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Reproductive Dynamics of Indian Squid, *Uroteuthis duvaucelii* (Cephalopoda: Loliginidae) of the Suez Gulf, Red Sea, Egypt

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ABSTRACT

The overall pattern of the reproduction of the commercially important Indian squid, *Uroteuthis duvaucelii* in the Suez Gulf was described in terms of sex ratio, mantle length at 50% maturity, gonado-somatic index as well as the morphological and histological changes in gonads of both males and females. The overall sex ratio (M: F) was 1.7:1 showing the dominance of males over females during the study period. Males grow to a larger size compared to females (maximum ML= 25.3 and 19.4 cm for males and females, respectively). But, females reach sexual maturity earlier than males ($ML_{50\%}$ = 13.5 and 10 cm ML for males and females, respectively). The gonado-somatic index values increased with the gonad maturation, having 2 beaks during spring (in April for both males and females) and autumn (in November for females and in October for males) and declined thereafter during winter. Thus, the reproductive period occurs frequently throughout the year, especially in warm months. Four stages (I - IV) of maturity were recognized according to their macroscopic appearance. No spent (stage V) individuals were observed morphologically throughout the study. The histological evaluation showed five stages (I – V) of gonadal development in male's testis. While six stages (I – VI) were determined in female's ovary. The appearance of various stages of gonadal development at the same time in most histological slides indicates that the spawning strategy in this species is iteroparous and spawns partially throughout the year. These findings regarding the reproductive dynamics of *U. duvaucelii* in the Suez Gulf are very important for preserving its stock and regulating its fishing strategies.

INTRODUCTION

Investigation of the reproductive traits is essential for the assessment of main population parameters, including the reproductive capacity of the stocks, growth rate, sexual maturation, productivity of generations, life history, etc. Studies involving the reproductive biology of fish include several approach possibilities, such as the understanding of gonadal development or the reproductive cycle of the species. Analyses of gonadal development can be evaluated at macroscopic, microscopic and ultramicroscopic levels. Gonadal development can be analyzed macroscopically and changes in shape, size, color, and texture of the gonads have been used as parameters for the classification of maturation status. However, the most used analysis is that of the microscopic characters, since it allows a more detailed and precise description of the transitions and morphological and structural transformations that happen during gonadal development. Studies on reproductive biology that incorporate the histological examination of gonads are usually suitable to determine the precise duration of the spawning period and whether spawning occurs more than once in a breeding season **(Duarte *et al.*, 2007; Muchlisin, 2014**; **Cassel, 2020)**.

Cephalopoda is a major class of phylum Mollusca; it includes several economically important species. Octopus, squid, cuttlefish, and chambered nautilus are the familiar groups of that class. They are characteristically soft-bodied, large, active, short-lived animals, with complex behavioral and physiological capabilities. They are exclusively marine, occupying a wide range of benthic and pelagic habitats. About 800 living cephalopod species belong to three main groups are represented by different orders. Squids belong to the Order Teuthoidea. There are some 290 species of squids and about 30–40 species with substantial commercial importance. The other main cephalopod groups exploited for food are the cuttlefish and octopus plus to a much lesser extent the sepiolids **(Boyle, 2001**; **Arkhipkin *et al.*, 2015).**

Squids of the family Loliginidae are commonly known as pencil, inshore or neritic squids; they include nearly 12 species and most of them are important species in trophic systems, fisheries and biomedical studies. Species in this family are widely distributed; they are found in very shallow waters in bays and estuaries, over grass flats and coral reefs, and in depths over 700 m; primarily in warm to temperate waters worldwide **(Roper *et al.*, 1984; Hunsicker *et al.*, 2010**; **Jereb & Roper, 2010**).

The Indian squid, *Uroteuthis duvaucelii* (d'Orbigny, 1835), is one of the twelve species of the family Loliginidae. It has several synonyms, while the most frequent ones are *Loligo duvaucelii* and *Photololigo duvaucelii*. It is an Indo-West Pacific species with a wide range that extends from the Red Sea in the Western Indian Ocean to Malaysia and the South China Sea. *U. duvaucelii* is a significant constituent in the commercial fisheries throughout its range (**Jereb & Roper, 2010**; **Arkhipkin *et al.*, 2015)**. This species is a well-known exploited marine resource in the Suez Gulf; usually, it can be found in depths of 15 - 50 m in soft, sandy, and muddy bottoms of the Gulf. It can be caught by different fishing gears, but most abundantly caught by demersal trawlers operating in the Gulf and contributing by about 195 tons (**GAFRD, 2019**). The Indian squid, *U. duvaucelii*, does not only play a key role in the marine food chains since it feeds on fish, crustaceans as well as other mollusks, and are preyed on by larger fish but also serve as a popular source of food for humans **(Navarro *et al.*, 2013)**.

Considerable attention has been paid to the biology of this species, due to its importance in the fisheries. The key aspect of the present study is to provide basic information on the reproductive variables of *U. duvaucelii* at Suez Gulf such as sex ratio, gonado-somatic index, and maturation stages through focusing on the analysis of gonadal development at macroscopic and microscopic levels; the data required for effective management and conservation of its stock.

MATERIALS AND METHODS

1. Sampling of squid specimens

Samples of the Indian squid, *Uroteuthis duvaucelii*, were collected from the Suez Gulf, Red Sea, Egypt. Four fishing ports were considered; namely, Salakhana, Ataka, Ras Gharib, and El-Tor, extending from the North to the South along the Suez Gulf. The cephalopods in the Suez Gulf are mainly caught by commercial bottom trawl nets. Squid samples were collected randomly from commercial trawlers which were found at Ataka fishing port at the Suez Governorate. They were monthly collected during the fishing season (2017/2018) and directly transferredin ice boxes to the laboratory for further investigation.

2. Biometric measurements

The body mantle length (ML) was measured using measuring tape (cm), and total weight (W) was weighed with an electronic balance (sensitive to 0.01 g).

