REPRODUCTIVE BIOLOGY OF THE FLATTENED PORCELAIN CRAB, *PETROLISTHES RUFESCENS* (PORCELLANIDAE: ANOMURA: CRUSTACEA) FROM AIN SUKHNA, GULF OF SUEZ, EGYPT

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Abstract

The reproductive biology of the porcelain crab *Petrolisthes rufescens* (Heller, 1861) population at Ain Sukhna (62 km south Suez City) was studied during the period from April 2000 to May 2001. Sexual dimorphism was determined based on morphological differences between sexes. Abdomen is semi-circular with four pairs of biramous appendages in females, but tapering with only two pairs of unequal uniramous appendages (pleopds) in males. Size at first maturity was determined using allometric growth occurring at puberty from the relationships between male's 2nd pleopodal length and chelae length against carapace length (C.L.) and by the appearance of distinctive ovaries in females. Overall sex ratio for this crab species was 1: 0.70 males to females, with highly significant Chi-square value ($X^2 = 7.486$, $P< 0.05$, df =1). It was not constant throughout the year particularly during breeding seasons, showing highly significant Chi-square value ($X^2 = 20.185$, df= 25, $P < 0.05$). Based on the appearance of ovigerous females, the breeding of *P. rufescens* is lengthy, start often at late winter and early spring and extends through summer to early autumn. Incubated eggs are oval or semi-oval, sometimes elliptical with polygonal surfaces, and varied in size from 400 to 1000 μm with an average of 640 ± 105 μm. These eggs are passed through five stages of maturation, and showed remarkably gradual decreasing in size throughout the period of incubation, reaching the minimum at last stages (stage V or the releasing larvae). The color of incubated eggs is also changed from bright yellow in newly laid eggs to grey at the last stage of development. The fecundity of ovigerous females was relatively low, varied from 35 to 450 with an average of 208 ± 92 eggs /female crab. It was correlated by a curvilinear relationship with carapace length only.

Introduction

Porcelain crabs are much alike true crabs (brachyurans) but they can easily distinguished from them by possessing uropods and their small, dorsally directed 5 th pair (last walking pairs) of legs under the carapace (Barnard, 1950; Fransozo and Bertini, 2001; Osawa and McLaughlin, 2010). These crabs belong to family Porcellanidae Haworth, 1825 (Anomura: Decapoda), which comprises about 27 to 30 genera including from 225 to 277 species (Melo, 1999; Fransozo and Bertini, 2001; Osawa and McLaughlin, 2010), most of them are worldwide in distribution, occupying a variety of littoral habitats living in hard substrates within crevices or under boulders and even in bottom covered by calcareous algae, excepting rare species living in the deep sea (Veloso and Melo, 1993).

Among Porcellanidae, genus *Petrolisthes* is the largest, accommodates about 102 species of the total belonging to this family, several of them are worldwide in their distribution (Haig, 1960; Hollebone and Hay, 2007; Ng *et al*., 2008; McLaughlin *et* *al*., 2007). In the Red Sea and its associated gulfs (Suez and Aqaba), genus *Petrolisthes* is represented by 8 valid species (Lewinsohn, 1969), most of them are found in the intertidal zones, but few extend into about 5 m depth in the shallow subtidal areas (Lewinsohn, 1969; El-Sayed, 2002; Fouda *et al*., 2003).

At the northwestern coast of the Suez Gulf, particularly Ain Sukhna, the porcelain crab, *Petrolisthes rufescens* (Heller, 1861), is a common intertidal species. It inhabits, with the co-existing less common porcelain species, *Petrolisthes leptocheles* (Heller, 1861), the rocky and mixed (rocky and sandy) shores. During low tides, the individuals of this species escape always under small stones and boulders to protect itself from the effect of heat, desiccation and terrestrial predators (Ismail, 2005). They contribute with a significant part in the composition of the intertidal marine fauna at that area and can be used as a biological indicator for the effects of human activities on marine environment.

Since the last decade of the 20^{th} century till now, several resorts, tourist villages, hotels and camp sites are either constructed or are under constructions along nearly every space at the western coastal-line of the Gulf of Suez extending from Suez City (north) to Za'afarana (south). Hence, the natural habitats at several intertidal areas became threatened as results of severe cleaning, dredges, filling and continuous changes for the natural profile of the coast-line, in addition to the profound effects of oil refiners and new constructed industrial commercial companies at Ain Sukhna. These man-made constructions associated with processes of reconstruction for the major areas of the shoreline and human activities, threat in turn and affect directly different types of littoral marine habitat with its associated fauna, particularly those distributed on the exposed intertidal and coastal zones. Therefore, particular attention needs to carry out detailed studies at that area, to be considered as baseline data available to follow up these changes and take in consideration the future studies which will be done later.

