Marine Ecosystem Enclosed Experiments

Proceedings of a symposium held in Beijing, People's Republic of China, 9–14 May 1987

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Abstract

This symposium on marine ecosystem enclosed experiments (MEEE) consists of nine review papers that describe various types of ecosystem enclosures and a series of papers resulting from enclosure experiments in Xiamen, People's Republic of China, and Saanich Inlet, BC, Canada. The reviews on types of enclosures include benthic enclosures for rocky and sandy shores and the effects of pollutants (primarily hydrocarbons) on bacteria, macroalgae, and invertebrates. The pelagic enclosures were used to study the control of phytoplankton blooms, the uptake and release of dissolved organic substances, and the effects of pesticides on freshwater ecosystems.

Six enclosure experiments were conducted in China and Canada from 1986–87. Some of these experiments examined the effects of contaminated sediments, primarily heavy metals, on bacteria, phytoplankton, and zooplankton and the pathways and fates of these heavy metals in the seawater. Other experiments studied the chemistry and biological effects of chemically dispersed oil.

Résumé

Ce compte rendu du symposium sur les expériences faites en écosystèmes marins comprend neuf communications qui décrivent les écosystèmes retenus et les expériences faites à Xiamen en République populaire de Chine et à Saanich Inlet, C.-B., au Canada. Les communications portent, notamment, sur les écosystèmes benthiques des littoraux rocheux et sablonneux et sur les effets des polluants (surtout les hydrocarbures) sur les bactéries, les grandes algues et les invertébrés. Les expériences sur le contrôle des brutales pullulations ("blooms") du phytoplancton furent menées dans les écosystèmes pélagiques, ainsi que l'absorption et le dégagement des substances organiques dissoutes et les effets des pesticides sur les écosystèmes d'eau douce.

Six expériences ont été faites en Chine et au Canada entre 1983 et 1987. Certaines ont porté sur les effets des sédiments contaminés, principalement par des métaux lourds, sur les bactéries, le phytoplancton et le zooplancton et sur le cheminement et le sort de ces métaux lourds dans l'eau salée. D'autres expériences portaient sur la chimie et les effets biologiques du pétrole dispersé chimiquement.

Resumen

Este simposio sobre Experimentos Marinos en Ecosistemas Cerrados (MEEE) consistió en nueve trabajos de análisis que describen varios tipos de enclaustramientos ecosistémicos y una serie de trabajos derivados de experimentos con estos enclaustramientos en Xiamen, República Popular de China, y en Sannich Inlet, Canadá. Los estudios incluyen enclaustramientos bentónicos para costas rocosas y arenosas, y los efectos de los contaminantes (fundamentalmente hidrocarburos) sobre bacterias, macroalgas e invertebrados. Los enclaustramientos pelágicos se utilizaron para estudiar el control de la reproducción del fitoplancton, la ingestión y expulsión de substancias orgánicas disueltas y los efectos de pesticidas en los ecosistemas de agua dulce.

Se realizaron seis experimentos en ecosistemas cerrados en China y Canadá, de 1983 a 1987. Algunos de estos experimentos examinaron los efectos que ejercen los sedimentos contaminados, fundamentalmente los metales pesados, sobre bacterias, fitoplancton y zooplancton, y el ciclo y destino final de estos metales pesados en el agua de mar. Otros experimentos estudiaron los efectos químicos y biológicos de los aceites crudos dispersados por medios químicos.

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Phosphate Limitation of Phytoplankton Growth in Coastal Estuarine Waters of China and Its Potential Interaction with Marine Pollutants

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Mesocosm experiments conducted in estuarrine coastal waters at Xiamen, People's Republic of China, indicate that phosphate is the nutrient that limits phytoplankton production. During these experiments, phytoplankton grew in marine enclosures and phosphate always reached undetectable levels several days before inorganic nitrogen (NO_3^- + NO_2^- + NH_4^+). This observation was supported by the N:P ratio in the water at the start of the experiment. The N:P ratio was nearly 80:1 (by atoms), indicating a fivefold excess of nitrogen, assuming that phytoplankters take up nitrogen and phosphorus in a N:P ratio of 16:1. Laboratory bioassay experiments with the diatom Chaetoceros calcitrans *also confirmed that phosphorus was limiting to phytoplankton growth in Xiamen Bay. These estuarine coastal waters are unique because marine waters are usually nitrogen-limited. Several other estuaries along the coast of China also have N:P ratios ranging from 30:1 to greater than 80:1. These observations suggest that many of China's large rivers appear to act like giant nutrient pumps, delivering excess nitrogen, relative to phosphorus, to the coastal waters. The consequences of phosphorus limitation in relation to eutrophication and the interaction of phosphorus-limited phytoplankton with pollutants such as oil and heavy metals are discussed.*

