

RELATIONSHIP BETWEEN PHYTOPLANKTON FUNCTIONAL GROUPS AND ENVIRONMENTAL FACTORS IN THE HARBIN SECTION OF SONGHUA RIVER, NORTHEAST CHINA

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Abstract. In this study, samples were collected from 8 sites in the Harbin section of Songhua River, northeast China. We aimed to analyze how phytoplankton functional groups and environmental factors change from May to October during 2018. We identified 16 functional groups (C, D, F, G, H1, J, L0, MP, P, S1, TB, W1, W2, X1, X2 and Y) from 84 species and 7 (C, D, MP, W1, W2, X1 and X2) of them were dominant. The seasonal succession of dominant phytoplankton functional groups was the following: X2/C→X2/C/MP→X2/C/X1/MP. X2 and C always played a dominant role throughout the year. Results of the one-way ANOVA test and RDA revealed that WT, SD, NTU, Chl-a, DO, BOD5, NH4⁺, TP and Fe³⁺ were the major influencing factors. This study revealed that the dominant functional groups were positively correlated with SD and Chl-a and negatively correlated with NTU and TP.

Keywords: *phytoplankton functional groups, environmental factors, Songhua River, RDA, spatial and seasonal variation, biomass*

Introduction

Phytoplanktons are microbes that provide the foundation for aquatic food chains, and include zooplankton, fish and other aquatic animals in water environments and most important biological component in the River ecosystem. As a key part of material circulation and energy conversion in river ecosystem, phytoplankton has a very important impact on the status of aquatic ecosystem (Qiu et al., 2012). Meanwhile, phytoplankton function groups are communities of phytoplankton with similar functions in aquatic ecosystems (Reynolds et al., 2006; Crossetti et al., 2013; Huszaret al., 2015; Qu et al., 2019). It could be more directly described the growth of different species within a group in phytoplankton communities

through the relatively simple classification of phytoplankton groups with observed data (Maggio et al., 2016). At present, Several studies have reflected the responsiveness of phytoplankton to environmental abiotic factors (Kruk et al., 2002; Bohnenberger et al., 2018), including water temperature (Hong et al., 2014), acidity (Klug et al., 2001), water level fluctuations (Su et al., 2019), total phosphorus and dissolved carbon (Klug et al., 2001), nutrient loading (Cupertino et al., 2019), etc. In addition, phytoplankton community development is influenced by the inter-specific interaction of organisms (Kim et al., 2020). However, most previous studies on phytoplankton functional groups had focused on the effects of a single environmental factor or a few species in a phytoplankton communities (Wang et al., 2020). The coexistence and succession of different phytoplankton functional groups under the same or similar environmental conditions have not been fully explained. The seasonal variation of environmental and biological factors are quite dynamic, they have a cumulative effect on different phytoplankton functional groups successions (Chuo et al., 2019). As such, more research is needed to quantify the interactions of stress effects of different factors on aquatic organisms in the future. This study can also meet the growing demand for sustainable watershed management of aquatic biota.

Songhua River, one of the 7 largest rivers in China, is the largest river in Heilongjiang Province and one of the key monitoring regions of the aquatic biodiversity conservation in China. According to the Xinhua News Agency, Songhua River of 1,900-km-long meanders through an area of 557,200 km², spanning Jilin Province and Heilongjiang Province, northeast China. According to the topography and channel characteristics, Songhua River is divided into three sections: the upper, middle and lower reaches. Harbin section of Songhua River is located in the middle reaches of the Songhua River. Harbin section of Songhua River starts from Sanjiazhi in the upper reaches and ends at Dadingzi mountain in the lower reaches. There are two first grade tributaries, Ashi River and Hulan River. Along the Songhua River, there are Sanjiazitan wetland, Yangmingtan wetland, Jinhewan Wetland Park, Hulan estuary wetland park and other wetland parks. Songhua River would not only meet the domestic water demand of Harbin's industry and its people but also meet the ecological demand of landscape. Therefore, in order to ensure the healthy and sustainable development of the water ecological environment in the Harbin section of Songhua River, it is particularly important to investigate the changes of phytoplankton functional groups and analyze the relationship between phytoplankton functional groups and water environmental factors.

In this study, we monitored and calculated the biomass of the phytoplankton functional groups and the related water environmental factors in 2018. we analyzed the interaction between phytoplankton functional group and water environmental factors by redundancy analysis (RDA). This paper puts forward the experimental data support for the water environmental ecosystem and sustainable development of water resources of Songhua River, northeast China.

Materials and methods

Field sampling and measurements

According to the local climate and geographical characteristics, we investigated on the phytoplankton in the Harbin section of Songhua River, Heilongjaing Province, northeast China. Samples were collected seasonally (spring: May 8, 2018; summer: July 20, 2018; autumn: October 9, 2018) at eight sampling stations (*Fig. 1; Table 1*), respectively. Water samples were collected from 9:00 am to 5:00 pm.