Consequently, the present work aims to through light on the reproductive biology of the flattened porcelain crab, *Petrolisthes rufescens*, one of the common species inhabiting the intertidal zones at the study area, due to its sensitivity to environmental changes and its important ecological role in food chains for other co-existing species.

Materials and Methods

The specimens of porcelain crab, *Petrolithes rufescens* were collected, from the intertidal and uppermost borders of subtidal zones of the five selected study areas along the northwestern portion of the Gulf of Suez (Figure 1) during the period from June 2000 to May 2001. The intensive monthly work and specimen collection were carried out at site # 1, which is known as Al- Fangary Beach, located about 2 km south Ain Sukhna (Km 62 South Suez City). Additional specimens were collected irregularly during the same period from the other four localities including: Al-Khaima Beach, site # 2 (40 km South Suez City), Al-Semad Beach, site # 3 (32 km South Suez City), the shore line of the National Institute of

Oceanography and Fisheries, (Gulfs of Suez and Aqaba Branch) site # 4 and the most northern site # 5, at Suez City. At field, all observations on tidal cycle and changes in animal and plant communities were recorded; while collected specimens were preserved in 10 % seawater formalin solutions.

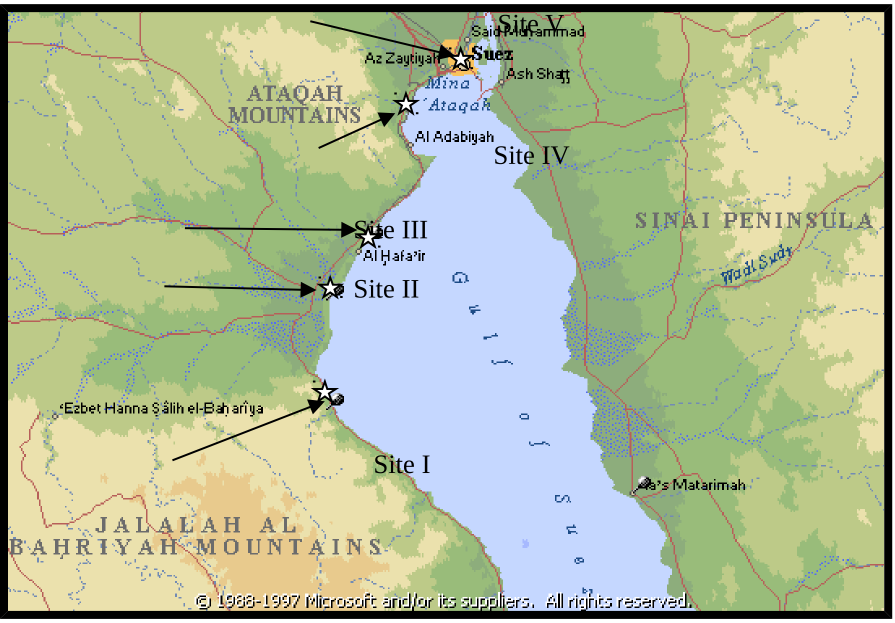


Figure (1): Shows the locations of the study sites.

At the laboratory, all preserved specimens of porcelain crab were sorted, sexually differentiated and counted, then weighed to the nearest 0.1 gm using an electric balance with accuracy of 0.01 g after removing excess water by blotting with absorbent tissues. Carapace length (CL), carapace breadth (CB), and second male pleopodal length (PL) were measured to nearest mm with a Caliper Verneer with accuracy of ± 0.01 mm. The adult (matures) and small immature or juvenile crabs were dissected, gonads were examined and Chi-square test was used for comparing sex ratios for all population throughout the study period.

The size at first maturity was determined for each sex by applying the relationship between males and females right chela propodal length and male's right second pleopodal length against carapace length according to Hartnoll (1982) and El-Sayed (1997).

The ovigerous (gravid) females were noticed and examined, and all incubated eggs were classified into five developmental stages according to methods of Subramoniam (1982), Schembri (1982) and El-Sayed *et al*., (1998). The colour and incubation period for these eggs were recorded at each stages of incubation, while the frequencies of ovigerous females were used as an indicator for spawning season according to Subramoniam (1982).

Egg diameters were measured to the nearest micrometer (µm) using Sterio-binoclear microscope (PZO Warsaw) provided with graduated screen. Eggs were separated from pleopods, and put in saline solution for two hours, then taken onto a slide and measured under the microscope at power of magnification of 80 X.

Fecundity denotes to the total number of incubated eggs per female. It was determined according to Subramoniam (1982) and Schembri (1982) by counting all incubated eggs carried by ovigerous females after separation from pleopodal setae by a dissecting needle in a divided Petri- dish. The relationship between mean egg number (absolute fecundity, F) of each size class and female carapace length (CL) was calculated according to the following formula:

$$\text{Log } F = \log a \pm b \text{ Log } CL$$ (Haynes *et al*., 1976)

Where F = absolute fecundity, CL = independent variable represented by carapace length in mm, "a" is a constant equal to the intercept of straight line with Y-axis and "b" is an exponent, equal to the slope of fitted line.