3. Sex determination

Males were distinguished from females primarily by the possession of hectocotylus [a modified arm of the male, functioning as a reproductive organ in transferring spermatophores to sperm receptacle of female] in the left arm IV (Fig. 1). Additionally, sex was determined by dissecting each specimen and examining gonads in the posterior part of the mantle cavity to be hence defined as testis or ovary, as well as determining the presence of Nidamental glands in females (Fig. 2). Notably, sex can be determined easily based on the presence of Nidamental glands even in immature individuals.

Fig. 1. Investigation of the hectocotylus arm of male Indian squid, *U. duvaucelii*, for quick sexual identification

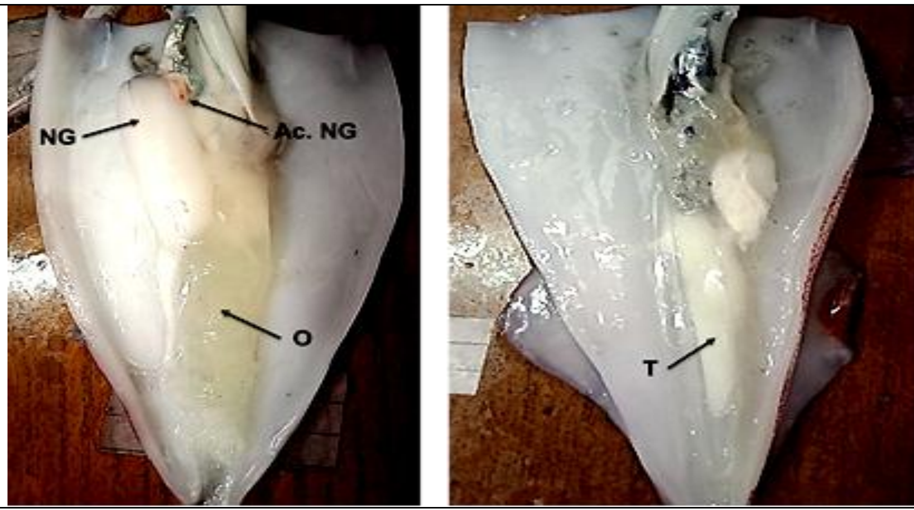

Fig. 2. Photographs of dissected Indian squid, *U. duvaucelii*, showing the internal reproductive organs of for sex determination **(O:** Ovary; **NG:** Nidamental gland; **Ac. NG:** Accessory Nidamental gland**; T:** Testis**).**

4. Sex ratio

The sex ratio; males: females [M: F] was calculated monthly and annually using the following formula: total number of males: total number of females **(Vazzoler, 1996**). The chi-square (χ^2) test was used to verify the existence of significant differences between the sex ratio of *U. duvaucelii* and the commonly expected 1:1 sex ratio.

5. Mantle Length at 50% Maturity ($ML_{50\%}$)

Mantle length at 50%maturity refers to the length at which 50% of individuals in the population were sexually matured. Mantle length at 50%maturity of *U. duvaucelii* was estimated separately for each sex according to **Bakhayokho (1983)** as follows: the mantle length corresponds to 50% mature and fully mature.

6. Gonado-Somatic Index (GSI)

Gonads were removed from each specimen, and their weights were recorded to the nearest 0.001 g. The gonado-somatic index (GSI) was calculated for both sexes following the formula in the study of **Anderson and Neumann (1996**) as follows:

GSI = [Gonad weight (g) / Total body weight (g)] x 100

7. Stages of gonads maturation

Stages of gonads maturation during the reproductive cycle of *U. duvaucelii* were determined macroscopically for all specimens and were ascertained microscopically.

7.1. Macroscopic investigations of maturity stages

The macroscopic assessment (with the naked eye) of each maturity stage throughout the gonadal development in males and females of *U. duvaucelii* in the Suez Gulf during the present investigation was determined based on different criteria described in the study of **Sauer and Lipinski (1990)**. For females, the stages were defined according to 1) the size and color of the ovary; 2) the size and color of the Nidamental glands as well as Accessory Nidamental glands; and 3) the presence of fully mature oocytes in the oviduct (Table 1). On the other hand, male maturity stages were defined on basis of 1) the size and color of the testis; and 2) the development of the Spermatophoric complex {development of Needham's Sac [NS] or Spermatophoric Sac, Spermatophoric organ [SO], and Penis [P]}; and 3) the presence of sperms in the Needham's sac (Table 1).

7.2. Histological investigations of maturity stages

The procedures of gonad histology were carried out according to **Drury and Wallington (1980)**. A subsample of ten specimens was taken from each monthly sample. The gonads were removed and fixed immediately in Bouin's solution (75% saturated picric acid, 25% formalin, and 5% glacial acetic acid) for 24 hours. Specimens were then transferred to an ethyl alcohol series of ascending concentrations (70, 80, 90, 95, and 100%) for dehydration. After that, they were maintained in Methyl Benzoate overnight. Then, they were cleared by xylene, and embedded in paraffin. The paraffin-embedded specimens were cross-sectioned at 5 µm and stained with Mayer's hematoxylin-eosin (H &E) for microscopic examination of the gonadal features with a light microscope (Leica ICC50 HD). The developing stages of the oocytes and spermatocytes were defined following the index proposed by **Lipinski and Underhill (1995)** that is given in Table (2).

Table 1. The macroscopic sexual maturity scale applied for *U. duvaucelii* as described by **Sauer and Lipinski (1990)**

Maturity Stage		Morphological Characters	
		Males	**Females**
I	**Immature**	Testis membranous, no sperm in spermatophoric sac.	Small ovary, transparent membranous, not granulate structure. The nidamental gland is noticeable.
II	**Maturing**	Testis is visible. Seminal vesicle and spermatophoric sac well developed. Spermatophoric sac with few sperm, soft, whitish, and structureless particles.	Nidamental gland and accessory nidamental gland small to large, ovary with granulate structure, whitish opaque.
III	**Mature**	Testis compact and voluminous. Sperm develop in the spermatophoric sac, but not full.	Nidamental gland and accessory nidamental gland large. Two different stage eggs (oval or polygonal, whitish opaque, and round, reticulated pale-yellowish).
IV	**Fully Mature**	Testis is rigid. Sperm densely packed in spermatophoric sac and noticeable in base and tip of penis.	Nidamental gland and accessory nidamental gland very large, accessory nidamental gland bright red color. Ovary enlarged to fill dorsal portion of mantle cavity with reticulated pale-yellowish eggs. Oviduct gland filled with fully mature eggs.
V	**Spent**	Testis long and thin, few sperm in spermatophoric sac.	Gonad small, nidamental gland relatively large and soft. A small number of large eggs in ovary.

