

**BACTERIAL NUMBER, HETEROTROPHY AND EXTRA CELLULAR
ENZYME ACTIVITY IN THE SEA WATER OF ALEXANDRIA HARBOUR,
EGYPT**

BY

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ABSTRACT

To study the structure and function of bacterial population in Alexandria harbour which is located between 29° 50' - 29° 53' longitude E and 31° 9' - 31° 12' latitude N. sampling stations. Samples were chosen. Samples were collected from surface and bottom sea water in the years 2001-2002.

The number of total bacteria in sea water was between 2.3×10^4 cells / ml and 1.4×10^5 cells / ml and the total number of saprophytic bacteria was significantly low with regard to the total bacterial number. Turnover times of glucose and leucine were extremely variable depending on the sampling station and the water depth.

In deep sea water the enzyme activity of α -glycosidase, N-acetyl β -glucosaminidase and amino peptidase of the slow growing bacterial population were higher then those of the fast growing bacterial population, B- glycosidase activities, however, were higher in the fast growing bacterial populations.

INTRODUCTION

The Mediterranean Sea is an area of outstanding scientific interest. Bacterial heterotrophy was considered negligible in sea water, but now, it is starting to appear as an important pathway of secondary food chain production. Detrital material particularly that are derived from tiny crustaceans organisms appears to enter the food chain at different trophic levels (Vincent 1986). Bacteria in seawater exhibit a number of interesting properties and additionally play an essential role in the cycling of nutrients (Nessim 1990). and the bacterial load in its water varies according to factors like sewage disposal and maritime activities.

Area of Investigation

The western harbour lies between $29^{\circ} 50' - 29^{\circ} 53'$ longitude E and $31^{\circ} 9' - 31^{\circ} 12'$ latitude N and occupies an area of about 1862 ha., the harbour is shallow (water max. depth of about 16 m.) and opens to the sea by a narrow canal called "El-Boughaz" (Fig.1).

