Korean J. Limnol. 41 (4): 530~540 (2008)

Spatio-temporal Fluctuations of Size-structured Phytoplankton over an Annual Cycle in the Youngsan Lake

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The temporal and spatial variations of size-structured phytoplankton dynamics in Youngsan Lake were investigated to explore potential mechanims controlling the dynamics in the Youngsan Lake. Field data were collected monthly from February to October, 2003 at 6 stations along the axis of Youngsan Lake. In this study, phytoplankton (chlorophyll *a*) were categorized into three size classes: micro-size (>20 μm), nano-size (2～20 μm) and pico-size (<2 μm). Water temperature, light attenuation coefficients, PAR (photosynthetically active radiation) and suspended solids were measured to analyze relationship between physical-chemical properties and size structure of phytoplankton. Phytoplankton blooms developed during March, July and October in the upper region of the main stem whereas small-scaled spring bloom was observed in the lower region. The scales of phytoplankton blooms were higher in the upper regions than the lower region and blooms were predominated by micro-size class in upper region but predominated by nano-size class in lower region. Growth of size-structured phytoplankton appeared to be controlled by rather light availability than temperature-dependant metabolisms in the system. Phytoplankton growth may be also supported by ambient nutrients available in the water column from analyses of chlorophyll *a* vs. nutrient concentrations including nitrite+nitrate and orthophosphate. Growth of nano-sized phytoplankton alone appeared to be supported by orthophosphate as well as nitrite+nitrate indicating that response of phytoplankton to nutrient inputs may be size-dependent.

Key words : size-structured phytoplankton, Youngsan Lake, potential mechanism, light availability

INTRODUCTION

One approach to a better understanding of phytoplankton dynamics is to categorize phytoplankton community into different size classes since cell size determines both the response of phytoplankton communities to environmental variation (Malone and Chervin, 1979; Takahashi and Bienfang, 1983; Gieskes and Kraay, 1986; Joint and Pomroy, 1986; Oviatt *et al.*, 1989; Glibert *et al.*, 1992; Armstrong, 1994; Hein *et al.*, 1995), and associated impacts on aquatic food web structure and fisheries (Walsh, 1976; Lenz, 1992; Painting *et al.*, 1993). Over various time scales watershed inputs to aquatic systems may change both the quality (size structure) and quantity (biomass) of primary producers. In turn, these changes resulting from environmental disturbance may impact nutrient and dissolved oxygen (DO) distributions

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as well as heterotrophic consumers in the water column. Since cell size affects sinking (Michaels and Silver, 1988) and transport rates, it will determine where ungrazed biomass accumulates and undergoes microbial processing by bacteria and protozoa which in turn influences oxygen dynamics (Jonas, 1992) and nutrient remineralization (Caron, 1991). Remineralized nutrients may subsequently support primary production (Kemp and Boynton, 1984).

In past studies, phytoplankton were usually grouped into two size fractions: netplankton (20 ~ 200 μm) and nanoplankton (< 20 μm). In recent years picoplankton (0.2 ~ 2 μm), comprised of minute chroococcoid cyanobacteria and eukaryotic phytoplankton, have received attention in phytoplankton studies (Ray *et al.*, 1989; Lacouture *et al.*, 1990; Malone *et al.*, 1991; Iriarte, 1993). The Youngsan Lake was artificially formed in 1981 and has used for multiple purposes such as industrial and agricultural use primarily as well as use of drinking water for Kwangju and Chonnam regions since construction of the reservoir. The lake has been documented eutrophic based on the trophic state index and phytoplankton blooms occasionally developed (Lee *et al.*, 2006). Results on phytoplankton community (species) have also been reported for the reservoir (Kim and Choi, 1988; Kim, 1992; Choi *et al.*, 1995; Kim, 2003) but chlorophyll *a* content was not determined for the three different size classes. Therefore, to better understand phytoplankton processes and provide information useful for managing the water quality of the reservoir efficiently, studies on the spatial and temporal variations of size structure dynamics of phytoplankton is required. The principal goals of this study were to: (1) examine temporal and spatial variations in chlorophyll *a* of various size classes of phytoplankton in Youngsan Lake; (2) identify mechanisms including physical and chemical properties controlling size structure dynamics.

MATERIALS AND METHODS

1. Study site and sample collection

Youngsan River is composed of the Hwangryong, Jisuk, Komakwon and Hampyung Rivers as well as Youngsan Lake. Watershed area of the river is 3,455 km^2 and waterway length is 129.5 km. Surface area of the Youngsan Lake is 34.6 km^2 and length from embankment to Mongtan is 23.4 km. Total population in the Youngsan River watershed is 1,846, 834 (persons) based on the record of 2006. Six stations along the axis of the Youngsan Lake (Fig. 1) were sampled monthly over one annual cycle from February, 2003 to October, 2003. Samples were collected from 1 m below the surface and 1 m above the bottom using Nanssen bottle.

