Mansour Galal G.Y., Osman S. Khalefa and M. Maamoun Zoo.Dept., Fac. of Sc., Menf. Univ., Shebin El-Koum.

ABSTRACT

The present investigation proved that *P. clarkii* at less-polluted area recorded higher numerical densities and body weights than polluted ones. It was found that the minimal water temperature was more or less similar at less-polluted and polluted areas, while its maximal values were higher at polluted ones. This could be referred to the cumulative thermal effect of both weather and pollution. At the same time, the lower values of dissolved oxygen were similar (5 mg/L) at both examined areas, while the higher values showed a slight tendency towards less-polluted areas (8 and 7 mg/L at less-polluted and polluted regions, respectively). Data of the present study proved that river water near El-Semad factory was more alkaline than that near the electrical power station at Talkha. Simultaneously, ammonia levels were higher at the polluted areas (0.13 - 0.55 mg/L) as compared with those belonging to the less-polluted area (0.06 - 0.3 mg/L).

Finally, it was exhibited that the reproductive capacity of *P. clarkii* in this study was higher than those of the other countries in the world particularly USA. This gives a good opportunity to rear this crustacean animal in a large scale to provide an efficient and cheap edible animal protein in Egypt. Therefore, China began to make a project near the River Nile in order to manufacture salty caned crayfish.

Key words : Crayfish, Numerical densities, P. clarkii.

INTRODUCTION

Procambarus clarkii have been introduced recently to Egypt in the early 1980's, when the first immigrants of this species were introduced from USA. The ecological surveys indicated that the crayfish had flourished and widely spread all over most of the River Nile and its tributaries (Ibrahim *et al.*, 1995). *P. clarkii* usually live in marshes, rivers, slow flowing water, reservoirs, irrigation systems, and rice fields. It is an aggressive competitor and its burrowing behavior may cause significant agricultural problems (Ibrahim and Khalil, 2009).

According to Emam and Khalil (1995), *P. clarkii* is heavily exploited as a fishery product. They mentioned that the average annual yield of *P. clarkia* in Egypt achieved 4.6tones/ year. More extensive ecological and experimental studies on this crayfish were made in Egypt (Emam and Mubarak, 1999; Ibrahim *et al.*, 2000; Emam, 2002 and Ibrahim *et al.*, 2005). From the medical point of view, *P. clarkii* might serve in controlling certain human diseases caused by helminthes since it has shown to subsist on the vectors of such pathogens (Hunner & Barr, 1991 and Hofkin *et al.*, 1991).

MATERIALS AND METHODS

Weekly sampling during the study period (November 2009 to December 2010) was carried out at three different stations on the Nile River at EL Mansoura city, Dakahleyia province (study area of about 4 km length and 150 to 200 meter width). These stations

represent a less-polluted area at EL Mashaia in the middle of the River, a polluted area near El-Mansoura fertilizer factory (EL Semad factory) and another polluted one connected to Talkha Electrical power station (Fig. 1). Numerical densities, weights and dimensions of the specimens belonging to ten catches per each location were tabulated. Simultaneously, water samples were collected in tightly closed clean glass vials from the same sampling regions to measure some physico-chemical parameters.

RESULTS

It was proved that the maximum number of *Procambarus clarkii* at less-polluted area achieved 640 animals on the second week of July, while that of polluted regions was 240 animals during the 4th week of April (Fig. 2). On the other hand, the minimum number of *P. clarkii* at less polluted region was 60 animals on the 4th week of January, while that of polluted area was 10 animals at the middle of November 2010.

Regarding the monthly numerical densities of total, male and female *P. clarkia* at various sampling regions, the maximal values were achieved during July and May at less-polluted and polluted regions, while their minimal values were gained on December and January, respectively (Table 1). The size of males was larger than that of females and the range of body length (from rostrum to the end of telson) for both males and females was 9 - 14 cm and 9 - 13.5 cm, respectively. The length of the carapace from tip of rostrum to the posterior margin of the cephalothorax of both sexes varied between 4.6 and 7cm. The colour of the adults varied from deep red or reddish-brown to almost black with deep red sides, but that of young is greenish-brown

The relationship between the monthly total numerical density of *P. clarkii* and the studying period, using the regression analysis, was proved not statistically significant (P > 0.05) at both less-polluted and polluted areas. The seasonal densities of this animal (Table 3) at less-polluted area ranged between 6920 on Summer and 1330 animals during Winter, while that of polluted regions varied from a maximum of 2460 on Spring and a minimum of 350 individuals on Autumn and these relations were proved not to be statistically significant (P>0.05).