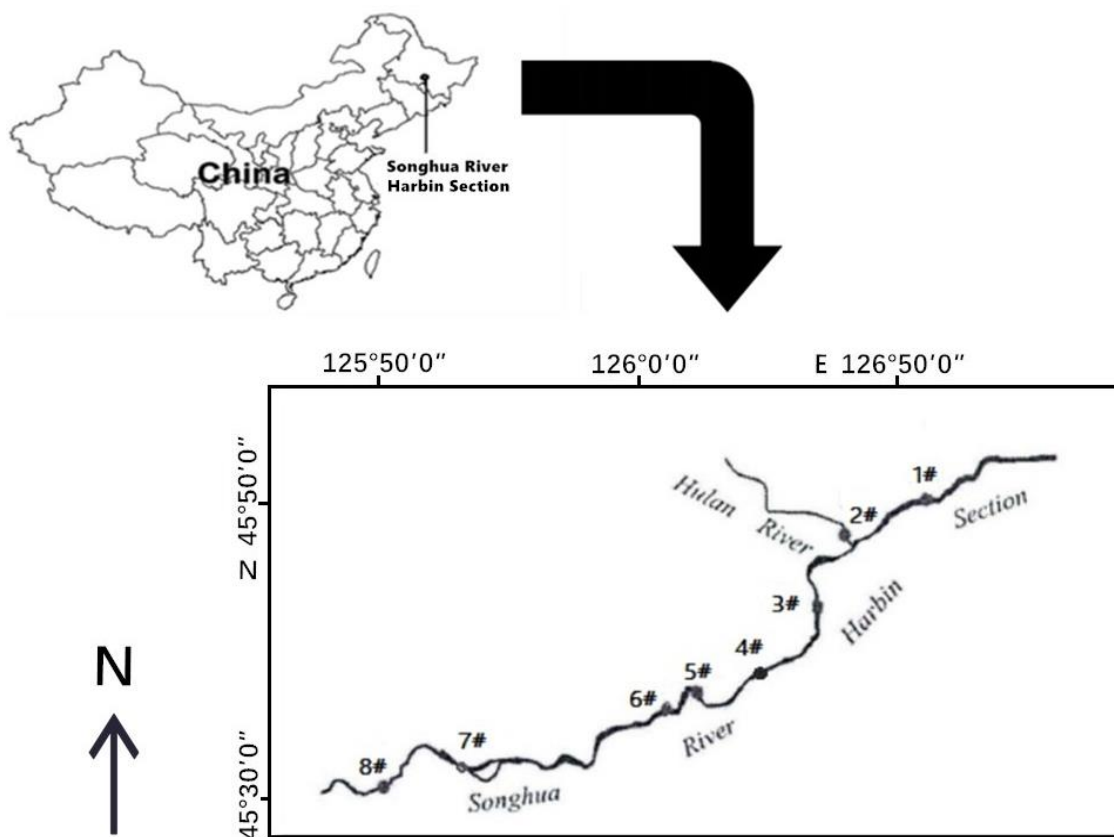


Figure 1. The sampling stations in the Harbin section of Songhua River, Northeast China

Table 1. The number, GPS and water depth of each sampling station in the Harbin section of Songhua River, Northeast China

Number	Station	GPS	Water depth (m)		
			Spring	Summer	Autumn
1#	Daliangzi village	N : 45°58'30" E : 126°51'39"	4.50	8.30	26.80
2#	Hulan estuary wetland park	N : 45°55'28" E : 126°47'20"	3.30	9.00	5.70
3#	Sanjiazitan wetland	N : 45°54'40" E : 126°45'52"	7.50	3.50	4.50
4#	Cement plant	N : 45°51'44" E : 126°42'23"	13.20	12.10	10.20
5#	The second water source	N : 45°45'23" E : 126°32'59"	9.80	11.70	12.70
6#	Yangmingtan wetland	N : 45°46'12" E : 126°31'13"	7.90	15.30	13.70
7#	Jinhewan wetland park	N : 45°46'10" E : 126°28'55"	2.50	3.10	2.20
8#	Sanjiazi village	N : 45°32'23" E : 125°53'11"	1.30	5.50	1.80

Sample collection and determination

Phytoplankton samples were collected with a 5-L water sampler, and mixed samples of the upper water layer, the mid-water layer and the bottom water layer. Integrated 1000 mL water samples were preserved with acid Lugol iodine solution immediately and stored in the dark. Water sample was Siphoned to 30 mL into polythene plastic bottle after 48 h. And then qualitative and quantitative analysis was performed with 40 times magnification by optical microscope (Motic BA210, Motic Inc., China) ($V = 0.1$ mL) (Utermöhl, 1958). Phytoplankton species were identified using the references from different literature (Hustedt, 1930; Prescott, 1954; Patrick et al., 1966; Komarek, 1983; Lange-Bertalot, 2001; Hu et al., 2006). The species were grouped into functional group as described by literature of science (Reynolds et al.; 2002; Reynolds, 2006; Padisak et al., 2009).

Transparency (SD) was estimated with a 20-cm diameter, black-white Secchi disc. Water depth and Flow rate (FR) were measured with the LCD digital sounder (HONDEX PS-7, HONDEX Inc., Japan) and hand-held current meter of YSI (YSI FlowTracker, YSI Inc., USA), respectively. Water temperature (WT) was measured with thermometer. PH, turbidity (NTU) and dissolved oxygen (DO) were recorded real-time by a multi-parameter water quality sonde (YSI 6600, YSI Inc., USA). Chlorophyll a (Chl-a), potassium permanganate index (COD_{Mn}), chemical oxygen demand (COD_{Cr}), 5-day biochemical oxygen demand (BOD_5), ammonium nitrogen (NH_4^+), total phosphorus (TP), total nitrogen (TN), fecal coliform (FC), and Fe^{3+} were checked and analyzed according to standard examine methods on drinking water (GB/T5750-2006) of the Chinese standard methods proposed by Ministry of Environmental Protection of People's Republic of China (MEP, 2006).

Statistical analyses

For this analysis, these phytoplankton species we selected were identified the dominant species ($y \geq 2$) of the study period, and their relative abundance > 1 at least in one sampling station (Xu et al., 1989; Lopes et al., 2005). The dominant species of phytoplankton (Y) was calculated by *Equation 1* (Lampitt et al., 1993):

$$Y = \left(\frac{n_i}{N} \right) f_i \quad (\text{Eq.1})$$

n_i is an individual amount of the phytoplankton species, N is the sum of all individual amount (Gafan et al., 2005), f_i is the appearance frequency of the occurrence of i species in all phytoplankton samples, i is considered as the dominant species if $y \geq 0.02$.

