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RED TIDE IN GREEK WATERS

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MARÉE ROUGE
ÉLÉMENTS NUTRITIFS
GYMNODINIUM
GRÈCE

RÉSUMÉ. — Durant l'été 1978 une marée rouge entraînant une mortalité de Poissons se déclara en quelques points de la côte orientale du Golfe pollué du Saronikos (Grèce). Elle est due au Dinoflagellé *Gymnodinium breve*, dont la densité cellulaire croît près de la côte, se traduisant par une augmentation au niveau de la demande chimique en oxygène (DCO). Dans les premiers stades, le taux de nitrate atteint des valeurs élevées qui tendent à diminuer vers la pleine mer. Ensuite, il décroît considérablement et inverse cette tendance. Les variations des concentrations de phosphate et de silicate suivent celles du DCO, diminuant vers la mer pendant toute la durée du « bloom ». Seule, la teneur en ammonium ne présente pas de profil distinct. Il semble que la prolifération des organismes résulte de l'eutrophisation dans un environnement riche en phosphate et quelque peu toxique. Le nitrate, pratiquement absent près du rivage vers la fin du phénomène, semble constituer le principal facteur limitant. L'excès d'éléments nutritifs provient en grande partie de déchets déversés par un ruisseau voisin. Il est retenu aux alentours en raison de la configuration de la côte et de courants favorables.

RED TIDE
NUTRIENTS
GYMNODINIUM
GREECE

ABSTRACT. — During the summer of 1978, a red tide involving fish mortality broke out at some locations on the eastern coast of the polluted Greek Gulf of Saronikos. It was caused by the Dinoflagellate *Gymnodinium breve*, the cell density of which increased near the land and was reflected in the level of the chemical oxygen demand (COD). In the early stages, the amount of nitrate reached high values, tending to decline towards the open sea. Later on, it fell dramatically and reversed the trend. The phosphate and the silicate concentrations followed that of the COD, diminishing seaward throughout the bloom. Only the quantity of ammonium did not exhibit any distinct pattern. It seems that the proliferation of the organisms resulted from eutrophication in a phosphate-rich and somewhat toxic environment. The nitrate, nearly absent inshore towards the end of the phenomenon appeared to be the main limiting factor. The excess nutrient originated in great part from waste discharged from a neighbouring stream. It remained trapped in the vicinity because of the configuration of the coast and favourable currents.

INTRODUCTION

Red tide or red sea-water bloom formation has been known for a very long time. However, it was not described before 1832, when Darwin undertook his « Beagle » voyage. It results from an excessive production of phytoplankton organisms, most commonly dinoflagellates, but not always (Lassus *et al.*, 1980). Some conditions frequently accompany it (Ryther, 1955; Jacques et Sournia, 1978-1979) : calm,

sunny weather, rather elevated sea temperatures and, fairly often, enhanced nutrient levels close to large cities.

The attention of the authors was drawn towards such a signal event, rare in Greece, in March 1978, when port authorities reported the appearance of an orange-coloured patch off the harbour of Kavalla, in the northern Aegean Sea. Only a few months later, in August 1978, another fish-killing, red brown phytoplankton bloom occurred in the Gulf of Saronikos (Fig. 1), giving them an opportunity to study

it at close quarters. Its location (Fig. 2) is directly exposed to pollution, with the nutrient levels tending to increase over the years (Friligos, 1981, 1982). It comprises a series of natural and artificial coves.

