Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2015; 3(3): 339-343

Received: 15-04-2015
Accepted: 18-05-2015

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Effect of oil pollution on polychaete annelids in the Algerian East coast

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Abstract
This study was to assess the marine environment quality by hydrocarbon contamination along the east Algerian coasts using an approach based on biomarkers and hydrocarbon rate in the polychaete *Perinereis cultrifera.* Individuals were collected from two sites: Skikda considered as a polluted site and from El-Kala as a reference site. During this study two approaches have been maintained: the first was the analysis of physico-chemical parameters of the seawater, where no significant differences were recorded for temperature, salinity and DBO_5, unlike the rate of dissolved O_2, pH and oil content in the seawater. The second approach was devoted to measure the biomarkers activity of glutathione S-transferase as phase II enzyme in the polychaete *P. cultrifera.* This approach confirms that individuals from Skikda have been submitted to highly polluted environment.

Keywords: *Perinereis cultrifera,* Algerian east coast, marine pollution, total hydrocarbons, biomarkers

1. Introduction

The polychaete *Perinereis cultrifera* (Nereidid) was described for the first time by Grübe [1] from the Adriatic Sea. It occurs along the north-western coasts of Europe and the Mediterranean. This species has also been described in the Indian and Pacific Oceans [2, 3]. According to the geographical location of the populations, mode of reproduction differs largely [4]. Reproduction in the English Channel and the Atlantic is of an epitokous type [5,6], as in the Mediterranean Sea at Salammbô near Tunis, at Annaba on the Algerian Mediterranean coast near the Tunisian border and in the Venice Lagoon in Italy [7, 8]. However, on the west coast of Algeria in the Bay of Algiers, the reproduction has been described as atokous [9], as on the Moroccan Atlantic coast [10] and the Gulf of Marseille [11]. *P.cultrifera* is an intertidal poor disperser polychaete. It is reported to be a free-spawner.

The Mediterranean area has been classified by UNEP as one of the five regions of the world where environmental problems are most severe [12], while the Mediterranean Sea is ranked among the seven most threatened seas [13].The coastal marine environment, widely exploited for its economic wealth (fishing, mariculture) and tourism. The coastal marine environment is widely exploited for its economic wealth (fishing, mariculture) and tourism. The marine ecosystem sufferers from several types of pollution; from direct discharges (municipal and industrial effluents, oil spills) and indirect discharges (riverine and atmospheric) [14]. Algeria, a producer of fossil fuel (oil and gas), is strongly influenced by the different hydrocarbons. Skikda city remains one of the most illustrative exemple of the oil pollution in the country. Indeed, Skikda has a platform dedicated to the petrochemical industry and is the main source of hydrocarbon pollution. Accumulation in the marine environment of chemicals from human activities is a major concern in environmental biomonitoring [15].

Biomonitoring of environmental quality depends on bioindicators that are organisms or groups of organisms used to determine the presence, abundance and bioavailability of environmental contaminants through concentrations in these organisms in their totality body, one or more organs and tissues [16]. Polychaete annelids are well represented in marine environments and constituted a significant percentage of the total biodiversity and abundance of benthic macrofauna. Polychaetes are the dominant macrofauna within fine sediments [17] and are commonly used in toxicological studies [18].Currently scientists and environmental managers work for a more specific concepts of the marine environment biomonitoring based on the study of the biological response by measuring biomarkers in pollution bioindicator species [19].

Biomarkers measure the interaction between a biological system and an environmental agent.

The inhibition or induction *in vivo* biomarker is a good tool to assess environmental exposure and potential effects of xenobiotics on organisms [20]. The ability of an organism to adapt to an environment altered by anthropogenic contamination depends mainly effective mechanisms for the detoxification of endogenous and exogenous various compounds [21].

The aim of this study is to evaluate water quality at two sites by assaying total hydrocarbons in seawater, and the determination of enzymatic activity of Glutathione S-transferase in *P. cultrifera* (Annelida, Polychaetes) on the eastern coast of Algeria.