We selected one-way analysis of covariance (ANCOVA) to establish the significant difference among physico-chemical characteristics during the study period, and the ordinations to run using computer program SPSS 16.0. All biomass data of functional groups and continuous environmental factors except for pH were $\log_{10}^{(X+1)}$ transformed to stabilize variance and reduce the influence of dominant taxa on the ordination and subjected to redundancy analysis (RDA) using down weighing of rare species (Jongman et al., 1987; ter Braak, 1988, 1990, 1994). Relationships between the phytoplankton functional groups and the environmental factors were assessed using redundancy analysis (RDA), which is a linear method of direct ordination. RDA was performed using CANOCO for Windows 4.5 (ter Braak et al., 1998), with forward selection to

identify the environmental variables that explained the phytoplankton functional groups, which is a linear method of direct ordination (ter Braak, 1994). The significance of the variables and the first ordination axis was determined using Monte Carlo permutation testing (499 permutations), implemented in CANOCO.

This kind of technique can perform both regression and data ordination concurrently. The significance of the correlations between environmental factors and the species distribution was tested by one-way ANOVA test and redundancy analysis (RDA). Environmental factors measurement included WT (°C), pH, FR (m/s), SD (cm), turbidity (NTU), DO (mg/L), Chl-*a*/ (ug/L), COD_{Mn} (mg/L), COD_{Cr} (mg/L), BOD₅ (mg/L), NH₄⁺ (mg/L), TP (mg/L), TN (mg/L), FC (mg/L), Fe³⁺ (mg/L). Only the samples which most environmental data were used in the RDA analysis to study the correlations with environmental factors on phytoplankton distribution.

Results

Characteristics of phytoplankton functional groups

A total of 84 species of phytoplankton were identified in the 24 samples collected from Harbin section of Songhua River, northeast China, belonging to 6 phyla, 8 classes, 15 orders, 26 families and 50 genera. Among these species, 37 belong to Chlorophyta, 29 belong to Bacillariophyta, and 9 belong to Cyanophyta, which represent approximately 44%, 35%, and 11% of the total species, respectively. In addition, the samples included six species in Euglenophyta (7%), two species in Cryptophyta (2%), and one species in Dinophyta (1%). And then classified into 16 functional groups namely: C, D, F, G, H1, J, L0, MP, P, S1, TB, W1, W2, X1, X2, Y (*Table 2*). There were 4 important functional groups, which with relative biomass > 20% at each sampling stations: C, MP, X2, Y. And there were 7 dominant function groups: C, D, MP, W1, W2, X1, X2 (*Table 3*). The seasonal relative biomass of functional groups in this study, X2 and C were dominant at 38.30% and 29.68% in spring, respectively; X2, C and MP were dominant at 40.97%, 15.87% and 13.20% in summer, respectively; X2, C, X1 and MP were dominant at 47.53%, 12.92%, 10.70% and 10.51% in autumn, respectively. It could be seen X2 and C were dominant in this year. In particular, X2 > 50% in 2# and 3# of spring, 2# of summer, 1#,3# and 7# of autumn, respectively. The distribution variation of phytoplankton functional groups is shown in *Figure 2*. Phytoplankton abundance was (407.40-767.40)×10⁴ ind./L and its mean was 524.10×10⁴ ind./L in spring; it was (51.00-156.60)×10⁴ ind./L and its mean was 88.95×10⁴ cell/L in summer; it was (69.60-184.80)×10⁴ cell/L and its mean was 119.03×10⁴ cell/L in autumn. Biomass ranged from 3.06 to 5.10 mg/L and its mean was 3.85 mg/L in spring; it ranged from 0.45 to 1.34 mg/L and its mean was 0.79 mg/L in summer; it ranged from 0.50 to 1.79 mg/L and its mean was 1.02 mg/L in autumn.

Environmental variables

The average concentration of physico-chemical characteristics of Songhua River is shown in *Table 4*. WT ranged between 13.60-23.37 °C with maximum in summer and minimum in spring. WT showed distinguishing seasonal differences ($p < 0.001$) which were higher in summer and relatively lower in autumn. On the contrary, SD ranged between 10.50-58.50 cm with maximum in spring and minimum in summer. SD differed significantly seasonally ($p < 0.001$) with higher significant values in spring than

in summer and autumn. NTU ranged between 7.55-170.30 with maximum in summer and minimum in spring. The river's NTU also varied seasonally ($p < 0.001$) with summer recording the highest values. The Chl-a ranged between 0.45-21.10 with maximum in spring and minimum in autumn. Chl-a differed with higher significant values ($p < 0.001$) in spring than in autumn. DO ranged between 4.70-9.75 mg/L with lower significantly values ($p < 0.001$) in summer than in spring and autumn. BOD₅ was significantly higher in spring and relatively lower in autumn ($p < 0.001$). NH₄⁺ ranged between 0.07-0.83 mg/L with lower significantly values ($p < 0.001$) in autumn than in spring and summer. On the contrary, total phosphorus (TP) ranged between 0.04-0.19 mg/L with higher significantly values ($p < 0.001$) in autumn than in spring and summer. Fe³⁺ ranged between 0.10-1.64 mg/L with lower significantly values ($p < 0.001$) in spring than in summer and autumn.