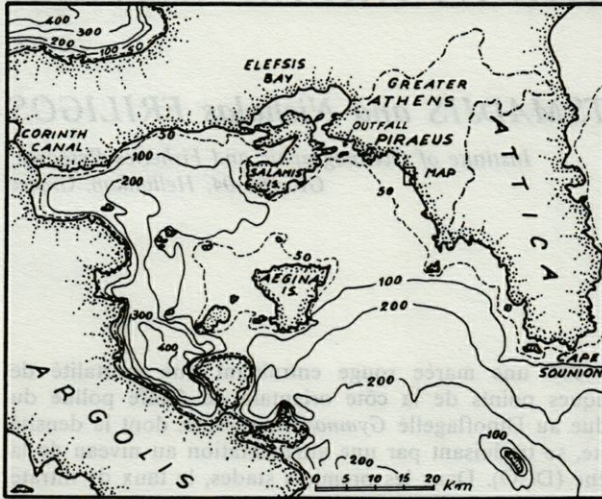


Fig. 1. — Bathymetric map of the Saronikos Gulf.

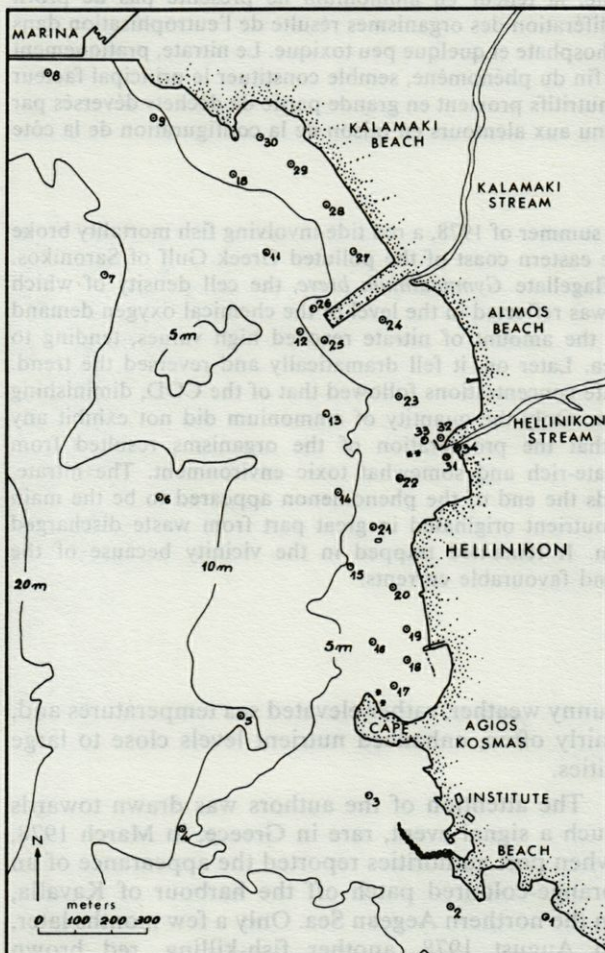


Fig. 2. — Sampling stations during the September 12th 1978 cruise.

Two brooks run into it, but they are dry most of the time. Its largest, northern part forms an open bay, with water mass dynamics considerably smaller than in other sections of the Gulf (Coachman and Hopkins, 1975). Its southern appendage, protected by its diminutive promontory, has even weaker currents. During summer, the weather is very dry, sunny, hot and with winds of moderate velocity, blowing predominantly from a NNE direction (Table I A, made from data published by the National Meteorological Service in Athens).

MATERIALS AND METHODS

Surface water samples were taken, on August 31st, near the Alimos Beach, on September 4th, off the Institute, on September 8th, by the Agios Kosmas paying Beach and, on September 12th, over the whole area, during a three-hour cruise, while a south-west wind blew with a velocity of just 0.7 to 2 m/s. The sea water was collected in 100 ml plastic bottles. Those for the determination of dissolved nutrients received immediately on filling one drop of 1% mercuric chloride solution as preservative. In the laboratory, they were stored in deep freeze. Colorimetric methods were used: Murphy and Riley's (1962) for reactive phosphorus, that of Armstrong *et al.* (1967) for nitrite, nitrate and silicate and Slawyk and MacIsaac's (1972) for ammonium. Processing took place on the Technicon CSM₆ Autoanalyser and the mathematical treatment of the data was carried out according to Satsmadjis (1978). The determinations of dissolved total phosphorus (TP) and COD were performed in conformity with the FAO manual. The samples were also examined for the identification of the main dinoflagellate species.