Results

A-Sexual dimorphism:

The adults mature individuals of the flattened porcelain crab, *Petrolisthes rufescens* are crab-like, have symmetrical abdomen, folded under thorax. The abdominal appendages are the main sex character for rapid differentiation between mature males and females of this species. However, it was very difficult to distinguish sex in specimens of 3 mm CL or less. Mature males have relatively tapering abdomen provided with two unequal pairs of uniramus pleopods. The second pleopods are well developed and large, compared with very short anterior ones. On contrast, in maturing females abdomen is more flattened, semicircular, provided with four biramus pleopods, usually fringed with entangled setae during spawning season.

On the other hand, chelipeds are slightly unequal in both sexes. Right chela is usually larger than left; appears clearly longer than carapace length in adult males and females, but tend to be slightly larger in males.

B- Sex ratio:

The results in Table (1) show that the overall sex ratio of *P. rufescens* during the present study is 1: 0.70 (males to females) with highly significant Chi-square (X^2 = 7.486, $P< 0.05$, df = 1). Monthly variations in sex ratio were clearly observed. In spite of the ratios of males to females were 1:1 during June, October and December 2000, the ratios of females were unstable and greatly varied during most months of this study, recording overdominance for males. However, female's ratios increased remarkably to 1:1.17 (males to females) during March 2001, and reached the maximum ratio, was 1:1.40 (males to females) during September 2000, but showing lowest ratio was 1: 0.26 (males to females), recorded in April 2001. The statistical analysis by Chi-square test showed significant differences in sex ratios between different months during this study (X^2 = 20.185, df= 25, $P < 0.05$).

C- The size at first maturity:

From the relationship between carapace length against right chela propdal length for males and females and right second pleopodal length in males it has been revealed that *P. rufescens* males and females attain sexual maturity between 4.0 and 6.0 mm CL (Figures 2-4). This is very clear from an inflection in the relations for both mature and immature individuals. It means that, all males and females below 4 mm CL are certainly sexually immature, while those have 6 mm CL or above become sexually maturing, but that lie between 4 and 6 mm are mixed from both groups.

Table (1): Total number of collected specimens, percentage of ovigerous females and sex ratios of *P. rufescens* from Ain Sukhna, Gulf of Suez

Number & % / Months	Total No.♀♀ &♂♂	Mature females			% of ovigrous females	Mature ♂♂	♀♀ & ♂♂ Juveniles	Sex ratio (♂♂: ♀♀)
		All females	Spent	Ovigerous females				
April 2000	47	19	7	12	63.16	28	-	1:0.68
May	Area did not visit							
June	24	12	6	6	50.0	12	-	1:1
July	20	9	1	8	88.89	10	1	1:0.90
August	23	9	3	6	66.67	12	2	1:0.75
September	27	14	6	8	57.14	10	3	1:1.40
October	14	7	7	0	0	7	-	1:1
November	12	3	3	0	0	9	-	1:0.33
December	11	5	5	0	0	5	1	1:1
January 2001	14	4	4	0	0	8	2	1:0.50
February	15	4	2	2	50.0	11	-	1:0.36
March	13	7	7	0	0	6	-	1:1.17
April	24	5	2	3	60.0	19	-	1:0.26
May	12	4	0	4	100	8	-	1:0.50
Total	256	102	53	49		145	9	1:0.70

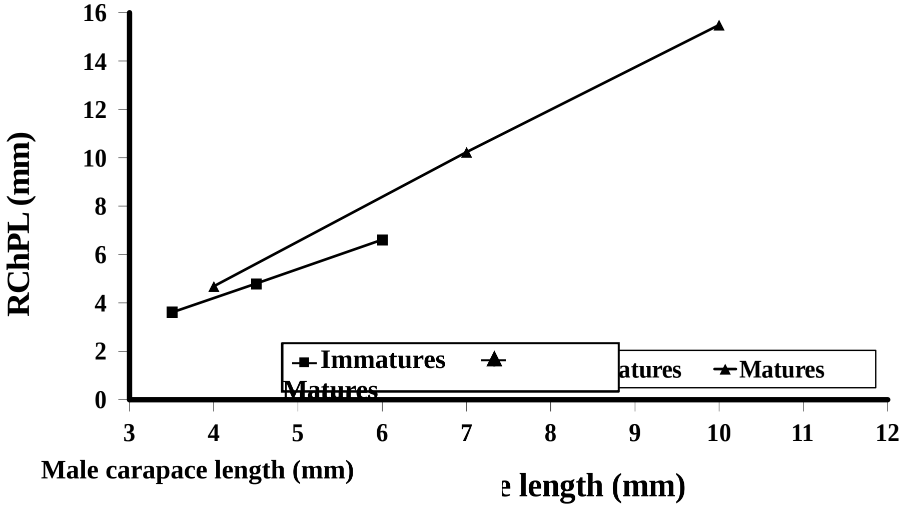


Figure (2): The relationship between male carapace length (CL) and right chela second propodal length (RChPL) of the porcelain crab, *P. rufescens* from Ain Sukhna, Gulf of Suez.