2. Chlorophyll *a* measurement

Phytoplankton were categorized into three size classes: micro-size (> 20 μm), nano-size (2 ~ 20 μm) and pico-size (< 2 μm). Phytoplankton were fractionated by filtration through 20 μm Nytex mesh (1 ~ 2 liters) and 2 μm membrane filters (1 liter) with low vacuum (< 150 mm Hg). For chlorophyll a determinations, 100 mL of non-fractionated whole water, 200 mL of 20 μm filtrate, and 200 mL of 2 μm filtrate were filtered through Whatman 25 mm GF/F glass fiber filters (0.7 μm) under vacuum (< 120 mm Hg). Sample filtration was performed in duplicate. The filters were placed in dark test tubes pre-filled with 8 mL extraction solution (90% acetone, 10% deionized water) After storage for 12 hrs at room temperature, fluorescence was measured on a Turner Designs10-AU Fluorometer. Two drops of HCl (2N) were added and the extracts re-read for determination of pheopigments following acidification. Grazers convert chlorophyll *a* to pheopigments,

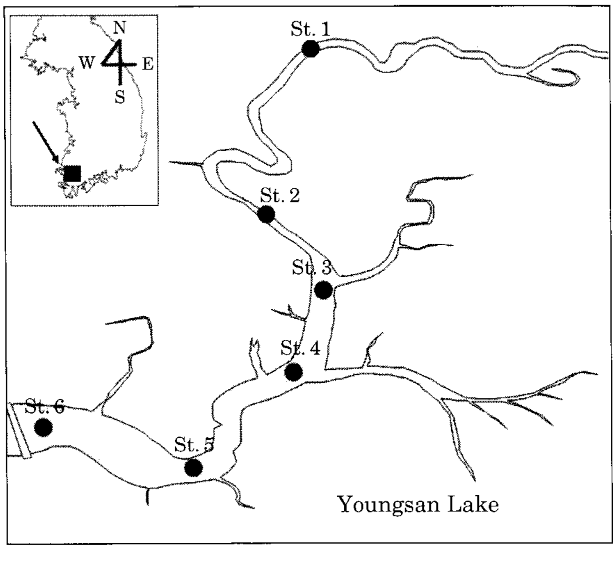

Fig. 1. Sampling stations located along the channel of Youngsan Lake.

which are released as egested fecal material and pheopigments can be used as indirect index for grazing effects: the lower the ratio, the higher the grazing activity (Welschmeyer and Lorenzen, 1985). Chlorophyll *a* in each size fraction was determined by consecutive subtraction of the <2 μm and <20 μm fractions from whole water chlorophyll *a*.

3. Measurement of physical-chemical properties

A YSI Model 85 S-C-T Meter was used to measure *in situ* temperature during field sampling. A LICOR PAR Quantum Radiometer was used to measure solar and submarine irradiance at depths of 10, 35, 60, 85 and 110 cm. Light attenuation coefficients were calculated using Beer's Law, $I_z = I_0 e^{-kz}$, where I_z is the intensity of light at z, the depth of interest, I_0 is the intensity at the surface, and k is the attenuation coefficient of water. Suspended solid concentrations were measured by filtering water samples with Whatman 25 mm GF/F glass fiber filters and by weighing the dry weight of materials on filters. Water depth was measured using a water depth sensor.

4. Other data collection and statistical analysis

Precipitation and duration (photoperiod) of solar radiation data were collected at the Korea Meteorological Administration, Mokpo, Chonnam. Nutrient data were collected by Chodang University and used to analyze relationship between chlorophyll *a* of size classes and nutrient concentrations (NH_4^+, $NO_2^-+NO_3^-$ and PO_4^{3-}). For the seasonal comparison, averages of data from winter (February), spring (March, April, May), summer (June, July, August) and fall (September, October) were reported. Linear simple correlation analysis were employed to investigate correlationships between phytoplankton size class chlorophyll a and the various physical-chemical variables.

RESULTS

1. Physical characteristics: precipitation, photo-period, temperature, & water clarity

Precipitation for the period from January 2003 to December 2003 (Fig. 2A) displayed a seasonality similar to that of other areas in S. Korea; high during summer and low during winter. Monthly sum of photo-period data collected in Mokpo also revealed a seasonal trend: highest during October and lowest during July (Fig. 2B). Photoperiod of sampling date, June 2 was longest whereas that of August 31 was shortest.

Surface and bottom water temperature at Stations 2, 4 and 6 was highest during August and lowest during February (Fig. 3A, 3B, 3C). Surface and bottom water temperature slightly increased as upstream. Evident seasonal pattern was observed for surface and bottom water temperature. Stratification of water column was ob-