Table 2. The microscopic sexual maturity scale applied for *U. duvaucelii* as described by **Lipinski & Underhill (1995)**

Maturity Stage	Histological Examination	
	Males	**Females**
I	First spermatogonia and first primary spermatocytes developed anywhere in gonad.	First oogonia developed anywhere in the gonad.
II	Tubules with primary spermatocytes inside clearly defined.	Follicle cells surround the oocyte anywhere in the gonad.
III	First spermatids develop anywhere in the gonad.	First invagination of follicular epithelium.
IV	First spermatozoa are formed anywhere in the gonad.	Yolk finishes displacing follicular folds in the gonad.
V	None	First mature oocytes found anywhere in the gonad.

RESULTS AND DISCUSSION

1. Sex Ratio

The overall sex ratio (M: F) regarding 622 sexed specimens of *U. duvaucelii* (389 males and 233 females) in the Gulf of Suez during the study period was 1.7: 1 [$\chi^2 = 2.67$] (Table 3). This value is departing from the expected sex ratio of 1:1, where males were significantly more abundant than females during the study period; showing a predominance of males in the population (with a percentage of 62.5 % for males and 37.5 % for females). The monthly variations of the sex ratio of *U. duvaucelii*, collected from the Gulf of Suez (September 2017 – April 2018) are represented in Fig. (3). Monthly sex ratios of *U. duvaucelii* were also not equal throughout the year during the study period. The Chi-square test revealed that the monthly sex ratio varied significantly in all months with chi-square values less than the critical value ($\chi^2 = 14.17$).

The predominance of *U. duvaucelii* males was also observed in the study of **Mohamed *et al.* (2014)** for *U. duvaucelii* in the Suez Canal [M: F ratio = 1:0.81]. Moreover, **Gewida *et al.* (2021)** assessed the predominance of males with respect to *Loligo duvaucelii* in the Suez Gulf [M: F = 1.6:1] for specimens collected monthly during the fishing season 2014/2015. Whereas, females were recorded predominant considering *Loligo duvauceli* in Thailand; the sex ratio of males to females was 1:2 (**Petsut & Kulabtong, 2012)**. Similarly, the predomince of females was detected in *U. duvaucelii* off the East Indian Ocean **(Arkhipkin *et al.*, 2015)**; the sex ratio of males to females was 1:1.3. The sex ratio is among the most basic demographic parameters as it indicates both the relative survival of females and males and the future breeding potential of a population. The observed sex ratio is a consequence of natural selection on the sexes and any anthropogenic effects of harvest. The sex ratio may vary from one species to the other from the expected 1:1,, or even in the same population at different times, being influenced by several factors such as adaptation of the population, reproductive behavior, food availability and environmental conditions. Additionally, this disparity could be attributed to the partial segregation of mature individuals through habitat preferences and migration or behavioral differences between sexes, thus rendering one sex to be more easily caught than the other.

Table 3. Sex ratio (M: F) of *U. duvaucelii*collected from Suez Gulf (September 2017 – April 2018)

Month	No. of specimens	No. of males	% of males	No. of females	% of females	Sex Ratio (M: F)	χ^2	p value
September	35	20	57.14	15	42.86	1.3: 1	0.44	0.01
October	153	99	64.71	54	35.29	1.8: 1	0.31	0.58
November	119	76	63.87	43	36.13	1.8: 1	0.10	0.95
December	103	62	60.19	41	39.81	1.5: 1	0.24	0.97
January	98	65	66.33	33	33.67	2: 1	0.60	0.96
February	29	16	55.17	13	44.83	1.2: 1	0.67	0.98
March	54	33	61.11	21	38.89	1.6: 1	0.05	1.00
April	31	18	58.06	13	41.94	1.6: 1	0.27	1.00
Total	**622**	**389**	**62.54**	**233**	**37.46**	**1.7: 1**	**2.67**	**0.91**

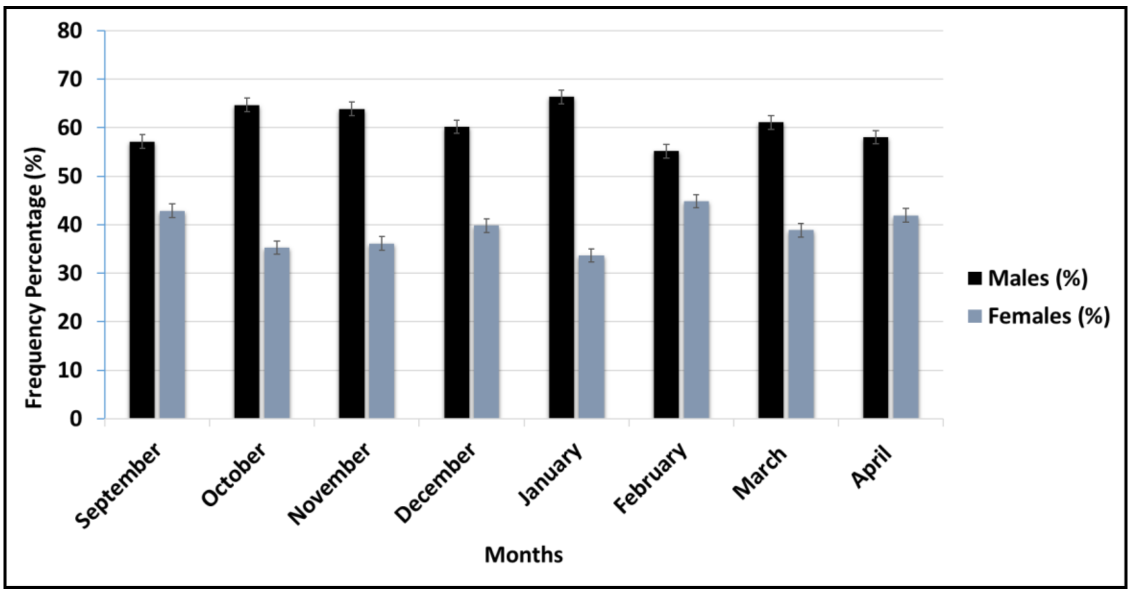

Fig. 3. Monthly variations of the sex ratio of *U. duvaucelii* collected from the Suez Gulf (September 2017 – April 2018)