RESULTS

Table I A indicates that the weather conditions prior to the outbreak were normal for the season. Table I B establishes that, a few days after the manifestation of the red tide, the sea off Alimos Beach held large amounts of phosphate, inorganic nitrogen and organic matter, with all the quoted parameters bar the silicate exhibiting a close dependancy. Table I C affords a similar picture. On the contrary, Table I D, although still displaying a good correlation between COD and all the nutrients except silicate, proves that the sea on the edge of the Agios Kosmas Beach was much cleaner, with the nitrate even well below normal. Table II shows that, save in the area directly influenced by the Hellinikon stream, the amount of organic matter, generally quite large, depended on both the eastward distance

Table I. — A, Meteorological features ; B, Nutrients in $\mu\text{g-at/l}$ and COD in mg/l in water samples collected off Alimos Beach on 31.8.78 ; C, Nutrients in $\mu\text{g-at/l}$ and COD in mg/l in water samples collected off the Institute on 4-9-78 ; D, Nutrients in $\mu\text{g-at/l}$ and COD in mg/l in water samples collected off Agios Kosmas Beach on 8.9.78.

IA		Mean air temperature °C			Mean wind velocity m/sec			Mean precipitation mm		
Year	July	August	Sept.	July	August	Sept.	July	August	Sept.	
1901 -										
1950	27.2	27.0	23.4	3.9	3.8	3.4	6.2	8.7	15.4	
1976	25.8	24.2	22.2	1.8	1.6	1.1	0.1	17.2	13.5	
1977	27.3	27.3	21.9	2.0	2.5	3.5	0.0	0.0	20.4	
1978	27.2	25.7	21.4	3.1	1.8	1.4	0.0	0.0	47.3	
1979	26.5	26.3	23.6	1.7	1.9	1.7	1.6	0.5	1.1	
1980	27.5	26.4	22.7	1.4	2.4	1.6	0.0	2.1	0.4	

Sample	Distance m	COD	NH ₄ -N	NO ₂ -N	NO ₃ -N	PO ₄ -P	TP	SiO ₄ -Si
IB								
1a	1	33.2	2.30	0.55	12.30	6.30	11.40	7.70
2a	50	32.6	1.60	0.40	10.00	5.60	9.30	2.00
3a	200	24.3	0.80	0.10	2.80	0.35	1.90	0.95
4a	30	33.8	1.30	0.35	9.60	5.20	8.20	3.40
5a	100	20.5	0.80	0.15	4.20	1.50	3.30	3.40
6a	1	32.6	1.50	0.10	9.30	7.90	12.50	4.60
7a	1	33.2	1.90	0.45	10.80	6.10	9.70	7.00
8a	1	41.4	2.80	0.65	13.50	9.00	13.80	3.80
9a	1	32.5	2.00	0.35	9.80	5.10	7.90	4.20
mean	40	31.6	1.70	0.34	9.10	5.20	8.70	4.10
IC								
1b	1	33.6	1.85	0.60	11.80	10.00	16.80	7.10
2b	100	6.4	1.10	0.30	3.20	5.00	8.50	3.40
3b	200	14.4	0.90	0.45	7.10	4.50	7.70	5.80
4b	200	4.8	0.85	0.20	2.50	3.10	6.00	3.00
5b	1	36.8	1.65	0.60	13.90	9.00	13.20	6.00
mean	100	19.2	1.27	0.43	7.70	6.30	10.40	5.10
normal	1	0.8	0.90	0.20	2.40	0.20	1.00	2.50
ID								
1c	1	8.0	0.60	0.10	0.25	0.85	2.80	3.90
2c	1	9.6	0.60	0.10	0.15	1.10	3.20	6.30
3c	1	8.0	0.60	0.20	0.25	1.10	2.80	7.20
4c	1	9.6	0.55	0.10	0.10	0.80	2.90	2.20
5c	1	6.4	0.60	0.15	0.30	1.10	2.40	4.40
6c	1	17.6	0.85	0.30	1.15	1.30	4.20	5.50
mean	1	9.9	0.63	0.16	0.37	1.04	3.00	4.90