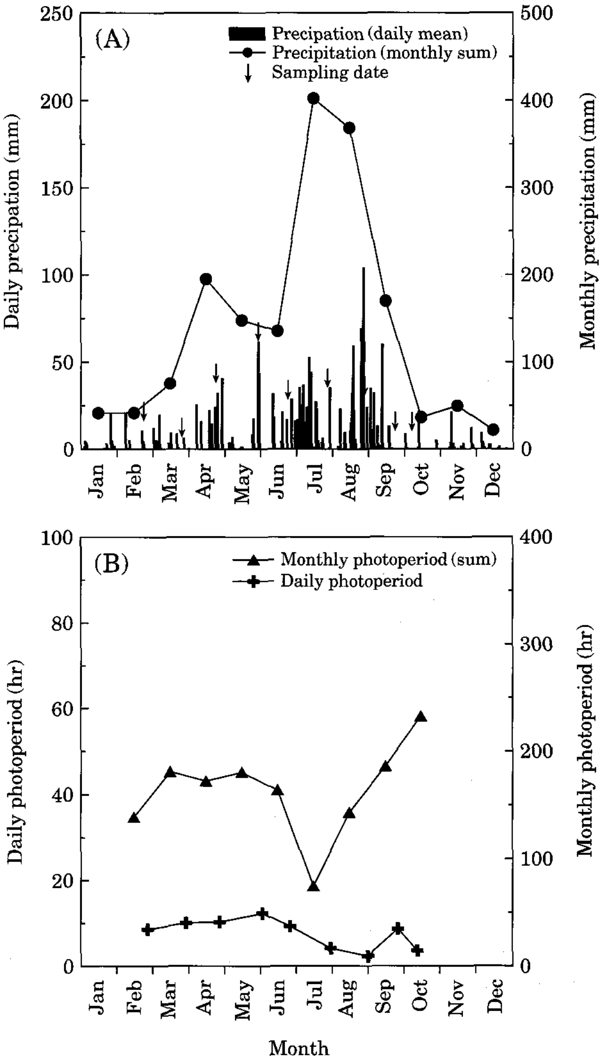

Fig. 2. Temporal variations of daily/monthly precipitation or rainfall (A) and daily/monthly totals of photoperiod (B) in Mokpo, Chonnam.

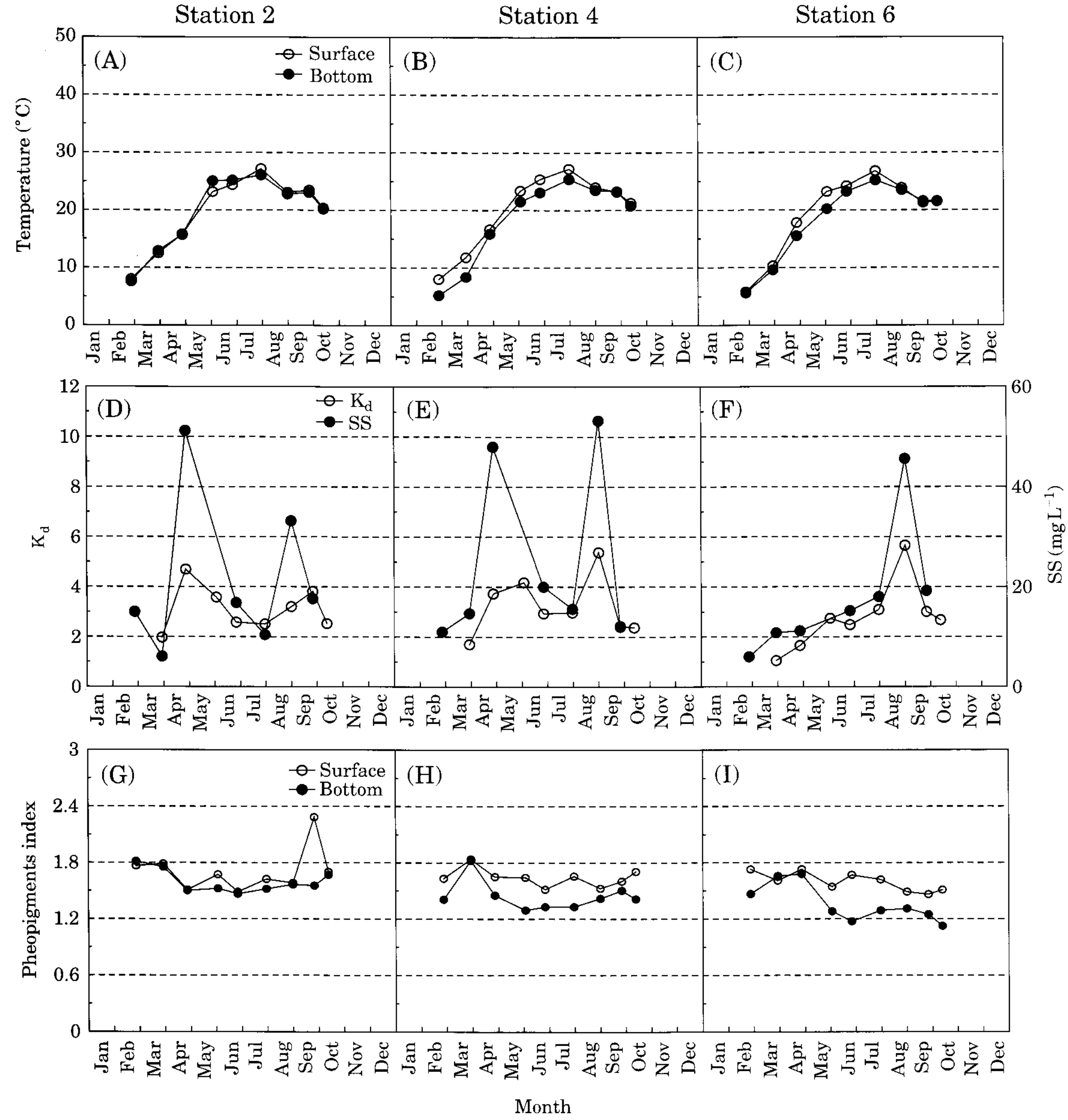

Fig. 3. Temporal distributions of water temperature, suspended solids, light attenuation coefficients (K_d) and pheopigment index (ratio of fluorescence after/before acidification) at Stations 2, 4 and 6.

