Journal of the Korean Society of Oceanography
Vol. 39, No. 4, pp. 234 — 242 December 2004

Analysis on the Pigment Composition of Phytoplankton Assemblages using HPLC (High Performance Liquid Chromatography) in the Adjacent Waters of Nuclear Power Plants in Spring

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The pigment composition and concentration of phytoplankton assemblages using HPLC in the adjacent waters of four nuclear power plants (Yonggwang, Kori, Wolsong and Ulchin) were investigated during the spring blooming in 2004. The mean concentration of chlorophyll *a* ranged from 563.8 to 2,949.0 ng l^{-1}, with the lowest concentration at Kori and the highest concentration at Wolsong. Among the carotenoids, the amounts of fucoxanthin and chlorophyll c_2 were relatively higher than those of other pigments in the study site. As minor pigments, zeaxanthin, chlorophyll *b*, 19'-butanoyloxyfucoxanthin, diadinoxanthin, 19'-hexanoyloxyfucoxanthin, chlorophyll c_3 and peridinin were detected. The results of pigment composition and concentration showed that diatoms had an important proportion of phytoplankton community when a spring bloom occurred. Cyanobacteria was present relatively low density at the Wolsong and the green alga such as chlorophytes and prasinophytes were abundant at the Yonggwang and Kori, while dinoflagellates characterized by peridinin were common at Ulchin and Kori. The pigment composition and concentration of phytoplankton after passing through the cooling-water system of nuclear power plant were highly variable. No distinct trend of the change of each pigment composition and amount was detected but the variation of fucoxanthin and chlorophyll c_2 highly coupled with that of chlorophyll *a*. We pointed out that the diatom controlled the overall variation of phytoplankton biomass during the spring season.

Key words: HPLC, Kori, Nuclear Power Plant, Pigment Composition and Concentration, Ulchin, Wolsong, Yonggwang

INTRODUCTION

Phytoplankton as a primary producer occupies an important position in marine ecosystem and its size distribution plays a fundamental role in determining the food web structure (Fenchel, 1988). Thus, the understanding community structure of phytoplankton is great help to assess marine environment. In studying the phytoplankton assemblage, chlorophyll *a* concentration has been used as the principal indicator of total biomass of phytoplankton in most studies. It has been measured by spectrophotometric or spectrofluorimetric method (Holm-Hansen *et al.*, 1965; Strickland and Parsons, 1972; Lorenzend and Jeffrey, 1980). Chlorophyll *a* concentration measured by these methods can be under- or overestimated due to the overlap of absorption and fluorescence bands of accessory pigments and degradation products of chlorophylls (Gieskes and Kraay, 1983; Trees *et al.*, 1986). To overcome some of these problems, the chemotaxonomic approach based on chromatographic pigment analysis of taxon-specific marker pigments, such as TLC (Thin-layer chromatography) and HPLC (High Performance Liquid Chromatography) which can accurately estimate chlorophylls and their degradation products and provide information on the phytoplankton assemblage in marine ecosystem (Jeffrey, 1976; Jeffrey and Hallegraeff, 1980; Letelier *et al.*, 1993; Wright *et al.*, 1991, 1996), has been employed to distinguish the main algal classes. Recently, advanced HPLC technique in phytoplankton pigment analysis enables the whole community approach. The pigment analysis using HPLC is a useful tool to evaluate the phytoplankton assemblage in the ocean, nevertheless,

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it scarcely has been used in the adjacent waters of Korea. Park and Park (1997) pointed out that the prymnesiophyte occupied an important biomass component of marine phytoplankton assemblages in southern waters of East Sea, and Song (1999) emphasized that diatoms and cryptomonads as primary producer were most important groups in the Kyeonggi Bay.

Meanwhile, the nuclear power plants (NPPs) which are located at the coastal areas use a lot of waters for cooling the system and then the cooling waters called as thermal discharges or thermal effluents enter the adjacent waters of NPPs (IAEA, 1974). These thermal effluents have an influence on marine ecosystem and often limit the distribution of marine organisms (Nayor, 1965; Langford, 1990; Suresh *et al.*, 1995). For these reasons, some studies were conducted on the phytoplankton community structure and the influence of thermal discharges on phytoplankton assemblage in the adjacent waters of power plants (Yoo and Lee, 1982; Yi and Chin, 1987; Yeo, 1992; Yeo and Shim, 1992; Lee and Lee, 1997; Cho, 1988; Shim *et al.*, 1991; Kim and Choi, 1995; Kang and Choi, 2001, 2002; Kang *et al.*, 2003). The studies conducted around four NPPs were focused on diatoms, so little was known on occurrence of other algal groups due to methodological difficulty in identifying and enumerating some species. Therefore, to establish whether the thermal discharge influences the phytoplankton composition, the differentiation of algal groups is necessary. In this study, we tried to analyze the pigment composition of phytoplankton assemblages using HPLC in the adjacent waters of four NPPs and gain information of the effect of entrainment on phytoplankton assemblage.