2. Mantle Length at 50% Maturity ($ML_{50\%}$)

The percentage of maturity distributions for each length group was applied as a marker to detect the onset of sexual maturity. The mean mantle length at 50% maturity ($ML_{50\%}$) of *U. duvaucelii* in the Suez Gulf was estimated separately for each sex. $ML_{50\%}$ of females (4.5 – 19.4 cm ML) was observed at 10 cm ML, whereas the males (3.4 – 25.3

cm ML) attained sexual maturity at 13.5 cm ML (Fig. 4). The results showed that females attain maturity at a smaller size compared to males.

These findings are in accordance with those reported by **Jereb and Roper (2010)** where *U. duvaucelii* females were found growing faster than males and they were heavier at the same length; however, males ultimately attain a greater size and age. The maximum reported size was 33 cm ML in the Indian waters and the size at 50% maturity ranged between 9 and 13 cm mantle length for females and 7 to 15 cm ML for males. In addition, the 50% maturity length was observed at 13 – 14.9 cm ML and 15 – 16.9 cm ML for *U. duvaucelii* females and males, respectively in the Suez Canal (**Mohamed *et al.*, 2014**). Moreover, **Gewida *et al.* (2021)** found that the females of *Loligo duvaucelii* in the Suez Gulf mature at smaller DML and attain a smaller adult length than that of the males; the mean length at maturity was 14.8 and 17.5 cm DML for females and males, respectively. On the other hand, the present results are dissimilar to those of **Petsut and Kulabtong (2012)** with regard to the average size of ML at 50% maturity, reaching approximate lengths of 4.3 cm and 10.0 cm in spawner males and females, respectively. Furthermore, **Arkhipkin *et al.* (2015)** reported that the males of *U. duvaucelii* off the East Indian Ocean (3 – 30 cm ML), mature at a size of 8 cm ML, and females mature at 9 cm ML. The different range in the size at 50% maturity may be attributed to the difference in temperature between the environments.

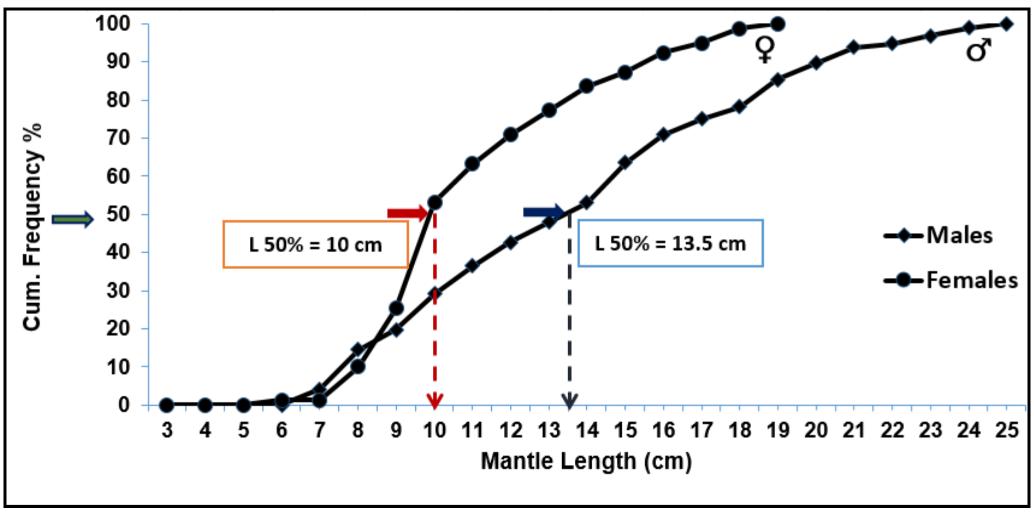

Fig. 4. Mantle length at 50 % maturity ($ML_{50\%}$) of males and females of *U. duvaucelii*

3. Gonado-Somatic Index (GSI)

Monthly variation of the gonado-somatic index is an indicator of the state of gonadal development of *U. duvaucelii* in the Suez Gulf, and hence, in determining the spawning season. The GSI for each sex was estimated and represented during different months of the study period (except for the closed season from May to August); all estimates are displayed in Fig. (5).

The graph of the gonado-somatic index elucidated that the females had an increase of GSI values during autumn (September, October, and November; GSI = 7.44, 8.48 and 9.20 %, respectively). Those values decline during winter (December and January; GSI =

5.36 and 5.77 %, respectively) then gradually increase during spring (March and April; GSI = 10.02 and 10.88 % respectively). On the other hand, the males had homogeneous GSI values throughout the reproductive cycle with a slight detected increase during autumn (September, October, and November; GSI = 4.71, 5.74 and 5.29 %, respectively) and a slight decrease during winter (December and January; GSI = 4.59 and 4.29 %, respectively) followed by a gradual increase once again during late winter and spring (February, March, and April; GSI = 5.75, 6.54 and 6.96 %, respectively). Consequently,it was noticed that, the females and males of *U. duvaucelii* in the Suez Gulf have two peaks in the year-round spawning period. The first is in April (GSI = 10.88 % and 6.96% for females and males, respectively) and the second is in November for females (GSI = 9.20%) and in October for males (GSI = 5.74%). It is worthy to mention that, during the present investigation, the spawning season of *U. duvaucelii* in the Suez Gulf occured in warm months (during spring and autumn). Missing GSI values of *U. duvaucelii* in the Suez Gulf during summer is due to the inability of obtaining samples because of the closed season (from May to August) applied in the Suez Gulf as a fishery management action.