served at Stations 4 and 6 during sampling period except September and October whereas small-scaled stratification was developed at Station 2 in August alone. Suspended solids at Station 2 peaked in April and were minimal in March (Fig. 3D). Similar pattern was observed for light attenuation coefficients (K_d). At Stations 4 and 6, maximum suspended solid and light attenuation coefficients were detected in August and minimum in February or March (Fig. 3E, 3F). Distribution of light attenuation coefficients generally followed that of suspended solid suggesting that K_d is closely correlated with suspended solids. Pheopigment index i.e., ratio of fluorescence before /after acidification was high February, March, September and October whereas the index was low during other seasons at Stations 2 and 4 (Fig. 3G, 3H). Pheopigment index was high in February, April and early June whereas the index was low during other periods at Station 6 (Fig. 3I). Difference between surface and bottom water was increased as downstream. Water depths at

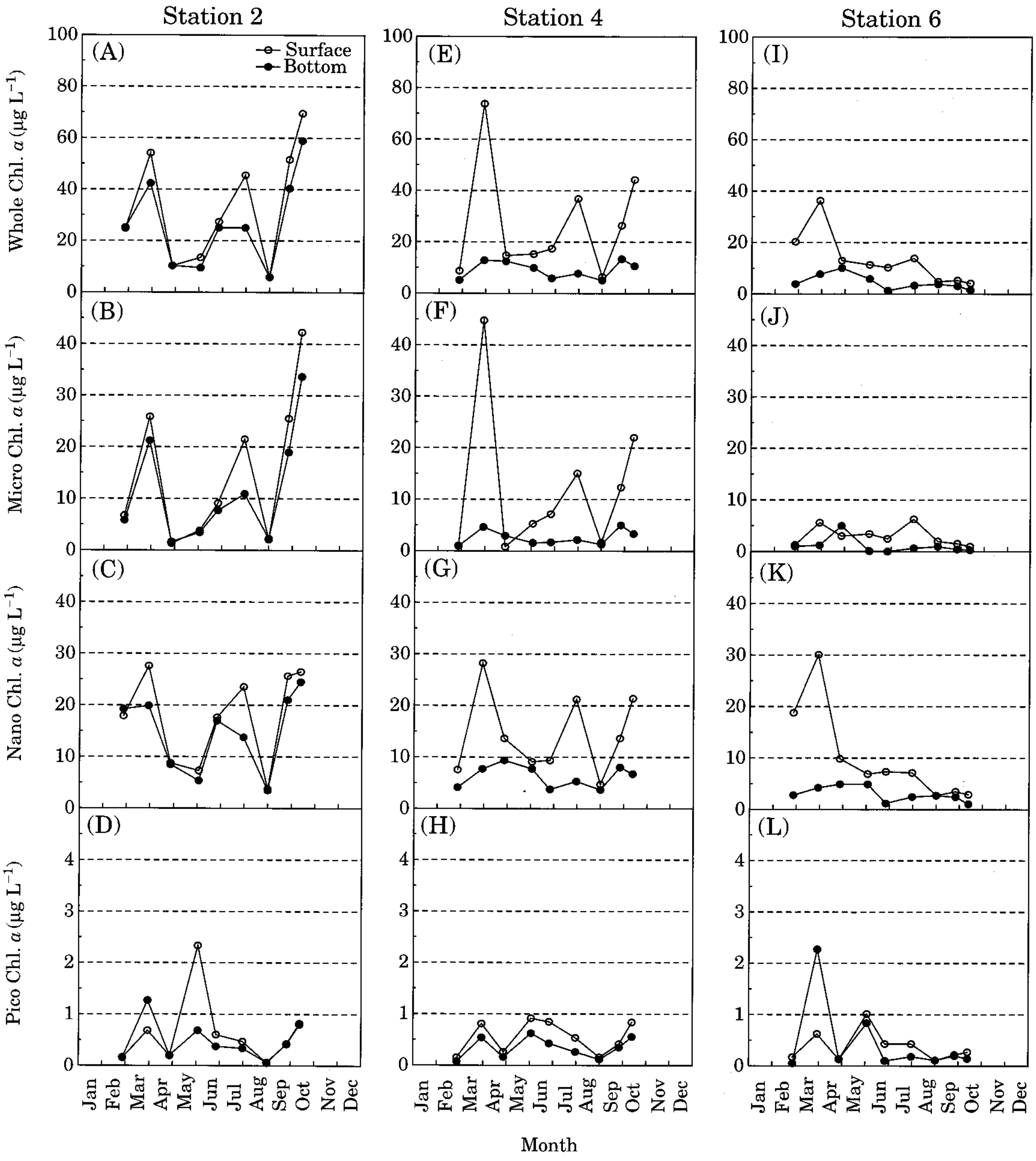

Fig. 4. Temporal variations of chlorophyll *a* in unfractionated water (whole-chl *a*), micro-sized chlorophyll *a* (micro-chl *a*), nano-sized chlorophyll *a* (nano-chl *a*), and pico-sized chlorophyll *a* (pico-chl *a*) at Stations 2, 4 and 6 along the axis of Youngsan Lake.