2. Materials and methods

2.1 Sampling sites

Sampling sites were chosen because of their geographical locations in eastern Algeria (Figure 1). Site selection was based on the level of pollution as well as ease of access to the study area and abundance of species. El-Kala (36°53'44" N; 08°26'35" E) is close to the Tunisian border (10 km). This site is part of a national park and is not urbanized, therefore, it was considered as the healthy reference site. And Skikda (36°45'0" N; 06°49'60" E) is located about 180 km from the Tunisian border. This site is near from Stora port that is characterized by an intense maritime traffic**.** This site is also polluted by PAH because of the presence of an important petrochemical complex and human activities.

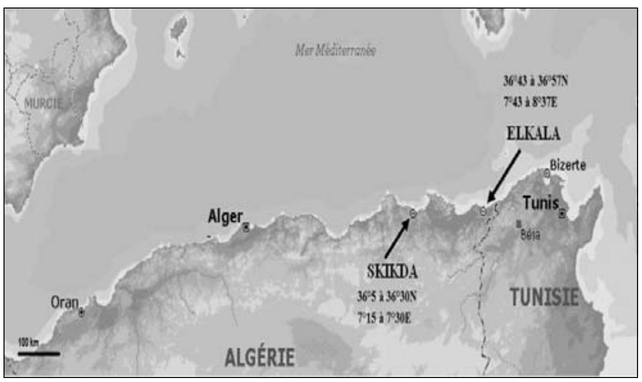

Fig 1: Location of the different sampling sites along the east Algerian coast: El-Kala and Skikda.

2.2 Collection of individuals

Individuals were collected in the intertidal zone within Rhodophyceae, in algal-covered hard bottoms monthly from February to July 2013.The region sampled at each site corresponded with the area of greatest density of individuals. The individuals occur low in the intertidal zone and extend down into the sublittoral; in consequence, the intertidal and shallow sublittoral hard bottoms were sampled methodically by scraping algae and looking for individuals [22].

2.3 Abiotic parameters

Seawater temperature, pH, dissolved O_2 and salinity were measured *in situ* with a Multiparameter-Oximeter (Multi 340 i/SET) at each site monthly from February to July 2013.

2.4 Hydrocarbons measurement

Three samples of seawater were taken at each site at about three meters from the seafront and a foot deep. For the determination of oil contents, samples were taken in black polyethylene bottles and transported in coolers at 4° C in order to avoid contamination of the samples (including the oil content) [23].

The extraction of oil from the samples is carried out according to standard AFNOR T 90-114 [24]. Total hydrocarbons consist of extractables by trichloromethane (CHCl3) in acid medium, then assayed after purification by ultraviolet spectrophotometry at a wavelength of 400 nm (according the Beer lambert's law).

2.5 Glutathione S-transferase activity determination

Individuals were homogenized at 4°C in 100 mM sodium phosphate buffer, pH 6. Homogenates were then centrifuged 13000 × *g* for 30 min at 4°C. The supernatant of each sample was stored at -20°C. Total protein content in the homogenate was determined according to Bradford [25] at 595 nm using Bovine Serum Albumin as standard.

Glutathione S-transferase (GST) activity was assayed by the method described by Habig *et al.* [26] using 1-chloro-2,4-dinitrobenzene as substrate and glutathione (1 and 4 mM final concentration, respectively) in 100 mM sodium phosphate buffer, pH 6. All GST activity assays were realized in conditions of linearity with respect to incubation time. The results were expressed as micromoles produced per minute per milligram protein.

2.6 Statistical analysis

Data were expressed as means ± standard deviation (S.D). The normality of the distribution was tested using the Shapiro-Wilk test. To assess multiple comparisons, a parametric one-way analysis of variance (ANOVA) was performed on data with a Tukey's test. A significant difference was assumed when $p < 0.05$. All statistical analyses were performed using Minitab statistical software (Version 16.2.0).

