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NITROGEN FIXATION IN POLLUTED SEA WATER OF ALEXANDRIA COAST, EGYPT.

By

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ABSTRACT

The inshore area of the Mediterranean water, west of Alexandria, Egypt, is subjected to different types of pollutants. Nitrogen fixation rate was studied in this area in relation to some environmental factors such as pH, oxygen, organic matter, total and inorganic phosphorus and ammonia. In addition, numbers of Cyanobacteria, as nitrogen fixers, were also determined.

In order to identify possible relationships between environmental conditions and N-fixation, correlation coefficients were determined for each environmental factor and rates of N-incorporation.

Temporal and spatial variations were observed in the rate of nitrogen fixation, related to a number of factors. Positive correlations were found between the rate of N-fixation and organic matter, total and inorganic phosphorus and the densities of Cyanobacteria in the region. On the other hand, N-fixation was negatively correlated with both ammonia and oxygen concentration.

INTRODUCTION

Nitrogen fixation is an important biological process in the marine habitat (Stewart *et al,* 1971; Goering and Parker, 1972 and Fogg, 1978). It is affected, as well as other biological processes, by different environmental factors such as oxygen concentration, organic matter,...., etc. (Billaud, 1968; Lallatin, 1972; Vanderhoef *et al,* 1975; Horn *et al,* 1979; El-Samra, 1980; and Moustafa, 1985) which in term are influenced by local land management practices.

According to the available literature N-fixation has not been studied so far in the Egyptian or other Mediterranean waters. The present study was conducted to follow the temporal and spatial variations of N-fixation in a polluted area of the inshore eastern Mediterranean water, west of Alexandria, from Al-Anfoushi to Agami. This area is considered as a perturbed marine ecosystem as it is subjected to several

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coincidental impacts (Dorgham *et al,* 1987) largely through waste waters loaded with variable concentrations of agricultural, industrial and domestic pollutants entering the region (El-Rayis *et al,* 1984 and Abul-Dahab *et al,* 1984).

MATERIALS AND METHODS

The study area is divided into three regions according to the effect of the different land based effluents; namely, Al-Anfoushi, Al-Mex and Agami (Dorgham *et al,* 1987). The Al-Anfoushi region is perturbed by municipal wastes from the main city sewer of Alexandria. Al-Mex water is affected by agricultural, industrial and domestic wastes discharged directly or indirectly into the region. The Agami region is more distant receiving little anthropogenic pollution (Fig. 1).

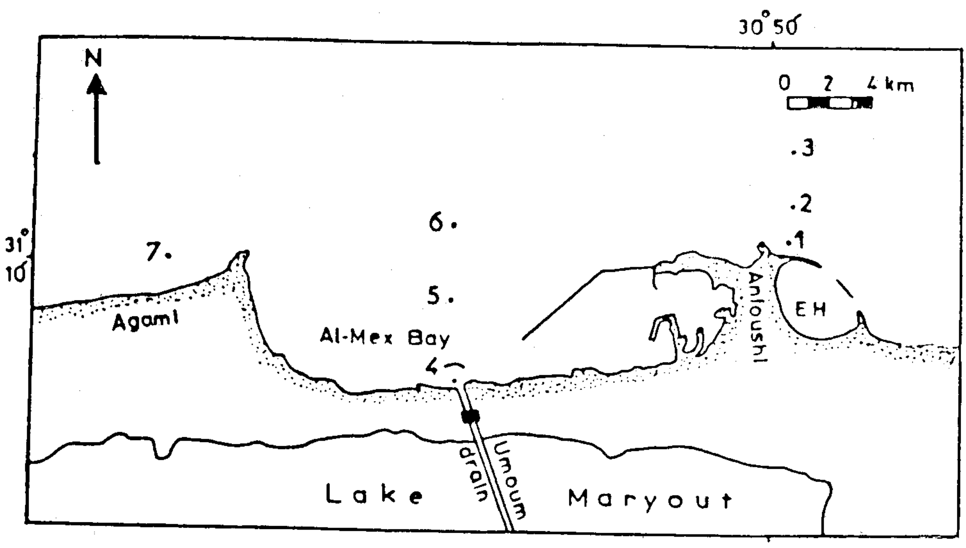


Fig. 1: The study area and locations of the sampling stations.