from the land and the position, rising inshore and northwards. The nitrate content, in contrast remarkably low, presented the opposite trend. The other dissolved nutrients had about the same concentrations as those existing previously around the corresponding spots, but their relationships with COD had weakened. Figures 3 A, B, C, D and 4 A, B, C depict the values of the various parameters during the September 12th 1978 cruise.

The microscopical examination of the samples implicated *Gymnodinium breve* as the only species responsible for the red tide, with concentrations of around 10^7 cells per litre.

DISCUSSION

Though red tide had never appeared to any marked extent in the Saronikos Gulf prior to 1978,

Table II. — Nutrients in $\mu\text{g-at/l}$ and COD in mg/l in water samples collected during the 12.9.78 cruise.

Sample	Distance	COD	NH ₄ -N	NO ₂ -N	NO ₃ -N	PO ₄ -P	TP	SiO ₄ -Si
1	50	1.4	0.95	0.10	0.85	0.50	1.80	2.90
2	280	0.7	0.85	0.10	0.85	0.35	1.60	2.00
3	200	0.2	0.90	0.05	0.90	0.75	2.10	1.90
4	740	0.2	0.45	0.05	0.90	0.10	1.30	1.00
5	320	0.6	0.75	0.05	0.85	0.20	1.90	1.20
6	850	0.1	1.85	0.15	2.35	0.30	1.60	1.10
7	800	0.6	0.85	0.05	0.90	0.15	1.30	0.80
8	250	3.0	0.75	0.05	0.60	0.75	1.90	1.30
9	180	4.6	0.60	0.05	0.40	0.90	2.40	1.50
10	280	4.2	0.75	0.10	0.70	1.10	3.00	2.20
11	340	23.2	0.90	0.05	0.85	0.20	1.50	0.90
12	440	6.4	0.85	0.05	0.90	0.45	1.60	1.50
13	420	15.2	0.40	0.05	0.35	0.70	1.60	1.80
14	360	10.4	0.50	0.05	0.30	0.35	1.20	1.30
15	180	10.4	0.60	0.10	0.25	0.40	1.00	1.60
16	150	2.4	0.50	0.05	0.10	0.60	2.40	1.80
17	140	13.3	0.50	0.05	0.05	1.05	2.30	2.30
18	120	8.8	0.40	0.05	0.05	0.85	2.00	2.20
19	50	10.4	0.40	0.05	0.05	0.50	2.40	1.80
20	80	24.0	0.50	0.05	0.05	0.95	3.20	2.30
21	120	16.0	0.60	0.05	0.10	1.15	3.70	3.20
22	180	26.4	1.10	0.40	0.35	4.00	5.70	7.20
23	220	29.6	1.00	0.10	0.25	4.00	8.50	3.50
24	200	12.8	0.50	0.10	0.05	1.10	2.90	3.70
25	380	15.2	0.60	0.10	0.10	1.00	2.50	2.70
26	250	16.8	0.65	0.05	0.10	0.85	3.00	2.70
27	100	28.0	0.85	0.05	0.05	2.25	6.50	4.10
28	80	17.6	1.10	0.10	0.25	1.05	3.40	3.00
29	100	14.4	0.75	0.05	0.05	0.55	2.80	3.00
30	120	15.2	0.75	0.05	0.05	0.65	2.90	3.00
31	20	12.0	11.20	2.20	11.20	3.00	5.60	13.90
32	1	16.8	0.65	0.05	6.40	0.75	2.70	5.50
33	40	19.2	0.65	0.10	0.40	1.00	2.90	3.70
34	0	15.2	4.80	0.55	13.10	1.10	1.40	17.60