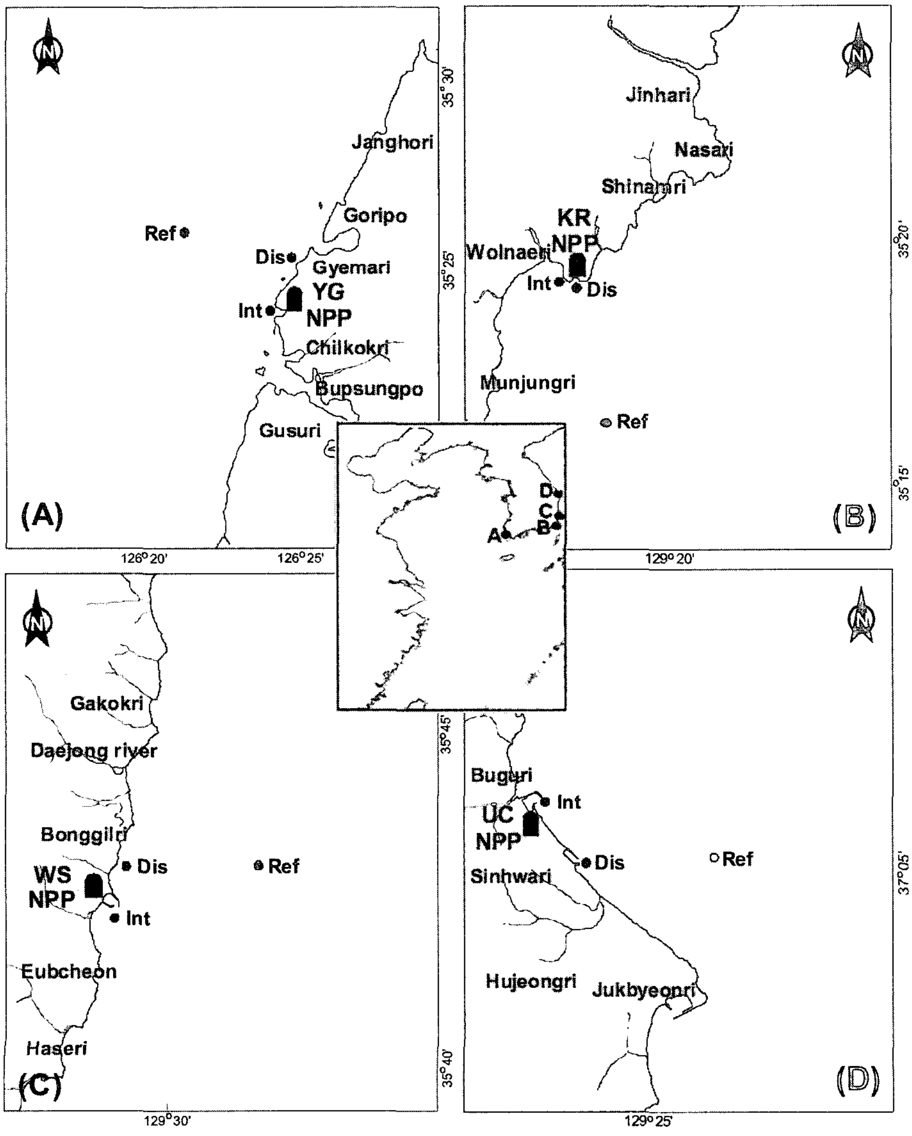

Fig. 1. Location of sampling sites around Yonggwang (A), Kori (B), Wolsong (C) and Ulchin (D) nuclear power plant (Int: Intake, Dis: Discharge, Ref: Reference).

MATERIALS AND METHODS

Samples were taken at three stations (intake, discharge and reference site) in the adjacent waters of each NPP and one sample for measuring the influence of entrainment on phytoplankton assemblages was taken at the discharge mouth (F-discharge) before being mixed with the ambient waters in April, 2004 (Fig. 1). The temperature and salinity of seawater were measured by HDMS (Horizontal Drew Monitoring System, YSI 6000) and CTD (IDRONAUT 316) at the surface within 10 km around each NPP. Water samples (500~1000 ml) for evaluating the pigment concentration were taken at the surface and filtered onto glass fibre filters (Whatmann GF/F, Φ 47 mm). Filters were wrapped with aluminum foil and stored in liquid nitrogen before extraction. Within one month, the pigments were extracted in 5 ml 100% acetone and 1 ml internal standard (trans-β-apo-8'-carotenal, Sigma Chemical Co.) in the dark for overnight. After extract was centrifuged at 3000 rpm for 10 min and filtered onto hydrophobic PTFE (Milipore 0.45 μm) to remove cellular debris and glass fibers, the 1 ml of clear extract was mixed with 0.3 ml DDW. And, 300 μl of extract was injected in the liquid chromatography. The HPLC instrument (Shimadzu VP series) consisted of a SCL-M10A*VP* system controller, a SPD-M10A*VP* diode array detector with wave lengths 436 nm. The column was a Rexchrom-S5-100-ODS (250×4.6 mm, 5 μm particle size). Identification of the pigment peaks at 436 nm and calculation of the areas to determine concentrations followed Park and Park (1997). Three solvents were used: A (methanol: 0.5 M ammonium acetate, 80:20), B (acetonitrile: H_2O, 90:10) and C (ethyl acetate, 100). The linear gradient was (min, solvent A%, solvent B%, solvent C%): (0, 100, 0, 0), (2, 0, 100, 0), (2.6, 0, 90, 10), (13.6, 0, 65, 35), (20.0, 0, 31, 69), (22.0, 0, 100, 0), (25.0, 100, 0, 0), (30.0, 100, 0, 0) and the flow rate was maintained at 1 mm min^{-1}. The signature of major pigments useful as markers of algal groups are showed in Table 1 (Jeffrey, 1997) and phaeophytin *b* was prepared by acidifying chlorophyll *b*.

RESULTS

Distribution of sea surface temperature and salinity

The sea surface temperature (SST) and salinity were measured in the adjacent waters of each NPP (Fig. 2). The SST in the adjacent waters of Yonggwang nuclear power plant (YNPP) varied from 11.3 to 21.7°C and the salinity was about 31.4 psu at the surface. The SST of the vicinity of discharge area of YNPP was about 10°C higher than that of ambient seawaters. At Kori nuclear power plant (KNPP), the range of SST was 12.7~21.7°C and the salinity was about 34.0 psu at the surface. The SST at the discharge area of KNPP was 9.0°C higher than that of ambient seawaters. The SST and salinity in the adjacent waters of Wolsong nuclear power plant (WNPP) were 13.1~18.3°C and about 34.3 psu, respectively. The SST around discharge area of WNPP was 5.2°C higher than that of ambient seawater. The SST in the adjacent waters of Ulchin nuclear power plant (UNPP) varied from 13.4 to 19.8°C and the salinity was about 34.2 psu at the surface. The SST of the discharge area of UNPP was 6.4 higher than that of ambient seawaters.