Nitrogen fixation, oxidizable organic matter, ammonia, total phosphorus, inorganic phosphorus and dissolved oxygen were measured bimonthly from April 1982 to April 1983 at 7 stations. Species composition and densities of cyanobacteria were also studied.

Measurements of N-fixation were carried out by the acetylene reduction technique (Stewart *et al,* 1968). Dissolved oxygen, ammonia, total and inorganic phosphorus were measured according to Strickland and Parsons (1968). The oxidizable organic matter was determined by the Standard Methods (1982).

For the determination of cyanobacteria densities 1-litre samples of the sea water were collected by Niskin bottle, preserved in 4 % neutralized formalin and concentrated to 100 ml by the sedimentaion technique (utermohl, 1936). I ml aliquotes were counted three times by the inverted microscope and the cyanobacteria densities were determined in colony/1 (Smayda, 1978).

Pair correlations between N-fixation and each environmental factor were made by computing Pearsons r (Meddis, 1975).

$$r = \frac{\leq n.\leq xy - \leq x \quad y}{\sqrt{(n.\leq x^2 - (\leq x)^2)\ (n.\leq y^2 - (\leq y)^2)}}$$

where r is a measure of correlation whose value lies between -1 and +1. A high degree of correlation exists when r approaches ± 1.

RESULTS AND DISCUSSION

Nitrogen fixation in the study area showed significant temporal variations. The early summer values were higher than those of the other seasons (Table, 1) reaching the maximum at most stations in June. The particularly high values (10.51, 11.03 and 12.33 uM/h) were observed at Stations 1, 4V AND 5, which are directly affected by the domestic and brackish water effluents. On theother hand, the lowest values of N-fixation were detected during winter with the minimum in February (0.01 - 0.03 uM/h). Such variations may be related to the seasonal variations in the water temperature (Table, 2). The present observations have agreed more or less with those of Stewart *et al* (1971) and Horne and Goldman (1972), who stated that N-fixation is an early summer process.

Table 1

Rate of nitrogen fixation (ug-at-N_2 $1^{-1}h^{-1}$) at different seasons in the study area.

Station	April	June	August	October	December	February	April
1	0.16	10.51	1.35	0.30	0.00	0.02	0.80
2	0.00	0.85	0.30	0.16	0.19	0.009	1.10
3	---	---	---	0.14	0.19	0.02	0.40
4	1.62	11.03	0.00	0.12	0.28	0.02	0.88
5	4.97	12.33	0.89	0.18	0.00	0.02	0.48
6	0.26	3.07	0.46	0.16	0.35	0.03	0.53
7	0.044	0.35	0.00	0.50	0.00	0.013	0.35

Table 2
The ranges of seasonal variations of the water temperature in the different regions.

Season	Al-Anfoushi	Al-Mex	Agami
Spring	19 - 20	19 - 20	20
Summer	23 - 28	23 - 28	23 - 28
Autumn	25 - 25.5	25.5 - 26.5	26
Winter	14 - 19	15 - 17.5	14 - 17

Regional distributions of the N-fixation indicated that the Agami region was most frequently characterized by the lowest values (Table, 1), which may be correlated with low cyanobacteria numbers in this region. In Al-Anfoushi and Al-Mex regions, on the other hand, N-fixation rates were higher and might be associated with higher cyanobacteria numbers or/and other N-fixers.