Similarly, **Jereb and Roper (2010)** reported that, the spawning period of *U. duvaucelii* along the west coasts of India appears to be prolonged, almost year-round, with peaks in different months, principally in spring and autumn. The values of GSI showed nearly similar results with those of **Mohamed *et al.* (2014)** for *U. duvaucelii* in the Suez Canal. The previous authors found that the spawning season takes place in late spring and summer. Addingly, **Gewida *et al.* (2021)** who addressed the monthly variations in the gonado-somatic index of *Loligo duvaucelii* in the Suez Gulf during the fishing season 2014/2015 clarified that, the spawning season extends from April to October (spring to early autumn) for males; and from April to November (spring to late autumn) for females. Whereas, the study of **Kilada and Riad (2010)** conducted on *U. duvauceli* in the Northern Red Sea showed that the spawning season is in winter and early spring. **Petsut and Kulabtong (2012)** demonstrated that the spawning season of the Indian squid in Thailand is found throughout the year; however, two peaks of spawning occur in two ranges, March-May and August-October. Furthermore, **Arkhipkin *et al.* (2015)** reported that in *U. duvaucelii* off the East Indian Ocean, spawning is likely to occur year-round, but peak spawning is observed during January–June, and August–December.

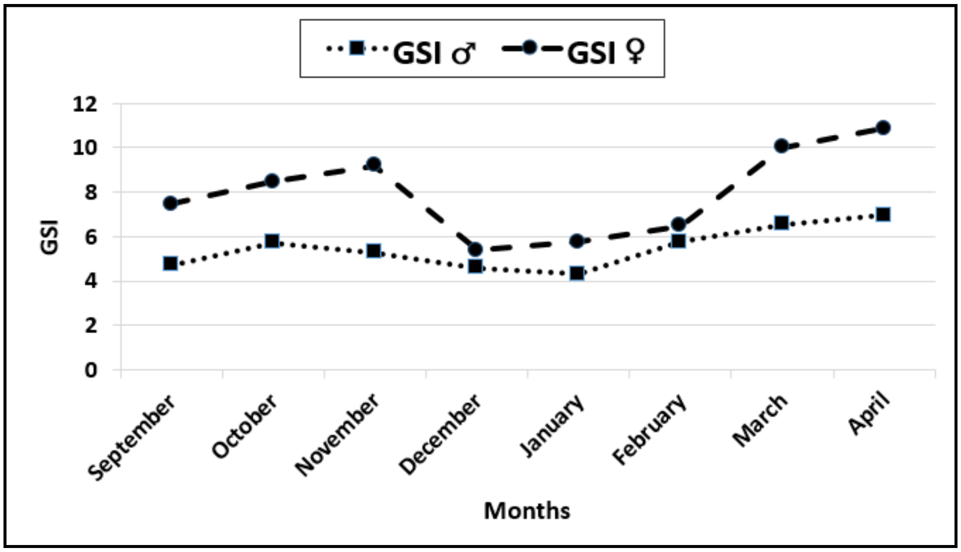

Fig. 5. Monthly variation of Gonado-Somatic Index in males and females *U. duvaucelii*

4. Gonadal Development

The morphology and histology of ovary and testis of *U. duvaucelii* in the Suez Gulf at all the stages of the reproductive cycle were investigated to describe and classify the stages of gonadal development.

4.1. Morphology of Gonads

Throughout the present study, ovaries and testis of *U. duvaucelii* were morphologically divided into four stages as follows: - stage I (Immature), II (maturing), III (mature), and IV (fully mature). No spent (stage V) individuals were obtained throughout this study. The main morphological features of the *U. duvaucelii* testis and ovaries in the different stages of the reproductive cycle were displayed in Fig. (6). The relevant explanations of these stages were:- for males: maturity stage (I) immature: Testis was undeveloped (thin, transparent and filmy), Spermatophoric complex (NS, SO, and P) was undeveloped; maturity stage (II) maturing: Testis was small, with a whitish color, Spermatophoric complex was small and well developed, Needham's Sac with few sperms; maturity stage (III) mature: Testis was large, soft, milky white, Spermatophoric complex was large and well developed, Needham's Sac was full of sperms; maturity stage (IV) fully mature: Testis was at its maximum size, compact, and milky white; the Spermatophoric complex was spacious; the Needham's Sac was filled with sperms, and moves out to the penis (Fig. 6).

Furthermore, for females, the same four maturity stages were maturity stage (I) immature: Ovary was transparent and unobvious, the Nidamental gland was very small, but easily discernible, the Accessory Nidamental gland was indefinite and was not yet developed; maturity stage (II) maturing: Ovary was obvious, small and pale yellow, the Nidamental gland was large, cloudy white, and Accessory Nidamental gland was hardly detected; maturity stage (III) mature: Ovary was large, yellowish in color; the Nidamental gland was large; the Accessory Nidamental gland was identifiable with orange color; maturity stage (IV) fully mature: Ovary was very large and broad, appeared like a jelly yellowish mass; the Nidamental gland was at an extreme size, and Accessory Nidamental gland was prominent with a reddish-orange color (Fig. 6).

This classification and description of different maturity stages of *U. duvaucelii* in the Suez Gulf during the present investigation is fit to that described by **Gewida *et al.* (2021)** for *Loligo duvaucelii* collected during the fishing season 2014/2015 from the Suez Gulf. The previous authors used the index of **Lipinski and Underhill (1995)** as a basis and gave a detailed description of four maturity stages; Immature (I), maturing (II), mature (III), and spawning (IV).