Simultaneously, the relationship belonging to the numerical densities of male *P*. *clarkii* and the studying time was not statistically significant (P > 0.05) at both less-polluted and polluted regions. The seasonal variations of total male number varied between 4152 and 798 animals on Summer and Winter for less-polluted area, while those of polluted zones were 1476 on Spring and 210 animals during Autumn respectively.

On the other hand, the numerical density of female *P. clarkii* at less-polluted area fluctuated from 1136 to 148 individual during January and December, while those of the polluted areas declined from 360 to 56 animals on May and December. The relationship between those densities versus the studying time was proved not to be statistically significant (P > 0.05) at both less-polluted and polluted areas.

Simultaneously, seasonal female numbers exhibited 2768 and 532 animals as maximum and minimum values on Summer and Winter, while those of polluted regions varied between a maximum of 980 (Spring) and a minimum of 140 animals (Autumn).

Regarding the weight yield, it was proved that the maximum total weight was 32 kg/week on July, while the minimum one achieved 3 kg/week during Dec. and Jan. at less-polluted area (Fig. 3). The maximal total weight was 12 kg/week by the end of April and the minimum one was 0.5 kg/week during November at both polluted areas. This relationship proved a statistically significant one (P < 0.05).

Dividing the whole data of the previously mentioned figure, it was found that the maximum weight of the ascending part of the curve at less polluted area achieved 32 kg during the second week of July, while that of the polluted regions was 12 kg in the fourth

week of April. On the other hand, the minimum weight of *P. clarkii* belonging to the ascending part at less-polluted and polluted areas was 3, 1.5 kg in the fourth week of January respectively.

The maximal weight of the descending part at less-polluted and polluted areas was 28 and 9 kg in the 3^{rd} week of July, while the minimum weight of *P. clarkii* at less-polluted and polluted areas was 3 kg on December 2010 and 0.5 kg in the middle of November.

The monthly total weight of *P clarkii* decreased from maximum value (142kg) on July to minimum of 18.5 kg on Jan. 2009 for less-polluted area, while those of polluted area varied from a minimum of 7 kg on December to 45 kg maximally during May 2010 (Table 2). Examining the relationship between body weights and the studying time was proved to be not statistically significant (P > 0.05) at both less-polluted and polluted regions.

The seasonal total weights (Table 3) of that crustacean animal varied between 66.5 kg during Winter and 346 kg on Summer at less-polluted area, while those of the polluted ones were 17.5 and 123 kg during Autumn and Spring respectively. Accordingly, the seasonal relationship of the weight of *P. clarkii* and the studying time was not statistically significant (P > 0.05) at both less-polluted and polluted areas.

It was found that the monthly weights of male *P. clarkia* declined from a maximum of 85.2 kg on July to a minimum of 11.1 kg on Jan. 2010 for less-polluted area, while those of the polluted regions declined from 27 to 4.2 kg during May and December respectively (Table 2). This relationship was proved to be not a statistically significant one (P > 0.05) at both less-polluted and polluted areas.

The seasonal weight yield for male *P. clarkii* varied between maximum of 207.6 kg during Summer and a minimum of 39.9 kg on Winter for less-polluted area, while those of polluted areas varied between 10.5 and 73.8 kg during Autumn and Spring respectively. It is necessary to mention that the relation between the seasonal total weights of males and the studying time was not statistically significant (P > 0.05) at less-polluted and polluted regions.

On the other hand, monthly weights of female *P. clarkii* fluctuated from a maximum of 56.8 kg during July to a minimum of 7.4 kg on Jan. 2010 at less-polluted area, while those at polluted area decreased from 18 kg to 2.8 kg on May and December. Accordingly, the seasonal variations of female weights ranged between 138.4 and 26.6 kg during summer and Winter for less-polluted areas, while those of polluted regions increased from 7 to 49.2 kg on Autumn and Spring. The relation between female body weights and the studying time was proved to be not statistically significant (P > 0.05) at both less-polluted and polluted areas.

It was found that the monthly water temperature (Table 4) at the less-polluted station (El-Mashaia) increased gradually from 18 to 30.2 0 C on January and August 2010, while the polluted stations exhibited more obvious variations (from 17 to 36.5 0 C on December and June 2010 at EL Semad station and from 22 to 41 0 C during December and June 2010 at the Electrical power station). The relationship between water temperature and the studying time was proved to be statistically significant (*P* < 0.05) at less-polluted area, while those of EL Semad and the Electrical power stations were not statistically significant (*P* > 0.05).