3. Results and discussion

3.1 Abiotic parameters and total hydrocarbons

The physico-chemical parameters were measured *in situ* throughout the duration of the study at the two study sites (Table 1). The evaluation of marine ecosystem quality could be done by the study of its abiotic and biotic components [27]. In fact, water temperature, salinity, pH and dissolved oxygen are important components of water quality [28].We did not observe any significant differences ($p > 0.05$) between the study sites for seawater temperature. The lower values were observed at Skikda (Bikini Djedid beach) (17.96 ± 2.7 °C); while the higher values of temperature were noticed at El-Kala (El-Morjène beach) (18.53 ± 1.9 °C). Ali and Soltan [29] have reported an increase of water temperature, a decrease of the dissolved oxygen and a neutral pH (due to alkaline effluents), in water samples taken from the polluted sites of Nile River. The same authors have highlighted that the increase of water temperature (caused by pollution) may be responsible for the reduction of dissolved oxygen concentrations, because oxygen is less dissolved in warm water. The mean values of pH was 7.88 ± 0.15 at El-Kala (El-Morjène beach) and 8.39 ± 0.1 at Skikda (Bikini Djedid beach). We observed trends ($p < 0.05$) of dissolved O_2 values, where the higher values was 8.08 ± 1.1 mg/l at El-Kala (El-Morjène beach) and 4.63 ± 0.55 mg/l at Skikda (Bikini Djedid beach). We noticed a slight decrease of the dissolved oxygen level in water samples collected from sites receiving pollutants Skikda. Dissolved oxygen is consumed during the heterotrophic oxidation of organic matter and respiration by aquatic flora and fauna.

We did not observe trends ($p > 0.05$) of salinity between El-Kala and Skikda, the mean values was 36.16 ± 0.3 g/l at El-Kala (El-Morjène beach) and 36.23 ± 0.46 g/l at Skikda (Bikini Djedid beach). The average content in the non-contaminated surface water is 8 mg /l and does not exceed 10 mg /L. Indeed, salinity with temperature and atmospheric pressure determine the solubility of oxygen. The data contents shows that dissolved oxygen in El-Kala is close to the average of 8.08 ± 1.1 mg /l, unlike at Skikda 4.63 ± 0.55 mg /l.

We did not observed variations of DBO_5 concentration between the two sites. The values were lower at El-Kala (El-Morjène beach) than Skikda (Bikini Djedid beach) with respectively 3.67 ± 0.85 mg/l and 4.1 ± 0.56 mg/l. Also, DBO_5 reflects the amount of molecular oxygen used by microorganisms during an incubation period of 5 days at 20° C to decompose the dissolved or suspended organic matter contained in one liter of water. According to Chapman & Kimstach [28], DBO_5 natural waters is less than 2 mg /l. Waters receiving domestic discharges are greater than 10 mg / l concentrations. However, the concentration of hydrocarbons in seawater was significantly higher ($p < 0.05$) at Skikda (4.78 ± 0.666 mg/l) than at El-Morjène (El-Kala) (2.87 ± 0.058 mg/l) (Figure 2).

Hydrocarbons, have adverse effects on the biota. Planktonic organisms are integral water masses, are particularly vulnerable to oil spills, which inhibit the photosynthetic activity of marine phytoplankton. The levels determined are well above 1 ppm, eliminating all the larvae of zooplankton in two or three days [30]. Rouidi [31] noticed high concentrations of hydrocarbons 75.92 mg/ l at the Oued Marhoum and 32.12 mg /l at the Oued Saf Saf.

Table 1: Physico-chemical parameters and hydrocarbons concentrations of seawater from El-Kala (El-Morjène beach) and Skikda (Bikini Djadid beach) during 2013.

	Temperature °C	pH	dissolved O_2 mg/l	Salinity g/l	DBO_5 mg/l	Hydrocarbons mg/l
El-Kala	18.53 ± 1.9	7.88 ± 0.15	8.08 ± 1.1	36.16 ± 0.3	3.67 ± 0.85	2.87 ± 0.058
Skikda	17.96 ± 2.7	8.39 ± 0.1	4.63 ± 0.55	36.23 ± 0.46	4.1 ± 0.56	4.78 ± 0.666

Mean ± SD, n=12.