Nitrogen is generally regarded as the limiting element for phytoplankton growth in the ocean (Ryther and Dunstan 1971; Goldman 1975). During recent mesocosm experiments conducted in estuarine coastal waters of the People's Republic of China, this general statement was found to be untrue. Experiments and bioassay results suggest that phosphorus is clearly limiting in the estuarine coastal waters of Xiamen Bay and may be limiting in several other estuarine areas along the coast.

Mesocosm experiments

Canada

In mesocosm experiments conducted at the Institute of Ocean Sciences in Saanich Inlet, British Columbia, and in Controlled Ecosystem Pollution Experiments (CEPEX) conducted in the same inlet (Grice and Reeve 1982), inorganic nitrogen was always exhausted in the mesocosms before inorganic phosphate (Harrison et al. 1986; Parsons et al. 1986). These observations along with the initial N:P ratio in the water (usually lesser than 16:1) provide evidence that nitrogen limits phytoplankton growth in Canadian coastal waters (Harrison et al. 1983).

China

The disappearance of nutrients in mesocosm experiments in China was very different from results obtained in Canada. During a sediment- and metal-addition experiment conducted at Xiamen in 1985 (Wu J. et al., this volume), with an initial N:P ratio of 32:1 in the seawater, phosphate went to undetectable concentrations 1 week before nitrate in the control mesocosm (Fig. 1). On day 10, when the phosphate concentration went to zero, there was still 15 μM nitrate left.

Further bioassay experiments were conducted to confirm that phosphorus was limiting to phytoplankton growth. Xiamen Bay water was collected in March and the diatom *Chaetoceros calcitrans* was used as the bioassay organism. The initial nitrate and phosphate concentrations were 17.0 and 0.21 μM, respectively, yielding a N:P ratio of 80:1 (by atoms). In one flask, only phosphate (0.8 μM) was added, giving a N:P ratio of 16:1. In another flask, only nitrate (122 μM) was added,

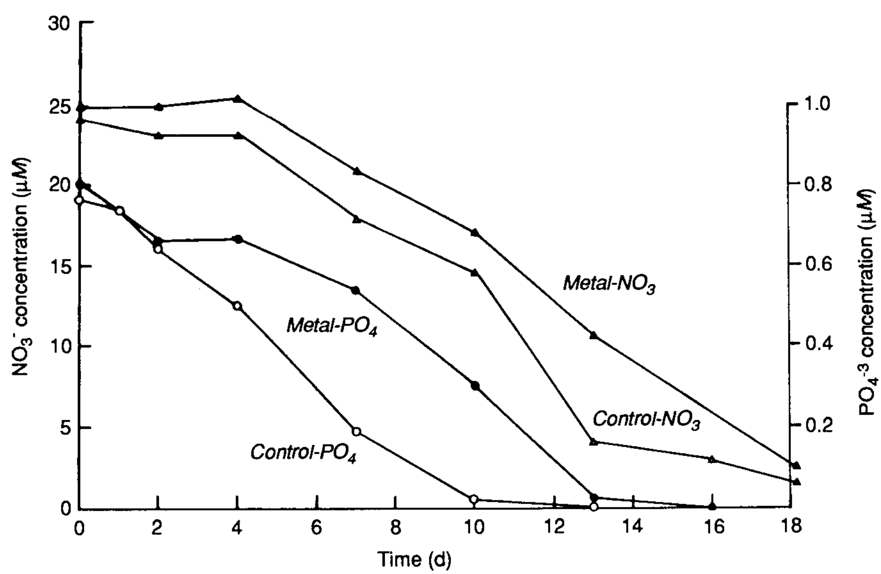

Fig. 1. Disappearance of nitrate and phosphate during a metal-addition experiment in mesocosms in Xiamen in 1985.

giving a N:P ratio of 656:1. In yet another flask, both N (122 μ*M*) and P (0.8 μ*M*) were added, giving a N:P ratio of 130:1. When N plus P and only P were added, fluorescence doubled compared with the control (no additions) (Fig. 2). When only N was added, fluorescence did not increase in over the control. Because only the addition of phosphate caused an increase in fluorescence, it was concluded that the water was phosphate-limited.