Minimal and maximal values of dissolved oxygen at less-polluted station were 5 and 8 mg/L during October and September 2010, while those of polluted stations achieved 5 mg/L at Electricity station and 7 mg/L at EL Semad factory station (Table 4). The relationships between dissolved oxygen and the studying time (monthly and seasonally) were proved to be not statistically significant (P > 0.05) at less-polluted and polluted areas.

The pH of the water samples belonging to the different regions was mostly alkaline as could be seen in Table (4). The monthly values of pH at the less-polluted station ranged from 7.53 to 7.8 on August and December respectively, while those of both polluted stations varied from 7.05 to 8.3 during April near by Electricity station and on November at EL

Semad factory station. This relationship was proved to be statistically significant (P < 0.05). Simultaneously, the seasonal levels fluctuated between 7.59 on summer and 7.74 during Spring at less-polluted station, while those of polluted ones changed from 7.47 on Spring at Electricity station to 7.93 on Autumn at EL Semad station and this relation was not statistically significant (P > 0.05).

The monthly levels of ammonia were changed from 0.06 to 0.3 mg/l on October and February, from 0.13 to 0.5 mg/l during April and July and from 0.2 to 0.6 mg/l on April and July at less polluted, El-semad and Electricity sampling stations respectively. This relationship was proved to be significant (P<0.05) at less polluted station and insignificant (P>0.05) at both polluted ones.

DISCUSSION

It is worthy to keep in mind that data of male, female plus their ascending and descending parts of both weekly numerical densities and body weights of this crustacean animal are statistically significant with the studying time (0.05 > P < 0.001) at both less-polluted and polluted areas.

Comparing the present data with others indicated that *P. clarkii* have different body colour, length and size in adults and youngsters. Huner and Romaire (1978) found that body size was directly influenced by habitat quality. Total body length of *P. clarkii* changed from place to anther. In Egypt a range varied from 4.4 cm to 12.4 cm for adult (Mubarak; 2001), but in Louisiana total length did not exceed 11.6 cm and 47 gram body weight (Penn, 1943), *P. clarkii* in pond near Zurich (Switzertand) was10.05 cm and its life span was 3 years (Frutiger *et al.*, 1999).

Sex ratio in the present study proved that the numerical density of male is more than that of females (1.5:1), while that of Frutiger et al. (1999) was 1.33: 1.0. The highest mortality (90%) occurred when all individuals were belonging to the same sex due to their aggressive behavior (Raafat, 2006). It was found that size of male was larger than that of female for two reasons; the lower frequency of moulting and the feeding inhibition in females during the studying period (Skurdal and Qvenlid, 1986; Pursiainen et al., 1987). Simultaneously, P. clarkii in the present study have two generations per year; the first generation was obtained during April and early May and the second happened on late September and October, while that in southern United States have one generation per year in early summer (Huner and Barr, (1991). This means that reproductive capacity for P. clarkii is very high in the Egyptian water (Soliman et al., 1998 b). This crayfish began the active season on late March when the temperature increases and the water level in different channels and ditches raised up after the winter closure period (Mubarak, 1996). Accordingly, water temperature accounts as one of the main factors that affect on life, growth, metabolic activity, food intake and reproduction for P. clarkii. At less-polluted area the water temperature had normal range (18 °C to 30.2 °C) but the polluted area had abnormal range of water temperature especially near the electrical power station where its values varied between 22 °C and 41 °C, while those near EL Semad factory changed from 17 °C to 36 °C. Espina et al. (1993) found that *P. clarkii* preferred temperature at 23.4°C, while a temperature range of 20 - 29° C had no effect on the developmental stages or the acclimation for P. clarkii. Habashy (2004) reported that, the relation between temperature and growth rate of *P. clarkii* in Egypt was significant (P <0.05) and the growth rate varied at different temperature levels (15, 20, 25 and 30 °C). He proved also that growth at higher temperature (30 °C) was almost three times greater than that at lowest temperature. Jussila and Evans, (1996) reported that growth rate was higher at optimum temperature and decreased when water temperature deviated from the optimum one. Peck (1985) mentioned that as water temperature affected the solubility of oxygen and many other factors in water, it influenced the abundance of the crayfish at various habitats. Mickie et al. (2006) proved that high temperature (35 °C) lead to a decreased metabolic rate. Low temperature acclimation (10 °C) resulted *in* 20% mortality in *P. clarkii*, while acclimations at 30 °C did not have any effect on the metabolic rate. Ackefors, (1999) reported that, in Lake Trasimeno optimal temperature was 21-27°C and growth inhibition occurred at temperatures below 12°C.

