

Analysis of microbial populations in River-lake ecotone of Poyang Lake

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Abstract: Using the independent -cultivated molecular biology method, By Next-generation DNA sequencing techniques, Gene sequences of microbial groups for water samples collected from River-lake ecotone of Poyang Lake. water samples were collected from River-lake ecotone of Poyang Lake to determine gene sequences of microbial, using the independent-cultivated molecular biology method and Next-generation DNA sequencing techniques. The data showed that Poyang Lake had the largest number of unique microbial population, followed by the Yao River, the Ganjiang River, and the Rao River. The Species difference was smaller between samples in Ganjiang River and Rao River, while significant differences were found among Lake Poyang and the three other rivers. Poyang Lake was a huge microbial's genes pool, including a large number of the DNA sequence from ammonia oxidizing bacteria to ammonia-oxidizing archaea. It's expected to separate functional microorganisms which may play an important role in the earth nitrogen cycle.

Introduction

Poyang Lake is China's largest freshwater lake, located at 28 ° 22 'N to 29 ° 45'N, 115°47 'E to 116 ° 45'E, on the southern bank of the middle and lower reaches of the Yangtze River in the northern Jiangxi Province. Poyang Lake swallows a large quantity of water coming from 5 tributaries, including Xiu River, Gan River, Fu River, Xin River and Rao River. Due to the unique hydrological regime and special geographical conditions, the water level of Poyang Lake is controlled by both its tributaries and the Yangtze River and has a great seasonal variation, This study takes the terrestrial - aquatic transverse zone of Poyang lake as a case, by Next-generation DNA sequencing techniques, Compared with the ordinary sequencing, the next -generation sequencing technology has the advantage of high flux, short test period, and is low-cost and repeatable^[1]. and has been widely used in all kinds of environments, such as soil^[2], ocean, hot spring, lakes and rivers^[3], the study sets the sampling points in Poyang Lake and Rao River ecotone (hx13 and hx11), Yao Lake and the Gan River ecotone (hu1 and he2), Based on the analysis of the characteristics of the microbial distribution, this work is helpful to enhance understanding between microbes population and environmental change.

Methods

Genomic DNA extractions from water samples

No. 1, 2, 3 and 4, bacterial genomic DNA were extracted from the water samples of Yao Lake, Gan River, Rao River and Poyang Lake, respectively. The molecular weight of genomic DNA

extracted from these water samples was approximately 23 kb, which was in agreement with the expected value.

Next - generation sequencing

The DNA was sequenced through the Illumina platform for Paired-end sequencing ^[4], and the low quality reads were removed under the machine data. The statistical data is shown in Table 1.

Table 1 Sample sequencing data statistics

Sample name	Reads length (bp)	Raw data (Mbp)	Adapter (%)	N base (%)	Ploy base (%)	Low quality (%)	Clean data (Mbp)	Data utilization ratio (%)
he2	250:250	251.23	0	0	0.017	6.438	225.22	89.65
hu1	250:250	192.11	0	0	0.009	8.307	166.66	86.75
hx11	250:250	227.56	0	0	0.015	17.255	158.38	69.60
hx13	250:250	167.94	0	0	0.014	5.146	153.64	91.48

In order to obtain high quality Tags, the splicing Tags sequence was processed. At the 0.97 level of clustering similarity for species classification of OTU (Operational Taxonomic Units), a total of 8657 OTU were obtained for the samples. Statistical results of sample OTU were in table 2.

Table 2 Sample OTU statistics

Sample name	Tag number	OTU number	OTU number (removing singletons)	Non singletons OTU ratio (%)
he2	353,270	1,851	1,493	80.66
hu1	283,182	2,813	1,681	59.76
hx11	265,326	1,491	1,169	78.40
hx13	241,012	4,390	3,047	69.41

Results and Discussion

The most abundant microbial populations was Proteobacteria, Followed by Bacteroidetes, then Actinobacteria, Acidobacteria, Verrucomicrobia, Firmicutes, Planctomycetes Etc. A number of 2974 Crenarchaeota was detected In Poyang Lake, and a little Crenarchaeota in Rao River(table 3).

Table 3 abundance of each sampling point of main microorganisms (Tags number)

Taxon	he2	hu1	hx11	hx13
Crenarchaeota	0	0	3	2974
Euryarchaeota	443	2	162	1
Other Archaea	103	2	70	7272
Acidobacteria	1265	35	400	57158
Actinobacteria	31728	50758	62200	13525
Armatimonadetes	529	4	3437	1018
Bacteroidetes	63464	69527	42588	28077
Chlamydiae	459	31	219	30
Chloroflexi	324	0	335	687
Fusobacteria	1596	50	153	0
Gemmatimonadetes	437	532	97	266
Nitrospira	26	0	20	462
Planctomycetes	3980	5178	1519	3949
Proteobacteria	189505	132484	107727	80613
Verrucomicrobia	22941	14143	20069	8332
Firmicutes	8832	1049	1974	2551

American scholars^[5] successfully isolated a strain of Crenarchaeota *Nitrosopumilus maritimus* from the sea for the first time in 2005, which contained ammonia oxidation that needs ammonia monooxygenase gene *amoA*. It can oxidize ammonia nitrogen to obtain the energy assimilation of inorganic carbon growth. Ammonia-oxidizing bacteria nitrosomonas were relatively rich in Poyang Lake, but it was not detected in the Yao Lake Water. *Nitrospira* has an effect on nitrification and nitrite oxidation^[6], and mainly distributed in the environment that is affected by human activities. The *Chlamydiae* was found in Ganjiang River water flowing through the Nanchang city, which is a kind of special bacteria that can only survive in the cytoplasm, and mainly includes two kinds of bacteria, *Chlamydia trachomatis* and *Chlamydia psittaci*. *Chlamydia trachomatis* usually only infects humans, while *Chlamydia psittaci* can infect many kinds of animal and birds, and lead to respiratory diseases, abortion, and arthritis. Archaea in Yao Lake were rare, and *Fusobacteria* was most abundant in the Ganjiang River water. Studies^[7] have found out that *Fusobacterium* is unusually active in colon cancer cells, and seems to coexist with tumor malignant degree. The planctomycetes also contained some species such as *Candidatus Brocadia*, *Candidatus Kuenenia* and *Candidatus Scalindua*. They are called anaerobic ammonia oxidation bacteria, as they can use nitrite under hypoxia (NO_2^-) oxidation of ammonium ion (NH_4^+) generated nitrogen to obtain energy. It has the vital significance to the global nitrogen cycle, and also important in wastewater treatment. On the whole, microbial components in different geographic space were both related and changed with different environments.

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