The high standing crop of the total freshwater planktonic algae in the study area was not always good indication of the N-fixation rate. At some stations, although the cell count of phytoplankton was relatively large (180,000 cell/1), N-fixation was moderately low (0.48 uM/h). This situation was observed several times, particularly when green phytoplankton dominated the assemblages. Such observations are consistent with an inability of green algae to fix atmospheric nitrogen. However, N-fixation showed a high positive correlation (r=0.74) with cyanobacteria densities (Fig. 2). The dominant cyanobacterians were: *Oscillatoria limnetica, Chroococcus turgidus and Spirulina princeps.*

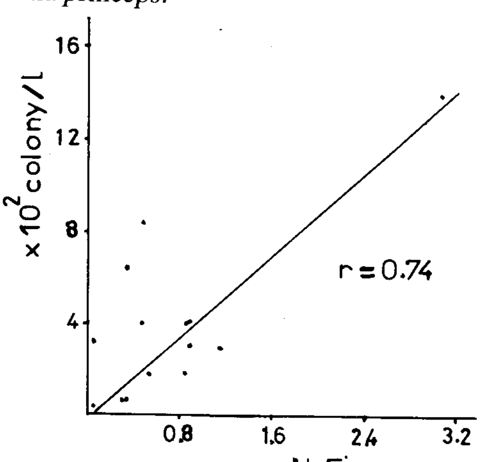


Fig. 2: Relationship between nitrogen fixation (ug-at N_2 $l^{-1}h^{-1}$) and numbers of cyano-bacteria.

Correlations between environmental conditions and N-fixation were determined to establish possible relationships. The correlation coefficients showed that N-fixation exhibited different responses to the environmental conditions. Relatively strong correlations were observed between N-fixation and the concentration of the total and inorganic phosphorus (r=0.42 and 0.77, respectively) (Figs. 3 & 4), and complementing results from Vanderhoef *et al.* (1975) and Horne *et al.* (1979). These researchers noted that Phosphorus (total and inorgnic) enhanced N-fixation in natural phytoplankton assemblages in two different systems. Such effect was also observed at Al-Anfoushi during summer and autumn. In winter and spring, an opposite pattern was found between N-fixation and concentration of total and inorganic phosphorus at the two regions. This situation may be attributed to the differences in the nature of the pollutant transeferred to the two regions. The same trend was also observed between organic matter and N-fixation with OM acting as source of energy required by the nitrogen fixers (El-Samra *et al.*, 1979); in the present study a positive correlation of 0.79 was noted between the N-fixation and the organic matter (Fig. 5).

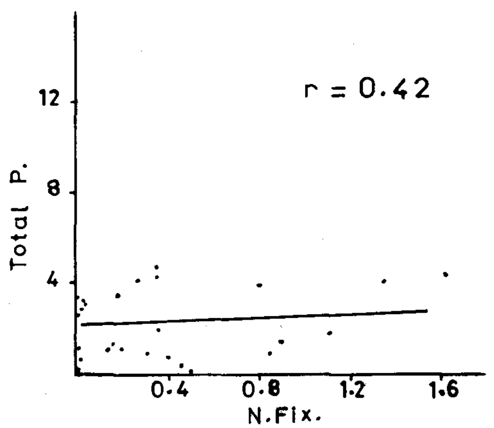


Fig. 3: Relationship between nitrogen fixation (ug-at N_2 $l^{-1}h^{-1}$) and concentrations of total phosphorus (ug-at l^{-1}).

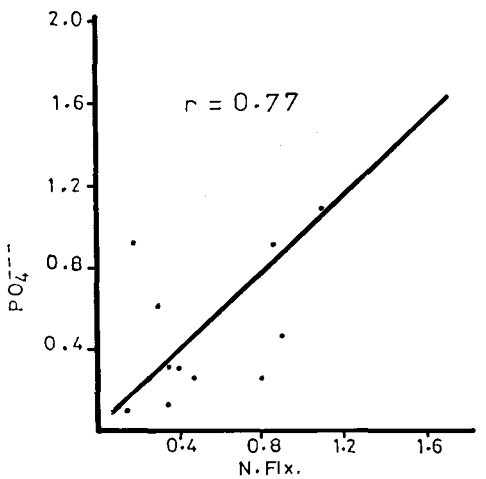


Fig. 4: Relationship between nitrogen fixation (ug-at N_2 $l^{-1}h^{-1}$) and inorganic phosphorus (ug-at l^{-1}).