Ibrahim et al. (2000) found that water temperature affected growth of P. clarkii by its influence on moulting interval; when temperature changed from 18 to 30 °C the moulting time interval decreased from 63.2to 21.4 days for mature and from 26.6 to11.6 days for immature animal. Huner (1988) reported that moulting rates increase in temperatures between 22 to 30 °C. Hartnoll (1983) mentioned that temperature and food supply affected the intermoult duration. Kumlu et al. (2000) reported that high temperature at certain point increased the moulting frequency and consequently growth. Soliman, et al. (1998) suggested that mating activity of P. clarkii was present all the year round, but increased in May. Females kept sperms in its seminal receptacle for about 2-8 months until time of fertilization. Richardson et al. (2007) observed that the ovarian development in mature females of this animal was temperature dependent. It was reported that temperature affected the period of egg hatching which usually last about 3 weeks (Gherardi, 2002). Simultaneously, Soliman, et al. (1998) found that the period of embryonic development usually spent about 10 to 20 days, depending on water temperature as well. Ibrahim et al. (2006) found that, in Egypt, temperature and photoperiod influenced egg laying in P.clarkii. Also, when temperature exceeds 30 °C it caused high morality, lower percentage of ovigerous female and increasing the time of egg laying. At 27 °C, P.clarkii took short time to egg laying and highest percentage of ovigerous at 29 °C.

France (1984 and 1985) and Berrill *et al.* (1985) showed that crayfish could tolerate high range of pH 6.5 to 8.5, while Ibrahim *et al.* (2006) found that the water pH should range from 6.5 to 7.5. France (1984) mentioned that low pH (< 5.5) could result in eventual population extinction due to mortality of juveniles. Jensen and Malte (1990) ; Ellis and Morris (1995) reported that *P. clarkii* could tolerate pH 2 for 24 hour which caused 100% mortality but pH 3 caused 50% mortality in four days, while McMahon and Stuart (1989) proved that *P. clarkii* could tolerate pH 4 for 60 days.

Dissolved oxygen is a basic requirement needed for healthy crustacean, but *P. clarkii* can depend on atmospheric oxygen to meet its oxygen needs by moving to more highly oxygenated water or by turning on their sides at the water surface in order to utilize atmospheric oxygen (Penn 1943, Konikoff 1977, Huner and Barr 1984). It was found that the dissolved oxygen at the less-polluted area ranged from 5 to 8 mg/l, while at polluted area it ranged from 5 to 7 mg/l (El-Semd factory) and from 5 to 6.2 mg/l near Talkha station. Moreover, Hobbs (1975) and Huner and Lindqvist (1995) mentioned that crayfish can adapt itself by increasing its gill filament surface and hence use the atmospheric oxygen. Vander (2003) mentioned that crustaceans could suffer from suffocation when dissolved oxygen concentrations declined to 3-4 mg/L.

Ammonia is toxic to a variety of aquatic organisms including fish (Harris *et al.* 1998). Also un-ionized form of ammonia account the most toxic form to aquatic organisms because it can readily diffuse through cell membranes, highly soluble in liquids causing impairment of cerebral energy metabolism and damaging gill, liver, kidney, spleen and thyroid tissue in fish, crustaceans and molluscs. (Smart, 1978). Reid (1961) reported that the concentration of ammonia (1 mg/l) has been given as an indicator of organic pollution and ammonia is toxic in concentration over 2.5 mg/l to aquatic organism. Simultaneously, the severe biological effect of nitrite on crustacean species was indicated by Rouse and Yeh (1995), since under the effect of nitrite, crustacean haemocyanin changes to methoglobin causing hypoxia and cyanosis.

