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Seasonal Changes of Chlorophyll *a*, Heterotrophic Bacteria, Particulate Organic Carbon, and Nutrients in Kagoshima Harbor

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Abstract

Seasonal variations of chlorophyll *a*, heterotrophic bacteria, particulate organic carbon, and nutrients in Kagoshima Harbor were studied from April 1990 to March 1991. The total amounts of chlorophyll in the water column was high in June and November, and low from December to May. The seasonal cycle of chlorophyll was strongly influenced by the nutrient standing stock. The viable counts of heterotrophic bacteria showed high values in June and December and these bacterial population peaks coincided with the peaks of particulate organic matter and phytoplankton. The level of particulate organic carbon in Kagoshima Harbor was up to ten times higher than that of the euphotic layer in the open Pacific Ocean. It was suggested that the seasonal cycle of particulate organic carbon was closely related to phytoplankton production.

Introduction

Oceanography in Kagoshima Bay has been studied by many investigators of Kagoshima University. Ōki (1989) briefly reviewed these published and unpublished oceanographic data obtained in Kagoshima Bay. However, information on biological oceanographic processes in Kagoshima Bay are still fragmentary. It is well known that there is a characteristic seasonal cycles of living organisms and non-living matter in temperate inshore waters. The seasonal cycles in Kagoshima Bay are not well described because most previous observations were done monthly or bimonthly. More frequent sampling is required to observe actual seasonal variation, especially in temperate water. In the present paper, we report the seasonal changes in chlorophyll *a* (phytoplankton biomass), heterotrophic bacteria, particulate organic carbon, and nutrients in Kagoshima Harbor in Kagoshima Bay. The point of discussion in the present work is to describe characteristic seasonal cycles of the above factors. Measurement were taken weekly or biweekly from April 1990 to March 1991. The basic data observed in the present study would be useful to an evaluation of environmental changes in Kagoshima Harbor.

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Materials and Methods

Study site

Kagoshima Harbor is located in a narrow and shallow strait west of Sakurajima in Kagoshima Bay. Although two sampling stations were selected for the study, the results of only one station is reported (Fig. 1), because both stations showed basically the same tendency in distribution of measured factors. As reclamation work is making progress in Kagoshima Harbor, the Kagoshima Environmental Research and Service has been monitoring water quality for a full two years. Our station is one of the sampling stations which has been observed by the Kagoshima Environmental Research and Service.

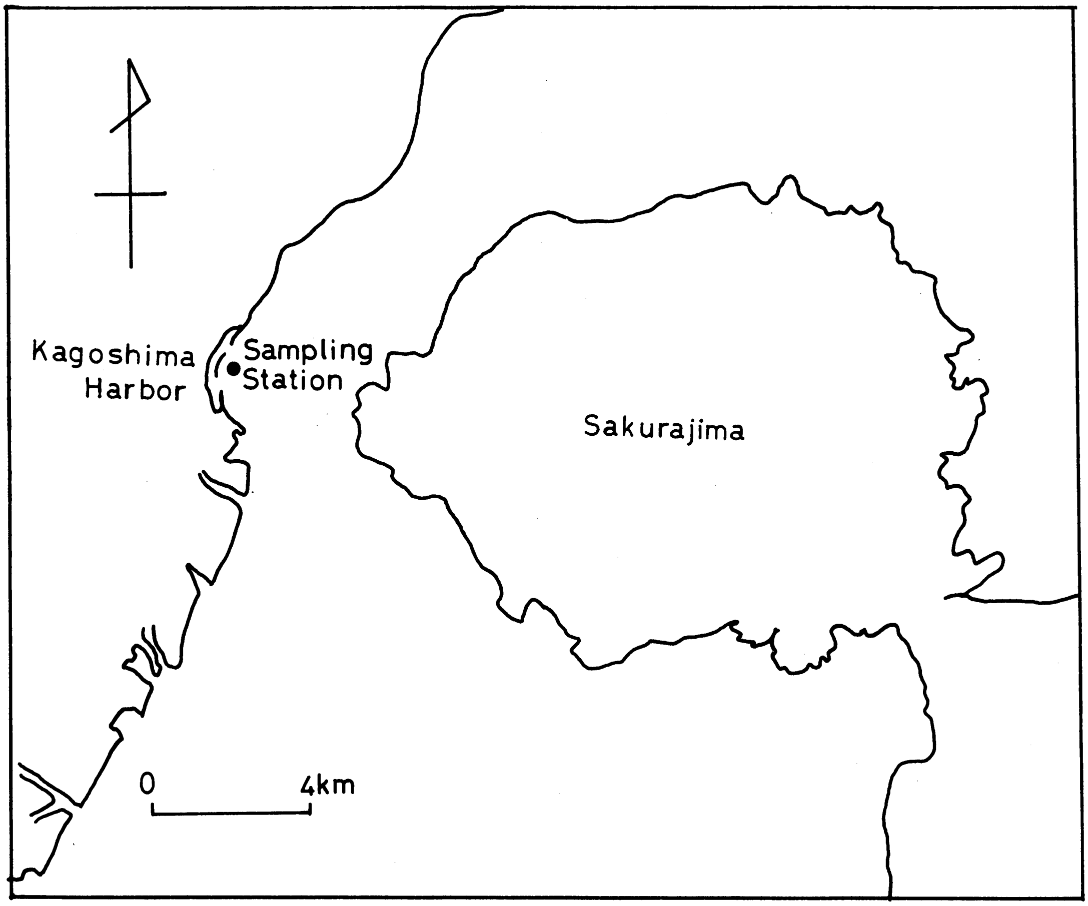

Fig. 1. Location of sampling station.