A plot of the seasonal variation in the N:P ratio in Xiamen Bay water and the Changjiang estuary (Yangtze River estuary near Shanghai) revealed that the ratio was high in the winter and spring (N:P = 30:1–40:1), decreased rapidly in late summer to less than 20:1, and then increased again in the fall (Fig. 3). It is important to note that all of these ratios are greater than 16:1, the Redfield ratio, except for one value recorded in July. The phosphate concentration decreased steadily from winter to spring, reaching a minimum in May (Fig. 3). In contrast, nitrate remained constant during the winter (Fig. 3). Reduced light due to the heavy suspended load in the water may affect nitrate uptake more than phosphate uptake during the winter.

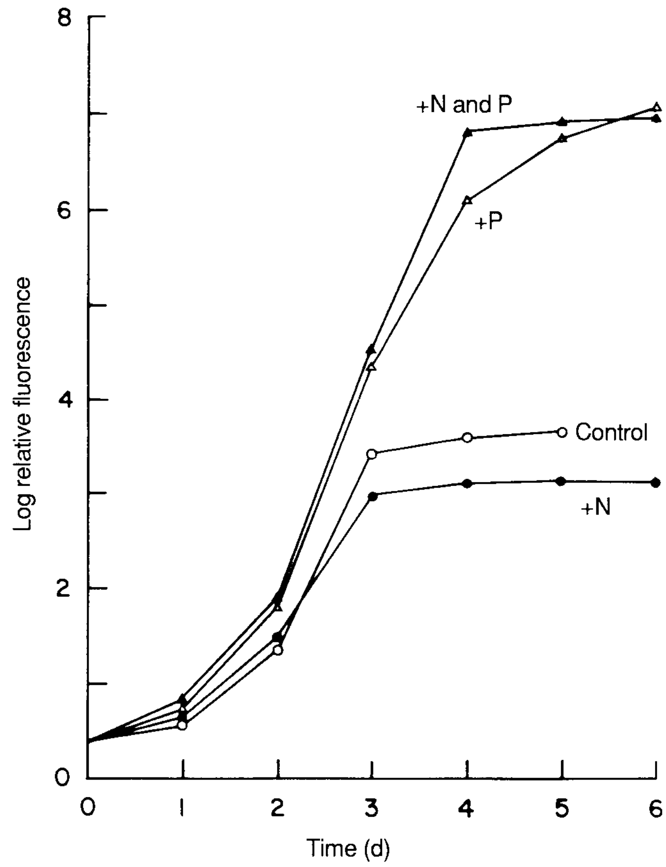

Fig. 2. Bioassay experiment in which water collected from Xiamen Bay in March 1986 was assayed for N or P limitation using a diatom, *Chaetoceros calcitrans*, as an assay organism. The control represents no nutrient additions, whereas +N = 122 μ*M* and +P = 0.8 μ*M*.