In contrast, negative correlations were found between N-fixation and ammonia (r = -0.81) and dissolved oxygen (r = -0.25) (Figs. 6 and 7); the rate of N-fixation was depressed at high concentrations (average, 15 uM) of ammonia or oxygen (10.06 mg/1). Horne *et al.* (1979) and El-Samra *et al.* (1979) noted that ammonia concentration higher than 7 ug-at/1 depressed nitrogen fixing activity. It is possible that at high levels, ammonia is taken up by cyanobacteria as a source of nitrogen preferentially to fixing it, as the former process requires less energy. Furthermore, the inhibiting impact of high oxygen content on N-fixation was detected by Stewart *et al.* (1971) and El-Samara *et al.* (1979).

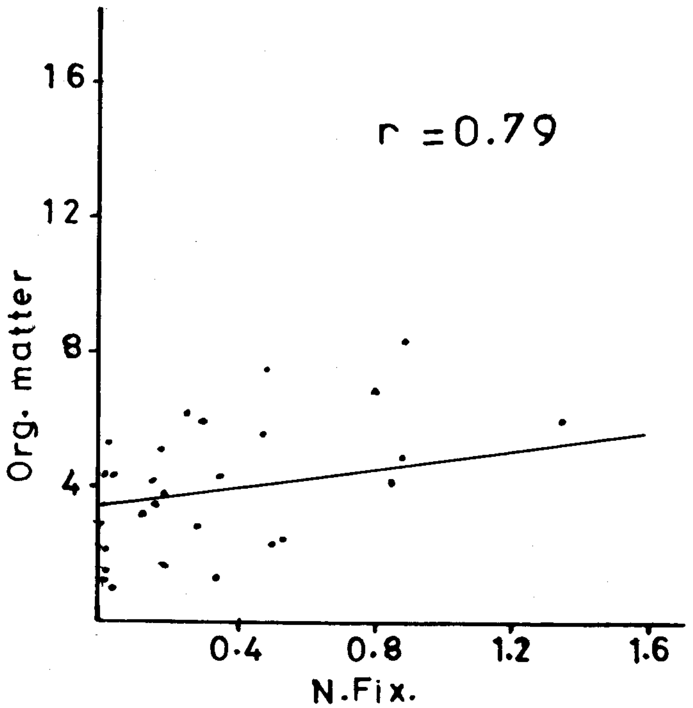


Fig. 5: Relationship between nitrogen fixation (ug-at N_2 $1^{-1}h^{-1}$) and concentrations of organic matter (mg 1^{-1}).

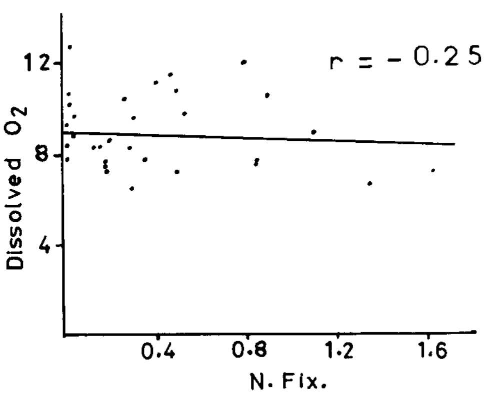
Regarding N-fixation at the different regions of the study area, intimate relationships were suggested between this process and the studied environmental factors. Seasonal effects on the N-fixation activity were noted in the local regions. Ammonia concentrations at Al-Anfoushi were higher than those at Al-Mex in spring and early summer (Table, 3) and could explain lower rates of N-fixation in the former region. This may be related to the preference of ammonia uptake by cyanobacteria rather than nitrogen fixation (Horne *et al.*, 1979; El-Samra *et al.*, 1979) particularly at low temperature. However, higher N-fixation rate was observed in Al-Anfoushi during late summer and autumn in spite of the high concentration of ammonia. Such observations may indicate the positive impact of the temperature on the N-fixation and its negative effect on the ammonia uptake. Otherwise, they might be attributed to other nitrogen fixers such as azotobacters and blue green picoplankton. However, these groups were not studied in the area and further research is required to identify other taxa contributing.