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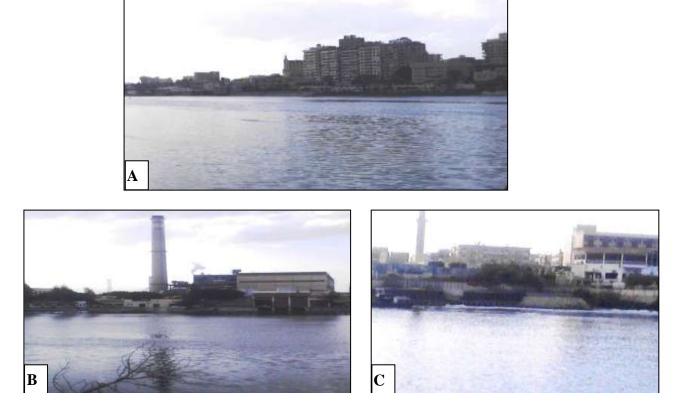
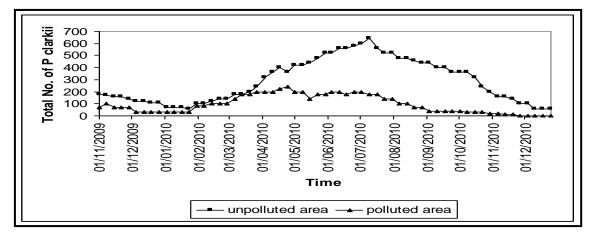
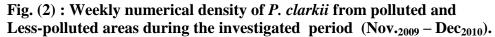


Fig. (1). Sampling stations at less-polluted and polluted regions at El-Mansoura city. A - Less-polluted area at El-Mashaia

B - Chemically polluted region at El- Semad factory

C – Thermally polluted area at Talkha





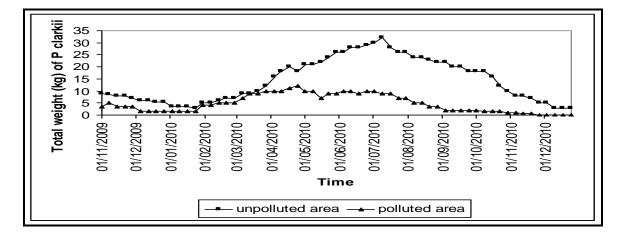


Fig. (3): Weekly variations of total weight (kg) of *P. clarkii* from polluted and less-polluted areas during the studying period.

 Table (1) Monthly numerical densities of male, female and total of *P. clarkii* at less-polluted and polluted areas during the present study.

Month Area	Nov. 2009	Dec. 2009	Jan. 2010	Feb.	Mar.	Apr.	May.	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
<u>Less-pollute</u> d Total no.	810	460	370	500	800	1440	2280	2220	2840	1860	1600	1480	560	280
Male no.	486	276	222	300	480	864	1368	1332	1704	1116	960	888	336	168
Female no.	324	184	148	200	320	576	912	888	1136	744	640	592	224	112
<u>Polluted</u> Total no.	380	140	205	380	700	860	900	780	840	340	160	150	40	0
Male no.	228	84	123	228	420	516	540	468	504	204	96	90	24	0
Female no.	152	56	82	152	280	344	360	312	336	136	64	60	16	0

Table (2): Monthly body weights of male, female and total of *P. clarkii* (Kg) at less-polluted and p olluted areas during the present study.

Month Area	Nov. 2009	Dec	Jan. 2010	Feb.	Mar	Apr.	May ·	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
<u>Less-polluted</u> Total wt.	40.5	23	18.5	25	40	72	114	111	142	93	80	74	28	14
Male wt.	24.3	13.8	11.1	15	24	43.2	68.4	66.6	85.2	55.8	48	44.4	16.8	8.4
Female wt.	16.2	9.2	7.4	10	16	28.8	45.6	44.4	56.8	37.2	32	29.6	11.2	5.6
<u>Polluted</u> Total wt.	19	7	10.25	19	35	43	45	39	42	17	8	7.5	2	0
Male wt.	11.4	4.2	6.15	11.4	21	25.8	27	23.4	25.2	10.2	4.8	4.5	1.2	0
Femle wt.	7.6	2.8	4.1	7.6	14	17.2	18	15.6	16.8	6.8	3.2	3	0.8	0

Table (3) Seasonal variations of the numerical densities and body weights of *P. clarkii* at less-polluted and polluted regions during the present study.

