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# RELATIONSHIP BETWEEN ALGOSPHERE AND FREE WATER BACTERIA IN SOLAR LAKE (SINAI)

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## Abstract

Cyanobacterial mats of solar lake were followed up during the four seasons of a year 1997 through day-light and night. Also, the free water microorganisms groups related to soil fertility were followed included: Total colony counts, asymbiotic-N-fixers, nitrifying bacteria, sulphur-oxidizing and reducing bacteria. The ratio between total phytoplanktons and bacteria in water was calculated as indicator for biological activity in the Solar Lake.

The obtained results showed that there are a positive correlation between the counts of free water bacteria and phytoplanktonic mass in the lake. The highest bacterial counts were recorded during day time of spring season. The ratio of algosphere to free water bacteria was generally more than one, reached highest value beginning of stratification period (autumn season). On the other hand at the highest figure of total aerobic bacterial counts and the lowest figure was recorded by sulphur reducing bacteria.

## Introduction

The dense of planktonic blooms of cyanobacteria are indicatory for eutrophication in water bodies; which often exhibit fluctuation between $O_2$ concentration during the day to brief sulfide buildup at night. Planktonic cyanobacteria have also been recorded in hypolimnonic water of stratified lake, where exposure to sulfide is prolonged during the stratification period. (slobodkin, and Zavargin, 1993).

Cyanobacteria are often found in sulfide rich environments in existence with ano-oxygenic photosynthetic bacteria. Sulfide is highly toxic to eucaryotic photosynthetic organisms and to non adaptive cyanobacteria. It inhibits the electron transport chain by reacting with cytochromes and heamoproteins and by binding with metal protein. The exposure to low redox potential may drastically inhibit $O_2$ evolution in some cyanobacteria. The occurance of oxygenic cyanobactera under sulfide, requires some adaptation to cope with toxicity. (Zhilina and Zavarzin, 1990).

Many autors stated that the occurrence of benthic cyanobacteria and sulphur-oxidizing becteria has close association with sulfide, which is very common in cyanobacterial mats occurred in hot springs and hypersaline lagoons and highly saline closed lakes. The diurnal fluctuations in sulfide and oxygen have been shown for both hot springs and hypersaline lagoons, where these bacteria are periodically exposed to alternating high concentrations of both sulfide and $O_2$. It was found that sulfide-dependent anoxygenic-photosynthetic bacteria was driven by photosystem I (PS/I) alone. These organisms are cabable to utilize $H_2$ as an alternative electron-doner to $H_2S$. (Smith, 1993).

On the other hand, several strains can carry out oxygenic photosynthesis under sulfide, indicating the continued operation of photosystem II, while other cyansobacteria may operate oxygenic and anoxgenic photosynthesis at the same time.

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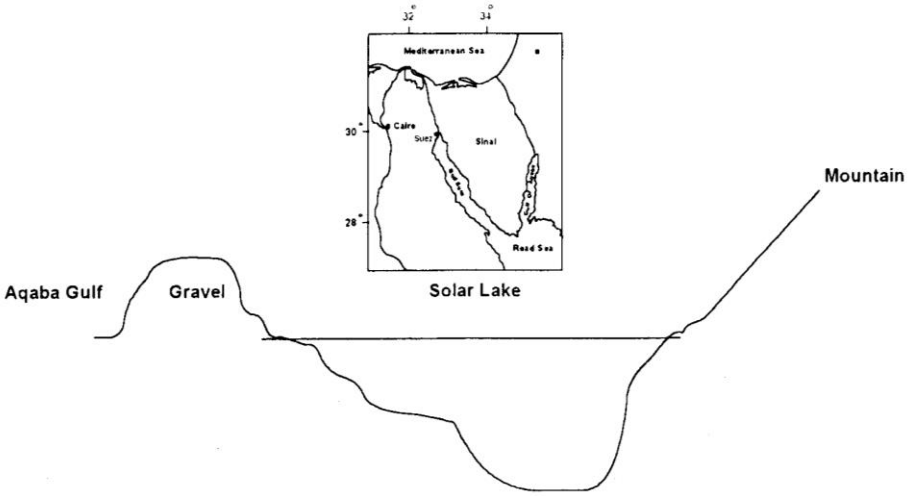
## *Materials and Methods*

### Sampling :

Water samples were collected periodically from Solar Lake during only one year four season of a year. The location was chosen, allocated on the map (Fig. 1) representing all the ecological factors affecting the microbiology of the lake.




