Diversity of eukaryotic plankton of aquaculture ponds with *Carassius auratus gibelio,* using denaturing gradient gel electrophoresis

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Abstract

PCR-denaturing gradient gel electrophoresis (DGGE) and canonical correspondence analysis (CCA) were used to explore the relationship between eukaryotic plankton community succession and environmental factors in two aquaculture pond models with gibel carp *Carassius auratus gibelio*. The main culture species of pond 1 were gibel carp and grass carp, and the combined density was 46224 fingerling/ha (gibel carp/grass carp/silver carp/bighead carp, 17:4:6:1). The main culture species of pond 2 was gibel carp, and the combined density was 37551 fingerling/ha (Gibel carp/silver carp/bighead carp, 52:1:1). Water samples were collected monthly. The results showed that the annual average concentrations of TP and PO_4-P in pond 1were significantly higher than pond 2 (p>0.05). The concentration of chlorophyll *a* (chl *a*) has no significantly difference between pond 1 and pond 2. DGGE profiles of 18S rRNA gene fragments from the two ponds revealed that the diversity of eukaryotic plankton assemblages was highly variable. 91 bands and 71 bands were detected in pond 1 and pond 2, respectively. The average Shannon–Wiener index of pond 1 was significantly higher than pond 2. Canonical correspondence analysis (CCA) revealed that temperature played a key role in the structure of the eukaryotic plankton community in both ponds, but the nutrient concentration did not affect it. Our results suggest that DGGE method is a cost-effective way to gain insight into seasonal dynamics of eukaryotic plankton communities in culture ponds, and the increase in the number of filter-feeding silver carp and bighead carp could increase the diversity of the eukaryotic plankton community.

Keywords: 18S rRNA genes, PCR-denaturing gradient gel electrophoresis, CCA, Eukaryotic plankton community, Environmental factors

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Introduction

Zooplankton and phytoplankton are two important biological components, and form the basis of the food web in aquatic ponds. They affect the color and transparency of water, ultimately affecting fish production. The abundance and diversity of phytoplankton and zooplankton vary in freshwater bodies. Previous studies have shown that several environmental factors have been suggested to affect plankton diversity in experimental and natural systems, which include water temperature and trophic state (Jensen *et al.*, 1994; Jeppesen *et al.*, 2000; Abrantes *et al.*, 2006; Lepère *et al.*, 2006). Compared with natural freshwater ecosystems, the ecology of culture ponds consists of many inter-dependent physical, chemical and biological processes, such as manures, fertilizer and feeds applied to the aquaculture pond to enhance production. During the past several decades, much research on aquaculture practice in water quality and its management have focused on reducing risks and improving production (Boyd and Tucker, 1998; Boyd, 2003; Zimba *et al.*, 2003; Rahman *et al.*, 2008) .

Gibel carp (*C. auratus gibelio*) is a popular and economically important aquaculture species in China, due to its good flavor and rapid growth compared with other crucian carps (Xue and Cui, 2001). It is a subspecies of goldfish or crucian carp. There are two main aquaculture pond models in Jiangsu Province of China. The first model is Gibel carp mixed with bighead carp and silver carp, and the other model is Gibel carp and grass carp mixed with bighead carp and silver carp. Nutrition and feed model were previously studied for Gibel carp (Xie *et al*., 2001; Xue and Cui, 2001). However, little research has been done on the ecology of ponds with *C. auratus gibelio*.

PCR-denaturing gradient gel electrophoresis (DGGE) fingerprinting was first used for profiling complex microbial populations by Muyzer *et al*. (1993). It has been widely used in environmental microbiology, and is a recognized method for elucidating differences and similarities of dominant populations of microbial communities (Lindstrom, 2000; Dorigo *et al.*, 2005). In recent years, DGGE has been used to analyze eukaryotic communities using 18S rRNA (Moon-van der Staay *et al*., 2001; Chen *et al*., 2011). The present study investigates the nutrient concentrations and eukaryotic plankton diversity in culture ponds with *C. auratus gibelio*. Canonical correspondence analysis (CCA) methods were used to explore whether there was a close relationship between eukaryotic plankton community succession and

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environmental factors in two aquaculture pond models with *C. auratus gibelio*.

Materials and methods

Site and sampling

The mudflat ponds (salinity: 1–3‰) were located in an aquatic farm in Yancheng, Jiangsu Province, China. The main culture species of pond 1 (33°41.629′N, 120°25.061′E) were gibel carp and grass carp, and the combined density was 46224 fingerling/ha (gibel carp/grass carp/silver carp/bighead carp, 17:4:6:1). The main culture species of pond 2 (33°39.816′N, 120°24.786′E) was gibel carp, and the combined density was 37551 fingerling/ha (gibel carp/silver carp/bighead carp, 52:1:1). *C. auratus gibelio* fingerlings and mixed fish were stocked in the two ponds in April 2011 and December 2010, respectively. The surface areas were 4.37 and 14.38 ha for pond 1 and pond 2, respectively. Water samples were collected monthly at each pond from January to December 2011, from surface water (0–0.3 m) with water samplers.