Pigment composition and concentration

The chlorophyll *a* concentration in the adjacent waters of YNPP ranged from 817.1 to 1,522.7 ng l^{-1}, with

Table 1. The signature of major pigments useful as markers of algal groups (Jeffry, 1997)

Pigment	Alagl Division/Class
Chlorophyll *a* (chl *a*)	All photosynthetic microalgae except prochlorophytes
Zeaxanthin (zeax)	Prochlorophyceae and cyanophyceae (prokaryotes)
Chlorophyll *b* (chl *b*)	Green algae such as chlorophyceae and prasinophyceae
19'-hexanoyloxyfucoxanthin (19'-hex)	Prymnesiophyceae
Fucoxanthin (fuco)	Bacillariophyceae (diatoms)
19'-butanoyloxyfucoxanthin (19'-bf)	Chrysophytes, some prymnesiophyceae and serveral dinoflagellates
Chlorophyll c_2 (chl c_2)	Most diatoms, dinoflgallates and prymnesiophytes
Chlorophyll c_3 (chl c_3)	Some prymnesiophytes, several diatoms and dinoflagellates
Diadinoxanthin (diad)	Diatoms, dinoflagellates, prymnesiophytes and chrysophytes
Peridinin (peri)	Dinoflagellates

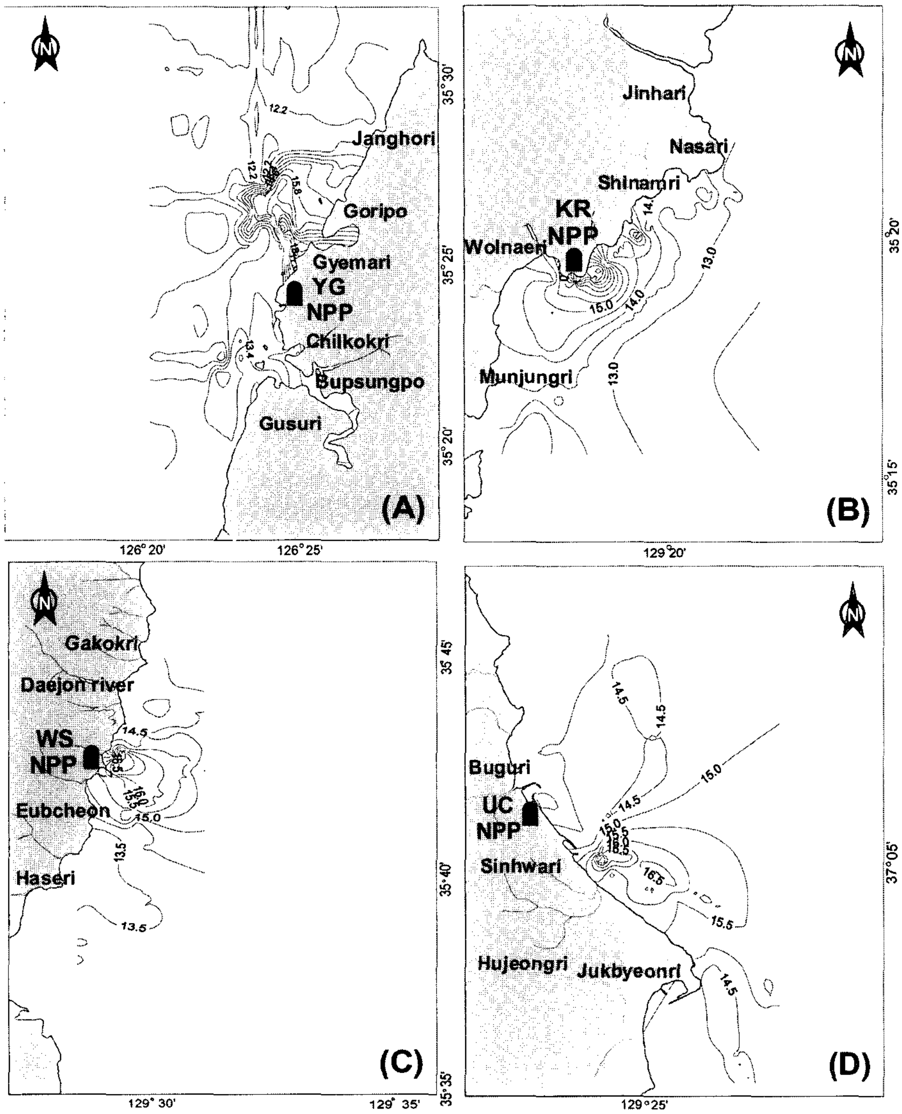

Fig. 2. Horizontal distribution of surface sea-water temperature in the adjacent waters of NPPs (A: Yonggwang, B: Kori, C: Wolsong, D: Ulchin).

the highest concentration at the intake site and the lowest concentration at the reference site (Fig. 3A). From among the carotenoid pigments, fucoxanthin concentration was the highest among all studying sites. It ranged from 178.7 to 378.3 ng l^{-1}, which was about 23.6% of chlorophyll *a* concentration (Fig. 4A). Chlorophyll *b* and chlorophyll c_2 concentrations were relatively high in the study area. Chlorophyll *b* was 8.4% of chlorophyll *a* concentration, which ranged from 60.6 to 95.1 ng l^{-1}. And, chlorophyll c_2 was 8.0% of chlorophyll *a* concentration, which ranged from 71.1 to 81.2 ng l^{-1}. Other pigments such as zeaxanthin, 19'-butanoyloxyfucoxanthin, 19'-hexanoyloxyfucoxanthin, chlorophyll c_3, diadinoxanthin and peridinin were also detected but their concentrations were relatively low. The concentrations of these pigments varied from 17.3 to 20.5 ng l^{-1}, from 14.5 to 24.3 ng l^{-1}, from 0.0 to 15.4 ng l^{-1}, from 23.4 to 38.2 ng l^{-1}, from 20.1 to 35.2 ng l^{-1} and from 0.0 to 31.1 ng l^{-1}, respectively.

In the adjacent waters of KNPP, chlorophyll *a* concentration ranged from 544.5 to 578.4 ng l^{-1}. The highest concentration was observed at the discharge site, while the lowest concentration occurred at the reference site (Fig. 3B). Fucoxanthin concentration was relatively high amount among the carotenoid pigments. It ranged from 155.7 to 206.3 ng l^{-1}, which was about 30% of chlorophyll *a* concentration. The highest concentration was observed at the discharge site and the lowest concentration was observed at the reference site (Fig. 4B). Chlorophyll c_2, chlorophyll *b* and peridinin were also abundant in the adjacent waters of KNPP. Chlorophyll c_2 was 10.7% of chlorophyll *a*, which ranged from 48.9 to 67.1 ng l^{-1}. Chlorophyll *b* was 7.3% of chlorophyll *a*, which ranged from 31.9