(Fig. 1)

The location was at the eastern side of the solar Lake under 30 cm from surface water of the eastern side of the lake. The water samples were collected in duplicate the first one at mid sunshine 12.00 a.m. while the other after mid-night at 4.00 am.

Sampling of the shallow-water mat prepared by cutting pieces from the central parts of the long cores (5 cm diam) and transfered to N.R.C. in ice-box.

Total nitrogen was determined in the algal mat by the micro-Kjedlahl method. Potassuim and cupper sulfate with selenium mixture (20:2:1) was used as a catalyst during the digestion with concentrated $H_2SO_4$. Boric acid (2%) solution was used for absorbing the evoluted ammonia. Sulphuric acid (1/70N) was used for the titration with bromocresol green-methyl red mixture as indicator (Ma and Zauzage, 1942). Blank determinations were run to correct for any traces of nitrogen present in the ingredients.

Total organic carbon was determined by using K-dichromate (1.0N.) The results were multiplied by 3 instead of 8 to obtain the figures of organic carbon in mg/L. (Piper. 1950).

**Microbioloical determinations :**

The collected samples were preserved in ice-box up to its analysis at NRC labs. The water samples were shaken for 5 min. Appropriate serial dilutions were prepared. Total colony counts, the counts of asymbiotic nitrogen fixing bacteria, nitrifiers, sulphur oxidizing bacteria, sulphur reducing bacteria and algoshere microorganisms were determined according to A.P.H.A. 1992 as follows :

- **Total colony count by poured plate technique** :
- The counts of asymbiotic nitrogen fixing bacteria enumerated by using of Ashby's medium. (A.P.H.A. 1992).
- **Nitrifiers** : Nitrosafication and nitrification bacteria were counted by using most probable Number (MPN) technique using Stephenson's medium. (Allen, 1953).
- The sulphur oxidizing bacteria, it was counted on starkey's medium. The (MPN) were obtained by using Hoskins tables.
- The sulphur reducing bacteria was enumerated on Van Delvin medium. The positive tubes were detected after incubation period by the production of gases ($H_2$, $H_2S$).
- **Algosphere microorganisms** : Water samples were shaked and filtered using Whatman 42 filter paper. The filtrate water cultivated on the nutrient agar medium (Allen, 1953) for counting total bacteria ($W_1$) and on Ashbey's medium for asymbiotic nitrogen fixers ($W_2$).
- The algal growth were removed from the filter paper by using sterilized water, then cultivated on the nutrient agar medium (Allen, 1953). Then the filtrated was cultivated on the previously mentioned specific media to count total colony counts, asymbiotic N-fixing bactria, nitrifiying bacteria, sulphur oxidizing bacteria and sulphur reducing bacteria.

## *Results and Discussion*

The present study concentrates on the relationship between algosphere and free water bacteria in Solar Lake (Siniai). The algal mat dominate in the shallow parts of the pond. Therefore we collected the samples from the shallow part of the lake shown in map (Fig. 1).

**Table 1. Organic carbon and nitrogen persentage in the shallow-water mat as dry weight basis**

| No. | Measurements | Aut. 96 | Wint. 97 | Spr. 97 | Sum. 97 |
|---|---|---|---|---|---|
| 1 | Total Nitrogen % | 0.4092 | 0.2012 | 0.1015 | 0.3092 |
| 2 | Total organic carbon % | 1.8081 | 1.2063 | 0.7053 | 1.6034 |
| 3 | C/N ratio | 4.4186 | 5.9955 | 6.9488 | 5.1856 |

The productivity measurements of the shallow-water mat were the total nitrogen and organic carbon. The highest photoassimilation of organic carbon was 1.8081 % recorded at Autumn 96. The total nitrogen of the mats through the four seasones are given in table (1) which shown general nitrogen decrease at spr. 97 reached 0.1015% then increased 0.3092% at summer and continued high up to autuin 0.4092 and at the winter started to decrease (0.2012%). These results of total organic materials in the shallow water mats of Solar Lake is very high, the concentrations of organic carbon and nitrogen are shown in Table (1). The carbon /nitrogen (C/N) ratios are low reached 4.42 in autumn 96, then the ratio continued increase through winter 97 (5.9955) up to spr. 97 (6.9488) and decreased in the summer 97 (5.1826). Cyanobacterial mats of the Solar Lake, this stromatic mats dominate the shallow parts of the pond cores taken of the cyanobacterial mate are in most cases undisturbed, regularly laminated with well documented carbonate interlayers. All the studies carried out on the various aspects of the Solar Lake mat showed that the organic matter in these mats is of marine derivative, (Cohen, et al. 1984).