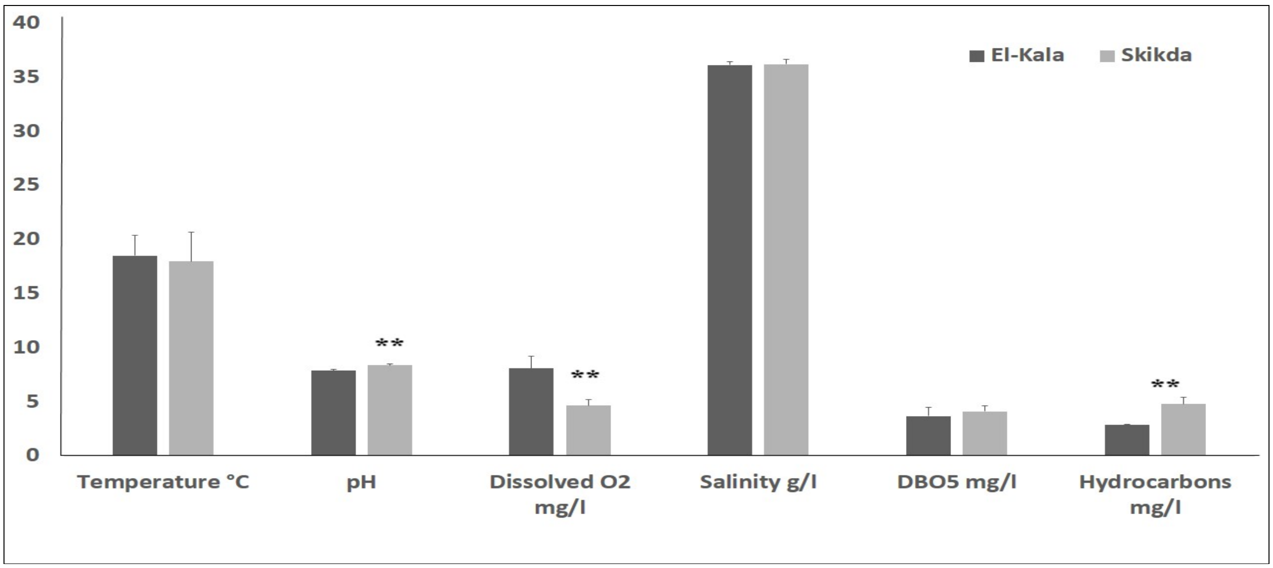

Fig 2: Physico-chemicals parameters and hydrocarbons concentrations of seawater from El-Kala (El-Morjène beach), and Skikda (Bikini Djadid beach) during 2013.

3.2 GST activity

We observed seasonal trends ($p < 0.05$) of GST activity at each sites. Globally, at El-Kala, GST activity was maximal in Spring at El-Kala, then it decreased in summer. In Skikda, GST activity increased in summer (Table 2). These enzymes are generally soluble (cytosolic) and present in several isoforms, some of which are inducible by type contaminants PAHs, PCBs and pesticides in the marine environment that make them less toxic [32]. The higher values of GST activity were observed in May 6.85 ± 0.5 µmol $min^{-1}mg^{-1}$ protein and in July 8.97 ± 0.83 $µmol^{-1}min^{-1}mg$ protein for individuals collected at El-Kala and at Skikda (Bikini Djadid beach) respectively, while the lower values were observed in February (5.67 ± 0.64; 7.78 ± 0.9 $µmol^{-1}min^{-1}mg$ protein) respectively at El-Kala (El-Morjène) and at Skikda (Bikini Djadid beach). We showed an increase of GST activity in worms sampled from altered sites. These findings are similar to those obtained for the worms *N. diversicolor* collected from polluted estuary of the Seine [33] and from the Oued Souss (Bay of Agadir, Morocco) before implantation of wastewater treatment [34]. GST activity was significantly lower ($p < 0.05$) in El-Kala compared to Annaba and Skikda throughout the studied period (Figure 3). GST activity was significantly more important ($p < 0.05$) in Skikda compared to Annaba throughout the studied period. GST is a phase II enzyme involved in the metabolism of lipophilic organic contaminants such as PAHs and PCBs detected at comparatively high levels in the Seine estuary [33]. In addition, this enzyme plays a role in cellular protection against oxidative stress which can be triggered by pollutants such as metals, PCBs and PAHs [35].