Chemical analysis

Water temperature was measured in situ. Water samples (5 L) for each point (five sampling points in each pond) were transported to the laboratory. Nutrients (ammonium (NH_4-N), nitrate (NO_3-N), total phosphorus (TP) and ortho-phosphorus (PO_4-P) were analyzed with a Multi-Parameter Water Quality (GDYS-201M) analyzer. Chlorophyll *a* (chl *a*) was measured after collection on glass fiber filters and extracted in 90% acetone in the dark, following Zhang and Huang (1991). For eukaryotic plankton analysis, samples (50 mL) were centrifuged at 4000 rpm for 30 min. The samples for DGGE analysis were fixed in 70% alcohol, and kept at −20°C until DNA was extracted, and the samples for morphological analysis were immediately fixed in 4% (final concentration) formalin. Zooplankton was identified and counted under the microscope according to Wang (1961) and Chiang and Du (1979). Phytoplankton species were identified according to Hu *et al.* (1979).

DNA extraction and PCR amplification

DNA was extracted using a nucleon DNA extraction kit (Easy Pure Genomic DNA kit, Beijing TransGen Biotech). Eukaryotic plankton 18S rRNA genes were amplified using the universal eukaryotic primers F1427-GC and R1616 (van Hannen *et al.*, 1998). Mixed DNA from five extractions was used as PCR templates. Polymerase chain reaction (PCR) conditions for each 50-μL reaction mixture were 1× PCR buffer, 2 mM $MgCl_2$, 3.0 U of Taq polymerase, 80 μM of each deoxynucleotide, 0.3 μM of each

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primer, and approximately 40 ng of mixed template DNA.

The PCR amplification was performed using an automated thermocycler (T-100, Bio-Rad) as follows: one cycle at 94°C for 5 min, 35 cycles at 94°C for 30 s, 64°C for 30 s, 72°C for 1 min, and a final extension at 72°C for 10 min. PCR products were visualized using 1.5% agarose gels stained with GelGreen. A negative control was prepared in the same manner as the samples except that DNA was excluded.

Denaturating gradient gel electrophoresis (DGGE)

A total of 800 ng of PCR product for each sample was loaded on a 10 % (w/v) polyacrylamide gel (37.5:1 acrylamide/bisacrylamide) with a denaturing gradient that ranged from 30% to 50%, where 100% denaturant is defined as 7 M urea and 40% deionized formamide. DGGE was performed with a Dcode system (Bio-Rad Laboratories, USA) using 1×TAE running buffer (20 mM Tris, 10 mM acetic acid, 0.5 mM EDTA, pH 8.0) at 60°C for 16 h at 75 V. The gel was silver stained and photographed using Gel Doc XR+ System (Bio-Rad) under UV light. DGGE were performed separately for pond 1 and pond 2.

Data processing and statistical analysis

Independent sample *t*-test by SPSS16.0 was applied to compare chemical and biological parameters between pond 1 and pond 2. Quantity one 4.6.2 (Bio-Rad Laboratories, CA, USA) was applied to analyze DGGE profiles based on position and intensity. The unweighted pair-group method using arithmetic averages (UPGMA) clustering was used to investigate the similarity between samples, and the Shannon–Wiener Index (H') was calculated to estimate changes in eukaryotic plankton community composition (Shannon and Weaver, 1963).

$$H' = -\sum_{i=1}^{n} p_i \ln p_i$$

Where p_i is the relative intensity of each band and n is the total number of bands in each lane. To reveal the relationships between eukaryotic plankton and environmental parameters, CCA was used because the length of the first DCA (detrended correspondence analysis) axis run on species data was >2. The tested environmental variables were as follows: T, chl *a*, TP, PO_4-P, NH_4-N and NO_3-N. All data were log (x+1) transformed. Environmental factors best describing the most influential gradients in community composition were identified by forward selection with 499 unrestricted Monte Carlo permutations. CCA was performed with the software CANOCO 4.5.

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Results

Physical and chemical characteristics

The nutrient concentrations (NH_4-N, NO_3-N, TP and PO_4-P), physical parameters (water temperature, water depth and area), chl *a* and abundances of crustacean zooplankton of each pond are shown in Table 1. The TP and PO_4-P concentrations in pond 2 significant lower than pond 1 ($p<0.05$), whereas the other nutrient concentrations and biological parameters showed no significant differences between the two ponds ($p>0.05$).

Water temperature was lower in the winter-spring months and higher in the summer-autumn months. The nutrients of the culture ponds with *C. auratus gibelio* showed a similar trend (Fig. 1). The nutrients showed considerable changes during the sampling period, especially in the summer and autumn months. Total phosphorus (TP) presented their lowest values in winter and began to increase in spring in both ponds, and peak values occurred in June and September in pond 1. Concentrations of PO_4-P were higher in the summer, autumn and winter months. Nitrogen levels were higher in the summer–autumn months. Concentrations of nutrients (TP, PO_4-P and NH_4-N) were lower in July. In both ponds, the concentration of chl *a* was significantly higher in April and May for pond 1 and pond 2, respectively. Chl *a* was positively correlated with TP and NH_4-N in pond 2 ($p<0.05$). TP was positively correlated with temperature in pond 1 ($p<0.05$).

Table 1: Physical, chemical, and biological parameters determined for culture for *Carassius auratus gibelio* in the two ponds from January to December 2011.