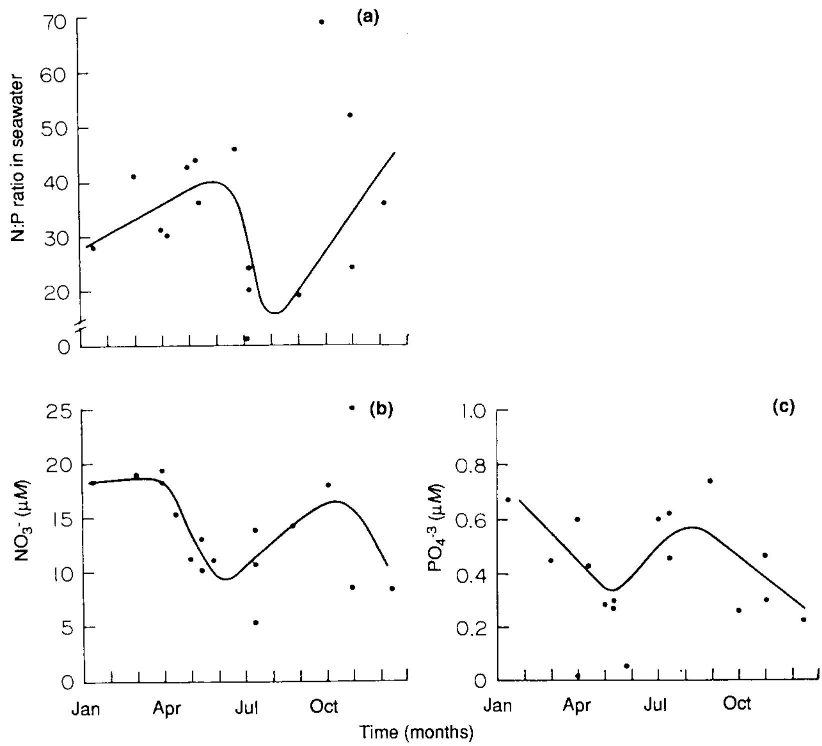

Fig. 3. (**a**) Seasonal variation in N:P ratios (by atoms) in seawater (nitrate + phosphate) from Xiamen Bay and Changjiang estuary; (**b**) Nitrate concentrations; and (**c**) Phosphate concentrations.

Why is phosphorus limiting in Chinese coastal waters?

Preliminary observations on nutrient concentrations are available for two estuaries in China. The Changjiang estuary shows a steep, decreasing gradient in nitrate, phosphate, and silicate concentrations as one proceeds from the river, through the estuary, and out to the open sea (Edmond et al. 1985; Shi et al. 1986; Sun et al. 1986). For June 1980, over the salinity range 0–32‰, the ranges of nitrate, phosphate, silicate, and ammonium concentrations were, 65–15, 0.7–0.0, 105–10, and 2–2 μ*M*, respectively (Edmond et al. 1985). The N:P ratio over the salinity range of 0-32‰ was about 90:1–150:1. In November, nitrate and phosphate in the river were 45 and 0.3 μ*M*, respectively, yielding a N:P ratio of about 140:1. In the summer, vigorous turbulence in the main channel of the inner estuary maintains high concentrations of suspended material (greater than 1 000 $mg{\cdot}kg^{-1}$; Milliman et al. 1985) in the surface layers, which suppresses biological activity. Phytoplankton blooms occur only on the inner shelf at salinities greater than 20‰.

Edmond et al. (1985) concluded that phosphate rather than nitrate was the limiting nutrient for biological productivity on the shelf, the reverse of the normal situation. They also concluded that the unusually high nitrate concentrations in the river were derived from agricultural sources. The yearly input of nitrate per square kilometre of drainage area for the Changjiang River is twice as much, whereas phosphate is half as much, as that of the Amazon River (Edmond et al. 1985). Measurements of nitrate farther offshore from the estuary reveal that the influence of nitrate input from the river extends out to the middle of the East China Sea (i.e., >500 km) (Shi et al. 1986). Beyond the estuary, primary productivity is high, ranging from 20–2 240 mg $C \cdot m^{-3}$ (Chai 1986).

The Huanghe (Yellow) River estuary in the southwest portion of the Bohai Sea was studied in July 1984 and in May and late August 1985 (Lu et al. 1985; Chen et al. 1986; Turner et al. 1987). Again, nitrate, phosphate, and silicate concentrations decrease dramatically as one moves from the river to the offshore region. The ranges in concentrations were as follows: nitrate, 140–1; phosphate, 1.0–<0.05; and silicate, 170–5 μM. The nitrate:phosphate ratios are shown in Fig. 4 as a steep gradient in contour lines ranging from >80:1 in the estuary to 10:1 in the Bohai Sea (Lu et al. 1985; Zou et al. 1985). Evidence indicating that nitrogen is most probably the limiting nutrient in areas beyond the estuary comes from the observation that cultivation of the seaweed *Laminaria japonica* is enhanced by adding nitrate rather than phosphate in this area (Tseng et al. 1955).

Moving offshore to the east, toward Taiwan, N:P ratios are generally less than 10:1 with nitrate values generally <5 μM and phosphate about 0.6 μM (Hung and