Water sampling

A total of 35 surveys were carried out for the present study from April 1990 to March of 1991 on board a research boat chartered by the Kagoshima Environmental Research and Service. Sea water samples were collected with a six liter Van Dorn water sampler at depths of 1, 5, 10, 15, 20 m, and near to the bottom (about 25 m). The water for counting viable bacteria was immediately transferred from the water bottle into a sterilized glass bottle and kept in a cold container. The water for chlorophyll, particulate organic carbon (POC), and nutrients was drained into five-liter plastic bottles. For chlorophyll and POC analyses, one liter of subsample was filtered respectively through a 47 mm Whatman GF/C glass fiber filter. All filters for POC determination were combusted in advance at 450 ℃ for 2 hours in a muffle furnace. The filters were stored in a deep freeze until later analyses. The filtrate was collected in a 250 ml plastic bottle and froze at −20 ℃ until nutrient analysis. The ammonia concentration in sea water was determined within several hours after sampling.

Chlorophyll *a* and POC

Chlorophyll *a* is the most useful indicator to estimate phytoplankton biomass in sea water. The glass fiber filter with precipitate was put in a glass mortar and chlorophyll was extracted with a 90% acetone solution. Chlorophyll concentrations were determined spectrophotometrically by the method of Parsons et al. (1984).

The particulate matter filtered onto the glass fiber filters included both living and non-living matter. The determination of POC is a widely used indicator to estimate total suspended particulate organic matter. POC in the present study was analyzed by a wet combustion method with dichromate and concentrated sulfuric acid based on the procedure by Parsons et al. (1984).

Heterotrophic bacteria

The viable counts of bacteria were done at three different layers (1, 10, 25 m). We basically followed the method developed by Yanagita et al. (1978). The water samples were diluted tenfold, and 4 ml of these diluted samples were filtered through 25 mm HA type Millipore filters fixed with Swinnex filter holders. The filters were placed on two kinds of agar plates in Petri dishes. Organic nutrient rich and poor media were prepared. The nutrient rich medium contained 10 g of polypeptone, 5 g of yeast extract, and 15 g of agar in one liter of filtered sea water, while the nutrient poor medium contained 10 mg of polypeptone and 15 g of agar. Duplicate or triplicate filtrations were carried out and the filters were placed on both the nutrient rich and poor media. The filters were incubated at 23 ℃ for two days for nutrient rich medium, and usually seven to ten days for the poor medium. After incubation, the numbers of colonies grown on the filters was counted with a binocular microscope.

Nutrient

Analyses of phosphate (PO_4-P), nitrate (NO_3-N), and nitrite (NO_2-N) were followed by the methods of Parsons et al. (1984). Inorganic phosphate was determined by the reduction of the phospho-molybdate complex with ascorbic acid. Nitrate was reduced to nitrite through a cadmium reduction column, and produced nitrite was measured spectrophotometrically. Ammonia was analyzed by the phenolhypochlorite method (Solórzano, 1969). Total dissolved nitrogen (both inorganic and organic) in the sea water was determined using persulfate digestion as described by D'Elia et al. (1976). Then, the dissolved organic nitrogen (DON) content was calculated as the difference between the total nitrogen and the sum of inorganic nitrogen (nitrate, nitrite, and ammonia).

Results

Fig. 2 shows the seasonal change in the Secchi disk transparency test. In spite of the shallow study site (about 27 m), the seasonal pattern of the observed transparency was essentially the same as that of the previous year. Secchi disk transparencies of this study ranged from 2 to 12 m. The lowest transparency of 2 m, due to a phytoplankton increase, appeared in June as shown in Fig. 3, and the transparency gradually increase with time to 9 m (September). The Secchi disk depth decreased again to 7.5 m in November at the second peak of chlorophyll. The transparencies in winter showed the highest values of 12 m, and suddenly dropped to 3.5 m at the end of March.

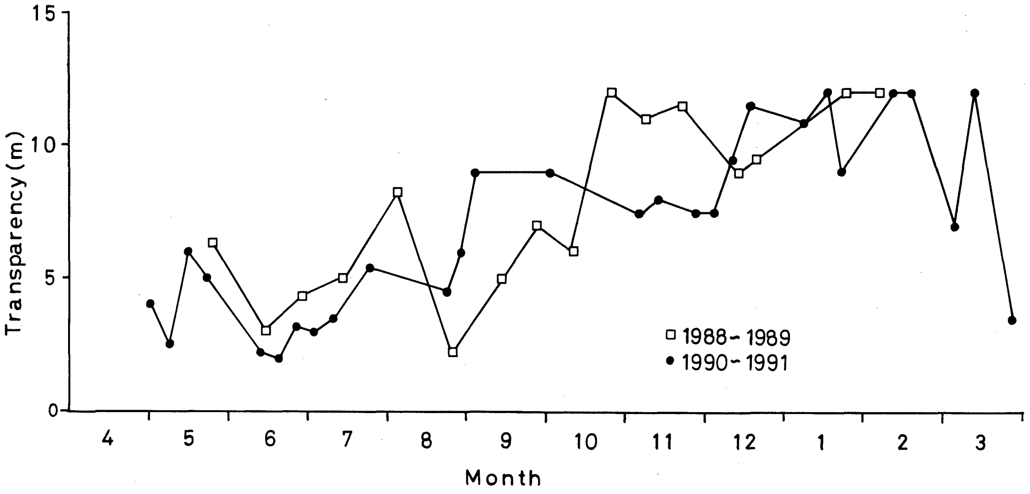

Fig. 2. Seasonal change in the Secchi disk transparency (m).