Environmental factors	Pond 1 mean (range)	Pond 2 mean (range)	*p*-value (t-test)
Water temperature (℃)	14.71(0-26)	14.71(0-26)	_
Depth (m)	2.1	2.0	_
Area (ha.)	4.37	14.38	_
Total phosphorus (TP, mg L^{-1})	0.40(0.11-0.83)	0.29(0.16-0.39)	0.002
Phosphate (PO_4-P, mg L^{-1})	0.24(0.04-0.62)	0.15(0.06-0.27)	0.031
Nitrate-nitrite (NO_3-N, mg L^{-1})	1.6(0.03-6.40)	2.24(0.03-9.37)	0.476
Ammonium (NH_4-N, mg L^{-1})	1.40(0.30-7.05)	1.03(0.11-3.09)	0.752
Chlorophyll a (μg L^{-1})	0.62(0.02-3.09)	0.93(0.01-4.64)	0.278
Alge ($\times 10^4$ mL^{-1})	1.48(0.14-3.02)	4.57(2.81-7.58)	0.857
Rotifera abundance (ind.m L^{-1})	1.05(0.06-3.50)	0.51(0.03-2.15)	0.072

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Continued Table 1:

Environmental factors	Pond 1 mean (range)	Pond 2 mean (range)	*p*-value (t-test)
Copepoda abundance (ind.m L^{-1})	0.027(0-0.08)	0.094(0-0.33)	0.143
Protozoa biomass (ind mL^{-1})	0.10(0.08-0.40)	0.21(0.07-0.73)	0.055
gibel carp（ind.ha^{-1}）	27460	36161	_
grass carp（ind.ha^{-1}）	6865	_	_
silver carp（ind.ha^{-1}）	10297	695	_
bighead carp（ind.ha^{-1}）	1602	695	_

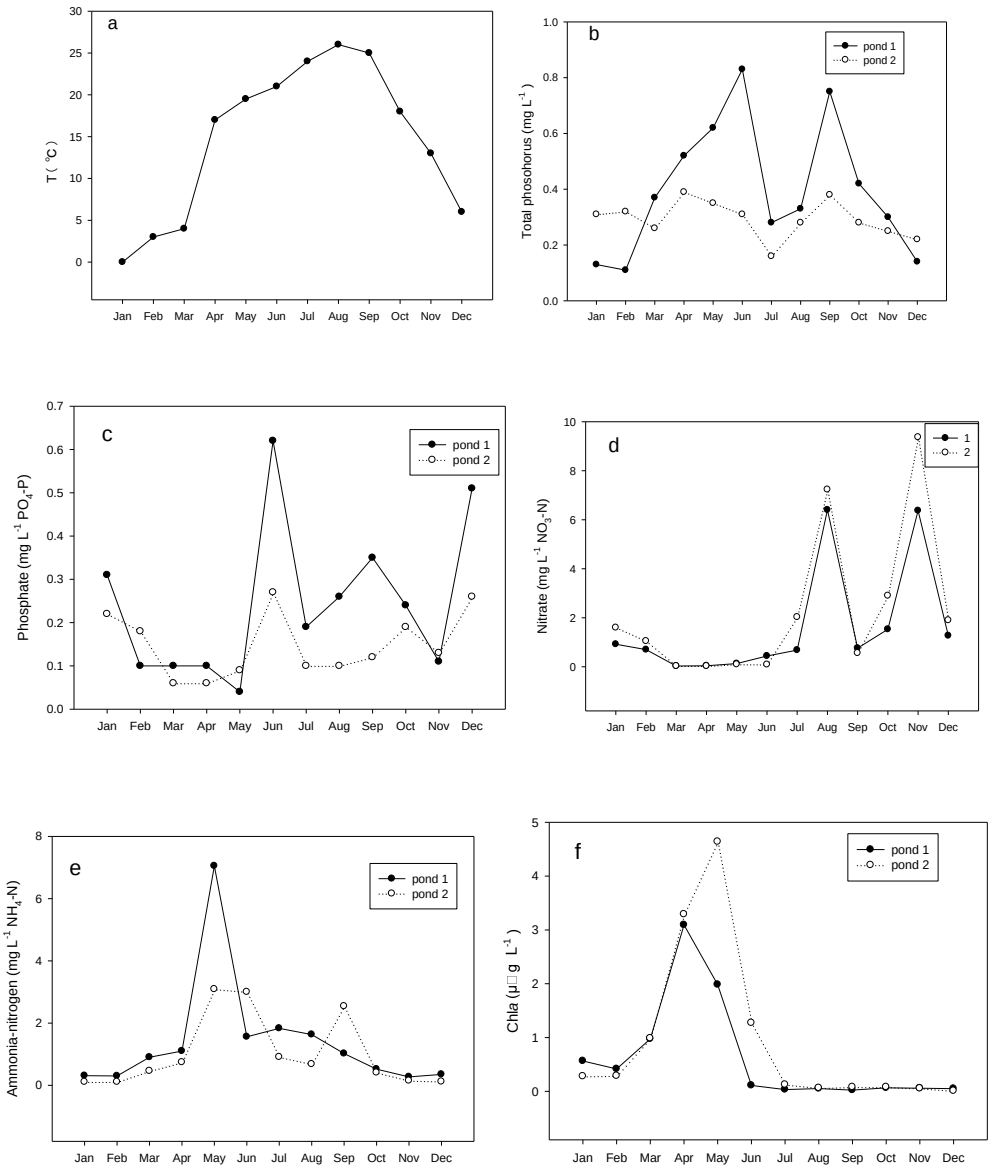

Figure 1: Seasonal variations in temperature (a), total phosphorus (b), phosphate (c), nitrate-nitrite (d), ammonium (e), and chlorophyll *a* (f) from January to December 2011 for culture of *Carassius auratus gibelio* in two ponds.

