PRELIMINARY STUDIES ON ECOLOGICAL DISTRIBUTION OF MICROORGANISMS IN WEST LAKE, HANGZHOU 2. Microorganisms in sediment

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Abstract

The abundance and distribution of microorganisms in the upper-layer sediment of West Lake were studied. Results showed that aerobic heterotrophic bacteria were 2.01 x $10^5/g$ and actinomycetes were 1.83 x $10^4/g$ on average. Both were more abundant in L. Yuehu. While the bacteria had negative correlation with the surrounding water temperature, actinomycetes had positive correlation. Aerobic cellulose decomposing bacteria were $1.88 \times 10^3/g$ on average, and more abundant in L. Xili and L. Yuehu L., peaking in July and May. Nitrite bacteria, nitrate bacteria, nitrate reducers and denitrifiers were $2.75 \times 10^5/g$, $1.81 \times 10^5/g$, $1.23 \times 10^5/g$ and $3.14 \times 10^3/g$ on average, respectively. Organic phosphorus decomposing bacteria were $2.38 \times 10^3/g$ on average, while calcium phosphate dissolving bacteria were very low. About 50.4% of heterotrophic bacteria were found to be *Bacillus* and about 23.9% were *Enterobacteriaceae*. Remain about 25.7% consisted of 11 other minor bacteria genus.

Key words: Lake sediment; Microorganisms; Distribution of bacteria

Introduction

West Lake, Hangzhou is a relatively small shallow lake. Its water surface area is approximately 5.67 Km², and its average depth is only 1.56m. Due to an abundant growth of plankton and pollution from tourism garbage each year, the nutrient content of the sediment has become very high. The content of total carbon (TC), total nitrogen (TN) and ammonium nitrogen (NH₄ –N) in the lake sediment had reached about 10.5%, 1.24% and 0.0097%, respectively. While total phosphorus (TP) and inorganic phosphate (PO₄-P) were 0.25% and 0.0016%, respectively (Li, 1990). C, N and P in the sediment of lake are gradually released from lake bottom into the water body, resulting in the water becoming more eutrophic state. Microorganisms in the sediment play an important role in nutrient release (Hattori, 1978), but studies on such microorganisms in the West Lake have yet to be reported. A joint research between China and Japan has been established in order to complehensively study several aspects of the lake ecosystem. The present paper deals with the distribution of microorganisms in the upper layer sediment of the West Lake.

Materials and methods

Sampling sites were selected as in part 1 of this study. The sediment samples were taken using an Ekman-Birge dredge from five sampling sites.

Using the diluted sediment of lake, heterotrophic bacteria, actinomycetes, cellulose decomposing microbe, ammonium oxidizing bacteria, nitrite oxidizing bacteria, nitrate reducers, denitrifiers and phosphorus decomposing bacteria were then cultured and counted according to procedures described in part 1 of this study (Xue and Zheng, 1986). The numbers of microorganisms (cfu) were determined in per gram of dry sediment (cfu/g dry sediment).

117 strains were screened from the sediments between October 1995 and March 1996, and were classified according to Bergy's manual (Buchanan and Gibbon, 1974).

Results and discussion

Table 1 shows the abundance of heterotrophic bacteria in upper layer sediment in May 1995. The aerobic bacteria (average 2.12×10^{5} /g) were almost 100 times more abundant than anaerobic bacteria (average 2.85×10^{3} /g). This implies that dissolved oxygen in the upper layer sediment was relatively rich. Inspection of the sediment confirmed this, and the dissolved oxygen (DO) level in the upper layer was more than 5.5mg/l. These results verify the oxidized state of upper layer sediments from a microbiological parspective.

the upper layer sediment of the West Lake							
Sub-lake	Beili L.	Yuehu L.	Xili L.	Outer L.			
aerobic-bacteria(10 ⁵ /g)	2.76	4.55	1.59	2.07			
anaerobic- bacteria(10 ³ /g) *	1.83	5.32	2.27	1.97			

Table 1. Aerobic and anaerobic heterotrophic bacteria in

*determined by the pyrogallic-acid-vaccum-pump method.

Figure 2 shows the aerobic bacteria distribution during July 1995 to May 1996. Of the sub-lakes, the density was highest in L. Yuehu (average $3.63 \times 10^5/g$) and lowest in L. Xili ($0.94 \times 10^5/g$). For seasonal distributions, the density was greater in March ($3.21 \times 10^5/g$) than in July ($1.49 \times 10^5/g$). These distributions may be determined by the contents of organic substances, the microbe can absorb and the density of zoobenthos fauna devouring microbes.

Figure 3 shows the distribution of actinomycetes in the West Lake sediment. The average density was $1.83 \ge 10^4$ /g, with the highest density occurring in L. Yuehu ($3.31 \ge 10^4$ /g). Medium density in L. Beili, L. Xili and L. Xiaonan , $1.78 \ge 10^4$, $1.68 \ge 10^4$, $1.55 \ge 10^4$ /g, respectively, and Outer lake having the lowest density ($0.84 \ge 10^4$ /g). In figure 3 it can also be seen that actinomycetes were most abundant in July ($3.69 \ge 10^4$ /g) and least abundant in March ($0.57 \ge 10^4$ /g). This suggests that they had positive growth correlation with temperature and a negative correlation with the amount of heterotrophic bacteria present.