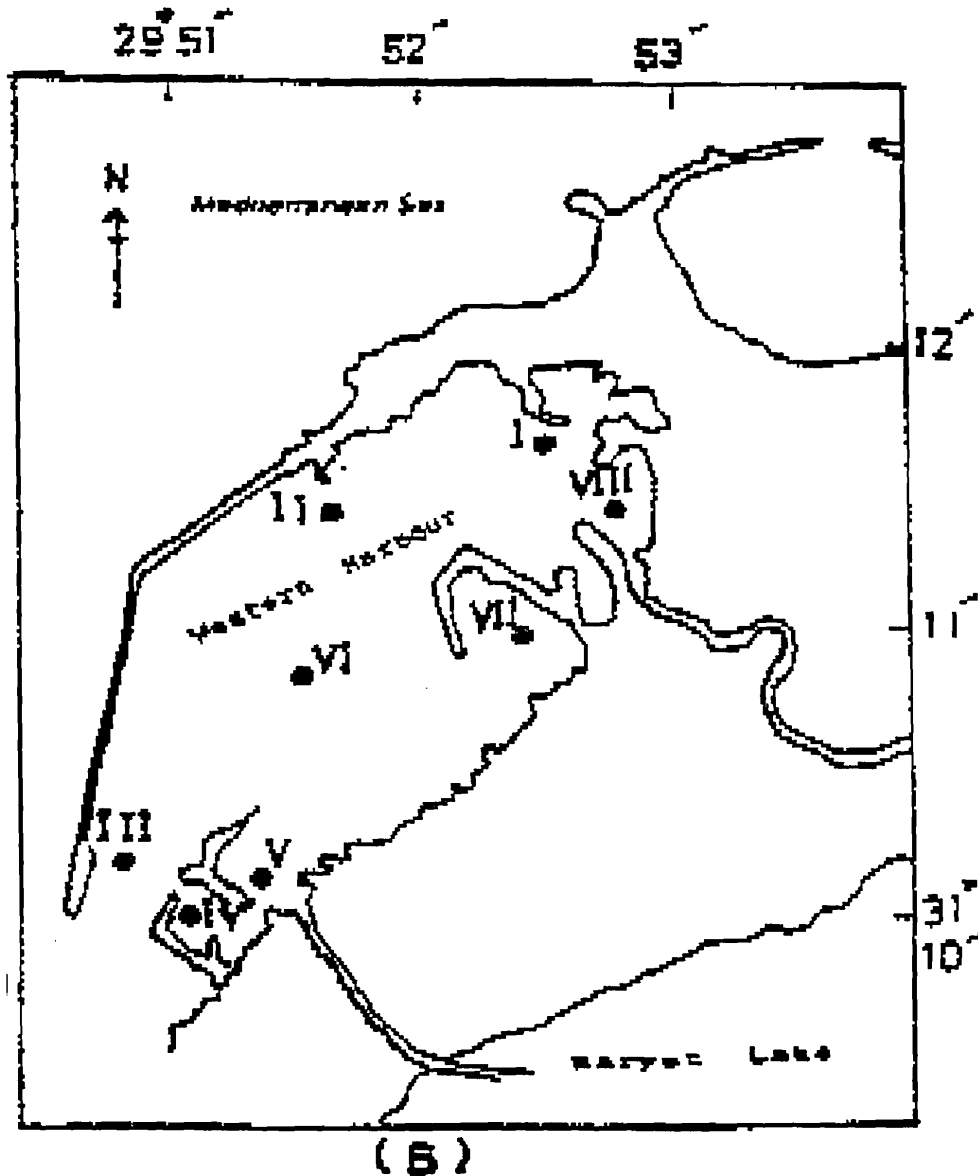


Fig. 1: Western harbour of Alexandria

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In addition to the pollution which results from different shipping activities, e.g. cargo, tankers and passengers ships the marine environment and biota of the harbour could also be subjected to other different types of pollutants, i.e. industrial, agricultural and domestic effluents from the following sources:

1. El-Noubaria Canal, which passes across lake Maryut discharges 9000 cubic meters daily of fresh waters, loaded with suspended substances (Shriadah & Tayel 1992).
2. Several outfalls, at El-mahmoudia Canal which introduces remarkable amounts of untreated domestic wastes.
3. El-Mex pumping station which discharges 6 million cubic meters daily of polluted brackish waters (Abdel Aziz 1997).

MATERIALS AND METHODS

Surface and bottom sea water samples were collected during day time at one meter depth and from near bottom water layer from eight stations in the western harbour (Fig. 1) seasonally during 2001-2002 in sterile two liter screw capped bottles.

To estimate total bacterial cell number's epifluorescent microscopic method was used (Zimmermann 1977, Pomroy 1984).

To enumerate total saprophytic bacterial number, the membrane filter method (pore size: 0.45 μ^2) and plate count method with Zobell 2216 E agar medium was used.

Zobell agar plates were incubated at 8°C for 15 days. The extracellular enzyme activities of X and β -glucosidase, N-acetyl, β -glucosaminidase and amino peptidase were determined by the method described by Kim and Hoppe (1986). For the assessment of microbial activity, turnover time was measured by the method described by Gocke et.al (1990) using ^{14}C -glucose and C^{14} leucine as substrates.

RESULTS & DISCUSSION

Table (1) and Fig. (1) show the number of total bacterial and total saprophytes in each station of Alexandria western harbour at both surface and bottom seawater. The number of total bacterial cells varied from 2.3×10^4 cells/ml to 1.4×10^5 cells/ml. In most stations the total cell number in the surface water layer was higher than that in the near bottom layer and this might be due to the high amount of nutrients in the surface sea water which plays an essential role in bacterial growth.

The total cell number obtained in the present investigation was found to be similar to that reported by Hanson et.al (1993) for the Antarctic waters with values ranging between 1×10^4 and 2×10^5 cells/ml.

Table (1) and Fig. (2) show the fluctuation of total saprophytic number which lies between 0.5×10^2 and 1.5×10^2 CFU/l during the sampling period, similar to that reported for the eastern harbour in 1995 by Siam (1998). Compared with the total bacterial number the saprophytes number were extremely low.

Table 1: Number of total and saprophyte bacteria in Mediterranean Sea Water collected from eight stations of western harbour in Alexandria

Station No.	Sampling Depth	Total bacteria number $\times 10^4$ cells/ml	Total saprophyte bacteria number $\times 10^2$ CFU/l
I	S	13.6	31
	B	8.3	11
II	S	7.5	24
	B	11.3	29
III	S	6.1	15
	B	2.3	0.5
IV	S	8.0	24
	B	5.8	8.5
V	S	5.6	29
	B	6.6	21
VI	S	9.8	11
	B	8.2	1.5
VII	S	7.0	30
	B	4.9	3.5
VIII	S	10.5	25
	B	8.1	30
S: surface water sample CFU: colony forming unit B: bottom water sample			