Table 2: Monthly variations of GST activity expressed as $µmol^{-1}min^{-1}mg$ protein for individuals collected in El-Kala and Skikda from February to July 2013.

	Février	Mars	Avril	Mai	Juin	Juillet
El-Kala	5.67 ± 0.64	5.91 ± 0.47	6.07 ± 1.05	6.85 ± 0.5	6.33 ± 0.8	6.53 ± 0.71
Skikda	7.78 ± 0.9	8.56 ± 0.92	8.96 ± 0.65	8.21 ± 0.78	8.59 ± 0.92	8.97 ± 0.83

Mean ± SD, n=4.

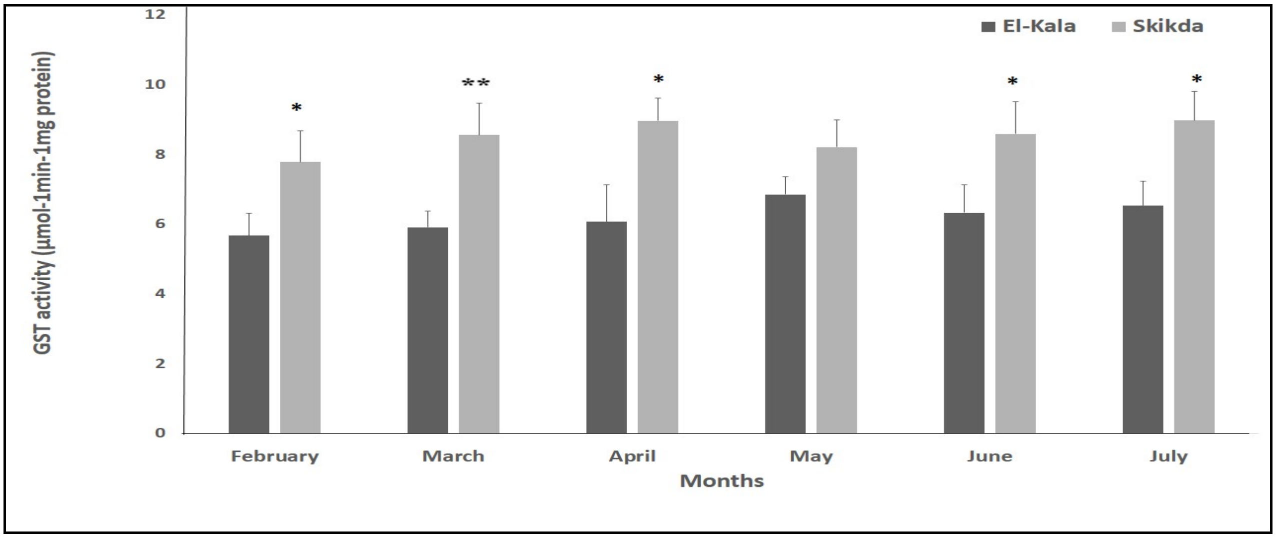

Fig 3: Monthly variations of GST activity expressed as µmol^{-1}min^{-1}mg protein for individuals collected in El- Kala and Skikda from February to July 2013.