Stations 1 from to 6 were 4, 4, 5, 15, 15 and 17 m respectively.

2. Temporal variations in chlorophyll *a*

At Station 2, total chlorophyll *a* concentrations in surface water were minimal (6 μg L^{-1}) during August and peaked (70 μg L^{-1}) during October (Fig. 4A). This same seasonal chlorophyll *a* signal characterized microphytoplankton size class at the station (Fig. 4B). The temporal variations of nano-size class were similar to those of the micro-size but chlorophyll *a* concentrations were higher in March than in October (Fig. 4C). Chlorophyll *a* concentrations of pico-size class were peaked in early June and relatively low during other seasons (Fig. 4D).

At Station 4, total chlorophyll *a*, micro-sized and nano-sized chlorophyll *a* concentrations in surface water were peaked in March and minimal in August or April (Fig. 4E, 4F, 4G). Different seasonal variations were observed for chlorophyll *a* concentrations of pico-size class displaying peak in early June (Fig. 4H).

At Station 6, a bloom of total and nano-sized a concentrations was developed in March and gradually decreased with time (Fig. 4I, 4K). Micro-size class showed a peak during July as well as during March (Fig. 4J). Two peaks of pico-sized chlorophyll *a* were developed in March and early June (Fig. 4L). Difference of surface and bottom chlorophyll *a* concentrations were generally increased as downstream.

Percentage contribution of micro-size class was high during March, July, September and October when phytoplankton blooms developed at Station 2 (Fig. 5A). Nano-sized phytoplankton were dominant during other seasons and pico-sized phytoplankton were increased in early June at the station. Similar seasonal pattern was observed at Sation 4 (Fig. 5B). At Station 6, nano-sized phytoplankton were dominant except for July when contribution of micro-sized phytoplankton peaked and pico-sized phytoplankton were incre-

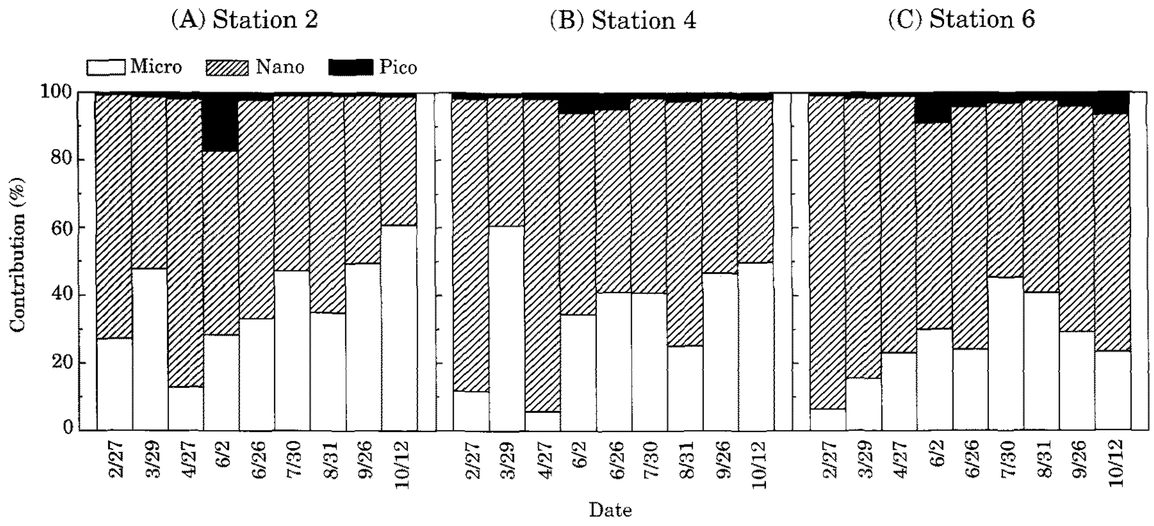

Fig. 5. Spatial variations of chlorophyll *a* in unfractionated water (whole-chl *a*), micro-sized chlorophyll *a* (micro-chl *a*), nano-sized chlorophyll *a* (nano-chl *a*), and pico-sized chlorophyll *a* (pico-chl *a*) during spring, summer, fall and winter.

Table 1. Results (r) of Pearson correlation analyses of surface chlorophyll *a* (μg L^{-1}) of each size class vs. physical and chemical properties including water temperature (T, °C), light attenuation coefficients (K_d), PAR at 1 m water depth (PAR, μEin m^{-2} s^{-1}), precipitation (Pr, mm), photo-period of sampling date (Photo, hour), and nutrients during the sampling period.