Table 2. Phytoplankton functional groups (FGs) and their relative biomass (%) in the Harbin section of Songhua River, Northeast China

FGs	Genera	Biomass (%)
C	<i>Asterionella formosa</i> , <i>Cyclotella meneghiniana</i>	24.73%
D	<i>Nitzschia palea</i> , <i>Synedra acus</i> , <i>Synedra amphicephala</i> , <i>Synedra tabulata</i> , <i>Synedra ulna</i>	3.24%
F	<i>Oocystis parva</i> , <i>Oocystis elliptica</i> , <i>Micractinium pusillum</i> , <i>Kirchnerilla lunaris</i> , <i>Selenastrum gracile</i> , <i>Westella botryoides</i> , <i>Westellopsis linearis</i> , <i>Stichococcus bacillaris</i>	0.62%
G	<i>Eudorina elegans</i> , <i>Pandorina morum</i>	0.20%
H1	<i>Anabaena variabilis</i> , <i>Anabaena circinalis</i>	0.01%
J	<i>Golenkinia radiata</i> , <i>Chodatella quadriseta</i> , <i>Chodatella ciliata</i> , <i>Tetraëdron trigonum</i> , <i>Tetraëdron regulare</i> , <i>Actinastrum hantzschii</i> , <i>Crucigenia aiculata</i> , <i>Crucigenia quadrata</i> , <i>Crucigenia tetrapedia</i> , <i>Tetrastrum staurogeniae forme</i> , <i>Tetrastrum elegans</i> , <i>Scenedesmus ovalternus</i> , <i>Scenedesmus platydiscus</i> , <i>Scenedesmus dimorphus</i> , <i>Scenedesmus abundans</i> , <i>Scenedesmus acuminatus</i> , <i>Scenedesmus bijuga</i> , <i>Scenedesmus quadricauda</i>	1.97%
L0	<i>Gyrosigma acuminatum</i> , <i>Merismopedia minima</i> , <i>Chroococcus minutus</i>	0.06%
MP	<i>Cocconeis placentula</i> , <i>Achnanthes linearis</i> , <i>Epithemia turgida var. granulata</i> , <i>Surirella capronii</i> , <i>Surirella angustata</i> , <i>Cymbella ventricosa</i> , <i>Pinnularia major</i> , <i>Pinnularia gibba</i> , <i>Navicula exigua</i> , <i>Navicula radiosa</i> , <i>Navicula dicephala</i> , <i>Navicula anglica</i> , <i>Diatoma vulgare</i> , <i>Diatoma vulgare var. linearis</i> , <i>Coelosphaerium kützingianum</i> , <i>Ulothris variabilis</i>	5.91%
P	<i>Fragilaria virescens</i> , <i>Fragilaria brevistriata</i> , <i>Fragilaria ca pucina</i> , <i>Melosira granulata</i> , <i>Melosira granulata var. angustissima</i> , <i>Melosira granulata var. angustissima f. spiralis</i> , <i>Closterium sp.</i>	1.09%
S1	<i>Planktothrix agardhii</i> , <i>Phormidium allorgei</i> , <i>Phormidium beggiatoiforme</i> , <i>Phormidium lismorense</i>	0.70%
TB	<i>Melosira varians</i>	1.59%
W1	<i>Heteronema discomorphum</i> , <i>Lepocinlis steinii</i> , <i>Euglena oxyuris</i> , <i>Trachelomonas cylindrica</i> , <i>Strombomonas urceolata</i>	1.92%
W2	<i>Trachelomonas granulosa</i>	2.35%
X1	<i>Ankistrodesmus angustus</i> , <i>Ankistrodesmus acicularis</i> , <i>Chlorella vulgaris</i> , <i>Schroederia nitzschoides</i> , <i>Schroederia robusta</i>	7.73%
X2	<i>Chlamydomonas ovalis</i> , <i>Chlamydomonas globosa</i> , <i>Chroomonas acuta</i>	40.34%
Y	<i>Glenodinium pulvisculus</i> , <i>Cryptomonas ovata</i>	7.53%