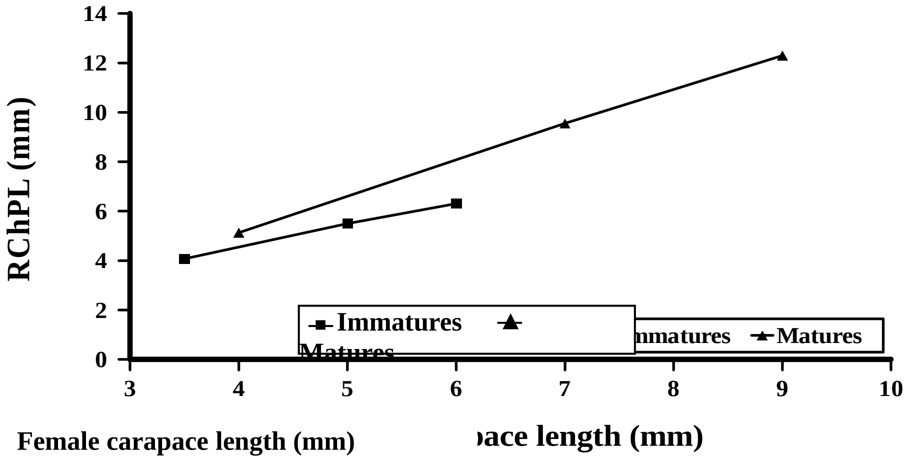
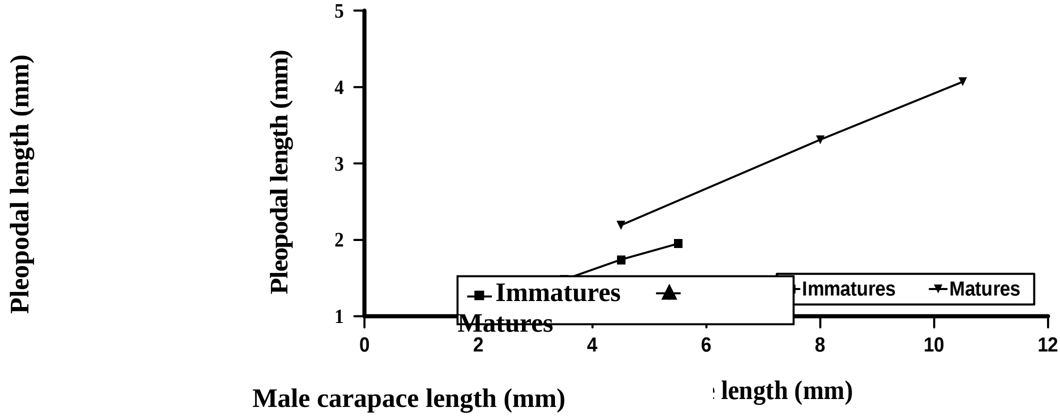


Figure (3): The relationship between female carapace length (CL) and right chela propodal length (RChPL) of the porcelain crab, *P. rufescens*, from Ain Sukhna, Gulf of Suez.



Fi… ace length and right second pleopodal length of the porcelain crab, *P. rufescens* from Ain Sukhna, Gulf of Suez.

The onset of smallest ovigerous females was also taken for determining the first maturity size. Field collection showed that the smallest ovigerous female measured 4.15 x 4.0 mm (CL x C B), captured in July 2000, which means that, most females attain sexual maturity between 4 and 5 mm CL, and may be become sexually maturing above 5 mm CL. Also, laboratory examination showed that the abdomen of immature females was tightly closed against sternum, with fine, delicate and slender pleopods, fringed with fine and weak setae. It becomes semicircular, provided with strong pleopods, fringed with dense marginal setae in full mature and adult females.

It is also worth to mention that, all individuals of juvenile equal to or below 3 mm CL were not sexually recognizable in their external morphology. It was also difficult to even identify them to their specific taxa due to the presence of juveniles of the coexisting less common porcelain crab species, *Petrolisthes leptocheles* in the same habitats.

