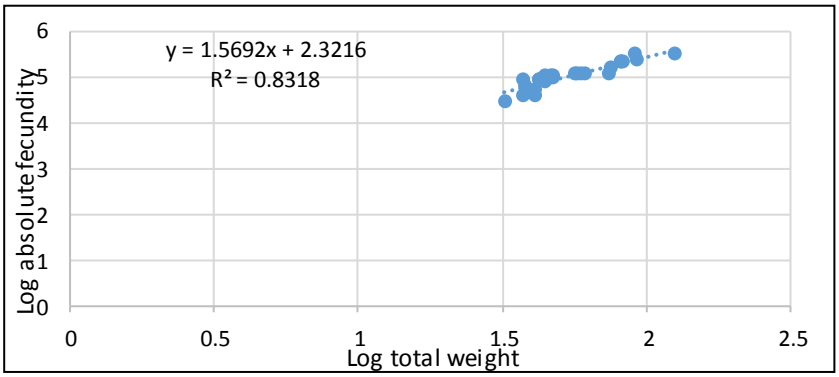

Fig. 10. logarithmic Relation between absolute fecundity and gutted weight of *A. djedaba* from the Mediterranean Sea.

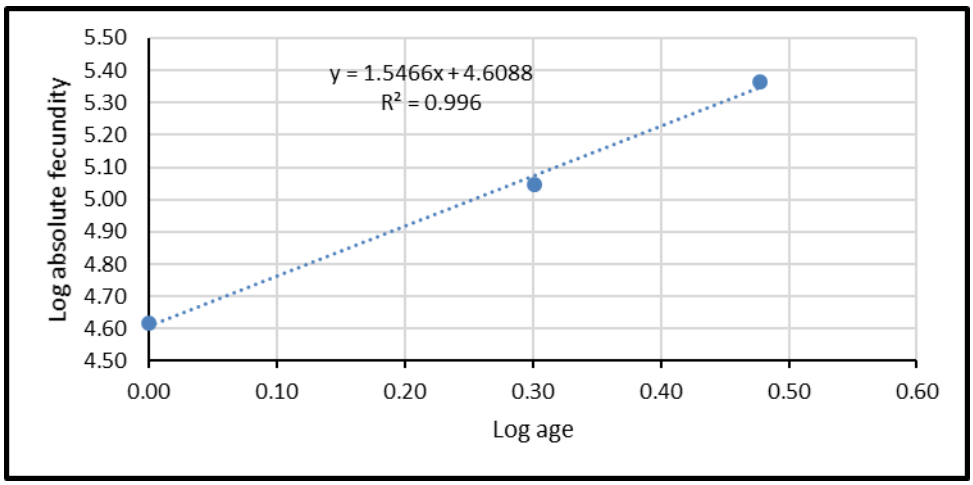

Fig. 11. logarithmic Relation between absolute fecundity and age of *A. djedaba* from the Mediterranean Sea.

DISCUSSION

Sex ratio is the ratio of males to females in the stock, which reflect the reproduction potential of the fish. The sex ratio of the shrimp scad population from the Mediterranean was 1.0:1.24 with significant difference between males and females. Shuaib and Ayub (2011) and Sajana *et al*., (2019) showed no deviation from the ideal sex ratio 1:1 of *A. djedaba* in Pakistan and south India. Sivakami (1990) and Raje (1993) reported that males *A. djedaba* were predominant in most of the months in the catches of Cochin, India.

Maturity stages show the degree of ripeness of ovaries and testes. Their determination is important to assess the beginning and ending of the spawning season (Osman *et al.*, 2020). In the current study, ripening of males and females shrimp scad individuals started from April to September, and the running stage started from May to November with highest value during May for males and June for females. The spent stage were recorded from July to January and then the maturing and developing appeared during December to March From these results it can be noticed that the spawning season extends from April to November. The GSI may refers to the gonad index (Batts, 1972) and according to (Wydoski and Cooper,1966) it may give indication of maturity coefficient or maturity index. The maturity stages development as well as GSI index revealed that *A. djedaba* in the Mediterranean Sea have prolonged spawning season with two peaks during May and October. This is agreed with Sajana *et al.* (2019); Raje (1993) ; Reuben *et al.,* (1992); Sivakami, (1990) and Shuaib and Ayub, (2011) who indicated that the spawning season of *Alepes djedaba* were observed almost throughout the year with two peaks of spawning, from March to April and August to December periods.

The size of fish at first sexual maturity (L_m)is a very important parameter for management of species fisheries, according to length at first maturity we can determine

the minimum legal size (MLS) of the fish which don't cause over exploitation of the stock (Osman *et al*., 2020). Length at first sexual maturity for pelagic fishes is usually close to two thirds of the fish maximum total length (Woodhead, 1978). This assumption is in agreement with our results as the maximum recorded size was 24.5 cm and the lengths at first sexual maturity were 16.0 and 16.7 cm for male and female respectively. Theses results indicated that the stock of *A. djedaba* in the Midetereanean Sea is over exploited as the length at first capture reported by El Ganainy *et al*., (2021) was 14.5 cm. Sajana *et al.,* (2019) found that L_m for *A. djedaba* were 19.3 and 17.4 cm for males and females respectively with maximum fish size of about 32cm. These results also agree with Sivakami (1990) and Reuben *et al.,* (1992).

A. djedaba is characterized by high fecundity as the absolute fecundity ranged from 31680 egg in 16.5 cm total length ripe sample to 336675 egg in 23.0 cm total length ripe sample. The average of absolute fecundity is 134940.6 egg. The relative fecundity ranged from 980.8 to 2683.9 oocytes g^{-1} with an average of 2152.1 oocytes g^{-1}. Fecundity was found to be in a positive linear correlation with the total length and weight in agreement with Sajana *et al.,* (2019). The results of this study give adequate information required to fishery management of *A. djedaba* species in the Mediterranean purse seine fishery.

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