**Table 2. Total colony count and asmbiotic $N_2$-fixing bacteria in the algosphere (Alg.) and free water (W.) (x $10^3$ org./ml)**

| Microbiol Counts in | Season Of Counting | Total colony count | | Asymbiotic $N_2$-fixing Bacteria | |
|---|---|---|---|---|---|
| | | Day | Night | Day | Night |
| (Alg.) Algospher | Autumn 96 | 5040 | 4810 | 46.8 | 32.1 |
| | Winter 97 | 423 | 394 | 100.8 | 80.2 |
| | Spring 97 | 260 | 282 | 85.4 | 64.7 |
| | Summer 97 | 1600 | 1590 | 66.2 | 42.1 |
| (W.) Free water | Autumn 96 | 182 | 231 | 6.6 | 7.2 |
| | Winter 97 | 342 | 326 | 14.2 | 16.2 |
| | Spring 97 | 221 | 342 | 9.6 | 10.1 |
| | Summer 97 | 192 | 270 | 8.6 | 9.2 |
| (Alg./W.) Ratio | Autumn 96 | 27.69 | 20.82 | 7.09 | 4.04 |
| | Winter 97 | 1.24 | 1.21 | 7.9 | 4.95 |
| | Spring 97 | 1.18 | 0.82 | 8.90 | 6.41 |
| | Summer 97 | 8.33 | 5.89 | 7.70 | 4.58 |

The bacterial counts indicated from table (2) that the total colony counts in the algosphere were generally higher than in the free water. This was more pronounced during autumn 1996 as shown in table (2) reached 5040x$10^3$ org./ml, at day time while at night reached 4810x$10^3$ org./ml. This is due to the higher organic load as we discussed before during these periods of the year, which could be used as a source of food materials for the different groups of heterotropghic bacteria in the water body. These results showed a positive correlation between the algosphere bacteria and the microbial activity in the mats. (Sieburth, 1993).

The asymbiotic $N_2$ – Fixing bacterial (Table 2) were generally higher in the algosphere of the algal mat of the lake, specially during winter 97 reached 100.8x$10^3$ org./ml at day time continued to spring 1997. These results are correlated with the ratio of Alg./w. counts of these specific groups of asymbiotic $N_2$- fixing bacteria. (Dannenberg et al. 1992).

On the other hand, there was no pronounced changes between the counts of $N_2$-fixing bacteria in both algosphere or free water organisms during the day and night periods of the sampling. Generally, the highest counts of $N_2$-fxing bacteria was reached at autumn season of the year, and it was higher in the algosphere than in the free water microorganisms. Therefore, ratio between Alo/w was higher than one continually, reached the lights value of 27.7 in the algosphere during autumn 96- these results are in accordance with the finding's of Binnerup et al (1992) who stated that Densification. dissimulator reduction of nitrate to ammonium and nitrification.

**Table 3. Counts of nitrifying bacteria in both algosphere (Alg.) and free water (W.) (x $10^3$ org./ml) during day and night**

| Microbiol Counts in | Season Of counting | Nitrifying bacteria | | | |
|---|---|---|---|---|---|
| | | Nitrosofication | | Nitrification | |
| | | Day | Night | Day | Night |
| (Alg.) Algospher | Autumn 96 | 3.2 | 2.0 | 2.8 | 2.2 |
| | Winter 97 | 10.1 | 12.1 | 17.6 | 14.2 |
| | Spring 97 | 3.4 | 1.9 | 10.2 | 15.1 |
| | Summer 97 | 6.8 | 2.8 | 12.4 | 10.1 |
| (W.) Free water | Autumn 96 | 0.2 | 0.1 | 2.1 | 1.7 |
| | Winter 97 | 1.1 | 2.1 | 13.1 | 8.2 |
| | Spring 97 | 0.8 | 1.8 | 8.2 | 6.1 |
| | Summer 97 | 0.6 | 1.7 | 6.2 | 5.1 |
| (Alg./W.) Ratio | Autumn 96 | 16.0 | 20.0 | 1.33 | 1.29 |
| | Winter 97 | 9.18 | 5.76 | 1.34 | 1.73 |
| | Spring 97 | 4.25 | 1.06 | 1.24 | 2.48 |
| | Summer 97 | 1.33 | 1.65 | 2.00 | 1.98 |

It is clear from Table (3), showing the counts of nitrifying bacteria in both algosphere and in water, that the highest counts of this group of microorganisms were higher in the algosphere during winter season 1997, while was less in free water reached maimum during autumn 96 (16x$10^3$).