Water temperature

Fig. 3 shows the seasonal cycle of temperature in Kagoshima Harbor. The general pattern of temperature distribution was characterized by the vertical isolines down to the bottom during

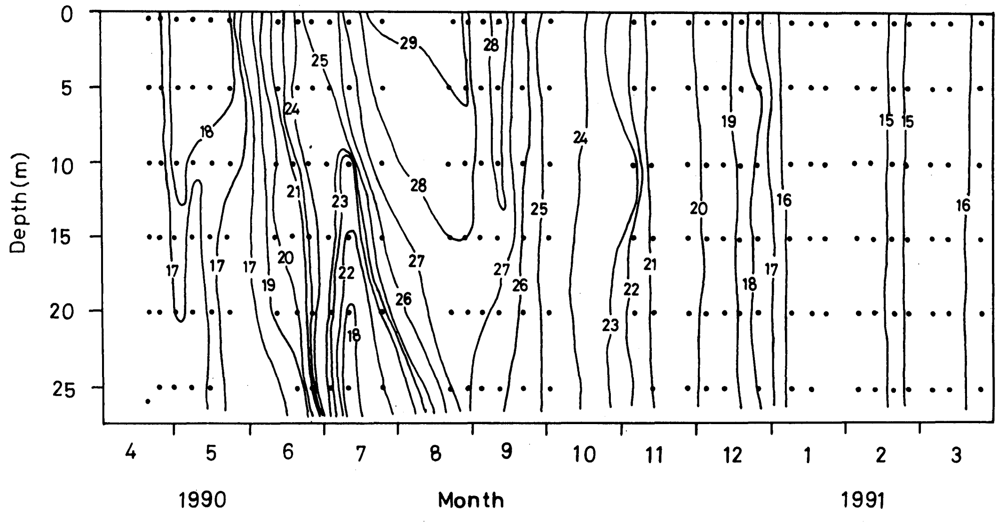

Fig. 3. Seasonal variation of water temperature (℃).

September to June. In this ten-month period, the water temperature in the water column was vertically homogeneous. The highest temperature of 29.2 ℃ was found at the surface layer at the end of June, and the temperature of the water column decreased towards winter. The lowest temperature of 14.6 ℃ was observed at the end of February, and the temperature increased as spring progressed. The temperature gap between the sea surface and the bottom was greatest (9 ℃) at the beginning of June. In late summer, a decline of the temperature resulted in the vertical circulation of water and the mixing continued to April with a gradual cooling of the water column. The duration of vertical circulation in the station may be variable in response to meteorological conditions. In July and August, water circulation seemed to be prevented by rapid heating of the surface layer from solar radiation.

Chlorophyll *a* (phytoplankton biomass)

Fig. 4 shows the seasonal cycle of chlorophyll *a* concentrations. Chlorophyll values in all of the observed samples ranged from 0.1 μg/l (July, 25 m) to 8.5 μg/l (June, 5 m). One of the characteristics in chlorophyll distribution was that distinct chlorophyll peaks appeared during the observation. High chlorophyll concentrations exceeding 5 μg/l was first observed in June. The concentration in the first peak attained to more than 8 μg/l (5, 10 m). After the peak, in the stratified summer, the chlorophyll contents dropped to low values and fluctuated about 1 to 4 μg/l. The chlorophyll maximum layer was often observed in 5 to 15 m of depth in July to November. A second chlorophyll peak appeared in November. The concentrations of the second peak were comparable to that of the first peak in July. After the second chlorophyll peak, concentrations ranged from 1-4 μg/l in the winter and spring. The chlorophyll concentrations in this circulation period were vertically homogeneous down to the bottom. Particularly, in the end of April, chlorophyll concentration in the whole water column showed very low values of less than 0.6 μg/l. The phytoplankton biomass gradually increased with time in late spring and attained the first peak described above.

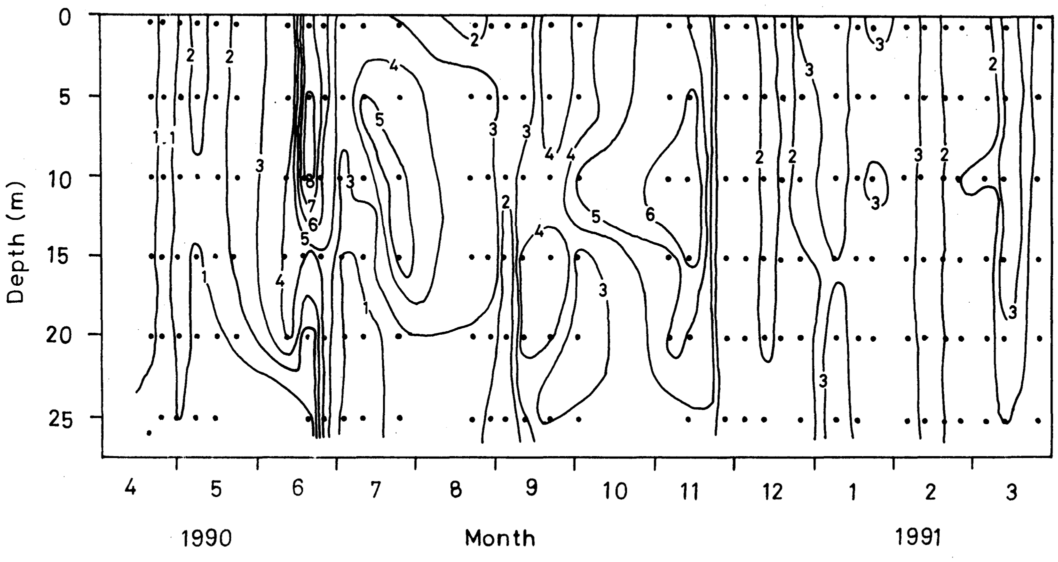

Fig. 4. Seasonal variation of chlorophyll concentrations (μg/l).