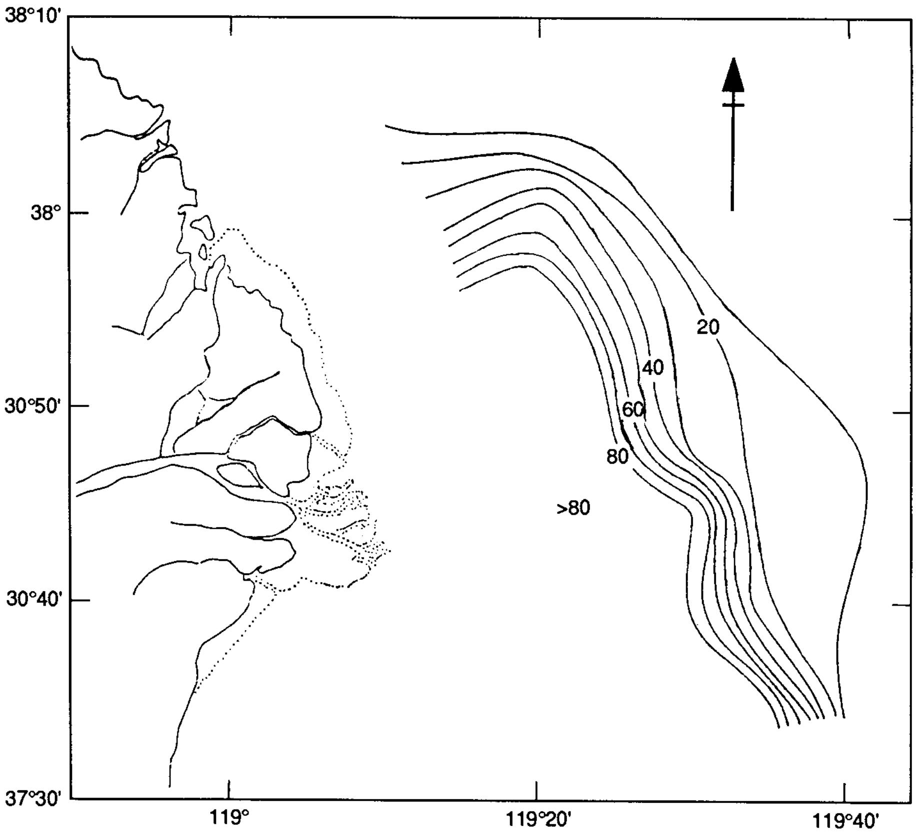

Fig. 4. Contours of nitrate:phosphate ratios (by atoms) in surface waters of the Huanghe River estuary (from Lu et al. 1985).

Tsai 1980; Hung et al. 1982, 1986). Similarly, off Hong Kong, N:P ratios were also <10:1, indicating that nitrogen was most likely limiting phytoplankton growth (Wear et al. 1984; Chiu et al. 1985).

Therefore, two of China's large rivers, the Changjiang and the Huanghe, appear to act like giant nutrient pumps, delivering excess nitrogen, relative to phosphorus, to coastal waters. At the outer edge of the estuary, therefore, phytoplankton growth is potentially limited by phosphorus, whereas well beyond the influence of the nutrients from the river (often >500 km offshore) nitrogen is limiting (the normal situation). In the inner part of the estuary, a high suspended load results in very turbid waters; therefore, primary productivity is light-limited and nutrients are never exhausted.

North of Xiamen Bay, where the mesocosm experiments were conducted, the Minjiang River (another large river in China) forms another large estuary. Nutrient distribution in the estuary has not been studied, but current measurements suggest that current flow from this area is often south along the coast. Therefore, nutrient input from this river could conceivably affect the Xiamen Bay area and could account for high nitrate relative to phosphate concentrations and the high N:P ratios (Fig. 3).

Similar observations have been made in the North Sea where polluted rivers have high N:P ratios (28:1–63:1); this is in contrast to unpolluted rivers where N:P ratios range from 8:1 (Amazon and Zaire rivers) to 21:1 (Niger River) (Wollast 1983). The nutrient-rich river water originating from the Scheldt, Meuse, and Rhine rivers mixes with seawater in such a way that 50% of the river discharge is transported in a 15-km wide strip along the Dutch and Belgian coasts. Therefore, the effects of nutrient input from rivers are often observed some distance from the estuary. At the edge of this 15-km strip, phosphate and silicate may limit phytoplankton growth in the spring.

Another explanation for phosphorus being limited in estuarine coastal waters is that phosphate may adsorb to suspended particles and sink to the sediments, carrying phosphate with it. This has been observed in the Changjiang estuary (Edmond et al. 1985) and in other large, turbid rivers, such as the Amazon River (Chase and Sayles 1980).

It has also been suggested that Chinese farmers use less phosphorus fertilizer because it is expensive and that agricultural land along the coast is low in phosphorus. This would help explain the low amount of phosphorus relative to nitrogen in the runoff.