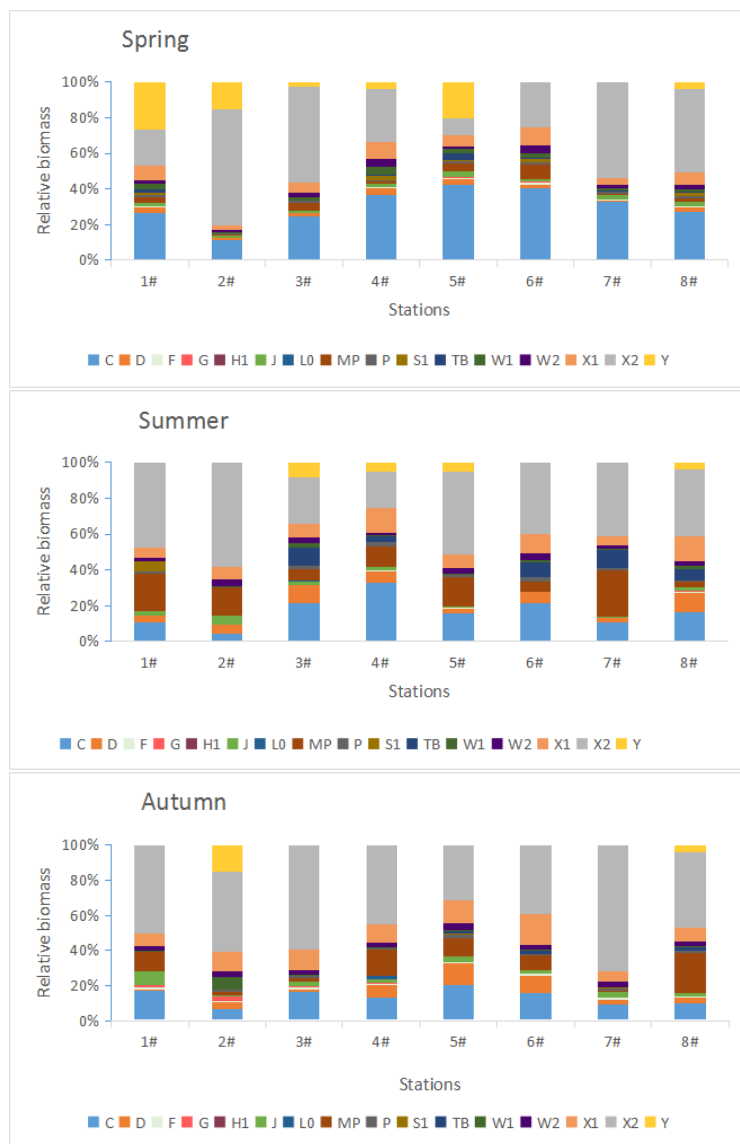


Figure 2. Distribution of relative biomass of phytoplankton functional groups in the Harbin section of Songhua River, Northeast China

Table 3. Appendix list of dominant genus of phytoplankton functional groups in the Harbin section of Songhua River, Northeast China

FGs	Phylum	Genus	Dominance		
			Spring	Summer	Autumn
D	Bacillariophyta	<i>Synedra acus</i>		0.03	0.03
C	Bacillariophyta	<i>Cyclotella meneghiniana</i>	0.47	0.32	0.25
W2	Euglenophyta	<i>Trachelomonas granulosa</i>	0.07	0.11	0.12
W1	Euglenophyta	<i>Trachelomonas cylindrica</i>	0.08	0.04	
X1	Chlorophyta	<i>Ankistrodesmus angustus</i>	0.03		
X1	Chlorophyta	<i>Chlorella vulgaris</i>	0.04	0.10	0.10
MP	Chlorophyta	<i>Ulothrix variabilis</i>			0.05
X2	Chlorophyta	<i>Chlamydomonas ovalis</i>		0.02	0.03
X2	Cryptophyta	<i>Chroomonas acuta</i>	0.08	0.11	0.11

Table 4. Environmental parameters, which contained water temperature (WT), pH, flow rate (FR), Secchi depth (SD), turbidity (NTU), Chlorophyll a (Chl-a), dissolved oxygen (DO), Permanganate index (COD_{Mn}), Chemical oxygen demand (COD_{Cr}), 5-day biochemical oxygen demand (BOD₅), NH₄⁺, total phosphorus (TP), total nitrogen (TN), Fecal Coliform (FC), Fe³⁺, in each season in the Harbin section of Songhua River, Northeast China

Parameters	Spring	Summer	Autumn	P value
WT (°C)	17.55 ± 1.63	22.9 ± 0.45	14.52 ± 0.4	0.000**
pH	7.86 ± 0.41	7.59 ± 0.18	7.72 ± 0.08	0.142
FR (m/s)	0.73 ± 0.58	0.97 ± 0.36	0.45 ± 0.24	0.070
SD (m)	37.5 ± 10.27	11.44 ± 0.82	18.5 ± 3.97	0.000**
NTU	15.91 ± 6.1	141.01 ± 26.9	43.9 ± 11.25	0.000**
Chl-a (ug/L)	14.27 ± 6.94	8.02 ± 2.45	5.98 ± 2.42	0.004**
DO (mg/L)	7.84 ± 0.93	6.6 ± 1.17	8.9 ± 0.29	0.000**
COD _{Mn} (mg/L)	5.56 ± 1	5.33 ± 0.7	4.65 ± 0.55	0.070
COD _{Cr} (mg/L)	12.25 ± 0.6	13.08 ± 1	12.53 ± 1.13	0.217
BOD ₅ (mg/L)	4.74 ± 1.08	2.96 ± 0.8	2.31 ± 0.48	0.000**
NH ₄ ⁺ (mg/L)	0.43 ± 0.3	0.18 ± 0.11	0.13 ± 0.06	0.010**
TP (mg/L)	0.07 ± 0.03	0.1 ± 0.02	0.15 ± 0.03	0.000**
TN (mg/L)	2.05 ± 0.79	2.26 ± 0.46	1.83 ± 0.11	0.288
FC (mg/L)	14050.25 ± 11553.15	26284.88 ± 12806.46	22707.88 ± 12551.8	0.149
Fe ³⁺ (mg/L)	0.19 ± 0.07	0.98 ± 0.22	1.23 ± 0.49	0.000**

**The environmental factors were significantly different at the level of 0.01

Correlation analysis of phytoplankton functional groups and environmental factors

The results of DCA performance by using CANOCO for Windows 4.5 showed that the Standard Deviation value was less than 3, so RDA analysis was carried out to analysis the relationship between phytoplankton functional groups and environmental factors in the Harbin section of Songhua River. The eigenvalue for RDA axis 1 (0.691) and axis 2 (0.156) together explained 84.7% of the functional groups variance out of which 69.1% of the total variability was from axis 1 (Table 5). The results of Monte Carlo test revealed that there were significant differences between the first axes and all canonical axes (F = 17.921, p = 0.0002, F = 4.109, p = 0.004). Axis 1 mainly positive correlated with SD (r = 0.8729), BOD₅ (r = 0.7026) and negatively correlated with Fe³⁺ (r = -0.8241), NTU (r = -0.8163). Axis 2 positively correlated with Chl-a (r = 0.2453) and COD_{Mn} (r = 0.2453) and negatively correlated with NH₄⁺ (r = -0.3812), TP (r = -0.3112).