4. Conclusion

This work allows the surveillance of physico-chemical parameters and their variations in the presence of oil pollution in Skikda. For costal biomonitoring, an assessment of the detoxification enzyme GST has been reported in *P. cultrifera*. This approach confirms there is an obvious correlation between contamination of the sites by hydrocarbons and the studied physico-chemical parameters. We can conclude that the biomarkers responses can reflect the pollution degrees of the site of Skikda and predict the ecological effects of pollutants.

5. Acknowledgement

I thank laboratory staff of the petrochemical complex of Skikda for their assistance during stage of the study.

6. References

1. Grübe E. Actinien Echinodermen und Würmer des Adriatischen und Mittelmeers. J. H. Bon, königsberg, 1840, 92.
2. Fauvel P. Polychètes errantes. In : Faune de France 4. Lechevaliers, Paris, 1923, 488.
3. Durchon, M. Problèmes posés par le comportement des néréidiens au moment de leur reproduction. Année Biologique 1957; 33: 31-42.
4. Scaps P, Rouabah A. Le complexe d'espèce *Perinereis cultrifera*: un exemple de spéciation entre l'Atlantique et la Méditerranée. Bulletin de la Société zoologique de France 2001; 126(1-2):35-48.
5. Herpin R. Recherches biologiques sur la reproduction et le développement de quelques annélides polychètes. Bulletin de la Société des Sciences Naturelles de l'Ouest de la France 1925 ; 4:1-250.
6. Durchon M. Les modalités de l'essaimage de *Perinereis cultrifera* Grübe (Annélide Polychète) à Luc-sur-Mer (Calvados). Archives de Zoologie Expérimentale et Générale 1951; 88:1-6.
7. Ansaloni I, Pellizzato M, Predevelli D, Zunarelli-Vandini R. Policheti di interresse economico nella laguna di Venezia. Nova Thalassia 1986 ; 8:641-642.
8. Zghal F, Ben Amor Z. Sur la présence en Méditerranée de la race *Perinereis cultrifera* (Polychète). Archives de l'institut Pasteur de Tunis 1989; 66:293-301.
9. Rouabah L, Rouabah A, Ferroudj S, Scaps P. Comparison of the life cycles of two populations of the polychaete *Perinereis cf. cultrifera* from the Bay of Algiers (Mediterranean Sea). Scientia Marina 2008; 72:769-778.
10. Rouhi A, Sif J, Gillet P, Deutsch L. Reproduction and population dynamics of *Perinereis cultrifera* (Polychaeta: Nereididae) from the Atlantic coast of El Jadida, Morocco. Cahiers de Biologie Marine 2008; 49:151-160.
11. Peres JM, Rancurel P. Observations sur la ponte de *Perinereis cultrifera* Grübe dans la région de Marseille. Bulletin de la Société zoologique de France 1948; 73:97-100.
12. Ramade F. Dictionnaire encyclopédique des sciences de l'environnement. Ediscience. Paris, 1993, 822.
13. Boudouresque CF. Impact de l'homme et conservation du milieu marin en Méditerranée. GIS Posidonie publ., 2ème édit., Marseille, 1996, 1-243.
14. Rudolph A, Rudolph MI. Activity of Benzo(a)pyrene Hydroxylase in Three Marine Species. Bulletin of environmental contamination and toxicology 1999; 63:639-645.
15. Abbes A, Chouahda S, Soltani N. Actes des 6ème journnées de l'INSTM. Tunisie. bulletin de l'institut national des sciences et technologies de la mer, 2003, 8.
16. Chambost-Manciet Y. Ampleur et effets biologiques de la contamination métallique (Cd, Cu,Fe, PB et Zn) des sédiments en Mer du Nord. Utilisation de l'étoile de mer Asterias rubens (L.)comme bioindicateur et biomarqueur (mémoire). Université Libre de Bruxelles, Bruxelles, 2002.
17. Fauchald K. The polychaete worms. Definitions and keys to the orders, families and genera. Natural History Museum of Los Angeles County, Science Series, 28, Los Angeles. 1977,188.
18. Lucan-Bouché ML, Biagianti-Risbourg, S, Arsac F, Vernet G. An original decontamination process developed by aquatic oligochaete Tubifex tubifex exposed to copper and lead. Aquatic Toxicology 1999; 45: 9-17.
19. Bouzeraa N, Abbes A, Soltani N. Actes des 7ème journnées de l'INSTM. Tunisie, bulletin de l'institut national des sciences et technologies de la mer 2004, 9.
20. Dembele K, Haubruge E, Gaspar C. Concentration Effects of Selected Insecticides on Brain acetylcholinesterase in the Common Carp (Cyprinus Carpio L.) Ecotoxicology