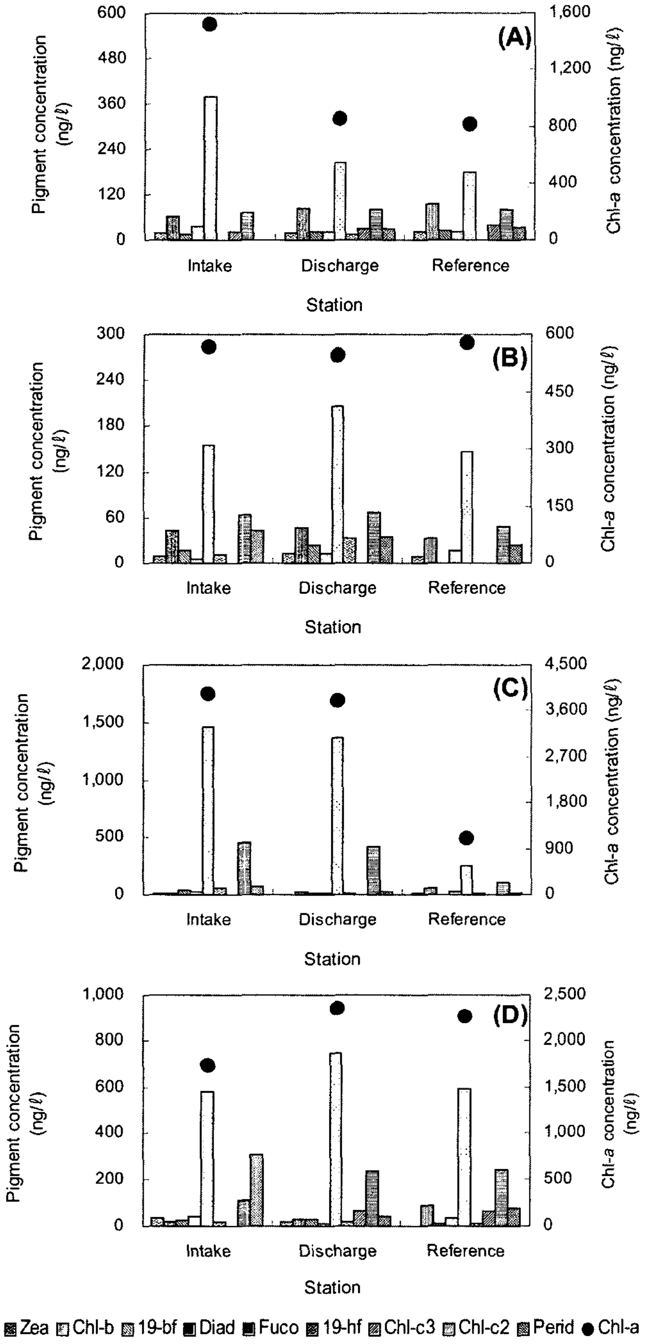

Fig. 3. Pigment and chlorophyll *a* concentrations at the surface sea-water in the adjacent waters of four NPPs. For explanations of pigment abbreviations see Table 1 (A: Yonggwang, B: Kori, C: Wolsong, D: Ulchin).

to 47.7 ng l^{-1}. And, peridinin was 6.1% of chlorophyll *a*, which ranged from 24.4 to 44.0 ng l^{-1}. Zeaxanthin, 19'-butanoyloxyfucoxanthin and 19'-hexanoyloxyfucoxanthin were also detected but their amounts were relatively low. The concentration of zeaxanthin, 19'-butanoyloxyfucoxanthin, diadinoxanthin and 19'-hexanoyloxyfucoxanthin varied from 8.2 to 12.7 ng l^{-1}, from 0.0 to 23.0 ng l^{-1}, from 6.1 to 15.5 ng l^{-1} and from 0.0 to 32.0 ng l^{-1}, respectively.

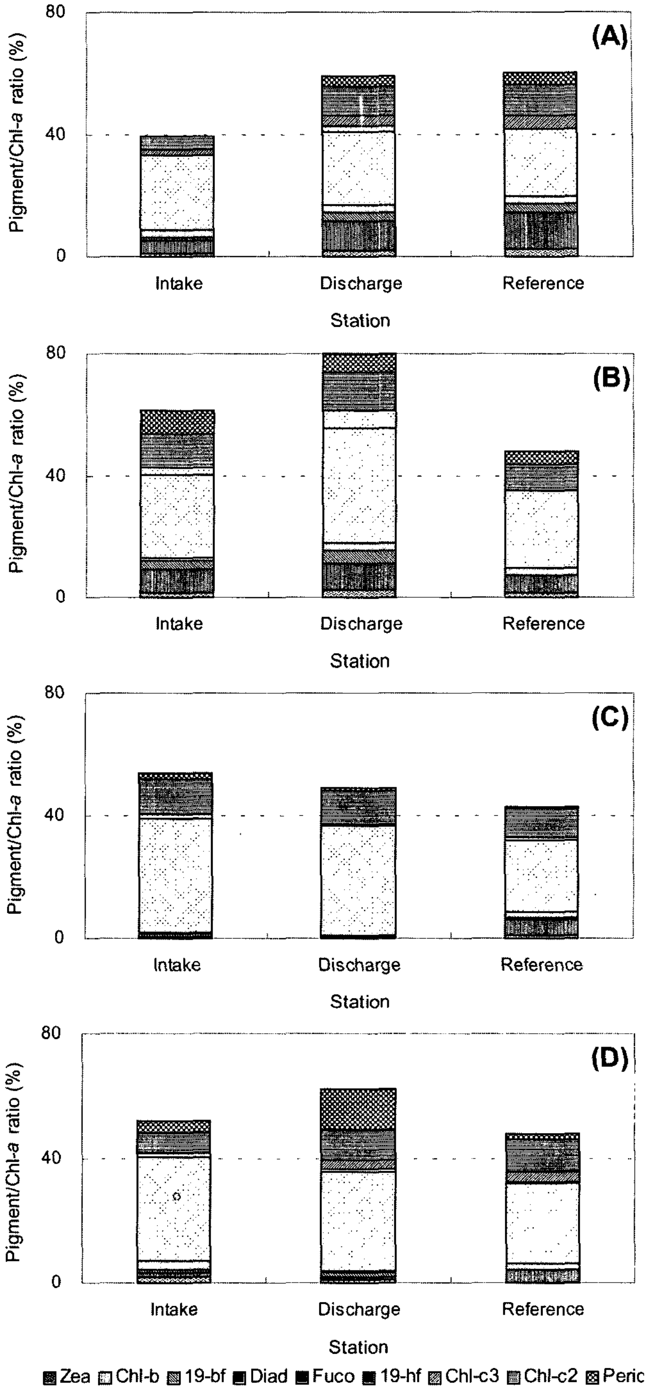

Fig. 4. Pigment/ chlorophyll *a* ratio at the surface sea-water in the adjacent waters of four NPPs. For explanations of pigment abbreviations see Table 1 (A: Yonggwang, B: Kori, C: Wolsong, D: Ulchin).