Table 5. Summary of redundancy analysis (RDA) of phytoplankton functional groups for the first two axes

Axes	I	II	III	IV
Eigenvalues	0.691	0.156	0.027	0.008
Species-environment correlations	0.972	0.838	0.930	0.893
Cumulative percentage variance of species data	69.1	84.7	87.4	88.2
Cumulative percentage variance of species-environment relation	78.1	95.7	98.7	99.7
Sum of all canonical eigenvalue	0.885			

Discussion

Variation of phytoplankton functional groups in the Harbin section of Songhua River, Northeast China

In previous studies, the use of classification of organisms into phytoplankton functional groups have been to be the best way to explain the influence relation between factors of phytoplankton community structure and aquatic systems (Heebert et al., 2016). The Padisak J's study also showed the changes of life-history and physiological activity of phytoplankton functional groups have limited by factors of nutrient, light condition, hydrodynamic character, foraging behavior of zooplankton and hydrology dynamic change characteristics (Padisak et al., 2009). In the recent past, phytoplankton functional groups based on the morphological, as the primary producer in aquatic ecosystem, is a reliable indicator to analysis on water environmental quality and illustrate the trend of spatial and temporal variations (Li et al., 2017). Many recently studies focused on the change trend of biomass of phytoplankton functional groups in relation to their surviving environment (Mwagona et al., 2018; Ma et al., 2019; Sun et al., 2019). During the study period, the average biomass of phytoplankton was 3.85 mg/L in spring, 0.79 mg/L in summer and 1.02 mg/L in autumn. The aforesaid experimental datum were lower compared to the datum of average biomass during spring (4.03 mg/L), summer (2.78 mg/L) and autumn (5.07 mg/L) recorded by Yan (2011) in the Harbin section of Songhua River, and also lower than those during summer (2.30 mg/L) and autumn (2.57 mg/L) of Li (2013) accepted for publication in the Harbin section of Songhua River. So far, the results showed that average biomass of phytoplankton reduce continuously.

Our research showed that the main dominant phytoplankton functional groups were group C and X2 in the whole year. The two phytoplankton functional groups which we investigated were also the dominant functional groups studied by An et al. in Small Xingkai Lake and by Ma et al. Sanhuanpao wetland reservation which located in the same province (An et al., 2016; Ma et al., 2016). Group C represents the eutrophic small and medium sized lakes, and it was suitable for the water environment of no stratification or suspension (Yang et al., 2014). According to Reynolds' phytoplankton functional group theory indicated, group X2, sensitive to the filtering food zooplankton, was inhibited when the filtering food zooplankton were rich (Reynolds et al., 2002). Sampling stations (1#, 2# and 3#) were located in the middle and upper reaches in the Harbin section of Songhua River recorded high relative biomass dominated by group X2 corresponding to low filtering food zooplankton (An et al., 2016; Feng, 2014). Group MP (mainly composed of *Ulothrix variabilis*) was important corresponding to frequently disturbed, inorganic and turbid water areas. In autumn, the wind was strong, the river was disturbed frequently, and the turbidity increased obviously, which was beneficial to the growth of MP (Yang et al., 2014). Therefore, the relative biomass of group MP is not only more than 10% in summer and autumn, but also one of the dominant functional groups in autumn.

Influence factors of water environment of Harbin section of Songhua River, Northeast China

Any environmental factors needed for phytoplankton growth, including temperature, light intensity and hydrodynamics, could be the limiting factors influenced the growth of phytoplankton, and may also be the environmental influencing factor for the

distribution of phytoplankton function groups (Xiao et al., 2011). The RDA analysis was used to explain clearly the relationship between crucial factors and phytoplankton functional groups and to further explore the driving factors changing the composition and distribution of phytoplankton functional group. The results of RDA analysis displayed that phytoplankton functional groups in the Harbin section of Songhua River were affected by SD, BOD₅, Fe³⁺, NTU, Chl-a, COD_{Mn}, NH₄⁺ and TP. Among them, SD was the key of environmental factors ($r = 0.8729$), which affected the change of functional groups throughout the year.

Studies of lakes and reservoirs in different regions of country showed that, such as Tianmu Reservoir of Shahe Reservoir located in north subtropical regions, Changshou Lake located in mid-subtropical regions, Yuqiao Reservoir located in warm temperate zone and Small Xingkai Lake located in temperate zone, SD was the main environmental factors influencing and changing the phytoplankton function groups (Cui et al., 2014; Yan et al., 2018; Liu et al., 2016; An et al., 2016). According to Cui et al. (2014), it was beneficial for phytoplankton growth to be in water bodies of low NTU and high SD, and it found a positive correlation between SD and the biomass of Bacillariophyta and Chrysophyta. Similarly, most of phytoplankton functional groups for this experiment were positive correlation with SD and negative correlation with NTU in the Harbin section of Songhua River. Li et al. (2013) also recorded that the water sediment content was high, SD was low and NTU was high in perennial water of Songhua River. Compared to other water bodies, SD in our research environment was remarkably lower. For instance, in Small Xingkai Lake located in Southeast Heilongjiang Province, An et al. (2016) recorded 0.46 m, 0.21 m, and 0.21 m mean values of SD for spring, summer and autumn respectively. In Aha reservoir, one of the three drinking water sources in Guiyang, SD mean value in spring, summer and autumn was 0.90 m, 0.83 m, and 1.19 m, respectively (Li et al., 2015). According to Ostos et al. (2008), SD of El Gergal reservoir located in southwest Spain ranged from 0.2 m to 8.0 m and its mean was 1.77 m between 1979 and 2003. According to Chai et al. (2016), SD of Pearl River Estuary located in southern China ranged from 0.2 m to 3.2 m and its mean was 1.2 m in spring (March 2012) and ranged from 0.5 m to 2.0 m and its mean was 0.8 m in autumn (November 2011). There were many factors that limited SD, including not only phytoplankton, but also the concentration of sediment, the speed of FR, the amount of suspended solids and many other factors in the water (Pace et al., 1992). The faster FR will increase the suspended matter in the water, and the increase of sediment content will reduce the content of DO in the water. It was not simply to describe this with a single linear correlation (Pace et al., 1992).