its advent had seemed likely. Thus, Friligos (1974) indicated the possibility there of monoculture phytoplankton bloom. Also, from the work carried out by Coachman *et al* (1976), as well as Dugdale and Hopkins (1978), the area between Aegina island and Piraeus harbour looked susceptible to dinoflagellate bloom. Furthermore, Moraitou-Apostolopoulou and Ignatiades (1980) reported *Gymnodinium breve* as a dominant phytoplankton species in the Gulf in May and June 1978, together with « *Exuviaella baltica* » and *Prorocentrum micans*. Ioannou (personal communication) has been observing the presence of *Gymnodinium breve* in the region since 1972. Nümann (1955-56) and Agara and Nalbandoglu (1960) also blamed the *Gymnodinium* group for the red tide in the Gulf of Izmir and suspected the phosphate in the domestic wastes of the city as the prime factor. Maretic *et al* (1978) offered the same explanation for the phenomenon, which occurred several times in ten years around the harbour of Pula in Yugoslavia. The involvement of phosphate is no surmise in the case of the *Noctiluca scintillans* bloom (10^5 cells/l) that appeared in March 1978 near the port of Kavalla, since it followed the wreck of a ship loaded with superphosphates. As the organism does not hold any chlorophyll, the sullied environment must have at first favoured the rapid multiplication of one or more photosynthetic phytoplankton species, which were feasted upon later. Anonymous (1971) and Lopez and Arte (1971, 1972) quoted a similar (0.38×10^5 cells/l) *Noctiluca scintillans* cell concentration during a bloom in 1971 on the north west coast