D- The incubated eggs and developmental stages:

The incubated eggs or eggs carried with ovigerous females are varied from oval or semi-oval, to elliptical with polygonal surfaces resulting from their crowding on the pleopods. Their overall size was varied according to developmental stages from 400 to 1000 μm with an average of 640 ± 105 μm. These eggs were classified into the following five stages:

- Stage I: It denotes to the newly laid eggs after recent ovulation. They appear bright yellow, larger in size, with an average diameter of 810 ±70 μm.
- Stage II: It refers to the early stage of incubated eggs, which have yellow, orange or brown colour, with an average size of 610 ±80 μm.
- Stage III: It denotes to the incubated eggs characterized by the presence of eyed-embryos, of pale orange or gray hues colour, with an average size of 605 ±75μm,
- Stage IV: This stage refers to incubated eggs having well developed embryos have whitish colour and an average size of 650±80 μm,

- Stage V or full complete embryo: It denotes to the releasing larvae, characterized by grey egg-cases attached to pleopod setae, with remaining few eggs with an average size of 640±70 μm

It was noticed that, the size of incubated eggs were remarkably decreased gradually with advances in maturation. The average egg size decreased sharply from 810 ±70 μm for the newly laid eggs, to 610 ±80, 605 ±75, 650±80, and 640±70 μm for the other following developmental stages of maturation, respectively, with a slightly increase in stages IV and V.

E- Incubation period:

The present results indicated that, the appearance of ovigerous females carrying eggs at late stages of incubation (complete embryos with eyes and appendages) were recorded from February to September. However, the appearance of all stages varied from the newly laid eggs to late stages with complete embryos during September, and their complete disappearance throughout October, November, December and the following January, implies that these eggs did not exceed than one month for incubation under female's abdomen and determines the incubation period which can be estimated about one month.

F- Spawning season:

The spawning season of this species is determined based on the appearance of berried or ovigerous females, characterizes by carrying clutches of fertilized eggs on their pleopods under her abdomen. The data in Table (1) and Figure (5) show that the porcelain crab, *P. rufescens,* has lengthy definite breeding season, starts often at late winter and early spring and extends through summer to early autumn.

The first appearance for ovigerous females was recorded in February, and its occurrence continued throughout spring and summer to early autumn (end of September). During 2001, the frequencies of ovigerous females increased gradually from 50 % in February, reaching 100 % during May of the same year. The same pattern was recoded in 2000, where ovegrious female's ratios increased from 63.16 % in April, and continued with high frequencies during summer, being 88.89 % in July, but declined to 66.67 % in August, showing more decline to 57.14 % during September, and disappeared completely during the period from October 2000 to the following January 2001 (Table 1 & Figure 5), in spite of an intensive survey which had been done in these months.

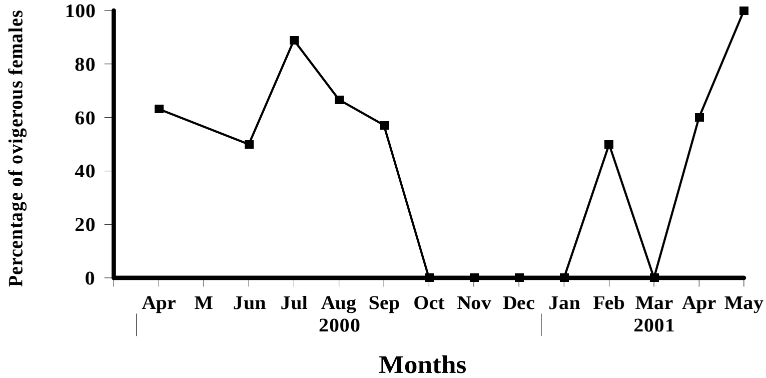


Figure (5): Monthly percentages for *Petrolisthes rufescens* ovigerous females showing spawning season.

G - Juveniles recruitments:

Results in Table (1) display the total number of collected specimens including maturing females (spent and ovigerous), maturing males and juveniles of both sexes. In spite of low percentage of juveniles, the data showed that the first appearance of recognizable juvenile of this species was recorded in July 2000 followed by continuous appearance throughout August and September as well as during winter (December 2000 and January 2001). The delaying of juvenile appearance in July in spite recording of late stages of eggs with complete embryos in February denotes to the relative long period for juvenile metamorphosis after hatching.

H -Fecundity:

The incubated eggs for 41 *P. rufescens* ovigerous females were counted. The egg number or fecundity for these females was relatively low, varying from 35 to 450, with an average of 208 ± 92 eggs /female. As previously mentioned, the smallest ovigerous female was 4.15 x 4.0 mm (CL x CB), collected in July 2000, and carrying 48 eggs, all at late stages of maturation with embryos having prominent eyes and well developed appendages. On the other hand, the largest egg number was 450 for female of 8.45 x 8.25 mm (CL x CB) collected in June 2000; whereas the lowest egg number was 35 for female of 4.65 x 4.55 mm (CL x CB), collected in August 2000. It is worth to mention that, 8 ovigerous females were found have egg clutches with very few eggs. These females may be subjected to egg loss. For example, the largest captured ovigerous female attained 9.0 x 8.85 mm (CL x CB), carrying egg clutches of 195 eggs, all were newly laid and at early stage of incubation, but several pleopd setae were ruptured and egg free.