It was reported by Li in 1990 that the total organic carbon content in the West Lake sediment reached about 10.5% (w/w). The dominant composition of these substances containing carbon

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was cellulose. Therefore, it can be assumed that an understanding the density and distribution of cellulose decomposing microorganisms can aid in clarifying the carbon cycle of the lake ecosystem. Table 2 shows the amounts of four kinds of cellulose decomposing microorganisms separated from the upper layer sediment in April 1995. Accordingly, aerobic bacteria were slightly more abundant than other types of decomposing microorganisms.

Sub-lake	Bacteria		Fungi		
	Aerobic	Anaerobic	Aerobic	Anaerobic	
Beili L.	1,041	28	1,041	139	
Yuehu L.	8,782	1,976	6,586	1,098	
Xili L.	22,585	1,004	2,258	1,225	
Outer L.	308	269	few	346	
Average	8,179	819	996	702	

Table 2: Cellulose decomposing microorganisms in the sediment of West Lake

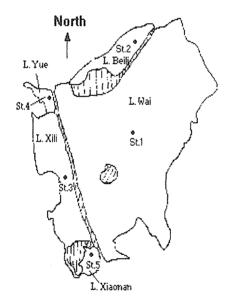


Figure 1. Schematic diagram of sampling stations at West Lake



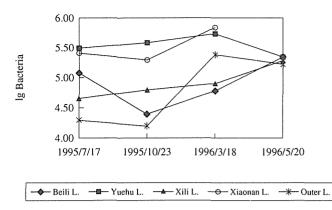


Figure 2. Distribution of aerobic heterotrophic bacteria in the sediment of West Lake

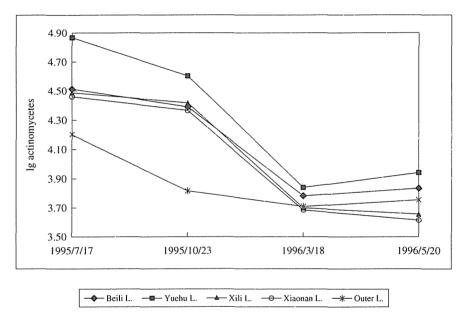


Figure 3. Distribution of actinomycetes in the sediment of West Lake

Table 3 shows the distribution of aerobic deleulose decomposing bacteria in West Lake during May 1995 to March 1996. These bacteria were rich in L. Xili and L. Yuehu, and more abundant in spring (May) and summer (July).

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Sub-lake	95.5	95.7	95.10	96.3	Average
Beili L.	1.10	3.08	0.20	0.31	1.17
Yuehu L.	1.92	6.82	2.06	0.40	2.80
Xili L.	9.09	1.08	0.34	0.27	2.69
Xiaonan L.	nd	4.92	0.59	0.32	1.85
Outer L.	2.95	0.19	0.23	0.16	0.88
Average	3.32	3.22	0.69	0.29	1.88

Table 3. Distribution of aerobic cellulose decomposing bacteria(x 10³/g) in the sediment of West Lake

nd: not determined

As nitrogen in lake ecosystem exists in several forms, nitrogen-utilizing bacteria are also varied. Apart from nitrogen fixing bacteria, the abundance and distribution of several kinds of nitrogen utilizing bacteria was surveyed. Ammonifiers are able to grow on nutrient agar, so their distribution can be seen in figure 1. The abundance and distribution of nitrifiers including nitrite bacteria which oxidize amino-compounds to nitrite and nitrate bacteria which oxidize nitrite to nitrate are shown in Tables 4 and 5, respectively. Nitrate reducers are shown in Table 6, and denitrifiers in Table 7.

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Sub-lake	95.5	95.7	95.10	96.3	96.5	Average
Beili L.	0.73	6.51	0.17	0.17	1.38	1.79
Yuehu L.	17.0	20.1	4.36	0.63	1.13	8.64
Xili L.	0.88	13.4	0.80	0.12	2.22	3.48
L.Xiaonan	N	361	17.6	4.95	N	128
Outer L.	3.52	19.2	7.42	0.19	13.4	8.75
Average	5.53	84.0	6.07	1.21	4.53	27.6

Table 4 Abundance and distribution of nitritebacteria (x 10⁴/g) in the sediment of West Lake

Sub-lake	95.5	95.7	95.10	96.3	96.5	Average
Beili L.	0.29	1.37	0.06	0.17	3.10	1.00
Yuehu L.	10.6	11.4	6.88	3.99	5.64	7.70
Xili L.		5.11	50.5	2.22	22.1	20.0
Xiaonan L.		6.56	0.22	0.01		2.26
Outer L.		3.46	19.5	187	40.2	62.5
Average	5.45	5.58	15.4	38.7	17.8	18.1

Table 5. Abundance and distribution of nitrate bacteria (x 10⁴/g) in the sediment of West Lake

Table 6. Abundance and distribution of denitrifier (x 10³/g) in the sediment of West Lake