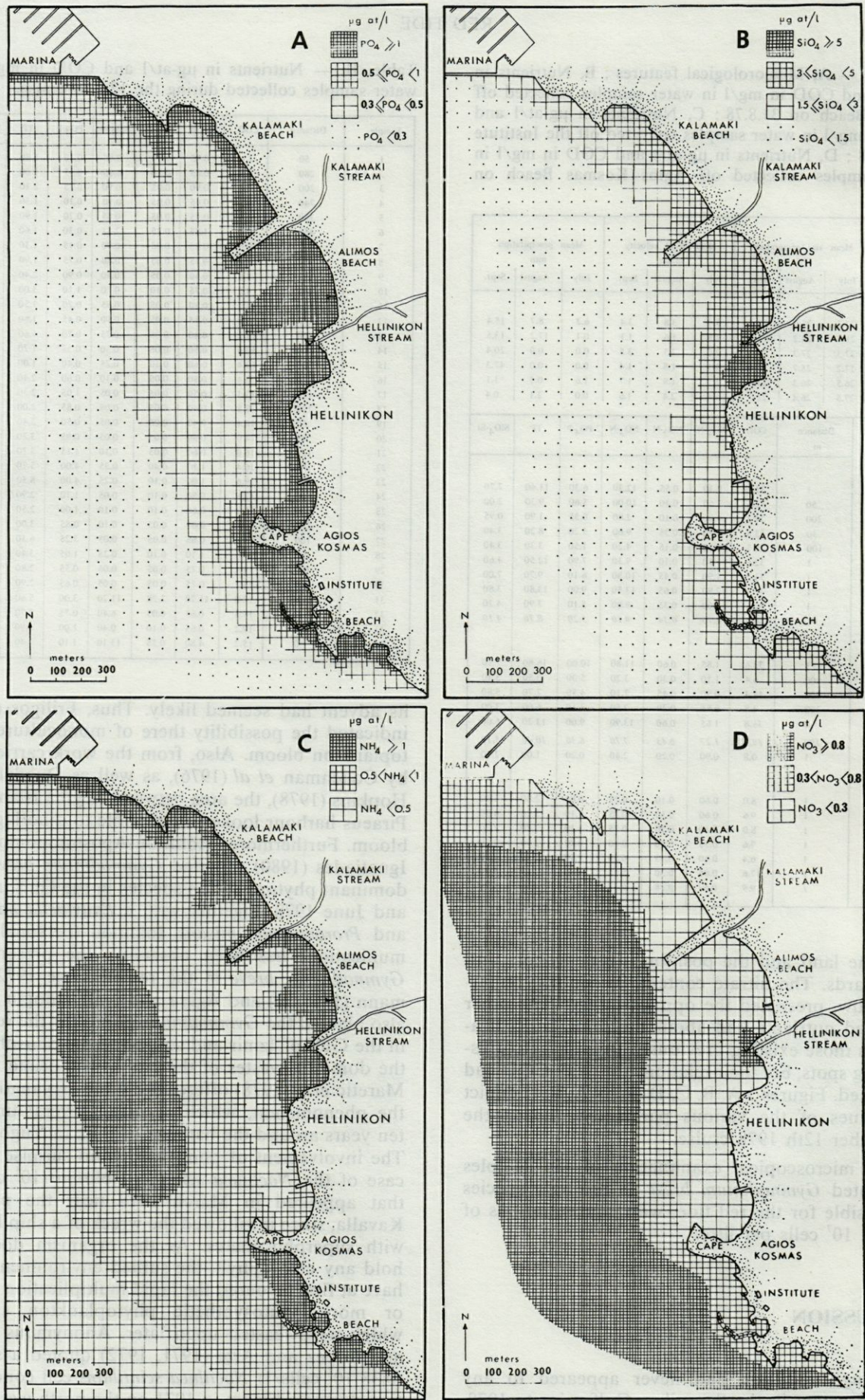


Fig. 3. — A, Distribution of the phosphate ; B, distribution of the silicate ; C, distribution of the ammonium ; D, distribution of the nitrate.