The chlorophyll *a* concentration in the adjacent waters of WNPP ranged from 1,096.4 to 3,939.5 ng l^{-1}. The highest concentration was found at the intake site and the lowest concentration was found at the reference site (Fig. 3C). Fucoxanthin was the most abundant among the carotenoid pigments. It was 32.0% of chlorophyll *a* concentration and ranged from 255.3 to 1,455.3 ng l^{-1}. Chlorophyll c_2 was also plenty in the adjacent waters of WNPP. It was 10.6% of chlorophyll *a* concentration, which ranged from 102.3 to 453.0 ng l^{-1} (Fig. 4C). The amounts of other carotenoid pigments such as zeaxanthin, chlorophyll *b*, 19'-butanoyloxyfucoxanthin, diadinoxanthin, 19'-hexanoyloxyfucoxanthin and peridinin detected around WNPP were relatively low. Their concentration varied from 0.0 to 8.7 ng l^{-1}, from 15.8 to 60.6 ng l^{-1}, from 3.9 to 31.7 ng l^{-1}, from 10.9 to 26.2 ng l^{-1}, from 10.8 to 63.4 ng l^{-1} and from 6.2 to 69.4 ng l^{-1}, respectively.

In the adjacent waters of UNPP, chlorophyll *a* concentration ranged from 1,732.2 to 2,353.9 ng l^{-1}, with the highest concentration at the discharge site and the lowest concentration at the intake site (Fig. 3D). The most abundant carotenoid pigment was fucoxanthin. Also, the amounts of chlorophyll c_2 and peridinin were relatively high. Fucoxanthin was 30.2% of chlorophyll *a* concentration and ranged from 581.5 to 743.8 ng l^{-1} (Fig. 4D). Chlorophyll c_2 and peridinin were 9.1%, 6.3% of chlorophyll *a* concentration and ranged from 113.0 to 241.3 ng l^{-1}, from 39.1 to 307.0 ng l^{-1}, respectively. Zeaxanthin, chlorophyll *b*, 19'-butanoyloxyfucoxanthin, diadinoxanthin, 19'-hexanoyloxyfucoxanthin and chlorophyll c_3 were also detected. Their amounts varied from 0.0 to 35.6 ng l^{-1}, from 18.7 to 90.7 ng l^{-1}, from 9.1 to 32.1 ng l^{-1}, from 11.9 to 42.2 ng l^{-1}, from 11.2 to 19.1 ng l^{-1} and from 0.0 to 66.3 ng l^{-1}, respectively. The amounts of phaeophytin *a* and *b* were very low or under the detection limit at the all stations (data not shown).

The alteration of pigment concentration after passing through the cooling-water system

The fluctuation of pigment concentration was shown after passing through the cooling-water system, and it varied with pigment composition and study site (Fig. 5, 6). In the YNPP, the chlorophyll *a* concentration at the discharge mouth was 9.7% lower than at the intake site. Among the carotenoid pigments, the amounts of chlorophyll *b*, diadinoxanthin and fucoxanthin were reduced 22.1%, 45.3% and 6.1%, respectively. In case of zeaxanthin, 19'-butanoyloxyfucoxanthin, chlorophyll c_3 and chlorophyll c_2, their amounts were slightly increased 4.2%, 15.9%, 28.4% and 22.3%, respectively (Fig. 5A, 6A).

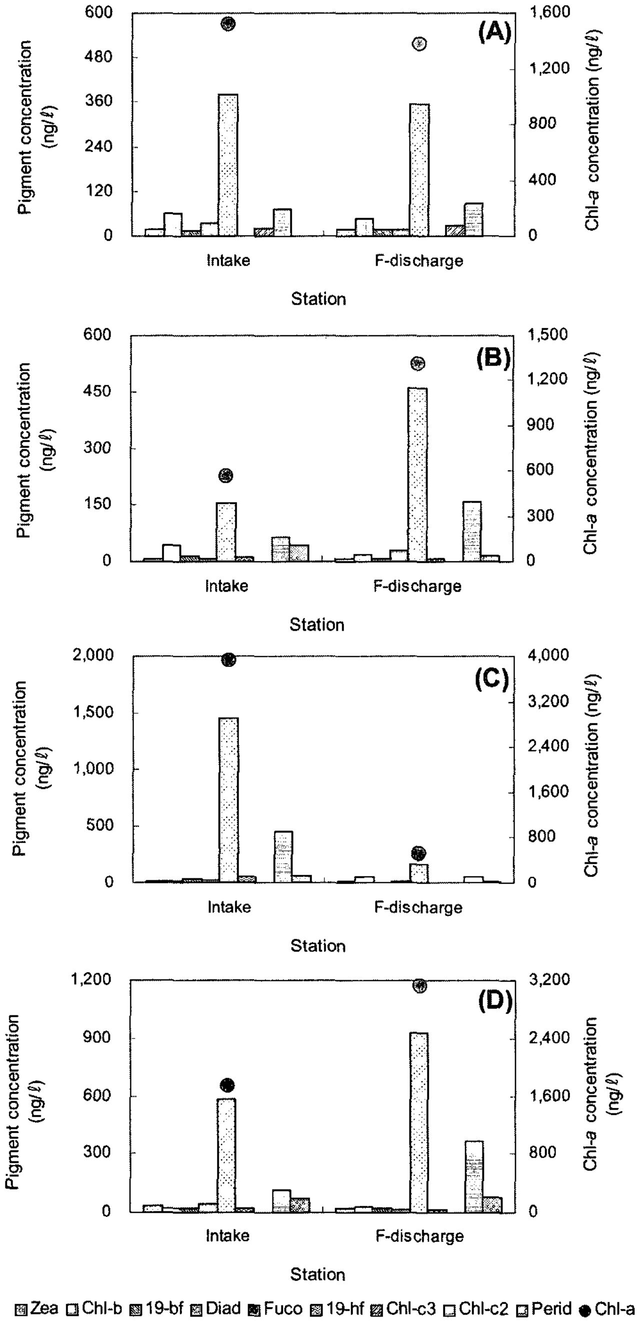

Fig. 5. Pigment and chlorophyll *a* concentrations at the intake and discharge mouth in the adjacent waters of four NPPs. For explanations of pigment abbreviations see Table 1 (A: Yonggwang, B: Kori, C: Wolsong, D: Ulchin).