	Whole	Micro	Nano	Pico	T	K_d	PAR	Pr	Photo	NH_4^+	$NO_2^-+NO_3^-$	PO_4^{3-}
Whole	1	$.962^b$	$.915^b$	$.507^b$	−.116	$-.480^b$	$.460^b$	-0.302^a	.042	−.044	$.400^b$	$.289^a$
Micro		1	$.772^b$	$.468^b$	−.043	$-.382^a$	.293	−0.225	−.007	−.093	$.287^a$	.224
Nano			1	$.456^b$	−.219	$-.549^b$	$.641^b$	-0.366^b	.091	.040	$.494^b$	$.354^b$
Pico				1	.139	−.239	.181	-0.319^a	$.351^b$	−.150	$.528^b$	.001
T					1	$.349^a$	$-.527^b$	0.598^b	$-.326^a$	$-.272^a$	−.178	$-.524^b$
K_d						1	$-.754^b$	0.516^b	−.222	−.126	−.167	−.208
PAR							1	-0.401^a	.196	−.050	.294	.301
Pr								1	-0.485^b	.395	.257	−.137
Photo									1	−.073	.260	.005
NH_4^+										1	.128	$.451^b$
$NO_2^-+NO_3^-$											1	$.379^b$
PO_4^{3-}												1

$^a P<0.05$; $^b P<0.01$

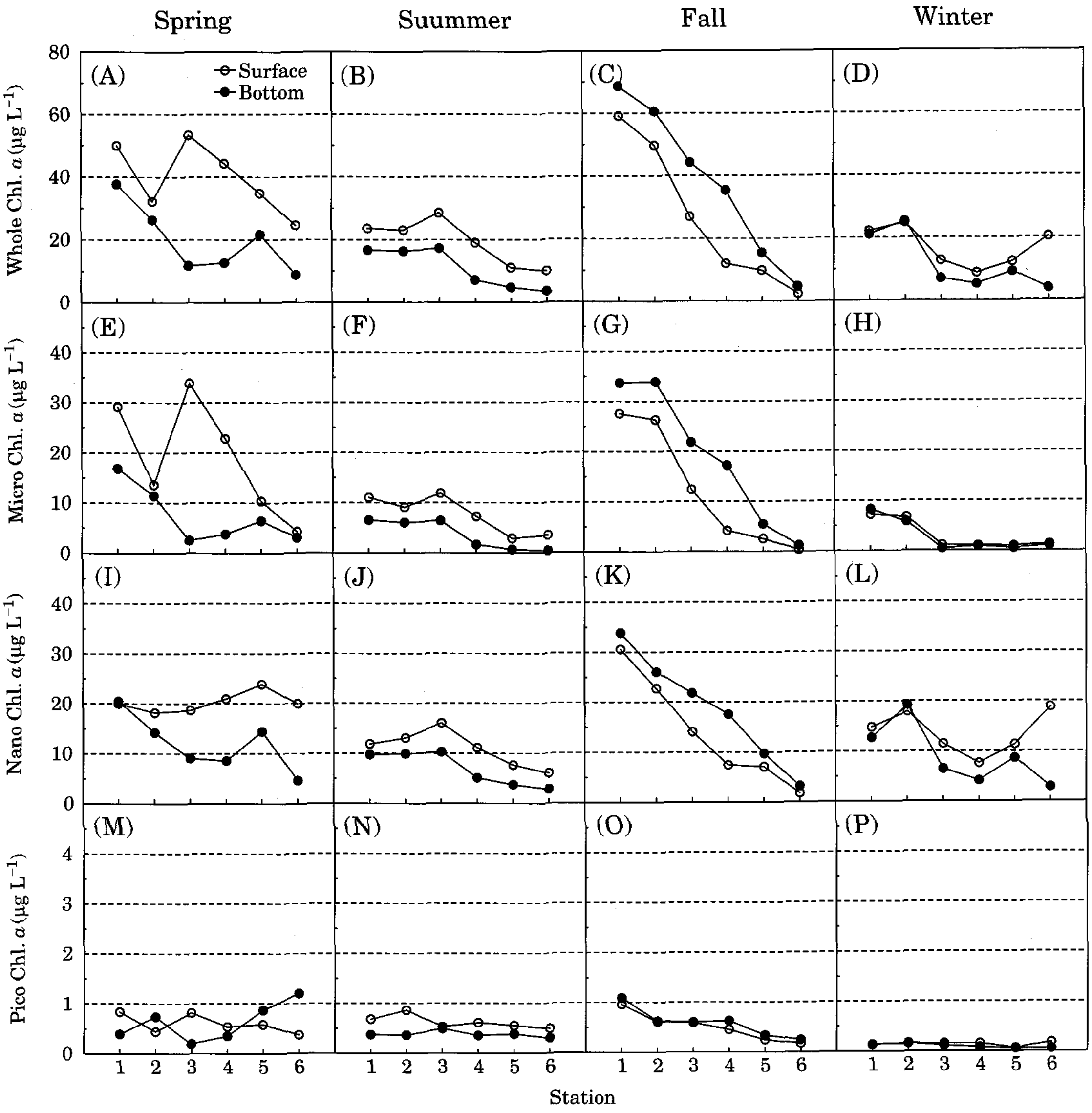

Fig. 6. Temporal variations for percentage contributions of three size classes (micro, nano and pico) to the total chlorophyll a in the surface water at he Stations 2, 4 and 6.

ased in early June (Fig. 5C).

3. Spatial variations in chlorophyll *a*

Total and micro-sized chlorophyll *a* concentrations during sampling period except winter revealed spatial pattern in surface water, low at upper regions (Stations 1 and 2) but high at lower regions including Stations 5 and 6 (Fig. 6A-6H). Nano-sized chlorophyll *a* concentrations were generally high at upper regions and low at lower regions whereas evident spatial variability was not observed during spring and winter (Fig. 6I-6L). Spatial variation was not clear for pico-sized chlorophyll *a* during the sampling period except for fall (Fig. 6M-6P).