The variation of the different stages of maturation of the gonads of *U. duvaucelii* in the Suez Gulf during different months of the study (except for the closed season from May to August) is represented in Fig. (7). All the maturity stages (I, II, III, IV, and V) were detected all over the months of the study period. The highest percentage of immature (I), maturing (II), mature (III), and fully mature (IV) male individuals was observed during March and December (59.38%; 48.39%), January and February (54.55%; 42.86%), April and February (34.62%; 23.81%) as well as September and April (34.78%; 30.77%), respectively. Furthermore, the highest percentage of immature (I), maturing (II), mature (III), and fully mature (IV) female individuals were observed during December and January (57.89%; 46.51%), November, and January (28.57% and 25.58%), September and April (38.89; 36.00%) as well as April and October (42.31%; 22.58%), respectively. Throughout the year, mature and fully mature males and females individuals of *U. duvaucelii* were present, suggesting that the spawning probably occurs the whole year. **Gewida *et al.* (2021)** recorded similar results pointing out that, the *Loligo duvaucelii* population in the Suez Gulf contained mature individuals mostly throughout the whole year during the fishing season 2014/2015.

4.2. Histology of Gonads

Concerning the histological examination of the *U. duvaucelii* gonads; the histological sections of the male's testis and female's ovary are displayed in Figs. (8 & 9) respectively. Results of examining the slides of testis of *U. duvaucelii* male individuals revealed that, the most abundant cell type was Spermatocytes (SC) at stage I (immature or undeveloped). In the early spermatogenic (stage II), the cell differentiation advanced where Sperms (S) and Spermatids (ST) were observed. Slides of the stage (III): mid-spermatogenic showed the abundance of Primary Spermatozoa (PSZ) and Spermatozoa (SZ). In stage (IV): late spermatogenic, the Lumen (L) and Spermatogonia (SG) were numerous; in stage (V): spent, the final sperm shape (FS), Spermatids (ST), Sperms (S) and Spermatogonia (SG) were scattered.

On the other hand, the investigation of slides of the ovary of *U. duvaucelii* female individuals revealed that, the abundance of Primary Oogonia (PO), Secondary Oogonia (SO), Oocyte at Primary Stage (PS) were detected in stage I (immature stage). Furthermore, the Follicle Cell (FC), nuclei (N), Connective Tissue (CT) were found in post immature stage I. In stage (II): pre-vitellogenesis, Advanced Follicle Cell (FC) and Primary Oocyte (PO) were abundant. In stage (III): mid-vitellogenesis, Early Yolk Oocyte (EY) and Follicle Cell Cap (FCP) were prevalent; Cuboidal Follicle Cells in Stage (IV) late-vitellogenesis were ample. In stage (V): post-vitellogenesis, Mature Oocyte (MO) and Late Vitellogenic Oocyte (LV) in stage V were abundant, whereas in stage (VI): spent or post-ovulatory, the Atretic Late Yolkless Oocyte (ALY) and Mature Oocyte (MO) predominated.

The present study gave more details in the classification and description of maturity stages than that described in the study of **Mohamed *et al.* (2014)**. They carried out a histological analysis of the gonadal development in the *U. duvaucelii* in the Suez Canal and determined only four stages of oogenesis depending on the degree of follicular cell development in association with oocytes and five stages of spermatogenesis according to the degree of development within the seminiferous tubules.

Various stages of gonadal development were represented in most histological slides affirming that the spawning of *U. duvaucelii* is iteroparous; where females *U. duvaucelii* lay their fertilized eggs and regenerate their gonads for a further reproductive event, indicating a partial spawning that occurrs throughout the year. This reproductive strategy is a conflict with those described by **Rocha *et al.* (2001)**, **Laptikhovsky *et al.* (2013)** and **Hoving *et al.* (2015)** where they reported that coleoid cephalopods; octopus, squid, and cuttlefish are considered semelparous, whereby females lay their eggs in one single spawning after which they die. Thus, coleoid cephalopods are thought to go through only one reproductive cycle in their life. Whereas, the findings of the present investigation agree with those of **Jereb and Roper (2010)** who reported that, the *U. duvaucelii* has an extended reproductive phase within the life cycle, i.e. not a strictly semelparous reproduction as in the case of other squids. In addition, these results agree with those of **Petsut and Kulabtong (2012)** who recorded that the Indian squid, *Loligo duvauceli*, in Thailand is a partial spawner.

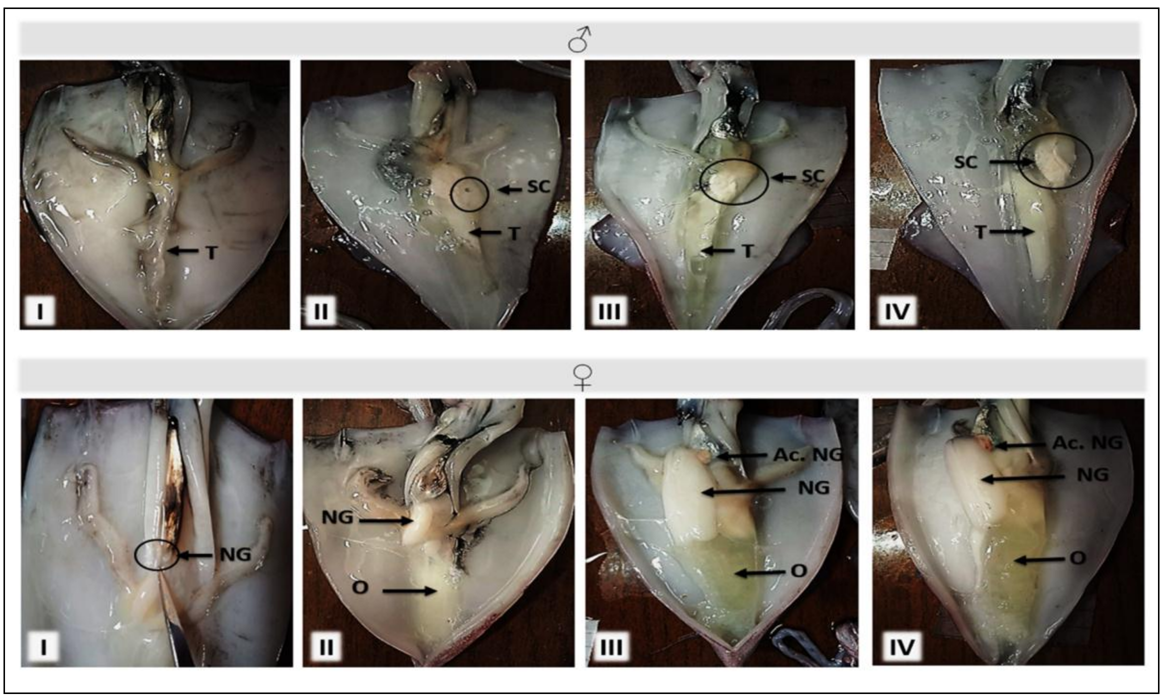

Fig. 6. Morphological variations in the reproductive organs of *U. duvaucelii* through sexual maturity stages (**O**: Ovary; **NG**: Nidamental Gland; **Ac. NG**: Accessory Nidamental Gland; **T**: Testis; **SC**: Spermatophoric Complex (**NS**: Needham's Sac or Spermatophoric Sac and **SO**: Spermatophoric Organ).