Heterotrophic bacteria

Fig. 5 shows the seasonal variation of viable counts of heterotrophic bacteria. Colony numbers of bacteria were counted by using two different media. We defined R and P bacteria in the present study that could grow on organic nutrient rich and poor medium, respectively. The pattern of seasonal variation of R bacteria was essentially the same as P bacteria. One notable feature was that higher concentrations of R and P bacteria were found in early summer (June and July). Especially the rapid increase in colony numbers in June coincided with the chlorophyll peak as shown in Fig. 4. In this bacterial peak, high concentrations exceeding 400 colonies/ml were observed in the whole water column. The highest concentrations exceeding 1000 colonies/ml were counted in the P bacteria at the

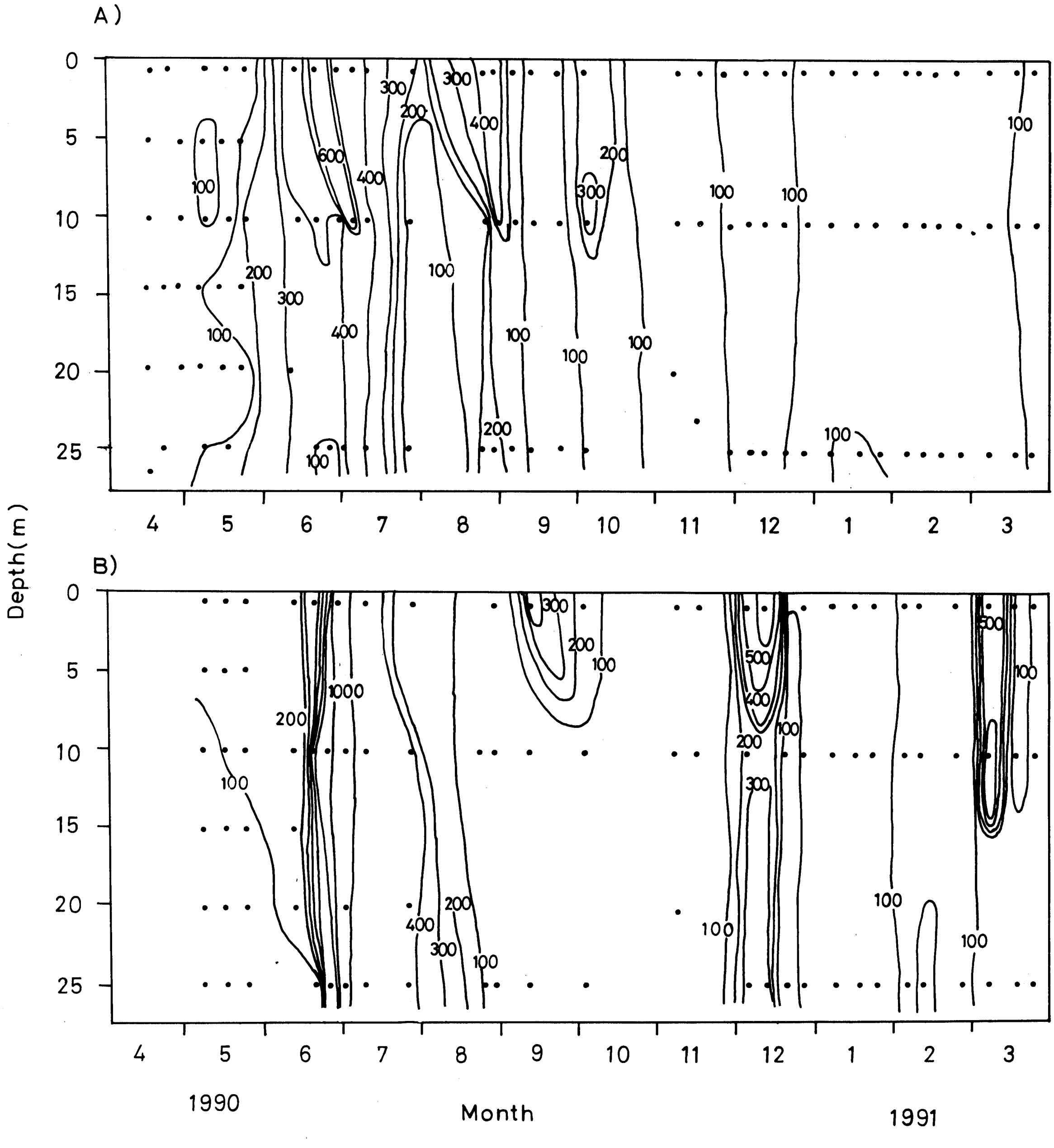

Fig. 5. Seasonal variation of viable counts of R (A) and P bacteria (B) in colonies/ml.

end of June. A pronounced second peak was found only in P bacteria in December, while the second peak of R bacteria was small. P bacteria increased more than fivefold in concentration. The second peaks of bacteria did not coincide with the chlorophyll peak observed in autumn, and the bacterial peaks occurred after the chlorophyll peak (Fig. 4). During winter and spring, both R and P bacteria showed low concentrations. Bacterial poor water of less than 10 colonies/ml was frequently found in winter. Except two bacterial peaks, the general range of the concentrations of R and P bacteria was the same order.

Particulate organic carbon (POC)

Fig. 6 shows the seasonal changes in concentration of particulate organic carbon. The carbon concentration ranged from 52 μgC/l to 899 μgC/l. The highest concentration was found at the sea surface in December and the lowest concentration was observed at 15 m in depth in May. One remarkable feature was that a seasonal variation in POC concentration was observed (Fig. 6). High values exceeding 400 μgC/l were found in early summer and in winter in the whole water column. The POC concentration in December was the highest, more than 600 μgC/l. A period of POC increase in June was observed during the period of chlorophyll increase, however, the POC peak in December followed the chlorophyll peak. Between POC peaks, POC concentrations generally fluctuated in the range of 100 to 300 μgC/l.