and Environmental Safety 2000; 45:49-54.
21. Jakanovic M. Biotransformation of organophosphorus compounds. Toxicology 2001; 166:139-160.
22. Rouabah A, Scaps P. Life cycle and population dynamics of the polychaete *Perinereis cultrifera* from the Algerian Mediterranean Coast. P.S.Z.N: Marine Ecology 2003b; 24(2):85-99.
23. Rodier J. L'analyse de l'eau: eaux naturelles, eaux résiduaires, eaux de mer. Chimie, physico-chimie, bactériologie, biologie. 8ème Ed. Dunod Bordas, Paris, 1996, 1135.
24. Norme AFNOR NT F. Détermination de la demande biochimique en oxygène (DBO), 1975. 90-103.
25. Bradford MM. A rapid and sensitive methods for the quantification of microgram quantities of protein utilizing the principe of protein-dye brinding. Analytical Biochemistry 1976; 72:278-254.
26. Habig WH, Pabst MJ, Jakoby WB. The first enzymatic step in mercapturic acid formation. Journal of Biological Chemistry 1974; 249:7130-7139.
27. Pai I. Ecology of Andaman Sea: past, present and future. Journal of Cell and Animal Biology 2007; 1(1):11-14.
28. Chapman D, Kimstach V. Selection of water quality variables. Water quality assessments: a guide to the use of biota, sediments and water in environment monitoring, Chapman edition, 2nd ed. E & FN Spon, London, 1996, 59-126.
29. Ali MM, Soltan ME. The impact of three industrial effluents on submerged aquatic plants in the River Nile, Egypt. Hydrobiology 1996 ; 340:77-83.
30. Michel P. La pollution des eaux marine « Pollution par les hydrocarbures: interaction avec les biocénoses ». Gauthier-Villars, Paris, 1976, 231.
31. Rouidi S. Cartographie de la pollution par les hydrocarbures totaux au niveau de la plate-forme industrielle de Skikda. Thèse de magistère en Ecologie. Université de Constantine, 2002, 110.
32. Boryslawskyj M, Garrood AC, Pearson JT, Woodhead D. Elevation of glutathione-S-transferase activity as a stress response to organochlorine compounds, in the freshwater mussel, Sphaerium corneum. Marine Environmental Research 1988; 24:101-104.
33. Durou C, Poirier L, Amiard JC, Budzinski H, Gnassia-Barelli M, Lemenach K *et al.* Biomonitoring in a clean and a multi contaminated estuary based on biomarkers and chemical analyses in the endobenthic worm Nereis diversicolor. Environmental Pollution 2007; 148:445-458.
34. Ait Alla A, Mouneyrac C, Durou C, Moukrim A, Pellerin J. Tolerance and biomarkers as useful tools for assessing environmental quality in the Oued Souss estuary (Bay of Agadir, Morocco). Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology 2006; 143:23-29.
35. Michel X, Narbonne JF, Mora P, Daubèze M, Ribera D, Lafaurie *et al.* Indicateurs biogéochimiques de pollution des écosystèmes côtiers: expérience du groupe interface chimie-biologie des écosystèmes marins (GICBEM). In: Lagadic L, Caquet T, Amiard JC, Ramade F. (Eds.), Utilisation de biomarqueurs pour la surveillance de la qualité de l'environnement Lavoisier Tec & Doc, Paris, New York, Londres, 1998, 9-30.