The chlorophyll *a* concentration in the KNPP was increased about 130.3% after passing through the cooling-water system. The amounts of diadinoxanthin, fucoxanthin and chlorophyll c_2 were also sharply increased about 339.7%, 194.2% and 152.7%, respectively. Other pigments such as zeaxanthin, chlorophyll *b*, 19'-butanoyloxyfucoxanthin, 19'-hexanoyloxyfucoxanthin and peridinin have slightly decreased 24.2%, 62.9%,

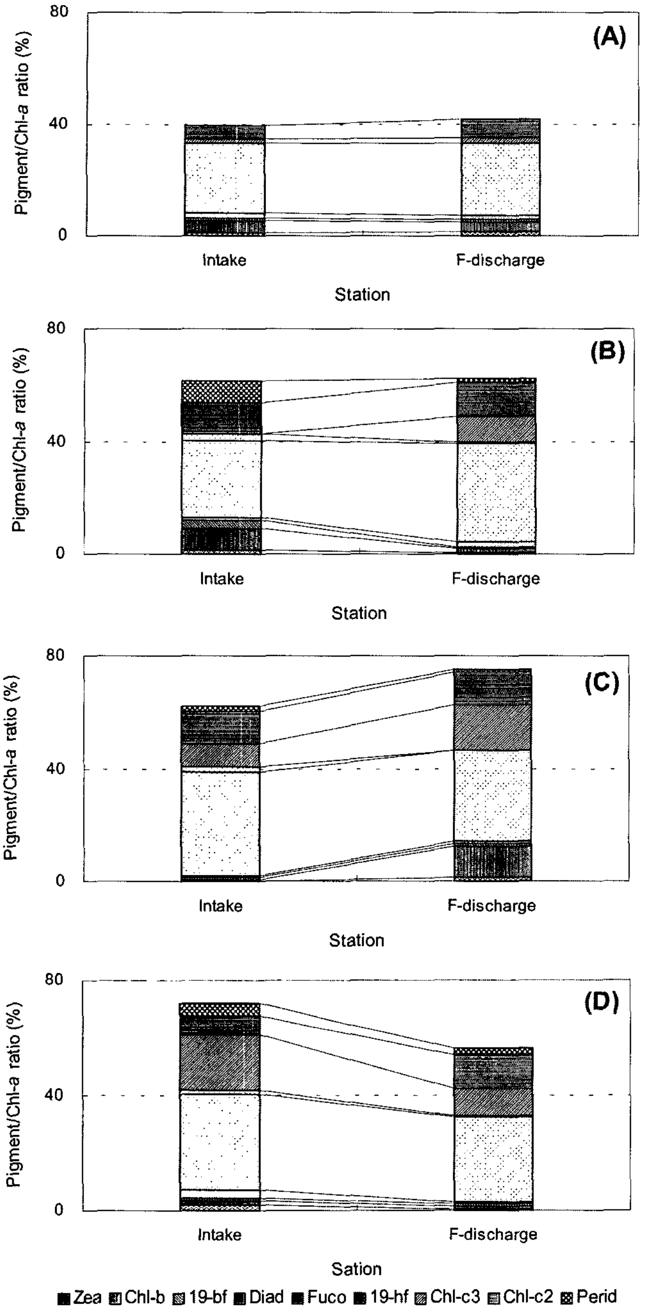

Fig. 6. Pigment/chlorophyll *a* ratio at the intake and discharge mouth in the adjacent waters of four NPPs. For explanations of pigment abbreviations see Table 1 (A: Yonggwang, B: Kori, C: Wolsong, D: Ulchin).

57.4%, 37.6% and 69.4%, respectively (Fig. 5B, 6B). In the WNPP, the chlorophyll *a* concentration in the discharge mouth was 86.5% lower than at the intake site. The amount of 19'-hexanoyloxyfucoxanthin has reduced about 100% and the other carotenoids such as 19'-butanoyloxyfucoxanthin, diatoxanthin, fucoxanthin, chlorophyll c_2 and peridinin have reduced about 86.3%, 73.0% 88.3%, 86.5% and 91.1%. On the other hand, the amounts of zeaxanthin and chlorophyll *b* have increased 36.5% and 262.8%, respectively (Fig. 5C, 6C). In case of UNPP, the chlorophyll *a* was slightly increased about 80.5%. In the majority pigments except for zeaxanthin, 19'-butanoyloxyfucoxanthin, diadinoxanthin and 19'-hexanoyloxyfucoxanthin, their amounts were increased. Chlorophyll *b*, fucoxanthin, chlorophyll c_2 and peridinin have increased about 56.3%, 58.9%, 224.1% and 15.6%, respectively. And, the amounts of zeaxanthin, 19'-butanoyloxyfucoxanthin, diadinoxanthin and 19'-hexanoylo xyfucoxanthin have slightly decreased 48.1%, 11.9%, 66.6% and 7.8%, respectively (Fig. 5D, 6D).