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Seasons Areas	Win.	Spr.	Sum.	Aut.	Win.	Spr.	Sum.	Aut.
<u>Less-</u> <u>polluted</u> Total no.	1330	4520	6920	3640	66.5	226	346	182
Male no.	798	2712	4152	2184	39.9	135.6	207.6	109.2
Female no.	532	1808	2768	1456	26.6	90.4	138.4	72.8
<u>Polluted</u> Total no.	725	2460	1960	350	36.25	123	98	17.5
Male no.	435	1476	1176	210	21.75	73.8	58.8	10.5
Female no.	290	980	784	140	14.5	49.2	39.2	7

 Table (4) Measurements of certain physico-chemical parameters at less-polluted and polluted areas of the Nile sector in El-Mansoura city.

	1- Water temperature													
Months	Nov. 2009	Dec.	Jan. 2010	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
ELMashia station (Control area)	21.4	19.1	18	18.7	20.5	24.5	26.5	27	28	30.2	27.5	27.2	25	22
EL Semad station (Polluted area)	21	21	18.2	22.9	19.8	26.7	26.5	36.5	31.2	35	32	25	22.5	17
Electricity station (Polluted area)	25.5	26	22.4	26.1	24.6	33.1	31.2	41	36	40	37.2	29	27	22

_	2- Dissolved Oxygen														
	ELMashia st.	6	6	6	5.7	6.1	5.9	7.3	5.8	5.3	5.2	8	5	6	6.8
	EL Semad st.	5.5	5.4	5.8	7	5.5	6	6.1	6.4	5.5	5.3	5.5	5.9	5.4	6
	Electricity st.	5	5.1	5.4	6.2	5	5.7	5.6	5.9	5	5.1	5.3	5.5	5	5.5

					3- pH									
ELMashia st.	7.75	7.8	7.7	7.7	7.7	7.72	7.8	7.61	7.62	7.53	7.6	7.57	7.74	7.65
EL Semad st.	8.3	7.9	7.75	7.79	7.85	7.3	7.8	7.9	7.7	8	8.1	7.8	7.89	7.9
Electricity st.	8.27	7.85	7.8	7.7	7.6	7.05	7.75	7.8	7.6	7.95	8	7.7	7.8	7.8

ELMashia st.	0.2	0.17	0.19	0.3	0.21	0.15	0.1	0.16	0.2	0.14	0.1	0.06	0.19	0.12
EL Semad st.	0.29	0.35	0.21	0.38	0.25	0.13	0.32	0.35	0.5	0.3	0.27	0.38	0.4	0.33
Electricity st.	0.39	0.46	0.35	0.55	0.38	0.2	0.41	0.52	0.6	0.4	0.32	0.45	0.48	0.39

4-Amonia

دراسات بيئية على استاكوزا الماء الغب بروكامبرس كلاركى فى نهر النيل بمدينة المنصورة

منصور جلال جمالات عثمان شيرين خليفة منى مأمون قسم علم الحيوان كلية العلوم جامعة المنوفية شبين الكوم

المستخلص

أجريت هذه الدراسة على حيوان استاكوزا الماء العذب "بروكامبرس كلاركى" فى فرع دمياط بنهر النيل بمدينة المنصورة - محافظة الدقهلية. ولقد ثبت من تلك الدراسة أن عشائر هذا الحيوان القشرى تكثر بصورة واضحة فى الأماكن الأقل تلوثآ سواء من ناحية الاعداد أو الأوزان. ولقد أثبتت هذه الدراسة أيضا أن قيم الحد الادنى لكل من درجة حرارة الماء والاكسجين المذاب تكون متساوية بينما الحد الاقصى لهما يكون اعلى فى المناطق الاقل تلوثآ. ومما هو جدير بالذكر أن المناطق الاكثر تلوثآ تكون كمية القلوية بينما الحد الاقصى لهما يكون اعلى فى المناطق الاقل تلوثآ. ومما هو جدير الامونيا. ولقد اتضح أيضاً من هذه الدراسة ان الكثافة العددية لهذا الحيوان كانت أعلى من مثيلاتها من الامونيا. ولقد اتضح أيضاً من هذه الدراسة ان الكثافة العددية لهذا الحيوان كانت أعلى من مثيلاتها فى الأمون خاصة أماكن تواجدها الاصلية فى الولايات المتحدة الامريكية. ولهذا يمكن التوصية باستزراع هذا الحيوان القشرى بمصر لتوفير مصدر للبروتين الحيوانى الرخيص ،ولقد بدأت دولة الصين بعمل مشروع بالقرب من نهر النيل لتعليب هذه التوفير مصدر للبروتين الحيوانى الرخيص ،ولقد بدأت دولة الصين بعمل مشروع بالقرب من نهر النيل لمكلار كي