In the surface water at upper region (Stations 1, 2), the contribution of large cells (microphytoplankton) to total chlorophyll *a* was greatly higher than lower region (Stations 5, 6) during spring whereas the opposite characteristics were observed for nano-sized phytoplankton (Fig. 7A). The similar pattern was observed during summer although % contribution of micro-size class was increased compared to spring (Fig. 7B). The percentage contribution of micro-size class were sli-

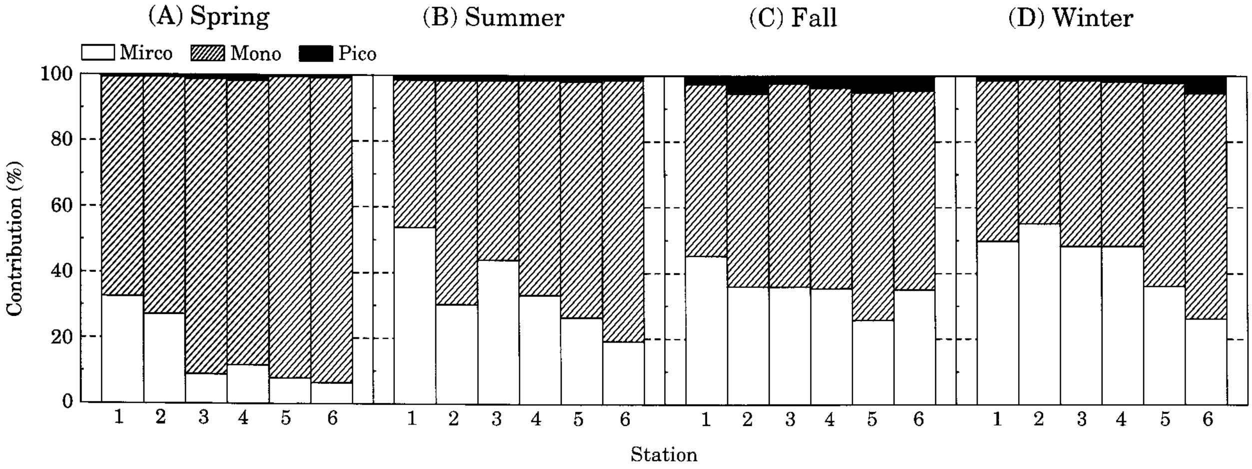

Fig. 7. Spatial variations of percentage contributions of three size classes (micro, nano and pico) to the total chlorophyll *a* during spring, summer, fall and winter in the surface water of the study sites of the Youngsan Lake.

ghtly decreased as downstream whereas small-sized phytoplankton (nano- and pico-size classes) slightly increased as downstream during fall and winter (Fig. 7C, 7D). Nano-sized phytoplankton were dominant during the sampling period especially for spring. Lower region (Stations 5 and 6) was dominated by nano-size class (Fig. 7A-7D).

4. Simple linear correlation analysis

Table 1 shows results (r) of correlation analyses of relationships between the chlorophyll *a* concentrations of three phytoplankton size classes and various physical-chemical properties of the Youngsan Lake, specifically water temperature (T), light attenuation coefficient (K_d), photo-period (Photo), precipitation (Pr) and nutrients including NH_4^+, $NO_2^-+NO_3^-$ and PO_4^{3-}. Total chlorophyll *a* were significantly ($P<0.01$) and positively correlated with micro-, nano- and pico-size classes. Micro-size class was also significantly ($P<0.01$) and positively correlated with nano- and pico-size classes and nano-size class was correlated with pico-class ($P<0.01$). Significant correlationship was not observed between size classes and water temperature. Light attenuation coefficients were significantly and negatively correlated with chlorophyll *a* concentrations of whole, micro- and nano-size classes ($P<0.05$). Pico-size class was also correlated negatively but relationship was not significant. PAR was significantly positively correlated with chlorophyll *a* concentrations of whole and nano-size class ($P<0.01$). Precipitation was significantly and negatively correlated with whole, nano- and pico-size classes ($P<0.05$). Photoperiod was significantly and positively correlated with pico-size class ($P<0.01$). Ammonium (NH_4^+) were not correlated with any size classes. Nitrite +nitrate ($NO_2^-+NO_3^-$) concentrations were significantly and positively correlated with all size classes ($P<0.05$) and orthophosphate (PO_4^{3-}) concentrations were significantly and positively correlated with whole and nano-size classes ($P<0.05$).

DISCUSSION

Temporal and spatial variations of size classes presented in Figures 4～7 showed that large phytoplankton (micro-sized) were more abundant in the upper region than the lower region whereas contributions of small phytoplankton (nano- and pico-sized) were increased as downstream in the Youngsan Lake. Phytoplankton blooms during spring and fall were mainly predominated by large cells i.e., micro-size class in the upper region whereas nano-size class was dominant during the spring bloom in the lower region such as Station 6 (Fig. 4I, 4K, 5C). This size structure of phytoplankton is different from that of Juam Lake where micro-size class was dominant during spring and fall blooms (Sin and Kim, 2003). Percentage contribution of pico-sized phytoplankton to total chlorophyll *a* concentrations was generally minor compared to other size classes but contribution of pico-size class was increased in early June when total chlorophyll *a* concentrations were minimal (Figs. 4, 6). Chisholm similarly reported that the percentage of small cells in the phytoplankton population increased as total chlorophyll *a* decreased (Chisholm, 1992). These results suggest that response of phytoplankton to environmental changes such as nutrient or turbidity fluctuations may be size- and location-dependent in the Youngsan Lake.