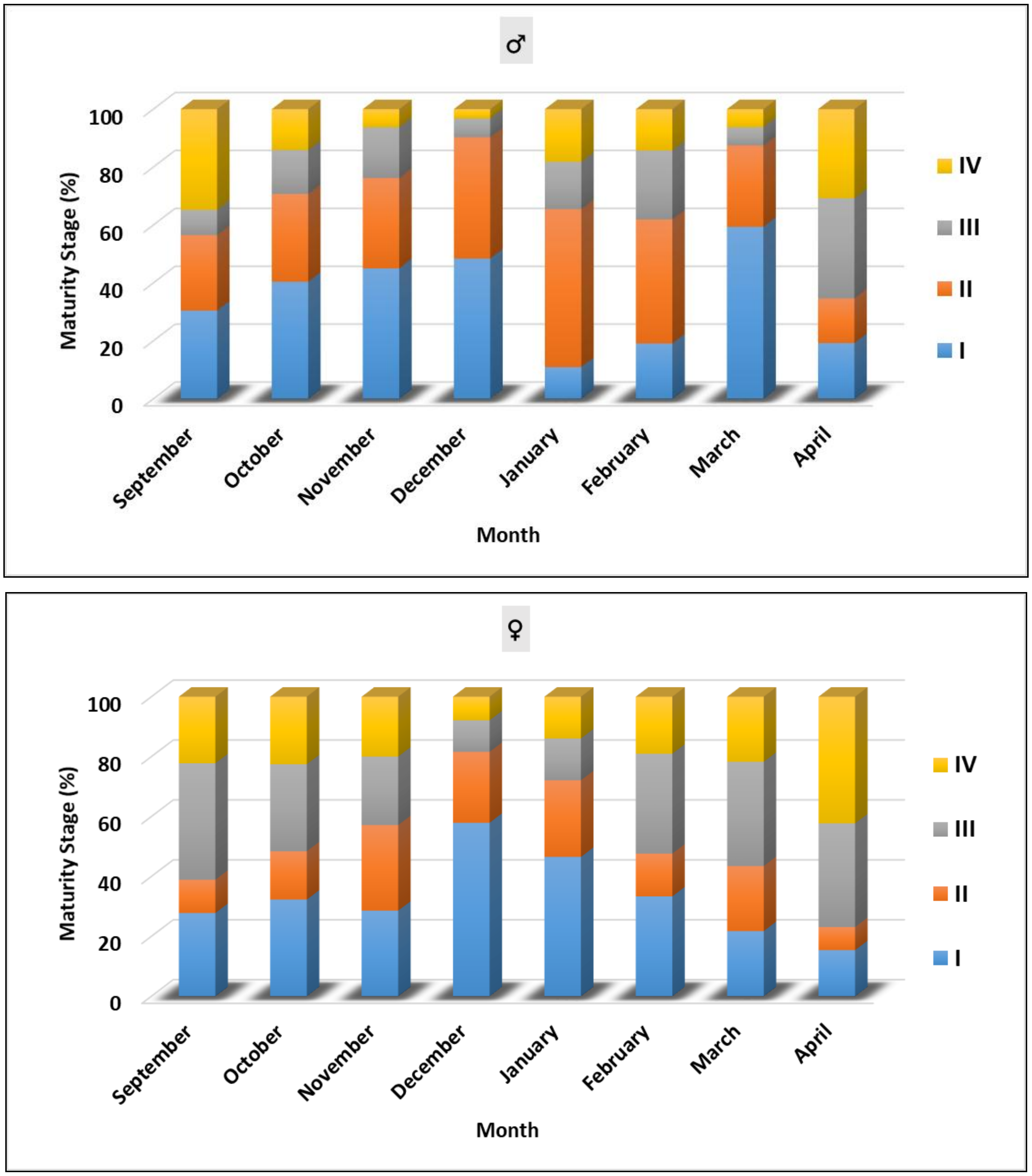

Fig. 7. Analysis of monthly distributions of different maturity stages for males and females *U. duvaucelii*, collected from the Suez Gulf (September 2017 – April 2018**)**