The logarithmic relationship between carapace length (CL) and total egg number or fecundity (F) for 27 ovigerous females (have nearly intact egg clutches) is illustrated in Figure (6), and represented by the following equation:

Log F = 0.455 + 2.21 Log CL

This relation is statistically significant, and curvilinear with high positive correlation coefficient “r” being 0.82, an allometric regression coefficient “b” 2.21, and positive intercept of Y-axis, “a” = 0.455.

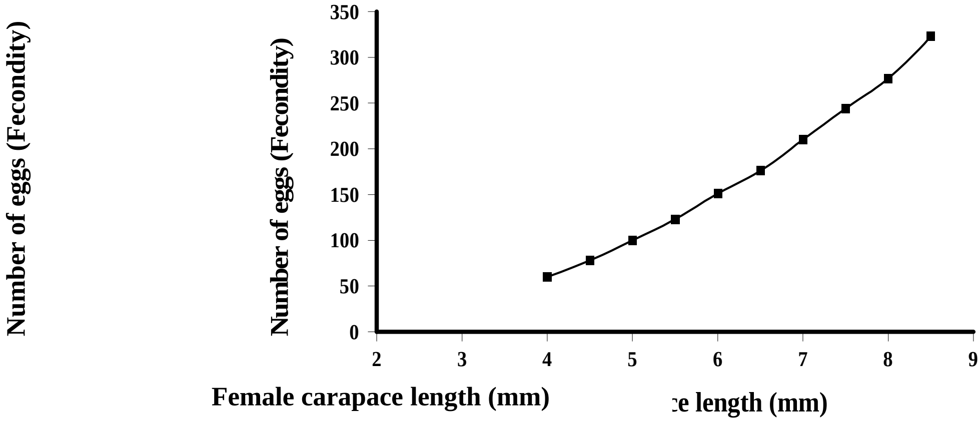


Figure (6): The relationship between carapace length and mean egg number of *P. rufescens* females from Ain Sukhna, Gulf of Suez

Discussion

It is well known that, the reproductive biology of decapod crustaceans is well documented, particularly for brachyurans (Warner, 1977; Sastry, 1983; Lowery and Nelson, 1988; El-Sayed *et al.*, 1998; Fouda, 2000; El-Sayed, 2004). However, little is known about the reproductive biology of Family Porcellainidae (Anomura) around the world (Ahmed and Mustaquim, 1974; Fransozo and Bertini, 2001; Hollebone and Hay, 2007; Monaco *et al*., 2010) and no information available till now about reproduction of *Petrolithes rufescens* from the entire Red Sea and its associated Gulfs (Suez and Aqaba) in spite of its abundance along the Red Sea coasts (Lewinsohn, 1969; El-Sayed, 2002: Fouda *et al*., 2003).

The results of the present study showed that the individuals of *Petrolithes rufescens* exhibited morphological sexual dimorphism helps for sex differentiation. The over all sex ratio for this species was 1: 0.7 (males to females), showing overdominance for males on females during most months of the study period, with only an increase in female's ratios during certain months. These results are in well agreement with those mentioned by Oliveira and Masunari (1995) on *Petrolisthes armatus* in Brazil and Hollebone and Hay (2007) on the same species in Gorgia, and that of Farnsozo and Bertini (2001) on *Pachycheles monilefer* belong to family Prcellanidae in Sâo Paulo coast (Brazil), where sex ratios remain nearly close to 1:1 throughout the year, but with seasonal fluctuations. In contrast, the present sex ratios are contradicted with that reported from South Carolina, which were male-biased at a ratio of 1.5:1 (Hartman and Stancyk 2001).

The sharp fluctuations in female's ratios particularly before or through spawning season may be coincides with the restriction in female's movements in their burrows or escaping into slightly deep water either for egg deposition (ovulation) and hatching, which agrees with results given by Lowery and Nelson (1988), El-Sayed *et al.* (1998), Fouda (2000) and El-Sayed (2004), on true crabs, or due to more catchability of ovigerous females than males during some months of spawning (El-Sayed, 1992, El-Sayed *et al*., 1998), or attributes to other environmental factors including differential mortality, migration or habitat selection, depth preference, changes in salinity, temperature and other environmental parameters which agrees with Hill (1975), Thomas (1975), and Potter *et al.* (1983) on true crabs and Farnsozo and Bertini (2001), Hollebone (2006), Hollebone and Hay(2007) and Monaco *et al*., (2010) on porcelain crabs.