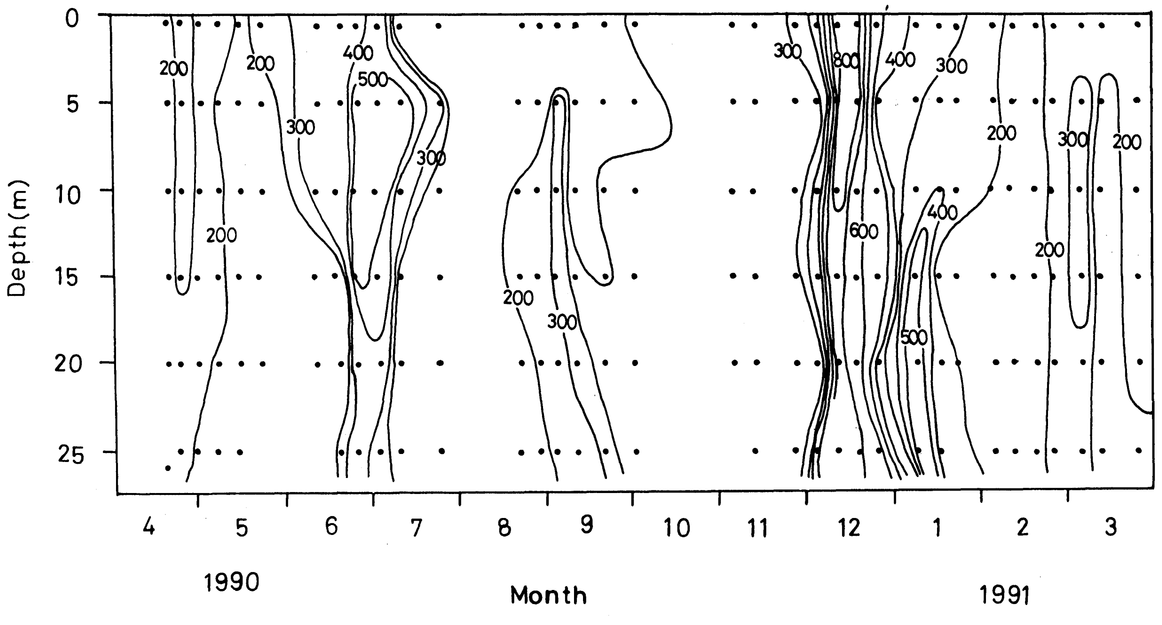

Fig. 6. Seasonal variation of particulate organic carbon (POC) concentrations (μgC/l).

Nutrients

Fig. 7 shows the seasonal variation of phosphate concentrations. The phosphate concentration in Kagoshima Harbor ranged from an undetectable amount to 2.8 μg-atP/l. Higher concentrations, ranging from 1.0 to 2.8 μg-atP/l, were observed in May before the rapid increase of phytoplankton, and lower values of less than 0.1 μg-atP/l were observed from June to September. Relatively high values of phosphate (0.5-1.3 μg-atP/l) were found in the winter season (circulation period).

Fig. 8 shows the seasonal changes of nitrate and nitrite. The nitrate concentrations varied from

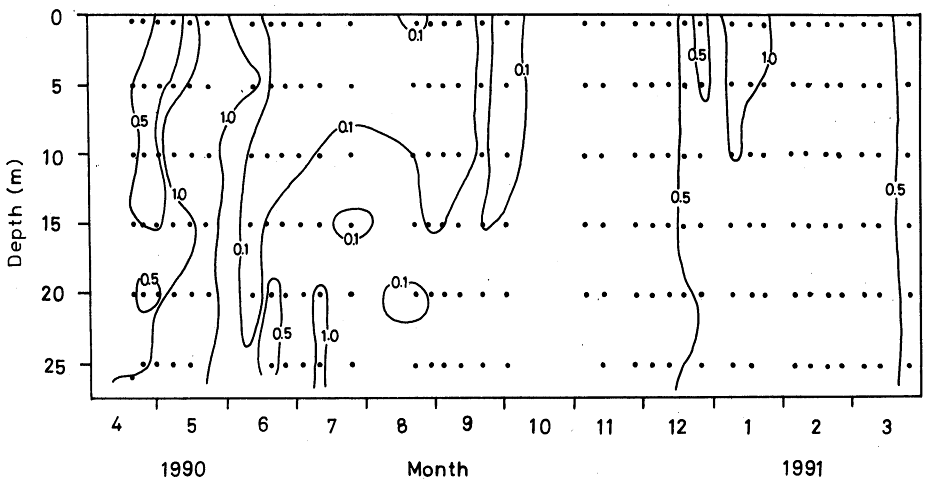

Fig. 7. Seasonal variation of phosphate concentrations (μg-atP/l).