DISCUSSION

In spring when the phytoplankton bloom was frequently occurred in these areas (Kang and Choi, 2002; KEPCO, 2003), the concentration of each pigment was highly variable and phytoplankton composition analyzed by HPLC was similar in the adjacent waters of four NPPs in this study. The highest and lowest chlorophyll *a* concentrations were observed in the adjacent waters of WNPP and KNPP, respectively. The chlorophyll *a* concentration in this study was higher than that of southern waters of East Sea and similar to the result from Kyeonggi bay (Park and Park, 1997; Song, 1999). The concentrations of chlorophyll *a* and carotenoids observed in the southern waters of East Sea, Kyeonggi bay and this study were summarized in Table 2. Among the carotenoids, the amounts of fucoxanthin and chlorophyll c_2 were relatively higher than those of other pigments in study area (Fig. 3, 4). The fucoxanthin/chlorophyll *a* ratio varied from 0.24 to 0.32 and the chlorophyll c_2/chlorophyll *a* ratio varied from 0.08 to 0.11. These results pointed out the dominance of diatoms all stations in spring. Except for fucoxanthin and chlorophyll c_2, the amounts of other pigments were relatively low. As minor pigments, zeaxanthin, chlorophyll *b*, 19'-butanoyloxyfucoxanthin, diadinoxanthin, 19'-hexanoyloxyfucoxanthin, chlorophyll c_3 and peridinin were detected. It showed the presence of cyanbacteria, green algae, chrysophytes, prymnesiophytes and dinoflagellates in the study area. Chlorophyll c_3, marking prymnesiophytes, was not detected around the WNPP and KNPP. In the adjacent waters of each NPP, the 19'-butanoyloxyfucoxanthin, diadinoxanthin and 19'-hexanoyloxyfucoxanthin/chlorophyll *a* ratio were not much different among the stations. However, in case of zeaxanthin, chlorophyll *b* and peridinin/chlorophyll *a* ratio,

Table 2. The comparisons of the pigment concentration (ng l^{-1}) of phytoplankton in the adjacent waters of Korea. For explanations of pigment abbreviations see Table 1.

Pigments	Park and Park (1997)[a]	Song (1999)[b]	This study (April, 2004)			
			Yonggwang	Kori	Wolsong	Ulchin
chl *a*	7.2 - 180.4	553.8 - 23,063.7	817.1 - 1,522.9	544.5 - 578.4	1,096.4 - 3,938.5	1,732.2 - 2,353.9
zea	3.9 - 34.1	n.d. - 65.6	17.3 - 20.5	8.2 - 12.7	0.0 - 8.7	0.0 - 35.6
chl *b*	22.7 - 53.7	n.d. - 1,448.3	60.6 - 95.1	31.9 - 47.7	18.7 - 90.7	15.8 - 60.6
19'-bf	3.9 - 16.7	n.d. - 247.4	14.5 - 24.3	0.0 - 23.0	3.9 - 31.7	9.1 - 32.1
diad	-	-	20.4 - 35.2	6.1 - 15.5	10.9 - 26.2	11.9 - 42.2
fuco	7.7 - 211.6	80.1 - 4,701.7	178.7- 378.3	146.7 - 206.3	255.3 - 1,455.3	581.5 - 743.8
19'-hf	5.4 - 75.0	n.d. - 493.0	0.0 - 15.4	0.0 - 32.0	10.8 - 63.4	11.2 - 19.1
chl c_2	3.3 - 58.5[c]	-	71.1 - 81.2	48.7 - 67.5	102.3 - 453.0	113.0 - 241.3
chl c_3	-	-	23.4 - 38.2	n.d.	n.d.	0.0 - 66.3
peri	-	n.d. - 5,279.8	0.0 - 31.1	24.4 - 44.0	6.2 - 69.4	39.1 - 307.0

*a was conducted at the southern waters of the East Sea on October, 1996.
*b was conducted at the Kyeonggi bay from December 1997 to November 1998.
*c included the concentration of the chl c_1.
*n.d.=not detected

some differences were shown. The zeaxanthin concentration around the WNPP was relatively lower than those of other NPPs. Chlorophyll *b*/chlorophyll *a* ratio was high in the adjacent waters of YNPP and KNPP. And, peridinin/chlorophyll *a* ratio was high around the UNPP and KNPP. These results indicated that the concentration of cyanobacteria was high around the NPPs except WNPP and the green alga such as chlorophytes and prasinophytes were abundant around the YNPP and KNPP in the spring. Also, dinoflagellates characterized by peridinin were common around the UNPP and KNPP in this season.

The input of artificial heat energy into marine environment is an important factor affecting the diversity, the community structure and production rate of marine organisms (Anraku and Kozasa, 1979; Langford, 1990). Phytoplankton community was known to be affected by elevated temperature, chlorination or dechlorination and mechanical effects (Langford, 1990). The reason for the main losses of cells of phytoplankton during the entrainment was due to the elevated temperature (Briand, 1975). Some studies on the effect of entrainment on phytoplankton community were conducted (Yi and Chin, 1987; Cho, 1988; Yeo, 1992), and they showed the reduction of the standing stocks, biomass and primary production of phytoplankton. Contrary to the these studies, the outfall productivity of phytoplankton as measured by the assimilation of ^{14}C increased about 300~400% below 25 (Langford, 1990). In this study, the chlorophyll *a* concentration was reduced after passing through the cooling-water system at YNPP and WNPP, but it was sharply increased at KNPP and UNPP (Fig. 5). Our results showed that the entrainment effect on the abundance and biomass of phytoplankton varied considerably. The entrainment effects on phytoplankton assemblages were highly variable because the phytoplankton species responded differently to various factors such as elevated temperature, chlorination and mechanical impacts. So, it seemed that the variation between the effects at each NPP was due to the differences of species composition of phytoplankton assemblages or the temperature in the cooling-water system. Detailed laboratory and field experiment is needed to reveal the effects of entrainment on phytoplankton assemblages. In this study, there were no distinct trend in the change of each pigment amount, but the variation of fucoxanthin and chlorophyll c_2 concentrations by entrainment was highly coupled with that of chlorophyll *a* concentration (Fig. 5, 6). We pointed out that the diatoms controlled the overall variation of phytoplankton biomass while passing through the cooling-water system.

REFERENCES

Anraku M. and E. Kozasa, 1979. The effects of heated effluents on the production of marine plankton (Takahama nuclear power station-II). *Bull. Plankton Soc. Japan.* **25**: 77–86.

Briand F.J.P., 1975. Effects of power-plant cooling systems on marine phytoplankton. *Mar. Biol.* **33**: 135–146.

Cho K.C., 1988. The effects of thermal effluents from nuclear power plant on the ecology of phytoplankton. MS thesis. Inha Univ. 103pp (in Korean).

Fenchel T., 1988. Marine plankton food chains. *Annu. Rev. Ecol. Syst.* **19**: 19–38.

Gieskes W.W. and G.W. Kraay, 1983. Unknown chlorophyll *a* derivatives in the North Sea and the tropical Atlantic Ocean revealed by HPLC analysis. *Limnol. Oceanogr.* **28**: 757–766.