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Composition of the eukaryotic plankton community

DGGE profiles of 18S rRNA gene fragments from the two ponds revealed that the eukaryotic assemblages were highly variable (Fig. 2). In pond 1, 91 bands were detected, and the average number of bands per sample was 40. However, 71 bands were detected in pond 2, and the average number of bands per sample was 23.75. The number of bands fluctuated strongly in each of the 12 months, especially in pond 1, ranging from 27 in June to 54 in October.

The Shannon–Wiener index calculated from DGGE bands of pond 1 varied from 3.06 to 3.73, while it varied from 2.44 to 3.08 in pond 2 (Fig. 3). The average Shannon–Wiener index of pond 1 was significantly higher than pond 2. The cluster analysis (UPGMA) dendrograms of eukaryotic plankton revealed remarkable seasonality in pond 2, and the eukaryotic plankton communities in various months from pond 2 (Fig. 4) were grouped into four defined clusters. The March sample formed its own separate cluster group, since it was very dissimilar to the other samples. The eukaryotic plankton communities from pond 1 also were grouped into four defined clusters, but its seasonality was not obvious. The August sample was dissimilar to the other samples, and also formed its own cluster group.

CCA

CCA based on DGGE data and environmental variables were carried out separately for the two ponds. According to the results of CCA (Fig. 5a), the differences in eukaryotic plankton were related to the water temperature ($p<0.05$) in pond 1. The two axes explained 60.7% of the observed variation in eukaryotic plankton. The first axis was positively related to the water temperature ($r = 0.75$) and the second axis was positively related to chl *a* ($r = 0.59$). Similarly, results of CCA (Fig. 5b) illustrated that the differences in eukaryotic plankton were also related to the water temperature in pond 2 ($p<0.05$). The two axes explained 64.7% of the observed variation in eukaryotic plankton.

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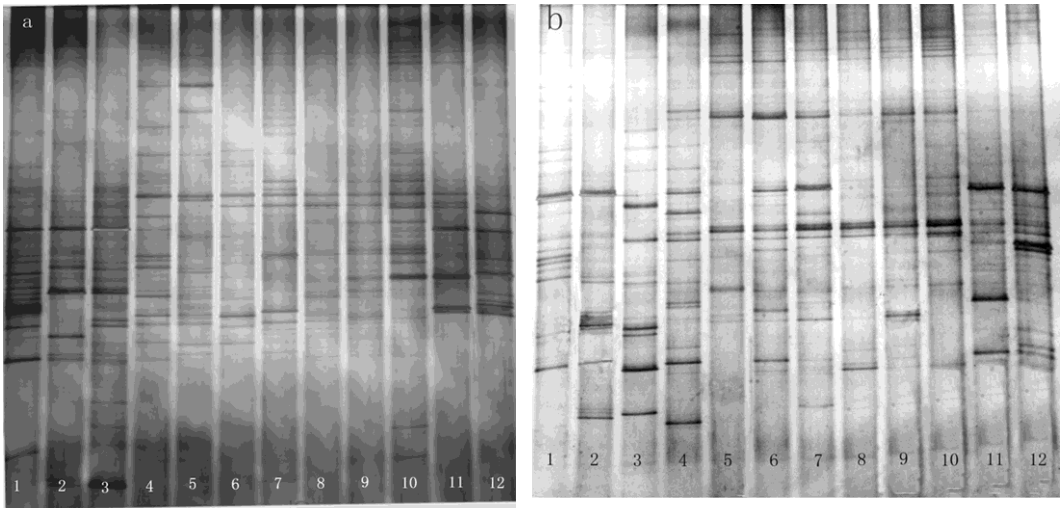

Figure 2: Images of DGGE gels containing eukaryotic 18S rRNA gene fragments from pond 1 (a) and pond 2 (b) culture of *Carassius auratus gibelio.*