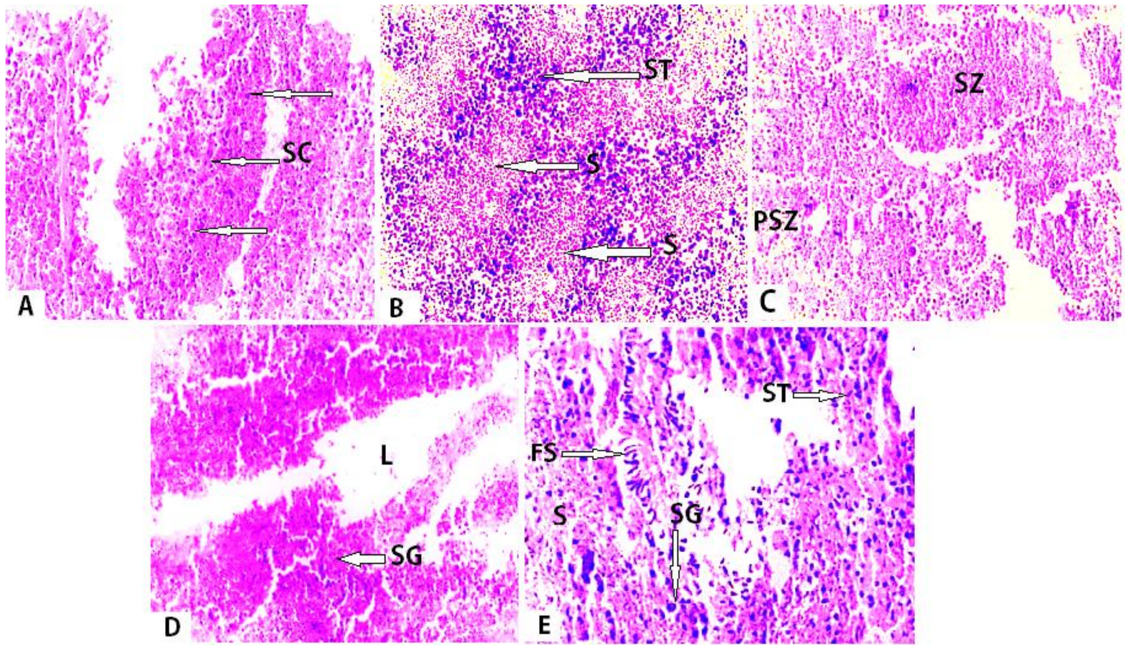

Fig. 8. Histological sections of the *U. duvaucelii* male testis showing Spermatocytes (SC & Arrows) in immature stage I **(A);** cell differentiation advanced, Sperms **(S),** Spermatids **(ST)** in early spermatogenic stage II **(B);** the Primary Spermatozoa **(PSZ),** Spermatozoa **(SZ)** in mid-spermatogenic stage III **(C);** Lumen **(L),** Spermatogonia **(SG)** in late spermatogenic stage IV **(D);** Final Sperm shape **(FS),** Spermatids **(ST),** Sperms **(S),** Spermatogonia **(SG)** in Stage V spent **(E) {H & E, 200 X}.**

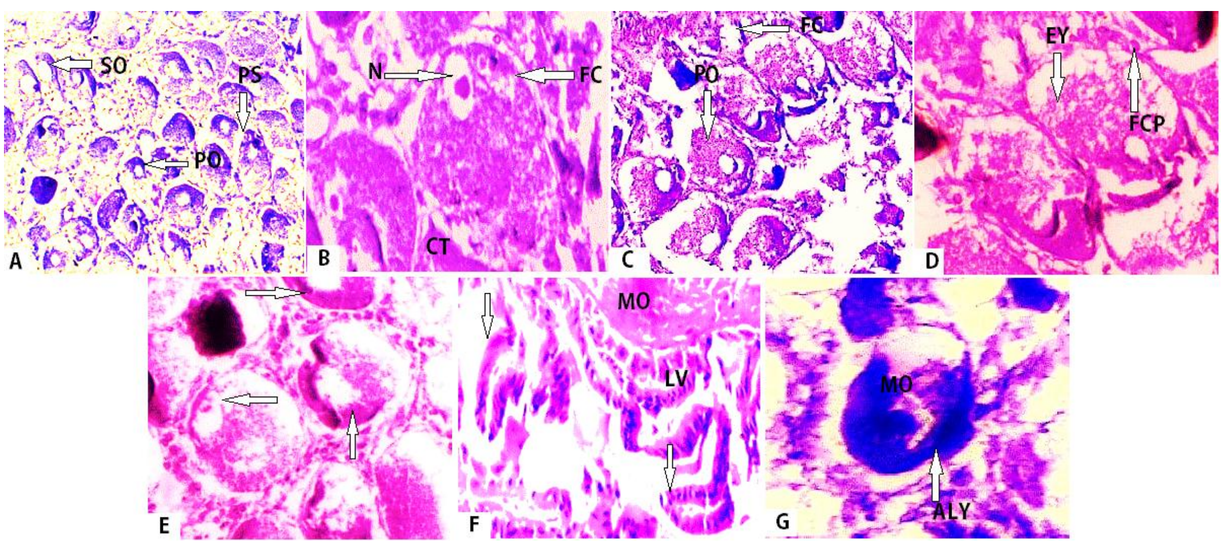

Fig. 9. Histological sections of the *U. duvaucelii* female ovary showing the Primary Oogonia **(PO)**, Secondary Oogonia **(SO)**, Oocyte at Primary growth Stage **(PS)** in immature stage I (A); Follicle Cell **(FC)**, Nuclei **(N)**, Connective Tissue **(CT)** in post immature stage I (B); Advanced Follicle Cell **(FC)**, Primary Oocyte **(PO)** in stage II (C); Early Yolk Oocyte **(EY)**, Follicle Cell Cap **(FCP)** in stage III (D); Cuboidal Follicle Cells **(Arrows)** in Stage IV (E); Mature Oocyte **(MO)**, Late Vitellogenic Oocyte **(LV)** in stage V (F); Atretic Late Yolkless Oocyte **(ALY)**, Mature Oocyte **(MO)** in stage VI (G) {H & E, 400 X}.

CONCLUSION

Reproductive biology is crucial for fishery management, where managers rely on size at first maturity and the onset and duration of spawning season for managing fisheries. The present investigation revealed that males of *U. duvaucelii* in the Suez Gulf are predominat compared to females. Estimation of the mantle length at 50% maturity clarified that females attain maturity at a smaller size than males. The convergence between GSI values indicates that this species has an extended spawning period extending throughout the year, with higher activity during warm months from spring to late autumn. Moreover, the presence of all maturity stages throughout the year suggests that the spawning probably occurs throughout the whole year. Morphologically, only four stages of sexual maturity in males and females were identified, whereas five stages of spermatogenesis in the males' testis and six phases of oogenesis in the females' ovaries were recognized microscopically. This finding confirms that the microscopic evaluation is the most reliable tool that accuretly defines the maturity stages. The present investigation of *U. duvaucelii* in the Suez Gulf serves as a yardstick for the identification of maturity stages throughout gonadal development and the prediction of the spawning season for sustainable management of this fishery in the Suez Gulf. In addition,it provides information that is potentially necessary for the commercial mariculture of this fast-growing species.

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