Holm-Hansen O., C.J. Lorenzen, R.W. Holmes and J.D.H. Strickland, 1965. Fluorimetric determination of chlorophyll. *J. Cons. perm. int. Explor. Mer.* **30**: 3–15.

IAEA, 1974. Thermal discharges of nuclear power stations: Their management and environmental impacts. International Atomic Energy Agency. Technical Report Series 155, Vienna.

Jeffrey S.W., 1976. A report of green algal pigments in the central North Pacific Ocean. *Mar. Biol.* **37**: 33–37.

Jeffrey S.W., 1997. Application of pigment methods to oceanography. Phytoplankton pigments in oceanography. S.W.Jeffrey, R.F.C. Mantoura and S.W. Wright, Paris, UNESCO. pp 127–166.

Jeffrey S.W. and G.M. Hallegraeff, 1980. Studies of phytoplankton species and photosynthetic pigments in a warm-core eddy of the East Australian Current I. Summer populations. *Mar. Ecol. Prog. Ser.* **3**: 285–294.

Kang Y.S. and J.K. Choi, 2001. Ecological characteristics of phytoplankton communities in the coastal waters of Gori, Wulseong, Uljin and Youngkwang I. Species composition and distribution (1992–1996). *Algae* **16**: 85–111 (in Korean).

Kang Y.S. and J.K. Choi, 2002. Ecological characteristics of phytoplankton communities in the coastal waters of Gori, Wulseong, Uljin and Youngkwang II. Distributions of standing crops and environmental variables (1992–1996). *J. Korean Soc. Oceanogr.* **26**: 77–82 (in Korean).

Kang Y.S., J.K. Choi and H.M. Eum, 2003. Ecological characteristics of phytoplankton communities in the coastal waters of Gori, Wulseong, Uljin and Youngkwang III. Distribution of dominant species and environmental variables. *Algae* **18**: 29–47 (in Korean).

KEPCO, 2003. Environmental survey and assessment around Gori, Wolseong, Uljin and Younggwang nuclear power plant (19862002). TM.C97TS01.S2003.617. 61pp (in Korean).

Kim Y.H. and S.I. Choi, 1995. Effects of cooling system at power plant on marine algal vegetation. *Algae* **10**: 121–141 (in Korean).

Langford T.E.L., 1990. Ecological effects of thermal discharges. Elservier Applied Science, London. 468 pp.

Lee J.H. and E.H. Lee, 1997. Seasonal variations of phytoplankton communities in the vicinity of thermal power plants (Boryung and Seochon). *Algae* **12**: 105–115 (in Korean).

Leterlier R.M., R.R. Bidigare, D.V. Hebel, M.E. Ondrusek, C.D. Winn, and D.M. Karl, 1993. Temporal variability of phytoplankton community structure based on pigment analysis. *Limnol. Oceanogr.* **38**: 1420–1437.

Lorenzen C.J. and S.W. Jeffrey, 1980. Determination of chlorophyll in seawater. *UNESCO Tech. pp. Mar. Sci.* **35**: 1–20.

Naylor E., 1965. Biological effects of heated effluent in docks at Swansea, S. Wales. *Proc. Zool. Lond.* **144**: 253–268.

Park M.O. and J.S. Park, 1997. HPLC method for the analysis of chlorophylls and carotenoids from marine phytoplankton. *J. Korean Soc. Oceanogr.* **32**: 46–55.

Shim J.H., H.G. Yeo and Y.K. Shin, 1991. Ecological effect of thermal effluent in the Korean coastal waters I. Significance of autotrophic nano and picoplankton in adjacent waters of Kori nuclear power plant. *J. Korean Soc. Oceanogr.* **26**: 77–82 (in Korean).

Song T.Y., 1999. Monthly variation of phytoplankton community structure and pigments in the Kyeonggi Bay, Korea. MS thesis. Inha Univ. 106 pp (in Korean).

Strickland J.D. and T.R. Parsons. 1972. A Practical Handbook of Seawater Analysis. 2nd ed., *Bull. Fish. Res. Bd. Can.* 167 p.

Suresh K, M.S. Ahamed, G. Durairaj and K.V.K. Nair, 1995. Environmental physiology of the mole crab *Emerita asiatica*, at a power plant discharge area on the east coast of India. *Environ. Pollut.* **17**: 161–165.

Trees C.C., R.R. Bidigare and J.M. Brooks, 1986. Distributions of chlorophylls and phaeopigments in the Northwestern Atlantic Ocean. *J. Plankton Res.* **8**: 447–458.

Yeo H.G., 1992. An environmental biological study on primary production system in thermal effluent region. Ph.D. thesis, Seoul Natnl. Univ. 155 pp (in Korean).

Yeo H.G. and J.H. Shim, 1992. Ecological effects of thermal effluent in the Korean coastal waters III. Abiotic environmental and the community structure of primary producers in the vicinity of Kori nuclear power plant. *Korean J. Environ. Biol.* **11**: 124–130 (in Korean).

Yi S.K. and P. Chin, 1987. Effects of cooling water system of a power plant on marine organisms I. Effects on primary production. *Bull. Korean Fish. Soc.* **20**: 381–390 (in Korean).

Yoo K.I. and J.H. Lee, 1982. Studies on the planktonic diatoms in the vicinity of Kori nuclear power plant. *Bulletin of KORDI* **4**: 53–62 (in Korean).

Wright S.W., S.W. Jeffrey, R.F. Mantoura, C.A. Llewellyn, F. Bjørnland, D. Repeta and N. Welschmeyer, 1991. Improved HPLC methods for the analysis of chlorophylls and carotenoids from marine phytoplankton. *Mar. Ecol. Prog. Ser.* **77**: 183–196.

Wright S.W., D.P. Thomas, H.J. Marchant, H.W. Higgins, M.D. Mackey and D.J. Mackey, 1996. Analysis of phytoplankton of the Australian sector of the Southern Ocean: comparisons of microscopy and size frequency data with interpretations of pigment HPLC data using the CHEMTAX matrix factorization program. *Mar. Ecol. Prog. Ser.* **144**: 285–298.

Manuscript received September 15, 2004
Revision accepted December 29, 2004
Editorial handling: Ik-Kyo Chung