Groups X2 were mainly comprised of minute-celled *Chlamydomonas ovalis* and *Chroomonas acuta*, and it was easier to be found it in waters with lower phosphorus levels (Laamanen, 1997). The study of Ma et al. (2019) also showed that group X2 with widely adaptability to nutrients was preferred to grow in slight alkaline water environment. We can see from the RDA biplot diagram in *Figure 3*, that groups X2 had a negative correlation with TP. On the one hand, the research of Siebielec et al. (2015) and da Silva and Fitzsimmons (2016) clearly proved that dissolved calcium ion will react with the phosphate in water forming insoluble calcium phosphate and other compounds that were not conducive to phytoplankton growth and propagation when the water environment was slight alkaline. On the other hand, the presence of large numbers of predators will reduce the number of filter feeders and the grazing pressure of filtering food zooplankton on group X2 (Padisak et al., 2009). *Asplanchna priodonta*, *Polyarthra trigla* and *Synchaeta*

stylata were the dominant species (rotifer) found by Li et al. (2013) in Songhua River. *Synchaeta stylata priodonta*, as one kind of macrofilter-feeders, ate Dictyotaceae, filamentous algae, dinoflagellate, and other 20-50 μm zooplankton. *Asplanchna* and *Polyarthra trigla*, as group RC (predators), ate macroscopic algae, protozoa, rotifer and microcrustacea (Wen et al., 2017). It was more suitable for them to living in the alkaline water like the Songhua River with low temperature and high dissolved oxygen (Li et al., 2013). A study even showed the number of filter feeders increased and the number of phytoplankton decreased when the SD exceeded 80 cm (Li and Li, 2001).

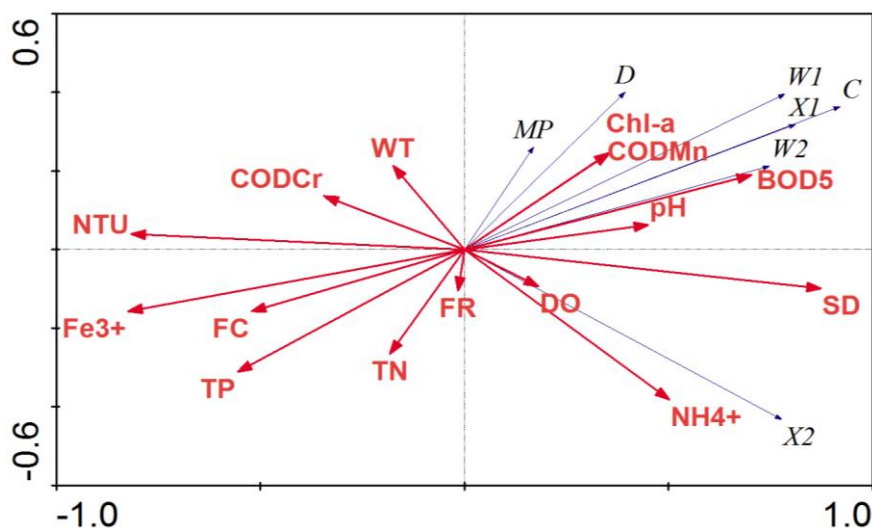


Figure 3. Correlation plots of the redundancy analysis (RDA) on the relationship between the environment variables and phytoplankton functional groups

Researchers had also noted that Chl-a, COD_{Mn} , BOD_5 , Fe^{3+} , NH_4^+ and FC were fundamental factors in shaping phytoplankton community composition. The study of Guo et al. (2020) in Xidayang Reservoir showed that phytoplankton density and biomass were significant positive correlation with Chl-a and COD_{Mn} ($P < 0.05$) and significantly negatively correlated with NH_4^+ ($P < 0.05$). Liu et al. (2014) pointed out that Chl-a, as the main pigment of phytoplankton photosynthesis, which growth was a significant positive predictor of the rapid growth of phytoplankton. In Muling River, the researchers analyzed the COD_{Mn} was positively related with the predominant phytoplankton functional groups (Sun et al., 2019). In Dalian Songshu Reservoir, the researchers analyzed the BOD_5 was positively related with the phytoplankton density (Yang, 2019). Arnaldo concluded that the growth rate of the dominant phytoplankton community structure limited by the concentration of Fe^{3+} in the upper waters layer of the Bransfield Strait in Antarctic Peninsula (D'Amaral et al., 2015). Similarly, from the RDA results, BOD_5 , Chl-a and COD_{Mn} were positive correlated with the dominant functional groups, while Fe^{3+} and NH_4^+ were negatively correlated with the dominant functional groups. Under the same experimental conditions, Mao et al. (1998) monitored the effect of phytoplankton with different densities or community composition on the amount of FC in the culture ponds. It was proved that there was a restrictive relationship between phytoplankton community composition and FC population. FC will release non-specific antibiotics substances to kill algae cells. When the amount of FC in the culture ponds was reduced, the densities of phytoplankton

continued to increase within 24 h. Same as the results in this experiment, the growth of FC and the biomass of phytoplankton had a negative correlation.