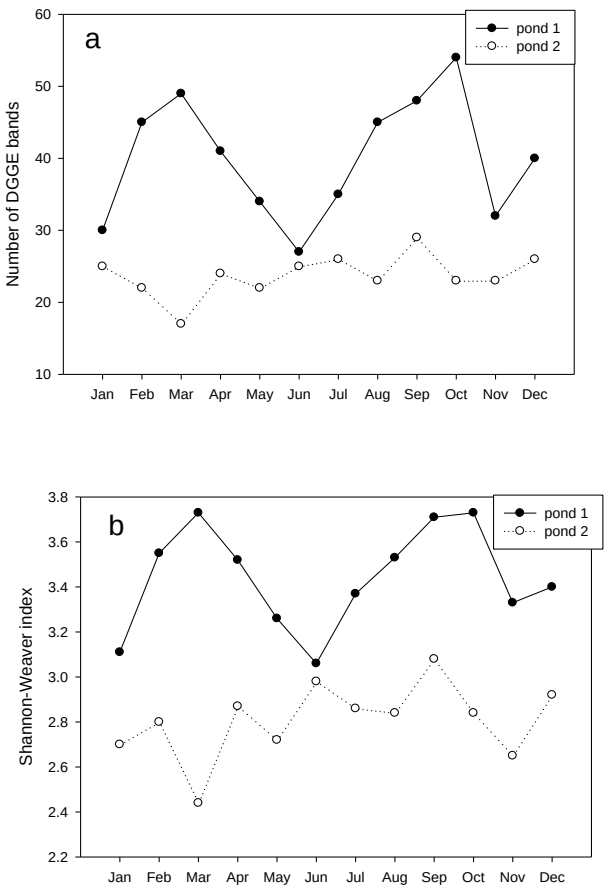

Figure 3: Seasonal variations in DGGE band number (a) and Shannon–Weaver index (b) of eukaryotic plankton from January to December 2011 in the culture of *Carassius auratus gibelio* in two ponds.

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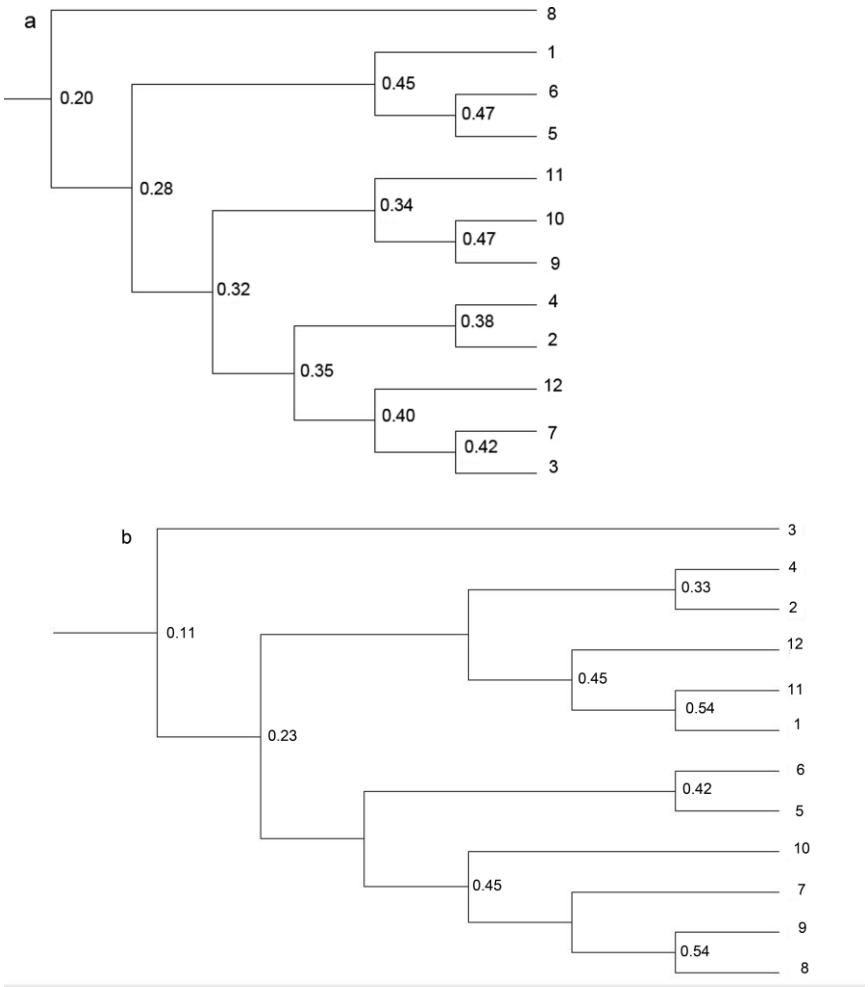

Figure 4: Cluster analysis of eukaryotic plankton based on DGGE profiles of pond 1 (a) and pond 2 (b) from January to December.

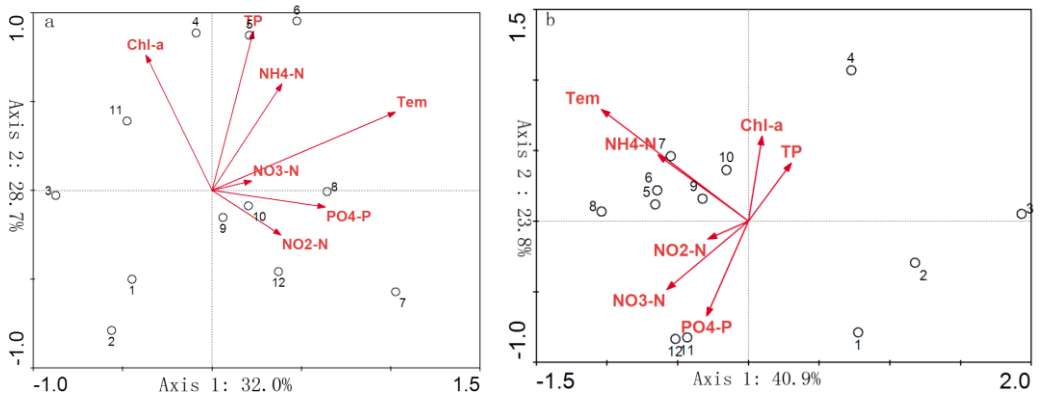

Figure 5: Correspondence canonical analysis (CCA) biplot based on DGGE data and environmental variables of pond 1 (a) and pond 2 (b) from January to December 2011.