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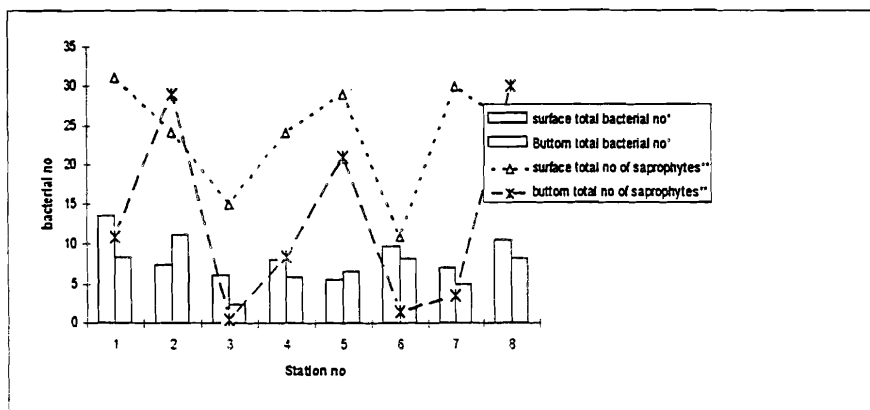


Fig. 2: Number of total and saprophyte bacteria in Mediterranean Sea Water collected from eight stations of western harbour in Alexandria

The turnover times for glucose and leucine in seawater samples from the Alexandria harbour are given in Table (2). These values were in the range of 41.3 – 2093.7 hours and 55.8 – 979.9 hours, respectively and varied depending on the sampling station and depth.

Fig. (3) shows that the turnover time of glucose in surface sea water in station iv, vi, vii and viii were higher than the turnover times of the near bottom water samples. It also shows that the turnover times of leucine were significantly shorter than those of glucose indicating that leucine could be taken up and remineralized faster than glucose in the water column of the Alexandria western harbour.

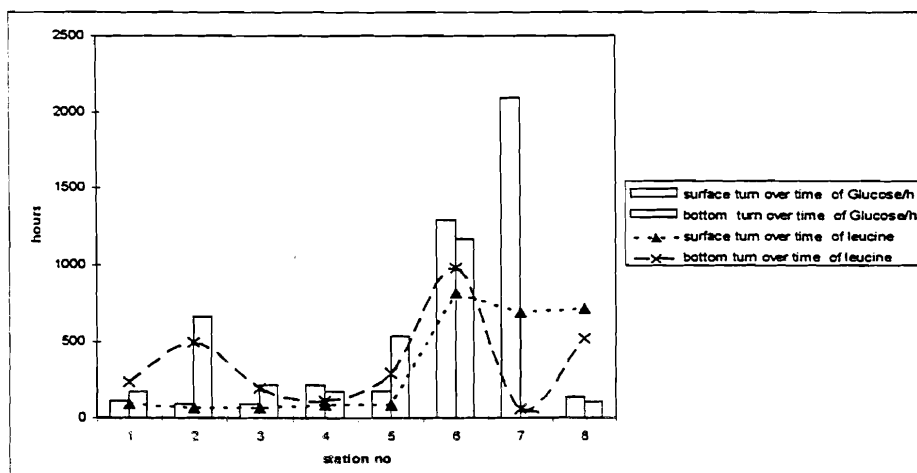


Fig. 3: Turnover time of glucose and leucine in seawater samples from the Alexandria western harbour

Table 2: Turnover time of glucose and leucine in seawater samples from the Alexandria western harbour

Station No.	Sampling Depth	Glucose (h)	Leucine (h)
1	S	112.1	92.3
	B	170.1	235.5
2	S	88.9	62.3
	B	664.6	495.1
3	S	90.7	62.3
	B	212.1	192.8
4	S	214.5	82.6
	B	167.6	110.2
5	S	178.2	84.2
	B	530.9	294.6
6	S	1291.8	816.6
	B	1168.2	979.9
7	S	2093.7	687.7
	B	41.3	55.8
8	S	138.2	713
	B	105.6	522

Water temperature is not likely to play an important role for the spatial distribution of heterotrophic bacteria potential as long as it remains unfluctuating in Alexandria western harbour (Tayel, 1997). Under this circumstance, the effect of total bacterial cell number and its metabolic activity (active or dormant) together with the concentration of organic nutrients on total heterotrophic potential may become dominant.