Using of relative allometric growth rates for *P. rufescens* individuals before and after puberty moult gave results very similar to that mentioned on other decapods particularly brachyuran species. The present results showed that, all males and females below 4 mm CL are often sexually immature, but those have 6 mm CL or above become sexually maturing. These results are in full agreement with the previous works on porcelanid green crab *Petrolisthes armatus* carried out by Boudreaux *et al*. (2006). The authors showed that collected females were sexually mature at just 3-4 mm carapace width; as well as with those reported by Jones (1977), Scelzo (1985), Oliveira and Masunari (1995), Fransoz and Bertini (2001) and Monaco *et al*. (2010) on species of some porcellaniid genera. These results are also in agreement with that carried out on true crabs by Somerton (1981), Schembri (1982), Potter *et al.*,(1983), Jewett *et al.*,(1985), El-Sayed (1997, 2004), El-Sayed *et al*., (1998), Fouda (2000).

The progressive increase in male chelae size after maturity is required for the enhancement of defense, feeding and reproductive functions (Hartnoll, 1974; El-Sayed *et al.*, 1998). In the flattened porcelain crab, chelae are used for defense and reproduction functions, but do not employ for feeding (Barnard, 1950; Rypien and Palmer, 2007). On the other hand, broadness of the females abdomen is useful to perform its function for carrying and protecting incubated eggs and helps maintain the continuous circulation of oxygenated water over the eggs carried on her underside (Hartnoll, 1974; Schembri, 1982; Oliveira and Masunari 1995; El-Sayed, 1997, 2004; Hartman and Stancyk, 2001; de Lastang, *et al*., 2003; Hollebone, 2006),

Like most Red Sea animals, *P. rufescens* population has definite lengthy breeding season, extending usually from February to the following September. It coincides by the appearance of ovigerous females from February to the end of September, and exhibits remarkable seasonality in breeding of this species as known as for most Red Sea invertebrates (El-Sayed, 1992, 1997, 2004; El-Sayed *et al*., 1998; Fouda, 2000) and agrees well with that mentioned by Ahmed and Mustaquim (1974) on the same species collected from Manora Island, Karachi, Pakistan, where gravid females appear annually from January to November, and with those recorded from subtropical (Fransozo and Bertini, 2001) and temperate decapods (Schembri, 1982).

The use of decapod ovigerous females as tool for determination of breeding seasons in anomurans had been employed in several species such as the tropical hermit crab, *Clibanarius clibanarius* (Varadarajan and Subramoniam, 1982) and hermit crab *Pagurus bernhardus* (Lancaster (1990) as well as in several species of porcelain crabs (Ahmed and Mustaquim, 1974, Fransozo and Bertini, 2001, and Hollebone and Hay, 2007).

The presence of *P. rufescens* ovigerous females with different developmental incubated egg stages throughout the period from February to September may indicate frequencies of spawning during the same breeding season. It also may define the length of incubation period, which may reach to nearly one month, which coincides with the results mentioned by El- Sayed (1997, 2004), El-Sayed *et al*., (1998), Fouda (2000) on some decapods from the Rd Sea and Gulf of Suez.

Hollebone (2006) explained that when *Petrolisthes armatus* eggs hatch they metamorphose through two planktonic zoeal stages and then one planktonic megalopal stage before finally becoming settlement-ready juveniles. The similar case is observed for *P. rufescens* in the present study where its juveniles are appeared in July, delayed for about 4 months after appearance of ovigerous females in February.

The present results showed that the fecundity of *P. rufescens* is low in comparison with that reported for other intertidal relatively same size decapod species such as *Leptosdius exaratus* from the same area (Fouda, 2000) and those from both the Red Sea (El-Sayed, 2004) and Indian Ocean (Siddiqui & Ahmed, 1992). The decreasing in fecundity of *P. rufescens* from the Gulf of Suez is attributed to several factors including the prevailing hazard environmental conditions, or according to semi-terrestrially habits for this species represented by relative large size of incubated eggs or due to the biological factors dominated by fighting and predations, or restriction of food resources (Warner, 1977).

The presence of good curvilinear relationship between fecundity and carapace length, with an allometric regression coefficient ‘b’(2.21), agrees well with that reported by Monaco *et al*., (2010) on *Petrolisthes granulosus*, Siddiqui and Ahmed (1992), El-Sayed (1997, 2004), El-Sayed *et al*.(1998) and Fouda (2000) on true crabs.