Table 3

The average seasonal variations of ammonia (ug-at 1^{-1}) in the different regions.

Season	Al-Anfoushi	Al-Mex	Agami
Spring	47.16	15.71	2.91
Summer	30.39	5.4	1.87
Autumn	18.01	19.66	1.50
Winter	20.71	31.80	3.45

In general, nitrogen fixation activity could be considered as a sum of cyanobacterial responses to many environmental factors. Thus, organic matter, phosphorus and, to a limited extent, warm water enhance nitrogen fixation in the region and increase contributions to nitrogen pools of the marine environment. High levels of ammonia and oxygen, on the other hand, inhibit cyanobacteria activity.



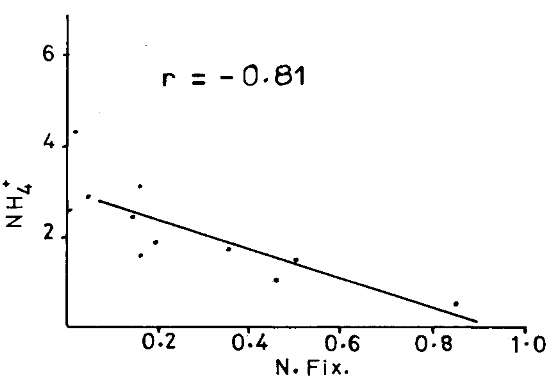


Fig. 6: Relationship between nitrogen fixation (ug-at N_2 $1^{-1}h^{-1}$) and ammonia concentration (ug-at 1^{-1}).

Fig. 7: Variation in nitrogen fixation (ug-at N_2 $1^{-1}h^{-1}$) with dissolved O_2 (mg 1^{-}

CONCLUSIONS

1 - Nitrogen fixation showed temporal and spatial variations related to seasonal variations of the environmental conditions but also to the polluted waste waters discharged to the area.
2 - Significant correlations were detected between N-fixation and numbers of cyanobacteria and concentrations of phosphorus (total and inorganic) and organic matter.
3 - Correlations between N-fixation and ammonia and oxygen were negative.

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تثبيت الأزوت في ماء البحر الملوث من شاطىء الأسكندرية ـ مصر

و ثناء مصطفى محمد محمود درغام ـ محمد ابراهيم السمره

يستقبل ماء البحر في منطقة البحث مخلفات صناعية وزراعية وصرف صحي محملة بأنواع شتى من الملوثات ، الأمر الذي يؤثر على عملية تثبيت الأزوت الجوي بواسطة البكتريا الخضراء المزرقة .

تتبع البحث التغيرات الموسمية والمكانية لسرعة هذه العملية مع التغيرات المناظرة في العوامل البيئية المختلفة . ولهذا أجريت الدراسة في الفترة من ابريل ١٩٨٢ إلى ابريل ١٩٨٣ ، حيث جمعت العينات وأجريت التجارب كل شهرين في سبع محطات تمثل أنواع المياه المختلفة في منطقة البحث .

تركـزت الدراسـة حول تعيين سرعة تثبيت الأزوت الجوي وربطها ببعض العوامل البيئية مثل الاكسجين الذائب ، المواد العضوية ، تركيز مركبات الفوسفور الكلية وغير العضوية والأمونيا ، بالاضافة إلى تقدير المحصول القائم للبكتيريا الخضراء المزرقة .

من النتائج تم حساب معامل العلاقة بين تثبيت الأزوت والعوامل المختلفة ومنها وجد أن هذه العملية تتأثر بتركيز المواد العضوية ومركبات الفوسفور واعداد البكتيريا الخضراء المزرقة .. ومن ناحية أخرى وجد أن سرعة تثبيت الأزوت في ماء البحر لاتتأثر إلى حد ما بالامونيا والاكسجين .