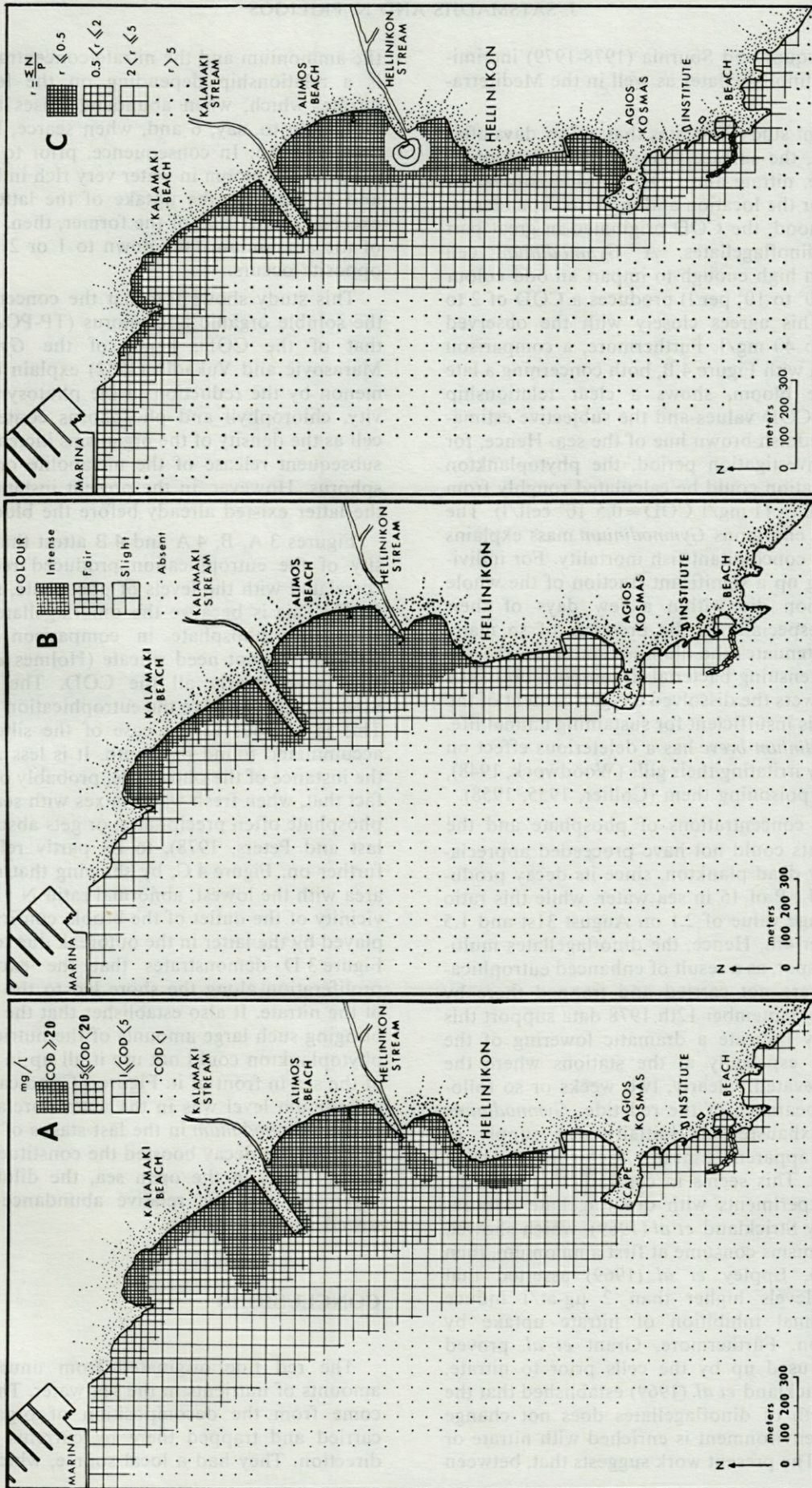


Fig. 4. — A, Distribution of the chemical oxygen demand (COD) ; B, subjective colour assessment ; C, distribution of the $\Sigma N : P$ ratio.

of Spain. Jacques and Sournia (1978-1979) incriminated other dinoflagellates as well in the Mediterranean.

The present study indicates that, a few days after the outbreak, the surface water contained amounts of phosphate, nitrate and COD considerably larger than usual for the location and in close dependency. In all likelihood, the COD originated in great part from the dinoflagellates. A *Gymnodinium* cell concentration high enough to impart an odd colour to the sea (10^6 to 10^8 per l) produces a COD of 2 to 200 mg/l. This agrees closely with the observed range of 4 to 40 mg/l. Furthermore, a comparison of Figure 4 A with Figure 4 B, both concerning a late stage of the bloom, shows a clear relationship between the COD values and the subjective estimation of the reddish brown hue of the sea. Hence, for the whole investigation period, the phytoplankton cell concentration could be calculated roughly from the COD values ($1 \text{ mg/l COD} = 0.5 \cdot 10^6 \text{ cell/l}$). The thus inferred enormous *Gymnodinium* mass explains the observed concomitant fish mortality. For individuals making up a significant fraction of the whole live population die within a few days of their generation, especially as the presence of so many organisms attenuates the illumination below the sea surface. The ensuing bacterial decomposition of the dead cells lowers the dissolved oxygen content of the water to levels insufficient for sustaining animal life. Also *Gymnodinium breve* has a deleterious effect on fish, either by irritating their gills (Woodcock, 1948), or, possibly, poisoning them (Collier, 1955, 1958).