Cyanobacteria or blue–green algae may be abundant in these waters. If they fix N_2, this would result in the depletion of phosphate. Dense blooms of *Trichodesmium* (*Oscillatoria*) have been reported in the Kuroshio current, but not in the East China Sea (Nishimura 1983).

Phosphorus limitation in estuarine areas along the coast of China is not totally unique (Smith 1984). Some estuaries along the east coast of the United States notably upper Chesapeake Bay, Hudson River, and Apalachicola Bay (Florida), are phosphorus-limited (Taft and Taylor 1975; Myers and Iverson 1981; Boynton et al. 1982). In Australia, the Peel–Harvey estuaries are phosphorus-limited in the winter and nitrogen-limited in the summer (McComb et al. 1981). Other larger areas, such as the Mediterranean Sea, have also been suggested as being phosphorus-limited (Berland et al. 1980; Maestrini and Kossut 1981; Azov 1986).

Phosphorus-limited growth of the macrophytes *Sargassum* spp and *Gracilaria tikvahiae* has been reported in the western North Atlantic off Florida (Lapointe 1986, 1987). Another macrophyte, *Macrocystis pyrifera*, has been reported as showing phosphorus-limited growth in the winter off California (Manley and North 1984).

The discharge of phosphorus and nitrogen transported by the Rhine River has increased sixfold since 1930. This increase in nitrogen and phosphorus and the essentially constant discharge of silicate has resulted in diatoms being silicate-limited, whereas flagellates are phosphorus-limited (Fransz and Verhagen 1985).

During blooms in Norwegian waters, phytoplankton communities are phosphorus-limited in fresh and brackish waters, and balanced or nitrogen-limited in marine waters. This results from the high N:P ratio in fresh or brackish water (<100:1) relative to the lower N:P ratio (12:1–16:1) in marine waters (Sakshaug and Olsen 1986).

Other estuaries in the United States are clearly nitrogen-limited, with N:P values <10:1 (Nixon 1981; Boynton et al. 1982; Pilson 1985). In these estuaries, dentrification has been reported to be a very important process where up to 50% of the nitrate may be lost to the atmosphere as N_2 (Pilson 1985; Smith et al. 1985).

Phosphorus versus nitrogen limitation — does it matter?

In most temperate and tropical areas, phytoplankton growth frequently becomes either phosphorus-limited or nitrogen-limited during part of the year. Therefore, it seems logical that cells that are already naturally stressed by nutrient limitation may be more sensitive to a pollutant (secondary stress) than cells that are healthy. It has been shown by Cloutier-Mantha and Harrison (1980) that ammonium-limited *Skeletonema* exposed to Hg had a higher Ks value for ammonium uptake and the cells could not deplete ammonium in the medium below 1 μM. Normal cells can take ammonium down to undetectable levels.

The question of whether phosphorus-limited cells are more sensitive to pollutants than nitrogen-limited cells has not been well studied. Only one study has attempted to examine this question and Karydis (1981) found that phosphorus-starved cells of *Skeletonema* were more sensitive to oil pollution than nitrogen-starved cells. This interesting question certainly deserves further study.

During normal growth, many phytoplankton store phosphorus intracellularly in polyphosphate granules (Jensen et al. 1986; Sicko-Goad and Lazinsky 1986). Recent studies have shown that heavy metals, such as Pb, are sequestered by these granules. During phosphorus limitation, the polyphosphate granules are degraded and the sequestered heavy metals are liberated internally. This may have adverse effects on cells that are already nutrient stressed. Observations such as this may help explain why phosphorus-limited cells appear to be more sensitive to pollutants than nitrogen-limited cells.

The second consequence of whether the ecosystem is phosphorus-limited or nitrogen-limited involves potential species changes if eutrophication is controlled or altered in the future. For example, a low N:P ratio (i.e., nitrogen limitation) is known to favour cyanobacteria, whereas phosphorus limitation may favour diatoms (Smith 1983). Therefore, if the N:P ratio in the runoff and sewage was altered in the future and the N:P ratio decreased, China's coastal waters would tend toward nitrogen limitation with potential shifts in the dominant species of phytoplankton. These species shifts could affect higher trophic levels.

Finally, most models of marine ecosystems are nitrogen based (Parsons and Kessler 1986b) or carbon based. For estuarine areas along the coast of China, nitrogen-based models, such as the one used to simulate mesocosm experiments in Canada (Parsons and Kessler 1986a), must be changed to a phosphorus-based model.

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