Conclusions

In this current study, a total of 84 species of phytoplankton species belonging to 16 functional groups (C, D, F, G, H1, J, L0, MP, P, S1, TB, W1, W2, X1, X2 and Y) were identified in the Harbin section of Songhua River, northeast China. There were 7 dominant functional groups: C, D, MP, W1, W2, X1 and X2. The seasonal dominant functional group succession: X2/C (spring)→X2/C/MP (summer)→X2/C/X1/MP (autumn). The result of one-way ANOVA test and RDA showed that WT, SD, NTU, Chl-a, DO, BOD₅, NH₄⁺, TP and Fe³⁺ were the major factors that influencing phytoplankton functional groups in the Harbin section of Songhua River, northeast China. RDA result revealed that SD and Chl-a were positive correlated with the dominant functional groups, while NTU and TP were negatively correlated with the dominant functional groups. This study revealed that the phytoplankton functional groups followed certain predictable pattern in the seasonal and spatial gradient. It could be useful for assessing and predicting the growth and development of phytoplankton in the future. At the same time, these environmental factors can also be considered as an important basis for the management and protection of rivers in northeast China for future studies.

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APPENDIX

Table A1. Environmental parameters in each season in the Harbin section of Songhua River, Northeast China

	WT (°C)	pH	FR (m/s)	SD (m)	NTU	Chl-a/ (ug/L)	DO (mg/L)	CODMn (mg/L)	CODCr (mg/L)	BOD5 (mg/L)	NH4+ (mg/L)	TP (mg/L)	TN (mg/L)	FC (mg/L)	Fe3+ (mg/L)
Spring1#	17.50	7.85	1.26	58.50	7.55	8.85	7.30	5.04	12.70	5.80	0.72	0.07	3.00	24196.00	0.12
Spring2#	18.00	7.88	0.83	48.00	10.00	12.25	7.00	6.00	12.40	4.30	0.83	0.10	3.32	24196.00	0.22
Spring3#	17.80	7.85	0.28	32.00	23.80	15.20	7.50	5.20	12.20	5.70	0.50	0.09	1.42	12997.00	0.10
Spring4#	18.10	7.99	0.39	31.50	15.15	19.75	7.00	5.84	12.20	5.60	0.63	0.10	2.20	24196.00	0.15
Spring5#	18.50	7.83	0.57	34.50	15.30	15.30	7.90	7.28	12.70	5.30	0.09	0.06	1.68	754.00	0.12
Spring6#	18.40	8.21	0.18	32.00	18.30	20.90	8.60	5.52	11.60	3.50	0.08	0.04	1.01	24196.00	0.23
Spring7#	18.50	8.31	1.89	29.00	24.60	21.10	7.70	5.84	13.00	4.80	0.11	0.06	1.61	393.00	0.22
Spring8#	13.60	6.95	0.41	34.50	12.55	0.80	9.75	3.76	11.20	2.90	0.48	0.04	2.18	1474.00	0.32
Summer1#	22.37	7.57	0.70	11.50	145.35	7.15	6.43	5.65	11.37	3.30	0.23	0.12	2.26	34305.00	1.00
Summer2#	22.93	7.58	0.87	11.50	170.30	10.05	5.93	5.23	13.20	3.53	0.25	0.12	2.17	36593.00	0.92
Summer3#	22.80	7.38	0.63	12.00	101.60	8.40	4.70	4.88	14.10	1.80	0.12	0.10	3.04	48392.00	0.64
Summer4#	23.30	7.64	0.64	10.50	157.65	8.45	6.63	4.85	13.27	3.73	0.38	0.11	2.01	28283.00	0.99
Summer5#	23.20	7.57	1.50	11.50	152.15	9.65	7.07	6.13	11.80	2.97	0.08	0.08	1.89	15291.00	1.28
Summer6#	23.10	7.34	1.05	11.00	146.70	8.90	6.20	4.56	14.10	1.70	0.08	0.09	2.90	17328.00	1.08
Summer7#	23.37	7.74	1.50	13.00	157.30	9.20	7.07	4.85	13.37	3.03	0.09	0.11	1.94	18975.00	1.20
Summer8#	22.10	7.88	0.84	10.50	97.00	2.35	8.80	6.51	13.47	3.63	0.22	0.06	1.88	11112.00	0.69
Autumn1#	14.55	7.73	0.23	19.00	30.30	6.05	8.60	4.56	14.00	2.20	0.19	0.08	2.05	36294.00	0.61
Autumn2#	14.35	7.72	0.29	19.00	34.60	5.65	8.75	4.48	12.95	2.05	0.23	0.16	1.84	32860.00	1.56
Autumn3#	14.50	7.78	0.29	20.00	47.50	6.80	9.30	3.76	11.80	2.60	0.07	0.14	1.70	17329.00	0.56
Autumn4#	14.75	7.68	0.66	12.50	64.65	6.75	8.75	5.36	10.60	2.95	0.16	0.19	1.86	36294.00	1.64
Autumn5#	14.90	7.63	0.45	14.00	49.45	8.65	8.75	5.20	13.55	2.90	0.09	0.15	1.89	11857.00	1.64
Autumn6#	14.00	7.84	0.94	18.00	46.80	7.30	9.40	4.08	11.50	2.30	0.07	0.17	1.72	4106.00	0.76
Autumn7#	15.10	7.77	0.29	20.00	45.40	6.20	8.75	5.04	12.95	1.55	0.18	0.17	1.82	13252.00	1.63
Autumn8#	14.00	7.59	0.47	25.50	32.50	0.45	8.90	4.72	12.85	1.95	0.09	0.16	1.79	29671.00	1.40