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Discussion

The annual nutrient cycle and chla

Phytoplankton is the primary producer of the organic matter on which nearly all other forms of life in any large body of water depend. Chl *a* concentration is a widely used measure of phytoplankton biomass. Many studies showed that chl *a* is regulated by many factors, among them nutrient concentration, especially nitrogen and phosphorus. Based on past literature of natural and artificial lakes, it may be expected that total phosphorus concentrations would explain a significant portion of chl *a* variation (Jones and Bachmann, 1976; Canfield Jr and Bachmann, 1981; McCauley *et al.*, 1989). Tucker and van der Ploeg (1993) suggested that TP concentrations were highly correlated with chl *a* in a catfish pond. In this study, the relationship between TP and chl *a* was also found in pond 2 culture with *C. auratus gibelio*. However, there was no significant correlation between TP and chl *a* (TP and chl *a* varied with no clear pattern) in pond 1, because TP and phosphorus levels in pond 1 were significantly higher than pond 2, and pond 1 had enough phosphorus not to limit phytoplankton growth (Dokulil *et al*., 2000). Tucker and van der Ploeg (1993) suggested that during the summer months, increases in water temperature, feed inputs, and solar irradiance were positively correlated with increased total organic matter, total nitrogen and TP in commercial channel catfish ponds, and chl *a* concentrations and numbers of phytoplankton were high in summer months, and decreased in the fall and winter. In the present study, temperatures increased during spring, and high concentrations of chl *a* occurred in April and May for pond 1 and pond 2, respectively. Then the subsequent decrease of chl *a* might be explained by the top-down effects of fish and zooplankton. *C. auratus gibelio* fingerlings and mixed fish were stocked in the pond 1 and pond 2 in April 2011 and December 2010, respectively. TP and phosphorus concentrations of two ponds were low from January to February, but the TP and phosphorus of pond 1 have a continuous rise from March to June. The peak values of TP and phosphorus of pond 1occurred in June, which were significantly higher than that of pond 2. The high concentration of TP and phosphorus of pond 1 could be related to the high breeding density and the decrease of phytoplankton. The decline in TP and phosphorus concentration in July and August might be explained by water exchange and continuing rainfall.

Environmental factors regulating eukaryotic plankton

Bacteria, phytoplankton and protozoans

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all have major roles in aquatic ecosystems. Previous studies indicated that limnological features and trophic state have been suggested as factors affecting the abundance and diversity of phytoplankton, zooplankton and bacterioplankton community composition in natural freshwater bodies (Lefranc *et al*., 2005; Lepère *et al*., 2006; Niu *et al.*, 2011). In the present study, CCA based on DGGE data and environmental variables was carried out separately for the two ponds. The results illustrated that the plankton abundance and diversity is mainly regulated by temperature ($p<0.05$). Our results agree with observations of several studies that temperature can significantly influence the seasonal variation in plankton (Abrantes *et al*., 2006; de Figueiredo *et al*., 2006). Plankton diversity was lower in winter and summer months than spring and autumn months in pond 1. It is well known that low temperatures limit the growth or occurrences of plankton. However, the decrease in the summer months can be explained by the fact that the dominant phytoplankton taxa changed as temperature and nutrient loads increased (Elliott *et al.*, 2006) .

Nutrient concentration and composition may directly influence phytoplankton biomass by affecting the growth of algae. Some researchers have hypothesized that phytoplankton diversity declines along with the eutrophication of lakes (Reynolds, 1984; Harper, 1992). Leibold (1999) documented a unimodal relationship between diversity of plankton (phytoplankton and zooplankton) and nutrient levels in fishless ponds, and observed that ponds with higher nutrient levels had lower taxonomic diversity. However, none of these factors were significantly related to eukaryotic plankton diversity in our study. Compared to natural lakes, the aquaculture ponds would be classified as hypereutrophic because of high fish biomass and allochthonous nutrients from feed entering the ponds (Zimba *et al*., 2003). Tucker indicated that the ponds cultured for channel catfish had a broad range of trophic states. Phytoplankton biomass in channel catfish ponds was limited by light availability rather than nutrients (Tucker and van der Ploeg, 1993).