The huge concentrations of phosphate and the other nutrients could not have proceeded appreciably from any dead plankton, since its decay produces a ratio N : P of 15 in sea water, while this ratio had an average value of 2.1 on August 31st and 1.5 on September 4th. Hence, the dinoflagellates multiplied on the spot, as a result of enhanced eutrophication; they were not carried and trapped there by currents. The September 12th 1978 data support this finding. They indicate a dramatic lowering of the nitrate level, especially at the stations where the COD was elevated. Clearly, two weeks or so following the appearance of the red tide, *Gymnodinium breve* had exhausted its initially rich supply of nitrate, the apparent limiting factor in its swift reproduction. This seems in contradiction with the results of experiments with dinoflagellate cultures performed by Strickland *et al* (1969), which showed that the organisms consume at first ammonium, then nitrate. Also, Eppley *et al* (1969) asserted that ammonium levels higher than $2 \mu\text{g-at/l}$ induce almost maximal inhibition of nitrate uptake by phytoplankton. Furthermore, Grant *et al.* proved that urea is used up by the cells prior to nitrate. However, Strickland *et al.* (1969) established that the rate of growth of dinoflagellates does not change whether the environment is enriched with nitrate or ammonium. The present work suggests that, between

the ammonium and the nitrate concentrations, there is a relationship depending on the level of the nitrate, which, when abundant, raises the NO_3 to NH_4 ratio to, say, 6 and, when scarce, lowers it to only, say, 0.5. In consequence, prior to the appearance of the bloom in water very rich in both nitrate and ammonium, the uptake of the latter probably exceeds greatly that of the former, then, as the level of the ammonium falls down to 1 or $2 \mu\text{g-at/l}$, the opposite occurs.

This study shows too that the concentrations of the soluble organic phosphorus (TP- PO_4) rose with that of the COD, hence of the *Gymnodinium*. Marasovic and Vukadin (1982) explain this phenomenon by the reduction in the photosynthetic activity, chlorophyll and phosphorus content of each cell as the density of the organisms increases and the subsequent release of the metabolite organic phosphorus. However, in the present instance, most of the latter existed already before the bloom.

Figures 3 A, B, 4 A and 4 B attest that the intensity of the eutrophication produced red tide was correlated with the levels of phosphate, silicate and COD. This is because the dinoflagellates consume very little phosphate in comparison with total nitrogen, do not need silicate (Holmes *et al.*, 1967) and cause nearly all the COD. The Hellinikon Stream contributed to the eutrophication of the area. This is obvious in the case of the silicate, which accumulated round its mouth. It is less apparent in the instance of the phosphate, probably owing to the fact that, when fresh water mixes with sea water, the phosphate often precipitates, or gets absorbed (Wollast and Peters, 1978), to be partly released later further on. Figure 4 C, by showing that most of the area with the lowest, abnormal ratio N : P lay in the vicinity of the outlet of the brook, confirms the part played by the latter in the outbreak due to pollution. Figure 3 D demonstrates that the excessive cell proliferation along the shore led to the exhaustion of the nitrate. It also establishes that the runnel was bringing such large amounts of the nutrient that the phytoplankton could not use it all up in the section of the sea in front of it. Figure 3 C indicates that the ammonium level was in the main unrelated to that of the *Gymnodinium* in the last stages of the bloom. It seems that decay boosted the constituent near the shore, while, in the open sea, the dilution of the organisms and the relative abundance of nitrate sowed its consumption.

CONCLUSIONS

The red tide originated from unusually large amounts of nutrients in the sea water. These did not come from the decomposition of organic matter carried and trapped there by currents of suitable direction. They had a local source, which added to

the load of the surrounding heavily polluted area, some spots of which presented also a bloom later on. The reason for the *Gymnodinium breve* monoculture could be the excessive concentration of phosphate from waste and the presence of toxic substances. The weather conditions (high temperature, strong insolation, negligible rain, moderate wind), though perfectly normal for the season, favoured the multiplication of the species. The organisms grew more numerous northward and inshore because the prevailing current tended to move to the north the nutrients poured out by the pollution source. Part of them dispersed into the open sea. The rest went to the coves, accumulating near the land, in places with minimal outgoing water velocity. Hence the degree of proliferation of the dinoflagellates depended upon the distance from the shore, the position, orientation and configuration of the coast, the presence of capes, breakers and piers.

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