The same trend was found in the ratio of this group of microorganism in algosphere and in free water. These results are in harmoany with the finding Asbton (1979) who stated that Nitrification of the nitrogen – Limited impoundment.

**Table 4. Counts of Sulphur Oxidizing and reducing bacteria in the Algosphere (Alg.) compared with those bacteria in free water (W.) (x $10^3$ org./ml), by day and night.**

| Microbiol Counts in | Season Of Counting | Sulphur Oxidizing Bacteria | | Sulphur Reducting Bacteria | |
|---|---|---|---|---|---|
| | | Day | Night | Day | Night |
| (Alg.) Algospher | Autumn 96 | 75.1 | 82.0 | 1.1 | 1.6 |
| | Winter 97 | 28.00 | 18.90 | 0.6 | 0.9 |
| | Spring 97 | 40.00 | 36.00 | 0.2 | 0.1 |
| | Summer 97 | 10.90 | 18.20 | 0.4 | 0.2 |
| (W.) Free water | Autumn 96 | 20.2 | 10.2 | 0.8 | 1.2 |
| | Winter 97 | 50.2 | 48.2 | 0.2 | 0.6 |
| | Spring 97 | 42.8 | 46.2 | 0.1 | 0.2 |
| | Summer 97 | 360 | 28.8 | 0.3 | 0.4 |
| (Alg./W.) Ratio | Autumn 96 | 3.72 | 8.04 | 1.38 | 1.33 |
| | Winter 97 | 5.58 | 3.92 | 3.00 | 4.50 |
| | Spring 97 | 9.46 | 7.79 | 2.00 | 0.50 |
| | Summer 97 | 3.03 | 6.32 | 1.33 | 0.50 |

The counts of sulphere-oxidizing and reducing bacteria in both algosphere and free water samples are shown in Table (4). The results revealed that these S-oxidizing organisms flourished during spring season , with no differences between the day and night of sampling . On the other hand, the counts of these organism in the free water reached maximum during winter 97 ($50.2x10^3$) with no difference between day and night sampling time.

On the other hand, the counts of Sulphur-reducing bacteria were very low as they are strickt anaerobes, with no differences between the counts in day and night sampling. The ratios of these organism enumerated in both algosphere and free water was higher than one, reached maximum of 9.46 during spring 1997. These results are in accordance with the findings of cohen (1993).

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# دراسة العلاقة بين النمو الطحلبي ونشاط البكتريا في مياه بحيرة الشمس

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تمت دراسة نشاط الطحالب الخضراء المزرقة في بحيرة الشمس (سيناء) خلال أربعة فصول لسنة ٩٦-١٩٩٧ وخلال النهار والليل وكذلك مجموعات البكتريا المرتبطة بخصوبة التربة في مياه البحيرة والتي شملت مثبتات أزوت الهواء الجوى لا تكافلياً ، ومؤكسدات الأمونيا والنيتريت ومؤكسدات المركبات الكبريتية ومختزلات المركبات الكبريتية أيضاً .

وقد تم حساب النسبة بين الاعداد الكلية للهائمات النباتية في الطبقة السطحية للبحيرة منسوبة الى الاعداد الكلية لمجموعات البكتريا تحت الدراسة وذلك لمقياس النشاط الحيوي في بحيرة الشمس .

وقد اوضحت نتائج البحث أن هناك علاقة موجبة بين الاعداد الكلية للهائمات النباتية والاعداد الكلية للميكروبات تحت الدراسة . وقد كانت أعلى أعداد لهذه الكائنات الدقيقة في فترة النهار وأقلها فترة الليل ، وكانت النسبة بين اعداد الهائمات النباتية الكلية والبكتريا الكلية أعلى من الواحد الصحيح في بدء فترة سكون الطبيعة المائية وذلك خلال الطبقة السطحية حيث وجدت الهائمات النباتية باعلى كثافة وذلك خلال فصل الخريف عام ١٩٩٦. وكانت اعلى اعداد للميكروبات المرتبطة بخصوبة التربة للبكتريا المثبة للأزوت الجوى في الطبقة السطحية للمياه وأقل اعداد كانت للبكتريا المختزلة للكبريت في الطبقات السفلية .