Compared to pond 1, the eukaryotic plankton diversity of pond 2 was relatively low and stable throughout the year, which might be related to the different aquaculture pond models. It has been suggested that the number of filter-feeding silver carp could decrease the netplankton (phyto- and zooplankton) and increase the nannoplankton which passes through the filter apparatus of the fish (Milstein *et al*., 1988; Radke and

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Kahl, 2002; Tucker *et al*., 2003). Smith (1985) suggested filter-feeders reduce algal biomass, but increase phytoplankton diversity. In this study, the stocked density of silver carp and bighead carp of pond 1 were about 15 times and two times greater than that of pond 2, respectively. They were able to substantially reduce phytoplankton and zooplankton biomass in the pond, and so the biomass of plankton and chl *a* of pond 2 was larger than pond 1, although the differences were no significant (Table 1). There was no obvious difference between the plankton species of the two ponds when observed by microscope. DGGE showed more plankton diversity than the microscopic method. Theoretically, each DGGE band represents a different species, and the intensity of each band is a qualitative measure of the proportion of that species in the DNA sample. So the DGGE technique has more advantages in revealing the occurrence of small microbial eukaryotes, which may be overlooked by the microscopic method, and the filter-feeding fish may increase the diversity of eukaryotic plankton.

Our results indicated that eukaryotic plankton assemblages, as profiled by PCR–DGGE of 18S rRNA genes in the two different culture pond models, underwent pronounced seasonal changes, and that these changes were significantly related to water temperature, but not related to nutrient concentration. Moreover, the eukaryotic plankton communities may be regulated by filter-feeding fish, and the increase in the number of filter-feeding silver carp and bighead carp could increase the diversity of the eukaryotic plankton community.

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References

Abrantes. N., Antunes, S.C., Pereira, M.J. and Goncalves, F., 2006. Seasonal succession of cladocerans and phytoplankton and their interactions in a shallow eutrophic lake (Lake Vela, Portugal). *Acta Oecologica*, 29, 54-64. doi: 10.1016/j. actao.2005.07.006.

Boyd, C.E., 2003. Guidelines for aquaculture effluent management at the farm-level. *Aquaculture*, 226, 101-112. doi:10.1016/S0044-8486(03)00471-X.

Boyd, C.E. and Tucker, C.S., 1998. Pond aquaculture water quality management. Springer-Verlag New

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[DOR: 20.1001.1.15622916.2016.15.4.13.3]

York.

Canfield Jr, D.E. and Bachmann, R.W., 1981. Prediction of total phosphorus concentrations, chlorophyll a, and secchi depths in natural and artificial lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, 38, 414-423. doi:10.1139/f81-058.

Chen, M.J., Chen, F.Z., Zhao, B.Y., Wu, Q.L. and Kong, F.X., 2011. Seasonal variation of microbial eukaryotic community composition in the large, shallow, subtropical Taihu Lake, China. *Water Research*, 45, 4169-4182. doi:10.1007/s10452-009-9254-7.

Chiang, S.C. and Du, N.S., 1979. Fauna sinica, crustacean: freshwater Cladocera. Academia Sinica. Science Press, Beijing, China

de Figueiredo, D.R., Reboleira, A.S.S.P., Antunes, S.C., Abrantes, N., Azeiteiro, U., Goncalves, F. and Pereira, M.J., 2006. The effect of environmental parameters and cyanobacterial blooms on phytoplankton dynamics of a Portuguese temperate lake. *Hydrobiologia*, 568, 145-157. doi:10.1007/s10750-006-0196-y.

Dokulil, M.T., Chen, W. and Cai, Q., 2000. Anthropogenic impacts to large lakes in China: the Tai Hu example. *Aquatic Ecosystem Health and Management*, 3, 81–94. doi:10.1080/14634980008656993

Dorigo, U., Volatier, L. and Humbert, J.F., 2005. Molecular approaches to the assessment of biodiversity in aquatic microbial communities. *Water Research*, 39, 2207-2218. doi:10.1016/j.watres.2005.04.007.

Elliott, J.A., Jones, I.D. and Thackeray, S.J., 2006. Testing the sensitivity of phytoplankton communities to changes in water temperature and nutrient load, in a temperate lake. *Hydrobiology*, 559, 401-411. doi:10.1007/s10750-005-1233-y.

Harper, D., 1992 Eutrophication of freshwaters: principles, problems and restoration. Chapman and Hall, London

Hu, H.J., Li, R., Wei, Y.X., Zhu, C., Chen, J., Shi, Z.X., 1979. Freshwater algae in China. Science Press, Shanghai, China.

Jensen, J.P., Jeppesen, E., Olrik, K. and Kristensen, P., 1994. Impact of nutrients and physical factors on the shift from cyanobacterial to chlorophyte dominance in shallow Danish lakes. *Canadian Journal of fisheries and aquatic sciences*, 51, 1692-1699. doi: 10.1139/f94-170

Jeppesen, E., Peder Jensen, J., Søndergaard, M., Lauridsen, T. and Landkildehus, F., 2000. Trophic structure, species richness and biodiversity in Danish lakes: changes

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along a phosphorus gradient. *Freshwater Biology*, 45, 201-218. doi:10.1046/j.1365-2427.2000.00675.x

Jones, J.R. and Bachmann, R.W., 1976. Prediction of phosphorus and chlorophyll levels in lakes. *Water Pollution Control Federation*, 48, 2176-2182.

Lefranc, M., Thenot, A., Lepere, C. and Debroas, D., 2005. Genetic diversity of small eukaryotes in lakes differing by their trophic status. *Applied and Environment Microbiology*, 71, 5935-5942. doi:10.1128/AEM.71.10.5935-5942.2005.

Leibold, M.A., 1999. Biodiversity and nutrient enrichment in pond plankton communities. *Evolutionary Ecology Research*, 1, 73-95.