Although the present results provide detailed information about the reproduction of porcelain crab, *P. rufescens*, as one of the most common intertidal decapods along the western coast of the Gulf of Suez, other studies are still required to assess their current status particularly at the same sites after exposing to negative impacts of the expanding constructions of tourist villages, resorts, camp sites and hotels, not only at the Gulf of Suez coasts but also allover the Red Sea Egyptian coasts. Therefore, these results represent the baseline data about this species in Ain Sukhna and can be used as a biological indicator for comparison with that will be taken at the present time after prevailing changes in natural habitats along the Gulf of Suez coasts through the first decade of the twenty first century.

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بيولوجية التكاثر في سرطان البورسيلين، بيتروليسثس روفيسينس(البورسيلينيدي، السرطانات ملتوية البطن، القشريات) من منطقة العين السخنة بخليج السويس، مصر

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الملخص العربي

أجريت هذه الدراسة على نـوع سـرطان البورسـيلين *Petrolisthes rufescens* المنتشر في المناطق المدية لشواطئ الساحل الشـمالي الغـربي لخليج السـويس في الفترة من أبريل 2000 إلى مايو 2001. وقد تم اختيار خمس منـاطق لإجـراء هذه الدراسة كانت من الجنوب إلى الشـمال: (1) جنـوب العين السـخنة، (2) بلاج الخيمـة، (3) بلاج السـماد, (4) شـاطيء المعهد القـومي لعلـوم البحـار والمصـايد بالسويس، (5) شاطيء مدينة السويس. ونظرا لتـأثر المنـاطق الشـمالية بكل من الأنشطة البشرية والتلوث البترولي معا كما بالمنطقتين 4&5، أو بزيادة النشـاط البشري والمعماري المتمثل بزيادة القرى والمنتجعات السياحة في المنطقتين 2& 3 جعل من الصعب متابعة إجراء الدراسات الحقلية بصـفة منتظمة بتلك المنـاطق، لذا فقد تم اختيار المنطقة الأولى (جنوب العين السخنة) لإجراء الدراسـات الحقلية والبيئية والبيولوجية بصـفة شـهرية نظـرا لكونها أبعد – حين ذاك عن تلك الأنشـطة سالفة الذكر وأقلها نوعا ما تأثرا بالنفط.

ولقد أوضحت النتائج المعملية والحقلية أن لنوع سرطان البورسيلين *P. rufescens* موسم تكاثر ممتد، يرتبط بظهور الإناث حاضنات البيض المخصب يبدأ مبكرا في أواخر الشتاء (فبراير) ويستمر طوال أشهر موسمي الربيع والصيف لينتهي في أوائل الخريف مع نهاية شهر سبتمبر، مع الزيادة الملحوظة في ارتفاع نسب تلك الإناث خاصة خلال فصلي الربيع والصيف، ودخول الأجيال الجديدة إلى بيئة الأطوار اليافعة (الآباء) بدءا من شهر يوليه ويستمر دخولها خلال موسمي الصيف والخريف. كما أشارت النتائج إلى الزيادة الواضحة في نسبة الذكور عن الإناث حيث كانت النسبة الشقية العامة 1: 0.70 (ذكور: إناث)، مع تأرجحها الموسمي واختلاف قيم أحد الشقين عن الآخر باختلاف المواسم . كما بينت النتائج وصول معظم الأطوار غير الناضجة من الذكور والإناث إلى مرحلة النضج الجنسي فيما بين 4- 6 مم من طول الدرقة لتصبح جميعها ناضجة جنسيا في الأطوار الأطول من 6 مم. وتظهرإناث هذا النوع إخصابية منخفضة نوعا ما، إذ تراوح عدد البويضات المحضنة تحت البطن فيما بين 35 و 450 بويضة بمتوسط 208± 92 بويضة / أنثى، وأظهرت التحاليل الإحصائية وجود علاقة ارتباط واضحة بين زيادة عدد البويضات وزيادة حجم الإناث. كما أوضحت القياسات أن قطر البويضات المحضنة تراوح فيما بين 400 و 1000 ميكرون بمتوسط عام 640 ± 105 ميكرون، مع ظهور تدرج في اللون مع تقدم مراحل النضج بدء من الأصفر ثم البرتقالي فالبني وصولا إلى الون الرمادي الشاحب لليرقات الكاملة قبيل الانطلاق.

ومما تجدر الإشارة إليه أن منطقة الدراسة بالعين السخنة كانت حينئذ شبه بعيدة عن تأثير الأنشطة البشرية، ولكن شوهد منذ الأشهر الأخيرة من الدراسة امتداد تلك الأنشطة إليها متمثلة في ردم الكثير من الأماكن الصخرية من أجل زيادة مواقع الاصطياف وكذلك تأثرها بزيادة التلوث النفطي مما يجعل هذه الدراسة بمثابة قاعدة بيانات يمكن الرجوع إليها لتقييم الوضع الراهن عن تلك المنطقة خاصة بعد أن أصبحت الآن تعج بالكثير من القرى السياحية وتعرضها للتلوث النفطي.