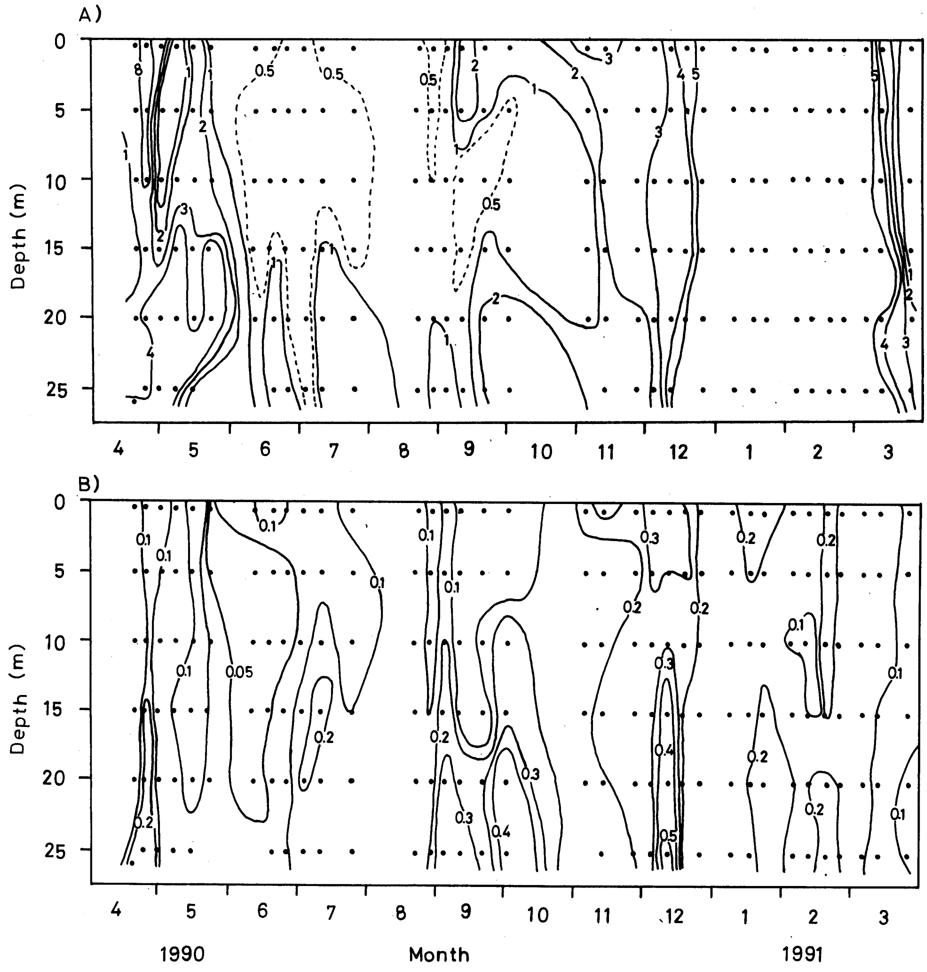

Fig. 8. Seasonal variation of nitrate (A) and nitrite (B) concentrations (μg-atN/l).

undetectable to 17 μg-atN/l. High concentrations exceeding 5 μg-atN/l were observed in the entire water column from December to the middle of March. During this period, remarkably high concentrations of greater than 10 μg-atN/l were often observed below 5 m in depth. The reason for these high values would be a result of nitrification since the nitrite concentration in the high nitrate layer significantly decreased. However, high nitrate concentrations in the surface layer would probably be an effect of run-off from the surrounding area. Low nitrate contents of less than 1 μg-atN/l were found from June to early November except near the sea bottom. Very low concentrations of less than 0.5 μg-at N/l were frequently observed in June and July during the period of chlorophyll increase. After the high nitrate season in the winter, the nitrate decreased to less than 1 μg-atN/l, and the nitrate again increased in April.

Nitrite concentrations ranged from negligible concentrations to 0.7 μg-atN/l. Low concentrations ranging from negligible to 0.1 μg-atN/l were observed in the entire water column from May to July and from March to April. Nitrite in September and early October showed low values down to 15 m in depth, however, it rapidly increased with depth to more than 0.4 μg-atN/l. Extremely low concentrations of nitrite of less than 0.05 μg-atN/l occurred in June. Nitrite contents in November to December were high, in the range of 0.2 to 0.6 μg-atN/l, and later in the circulation period they generally showed relatively high values of 0.1 to 0.3 μg-atN/l.

Fig. 9 shows the seasonal changes of ammonia. As the ammonia concentrations were variable, typical seasonal patterns were difficult to determine. The concentrations ranged from trace levels to 2.5 μg-atN/l. High ammonia contents of greater than 1.0 μg-atN/l seemed to appeared alternately with low concentrations of less than 0.5 μg-atN/l. The highest concentration of ammonia, 2.5 μg-atN/l, was observed in early July below 10 m in depth.