Lepère, C., Boucher, D., Jardillier, L., Domaizon, I. and Debroas, D., 2006. Succession and regulation factors of small eukaryote community composition in a lacustrine ecosystem (Lake Pavin). *Applied and Environmental Microbiology,* 72, 2971-2981.doi: 10.1128/AEM.72.4.2971-2981.2006.

Lindstrom, E.S., 2000. Bacterioplankton community composition in five lakes differing in trophic status and humic content. *Microbial Ecology,* 40, 104-113. doi:10.1007/s002480000036.

McCauley, E., Downing, J.A. and Watson, S., 1989. Sigmoid relationships between nutrients and chlorophyll among lakes. *Canadian Journal of Fisheries and Aquatic Sciences,* 46, 1171-1175. doi:10.1139/f89-152.

Milstein, A., Hepher, B. and Teltch, B., 1988. The effect of fish species combination in fish ponds on plankton composition. *Aquaculture Research*, 19, 127-137. doi:10.1111/j.1365-2109.1988.tb00415.x.

Moon-van der Staay, S.Y., De Wachter, R. and Vaulot, D., 2001. Oceanic 18SrDNA sequences from picoplankton reveal unsuspected eukaryotic diversity. *Nature,* 409, 607-610. doi:10.1038/35054541.

Muyzer, G., De Waal, E.C. and Uitterlinden, A.G., 1993. Profiling of complex microbial populations by denaturing gradient gel electrophoresis analysis of polymerase chain reaction-amplified genes coding for 16S rRNA. *Applied and Environment Microbiology,* 59, 695-700.

Niu, Y., Shen, H., Chen, J., Xie, P., Yang, X., Tao, M., Ma, Z.M. and Qi, M., 2011. Phytoplankton community succession shaping bacterioplankton community composition in Lake Taihu, China.

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Water Research, 45, 4169-4182. doi:10. 1016/j.watres. 2011.05.022

Radke, R.J. and Kahl, U., 2002. Effects of a filter-feeding fish [silver carp, *Hypophthalmichthys molitrix* (Val.)] on phyto- and zooplankton in a mesotrophic reservoir: results from an enclosure experiment. *Freshwater Biology*, 47, 2337-2344. doi: 10. 1046/j.1365-2427.2002. 00993.x

Rahman, M.M., Nagelkerke, L.A.J., Verdegem, M.C.J., Wahab, M.A. and Verreth, J.A.J., 2008. Relationships among water quality, food resources, fish diet and fish growth in polyculture ponds: A multivariate approach. *Aquaculture*, 275, 108-115. doi:10.1016/j. aquaculture.2008.01.027

Reynolds, C.S. (ed)., 1984. The ecology of freshwater phytoplankton. Cambridge University Press, Cambridge.

Shannon, C.E. and Weaver, W., 1963. The mathematical theory of communication. University of Illinois Press, Urbana.

Smith, D.W., 1985. Biological control of excessive phytoplankton growth and the enhancement of aquacultural production. *Canadian Journal of Fisheries and Aquatic Sciences*, 42, 1940-1945.

Tucker, H., Eversole, A.G. and Brune, D.E., 2003. Comparative Nile tilapia and silver carp filtration rates of partitioned aquaculture system phytoplankton. *Aquaculture*, 220, 449-457. doi:10.1016/S0044-8486(02)00614-2

Tucker, C.S. and van der Ploeg, M., 1993. Seasonal changes in water quality in commercial channel catfish ponds in Mississippi. *Journal of the World Aquaculture Society*, 24, 473–481. DOI:10.1111/j. 1749-7345.1993.tb00576.x

van Hannen, E.J., van Agterveld, M.P., Gons, H.J. and Laanbroek, H.J., 1998. Revealing genetic diversity of eukaryotic microorganisms in aquatic environments by denaturing gradient gel electrophoresis. *Journal of Phycology*. 34, 206-213. doi:10.1046/j.1529-8817.1998.340206.x

Wang, J.J. (ed), 1961. Freshwater rotifers in China. Science Press, Beijing, China.

Xie, S., Zhu, X., Cui, Y., Wootton, R.J., Lei, W. and Yang, Y., 2001. Compensatory growth in the gibel carp following feed deprivation: temporal patterns in growth, nutrient deposition, feed intake and body composition. *Journal of Fish Biology*, 58, 999-1009. doi: 10.1111/j. 1095-8649.

Xue, M. and Cui, Y.B., 2001. Effect of

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[DOR: 20.1001.1.15622916.2016.15.4.13.3]

several feeding stimulants on diet preference by juvenile gibel carp (*Carassius auratus gibelio*), fed diets with or without partial replacement of fish meal by meat and bone meal. *Aquaculture*, 198, 281-292. doi:10.1016/S0044-8486(00)00602-5

Zhang, Z.S. and Huang, X.F. (eds), 1991. Methods for the plankton study in freshwater. Science Press, Beijing, China.

Zimba, P.V., Mischke, C.C. and Brashear, S.S., 2003. Pond age–water column trophic relationships in channel catfish I*ctalurus punctatus* production ponds. *Aquaculture*, 219, 291-301. doi:10.1016/S0044-8486(02)00427-1

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