95.5	95.7	95.10	96.3	96.5	Average
6.29	1.03	0.27	0.17	3.10	2.17
9.41	6.82	6,88	6.30	>5.26	>6.93
4.16	2.42	0.48	0.02	2.22	1.86
	8.20	1.05	3.14		4.13
1.95	0.31	0.31	0.19	2.25	1.00
5.45	3.76	1.80	1.96	>3.21	3.14
	6.29 9.41 4.16 1.95	6.29 1.03 9.41 6.82 4.16 2.42 8.20 1.95 0.31	6.29 1.03 0.27 9.41 6.82 6.88 4.16 2.42 0.48 8.20 1.05 1.95 0.31 0.31	6.29 1.03 0.27 0.17 9.41 6.82 6.88 6.30 4.16 2.42 0.48 0.02 8.20 1.05 3.14 1.95 0.31 0.31 0.19	6.29 1.03 0.27 0.17 3.10 9.41 6.82 6.88 6.30 >5.26 4.16 2.42 0.48 0.02 2.22 8.20 1.05 3.14 1.95 0.31 0.31 0.19 2.25

From these tables, it can be seen that the numbers of nitrite bacteria had positive correlation with the surrounding temperature, that is, they were most abundant in July and least abundant in March. The other N-utilizing bacteria did not have such a relationship. The variations in the number of bacteria may be explained by the amounts of substrate in thr surrounding environment. The metabolic intensity of microbes has a positive correlation with the surrounding temperature to a certain degree, so rapidly metabolizing ammonifiers produce much more ammonia during the warm month of July. As ammonia is the substrate for nitrite bacteria, the numbers of nitrite bacteria are also higher in July. The West Lake is a shallow touring lake, however, disturbances to the water caused by boats enhances substance exchange between sediment and the water body. This causes the nitrite synthesized by bacteria to be easily released into the water body, resulting in a changeable nitrite content in the sediment. A similar case exists for nitrate. Of course, microorganisms in the upper layer sediment existed with other lake fauna and flora, whoich in turn would also have affected their numbers. For example, the microorganisms could have been devoured by zoobenthos.

Environmental factors such as DO, pH etc. would also have affected the growth cycle of microorganisms. Therefore, further study is required to give a more comprehensive explanation for the variations in the amounts of microorganisms in the sediment.

Sub-lake	95.5	95.7	95.10	96.3	96.5	Average
Beili L.	16.5	1.71	0.30	2.08	3.10	4.74
Yuehu L.	7.56	6.82	50.2	>58.8	1.69	25.0
Xili L.	41.6	2.42	5.05	1.23	1.23	10.3
Xiaonan L.		1.48	25.8	14.9		14.1
Outer L.	1.95	0.35	0.74	33.6	4.05	8.14
Average	16.9	2.56	16.4	>22.1	2.52	12.3

Table 7. Abundance and distribution ($x \ 10^4$ /g) of nitrate reducerin the sediment of West Lake.

From Tables 4 - 7, it can also be seen that nitrite bacteria were most abundant in L. Xiaonan, nitrate bacteria in Outer Lake and L. Xili, and nitrate reducers and denitrifiers were most abundant in L. Yuehu and L. Xiaonan. These abundance variations reflect the different nutrient conditions in the different sub-lakes of West Lake.

Phosphorus decomposing bacteria include organic phosphorus decomposers and inorganic phosphorus dissolving bacteria. A survey conducted in May 1996 showed that organic phosphorus decomposers were most numerous in L. Xili (5.42×10^3 /g), followed by L. Yuehu (1.69×10^3 /g). Outer Lake (1.35×10^3 /g) and L. Beili (1.04×10^3 /g). The number of calcium phosphorus dissolving bacteria was low in each of the four sub-lakes (note that L. Xiaonan was not tested for these bacteria).

117 strains of heterotrophic bacteria were randomly isolated from West Lake sediment between Oct. 1995 and March 1996. They were identified according to Bergey's manual. It was found that almost half were *Bacillus* (50.4%) and about 23.9% were *Enterobacteriaceae*. The remaining bacterial strains included *Aeromonas* (5.1%), *Acinetobater* (4.3%), *Micrococcus* (3.4%), *Cellulomonas* (3.4%), *Staphylococcus* (1.7%), *Pseudomonas* (1.7%), *Plesiomonas* (1.7%), *Alcaligenes* (1.7%), *Lactobacillus* (0.85%), *Agrobacterium* (0.85%) and *Microbacterium* (0.85%).

References

1. Li M. Z.: Studies on the Environment of Chinese Lakes. Beijin. China Environmental Science Press. 158-161. (1990) (in Chinese)

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2. Hattoli: Fundmental Microbiological Ecology. Japan: Tokyo University Press. (1978) (in Chinese)

3. Xue G. H. and Zheng H. Y.: Analysis manual of Soil Microbes. Beijin, Agriculture Press. 131-133 (1986) (in Chinese)

4. Buchanan R. E. and Gibbon N. E.: Bergey's Manual of Determinative Bacteriology (8th ed.). Baltimore, The Williams & Wilkins Company (1974)