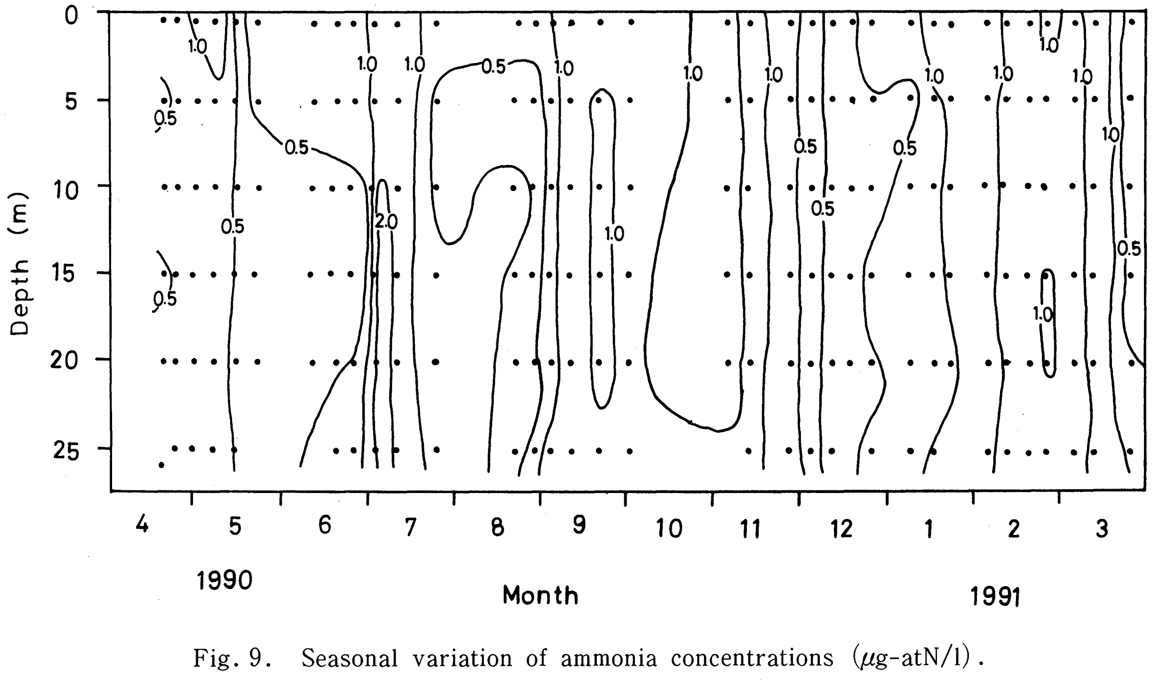

Fig. 9. Seasonal variation of ammonia concentrations (μg-atN/l).

Dissolved organic nitrogen (DON)

Fig. 10 shows the seasonal variation of the calculated concentrations of dissolved organic nitrogen (DON). Most of the DON values were in the range of 1 to 20 μg-atN/l. Extremely low DON concentrations were obtained where the nitrate values were remarkably high. DON concentrations were high, exceeding 15 μg-atN/l, in April and June. Relatively high DON values, about 10 μg-atN/l, were found in October to early November. DON concentrations in the summer and the winter showed relatively low values, 1 to 10 μg-atN/l. The DON level calculated in Kagoshima Harbor was almost similar to that observed in the open Pacific Ocean (Marumo, 1970).

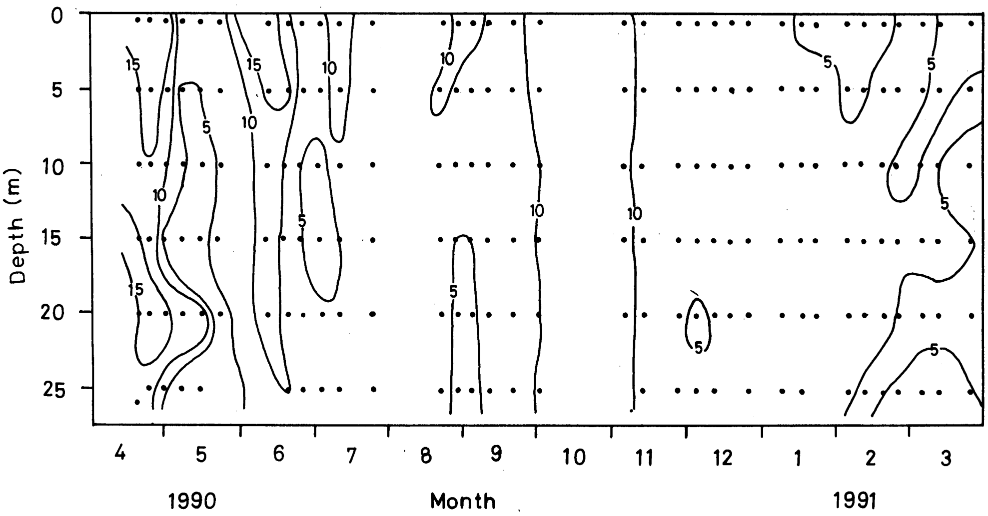

Fig. 10. Seasonal variation of dissolved organic nitrogen (DON) concentrations (μg-atN/l).

Discussion

It is evident that seasonal changes in the chlorophyll standing stock in Kagoshima Harbor were very marked; the fluctuations in the entire water column were fifteenfold in amplitude (Fig. 11).

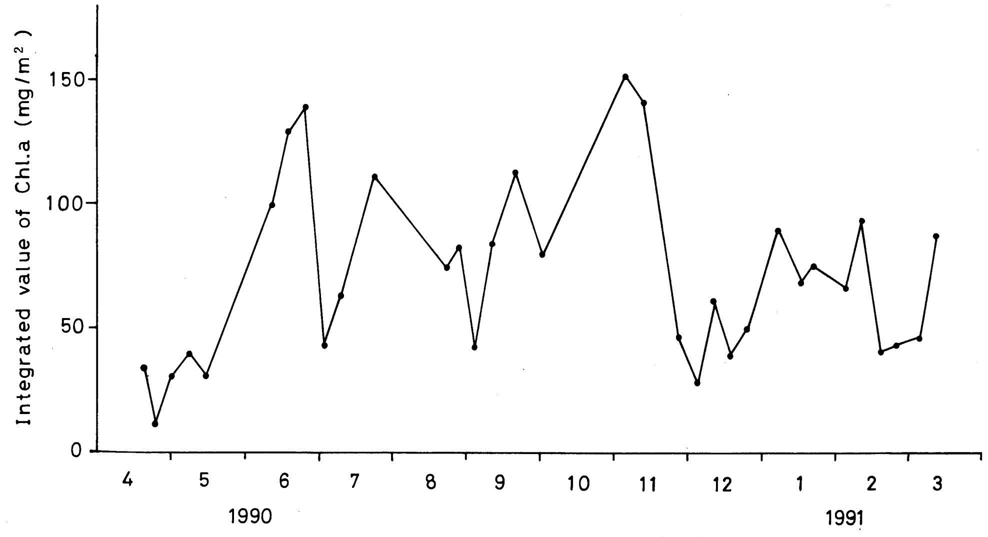

Fig. 11. Seasonal variation of total amount of chlorophyll *a* (mg) in a 1 m² of water column for 0 to 25 m depth.