Table (3) shows the percentage of colonies with specified enzyme activities to total colonies during different incubation periods of water samples collected at eight stations in Alexandria western harbour. The bacteria forming colonies within 5 days were classified as the fast growing populations and those growing to colonies after 6 days of incubation as the slow growing populations.

The enzyme activities of α -glycosidase, N-acetyl β -glucosaminidase and aminopeptidase in the fast growing population were higher compared with those in the slow growing population. In case of β -glycosidase activities, however, a reverse result was obtained with an implication that the slow growing bacterial population can play a major role for cellulose decomposition in the deep water environment of Alexandria western harbour.

Table 3: Percentage of positive bacterial colonies showing enzyme activities to total colonies during different incubation periods from near bottom seawater samples in the western harbour

Station	α -glucosidase			β -glucosidase			N-acetyl, β -glucosaminidase			Aminopeptidase		
	Within 0-5 days	Within 6-15 days	Total period	Within 0-5 days	Within 6-15 days	Total period	Within 0-5 days	Within 6-15 days	Total period	Within 0-5 days	Within 6-15 days	Total period
1	59	64	62	0	47	25	17	5	11	96	79	86
2	96	50	78	0	25	7	0	0	0	100	63	90
3	86	70	82	0	33	16	5	0	4	100	87	94
4	71	63	69	0	30	8	0	0	0	100	7	93
5	96	78	91	2	5	3	0	0	0	62	64	63
6	100	0	98	0	0	0	5	0	4	75	0	67
7	21	10	17	2	7	3	68	32	56	76	50	66
8	100	63	83	0	14	5	55	17	49	63	33	56
Average	78.62	49.72	72.5	0.2	20.13	8.4	18.8	6.75	15.5	84	56.6	76.9

REFERENCES

- Abdel-Aziz, N.E.M (1997) Zooplankton production along Egyptian Mediterranean Coast at Alexandria, with special references to life history of one copepod species. Ph.D thesis Faculty of Science, Mansoura University, 384 pp.
- Gocke, K., A.Heinanen, O.Kirstin, M.mac, G. Panov, and A. Tsiban (1990) Microorganisms In: Second periodic assessment of the state of the marine environment of Baltic sea 1984-1988. Background document, Baltic sea environment proceeding No. 35B, 303-329, Baltic marine environment protection commission, Helsinki commission 1990.
- Hanson, R.B., H. K. Lowery, D. Shafer, R. Sorocco and D.H. Pope, (1993). Microbes in cold water: vertical pattern of substrates uptake, productivity and biomass. *Polar biology* 2, 179-188.
- Kim, S. J. and H., G. Hoppe, (1986). Microbial extracellular enzyme detection on agar plates by means of fluorogenic methylumbelliferyl substrates. *Deuxieme colloque international de bacteriologie marine. CNRs, Brest, Actes de colloques* 3, 175-183.
- Nessim, R.B. (1990). Nutrient levels and chrophylla in Alexandria coastal waters, *Bull. Instit. Oceanogr. Fish. Egypt*, 17 (1): 129-140.
- Pomroy, A. J. (1984). Direct counting of bacteria preserved with Lugol-iodine solution. *Appl. Environ. Microbial* 47, 1191-1192.
- Shriadah M.M.A and Tayel F.T (1992) Environmental condition of the western harbour of Alexandria I. physico-Chemical characteristics. *The Bull. High Inst. Pub. health, Alexandria - Egypt* 213,28-34.
- Siam. E.E. (1998). A synoptic survey on number of bacteria and biomass in relation to physico-chemical parameters in Eastern harbour of Alexandria - Egypt. *Bull. Nat. Inst. Of Oceanogr. & Fish., A.R.E* 1998, vol. 24: 103-117.

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- Tayel, F.T.R. (1997). A comparative study on the state of Aromatic Hydrocarbon pollution In Abu Qir bay and the western harbour of Alexandria. Bull. Nat.Inst Of Oceanogr. & Fish., A.R.E.1997,vol 23:155-165.
- Vincent, W.F. (1986). Microbial ecosystem of Antarctica, Cambridge University press 15, 365-375.
- Zimmermann, R. (1977). Estimation of bacterial number and biomass by epifluorescence microscopy and scanning electron microscopy. In G. ein-Heimer (ed.) Microbial ecology of brackish water environment, Springer-Verlag, Berlin, 103-120.