The spring and fall blooms of phytoplankton cells were concomitant with decrease of suspended solids and light attenuation coefficients (K_d) in surface water (Figs. 3, 4). Chlorophyll *a* concentrations of phytoplankton were low when suspended solids and light attenuation coefficients (K_d) were increased, suggesting that light availability governed by turbidity controls phytoplankton variations in the surface water. This scenario is supported by the significant and negative correlation between light attenuation coefficients and chlorophyll of whole, micro- and nano-size classes (Table 1). PAR at 1 m water depth was also significantly and positively with chlorophyll *a* concentrations of whole, nano- ($P<0.05$) and micro-size ($P<0.1$) classes. PAR is affected by turbidity of water column in water column and the relationship was presented in the significant correlation between PAR and K_d in this study (Table 1). Lee *et al.* (2006) also documented that turbidity of the Youngsan Lake was high compared with other lakes in the Chonnam area suggesting that growth of phytoplankton may be limited by light in the Youngsan Lake.

Temperature was not correlated with phytoplankton size classes in the correlation analyses for data from all stations (Table 1) but whole and nano-size classes were significantly and negatively correlated with water temperature at Station 6; $r=-0.70$ and -0.87 ($P<0.05$) respectively (data not shown). This suggests that phytoplankton growth may be affected by water temperaure-dependent metabolism in lower region of the Lake.

Ammonium (NH_4^+) concentrations were not correlated with any phytoplankton size classes whereas nitrite+nitrate ($NO_2^-+NO_3^-$) concentrations were significantly and positively correlated with all size classes in the Youngsan Lake (Table 1). Orthophosphate (PO_4^{3-}) concentrations were also significantly and positively correlated with whole and nano-size classes in the lake. The results suggest that biomass of phytoplankton size classes especially nano-sized is influenced by ambient nutreints including nitrite+nitrate and orthophosphate in the water column. Trophic state index based on total phosphorus (TP) for Youngsan Lake is relatively high compared with Juam and Dongbok Lakes (Lee *et al.*, 2006). Sin *et al.* (2005) also reported that TP concentrations were gradually increased over 15 years from 1990 to 2004 and potential N or P limitation was observed based on molar ratios of dissolved inorganic N and P in the water column. This implies that eutrophication due to nutrients inputs may alter size structure of phytoplankton in the Youngsan Lake.

The ratios of fluorescence before and after acidification were analyzed as a proxy for grazing effect (top-down control) since herbivores convert chlorophyll *a* to pheopigments, which are released as egested fecal material. The ratio of chlorophyll *a* and pheopigments, calculated from the ratios of fluorescence before and after acidification is an indirect measure of grazing activity (e.g., Welschmeyer and Lorenzen, 1985). Suspended pheopigments can also be produced within phytoplankton cells during senescence as a result of poor growth environments or prolonged exposure to the dark (Daley and Brown, 1973). In the Youngsan Lake, the ratio was high when chlorophyll *a* concentrations were high (Fig. 3G-3I) suggesting that grazing pressure or stress from aggravated growth conditions were reduced resulting in phytoplankton blooms. However, measurements of direct grazing rates or abundance of zooplankton are required to estimate the grazing effects on phytoplankton size-structure clearly.

In summary, we investigated the temporal and spatial variations of size-structured phytoplankton dynamics in Youngsan Lake and potential mechanims controlling the dynamics focusing on physical-chemical properties in the Youngsan Lake. Phytoplankton blooms developed during March, July and October in the upper region of the lake whereas small-scaled spring bloom was observed in the lower region. The scales of phytoplankton blooms were higher in the upper regions than the lower region and blooms were predominated by large cells of phytoplankton (micro-sized) in upper region but predominated by small cells of phytoplankton (nano-sized) in lower region. Growth of size-structured phytoplankton appeared to be controlled by rather light availability than temperature-dependant metabolisms in the system. Phytoplankton growth may be also supported by ambient nutrients available in the water column from analyses of chlorophyll *a* vs. nutrient concentrations including nitrite+nitrate and orthophosphate. Growth of nano-sized phytoplankton alone appeared to be supported by orthophosphate as well as nitrite+nitrate indicating that response of phytoplankton to nutrient inputs may be size-dependent. Although further data analyses on grazing activity of herbivores are required to present conclusions on the relative importance between bottom-up and top-down controls of size-structured phytoplankton dynamics in the Youngsan Lake, the results from this study provide useful information to better understand phytoplankton dynamics in the Youngsan Lake.

ACKNOWLEDGEMENTS

This work was supported by Korea Research Foundation Grant funded by the Korean Government (MOEHRD) (KRF-2006-351-F00017).

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(Manuscript received 1 November 2008,
Revision accepted 10 December 2008)