High chlorophyll standing stock was observed in June (140 mg/m^2) and early November (150 mg/m^2), and low stock was found from December to May (28-92 mg/m^2). Between the two chlorophyll peaks, small peaks occurred in July (112 mg/m^2) and September (112 mg/m^2). The general level and the pattern of variation observed was essentially the same as in the previous year observed by Ichikawa and Tamotu (unpublished data). Based on these two years of observation, it is likely that there is a double peak in phytoplankton standing stock in Kagoshima Harbor.

It is generally accepted that one of the most critical factors controlling phytoplankton standing stock is nutrient supply, which is influenced by the physical (temperature) and biological (bacterial decomposition, zooplankton grazing) environment. During winter, despite the high level of nutrients, the chlorophyll standing stock was low (Fig. 11), due to the low water temperature and low radiation. The stock of phytoplankton increased from about 10 mg/m^2 (April) to 140 mg/m^2 (June), an increase of fifteen times in two months. Nitrate and phosphorus concentrations showed a decreasing trend from April with time, and an extremely low level coincided with the chlorophyll peak in June. Ammonia was at a relatively low level in April and May (about 0.5 to 1.0 μg-atN/l) and also decreased to an undetectable level related to the chlorophyll peak. But the ammonia concentration increased remarkably to 2 μg-atN/l after the chlorophyll peak. Although we have no data to explain this ammonia increase, it is possible that ammonia was regenerated by zooplankton excretion. The dissolved organic nitrogen suddenly increased in the chlorophyll peak and we feel this is related to the decomposition of organic matter by bacteria and excretion by zooplankton. The DON in sea water regenerates to nutrients and would supports the summer level of the phytoplankton standing stock. The chlorophyll standing stock can be high in summer, but probably due to lack of constant supply of nutrients due to seasonal stabilization of the water column, the phytoplankton biomass is controlled at a relatively low level. However, there were often small chlorophyll peaks in summer, this is associated with a disruption of the stratification by strong wind, the intensity of zooplankton grazing, and/or other local factors that influence nutrient supply. Summer stratification was completely broken at the end of September (Fig. 3) and the nutrients that had accumulated in the bottom layer were transported vertically in the water column, nutrient concentrations in the whole water column increased with decreasing temperature. This nutrient supply would be one of the important factors that caused the autumn peak (November) of chlorophyll. It is obvious that the nutrient standing stock in the water column was the most important factor in the seasonal change of chlorophyll in the observed area. Cushing (1975) indicated that the seasonal change of the phytoplankton standing stock was strongly influenced by the grazing activity of zooplankton. Zooplankton density data would be needed to describe the seasonal cycle of phytoplankton in Kagoshima Harbor.

The seasonal distribution of bacterial populations was highly variable (Fig. 5). Bacterial population peaks in June and December coincided with the two POC peaks observed. The bacterial peak in June also coincided with the chlorophyll peak, however, the second peak of bacteria followed the November chlorophyll peak. This suggests the heterotrophic bacteria in Kagoshima Harbor significantly depends on particulate organic matter including detritus and phytoplankton. Ichikawa (1983) found that the maximum bacterial layer in the open ocean frequently coincided with the maximum chlorophyll layer. Bell and Lang (1974) demonstrated that the growth of marine bacteria was stimulated or inhibited by the release of the extracellular products of phytoplankton. The present study suggests that one of the important factors controlling bacterial population size is the presence of living and non-living particulate matter in sea water.

Several workers reported the presence of bacteria in natural water that could not grow in nutrient rich medium such as ZoBell 2216 (Akagi et al., 1977; Mallory et at., 1977; Yanagita et al., 1978). Yanagita et al. (1978) found that colony counts were higher on nutrient poor medium (P medium) than on nutrient rich medium (R medium) in most of the samples studied, and experimentally showed that isolated bacteria from R medium produced similar numbers of colonies on R and P media. These results assumed that there was a group of bacteria that could not make colonies on nutrient rich medium. In the present case, the colony counts on P medium exceeded those on R medium in 50% of the samples, showing the presence of bacteria that could not grow on P medium. This situation was already indicated by Yanagita et al. (1978) in coastal eutrophic waters. Probably, the eutrophic waters in Kagoshima Harbor contained a significant population of bacteria that could grow only on nutrient rich medium.

Particulate organic carbon in inshore waters is derived from various sources such as phytoplankton, macroscopic algae, and terrestrial input. The relative quantitative importance of these sources in Kagoshima Harbor could not be precisely estimated, but it can be said that phytoplankton is the most important origin. POC concentration in eutrophic inshore waters is generally higher than that of oligotrophic offshore waters. The level of POC in Kagoshima Harbor is several to ten times higher than that of euphotic layers in the open Pacific Ocean (Ichikawa and Nishizawa 1975).

We suggest that the seasonal cycle of POC in Kagoshima Harbor is closely related to phytoplankton productivity. The POC peak in early summer coincided with the phytoplankton standing crop peak and the POC declined to low levels after the termination of the chlorophyll peak. The POC concentration sharply increased again in December following the chlorophyll peak in November. The delay in the POC peak would be expected because the process of particle formation is complicated and biological and physical processes may strongly affect the particle distribution. Riley (1970) observed that the number of small particles in Long Island Sound produced by physical adsorption from dissolved matter was larger in winter than in summer. In the shallow environment, resuspension of sediments would also affect POC concentration during the period of winter circulation.

Acknowledgment

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