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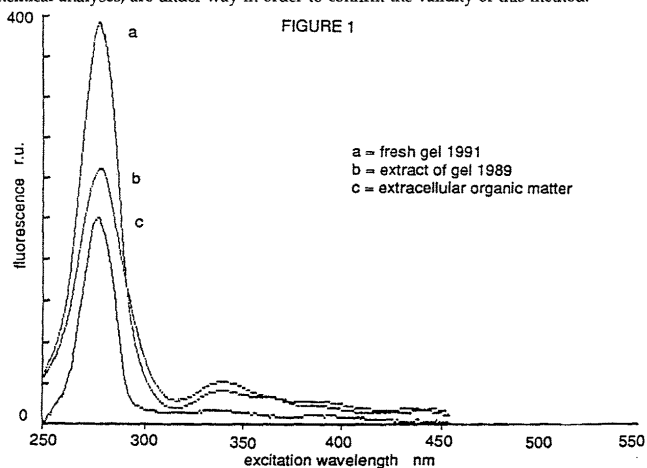
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The phenomenon of the appearance of massive quantities of gel aggregates recently affected the Northern Adriatic Sea in the summer seasons 1988-89-91, and covered a surface water area estimated at over 10,000 km<sup>2</sup>. Such gel formation is attributed to the hyperproduction of extracellular algal exudates (MARCHETTI *et al.*, 1989). The ability of phytoplankton to produce gel-forming material is genetically determined, but its formation in large amounts depends on the physiological state of the cells (FOGG, 1990). The increase in the production of organic extracellular material by planktonic marine diatoms is attributed by some authors to nutritional stress, such as an unbalanced N/P ratio (MYKLESTAD, 1989). However, the environmental factors which regulate the release of these substances have not yet been clearly defined. A need for further investigation aimed at a better understanding of the mechanism of potentially gel-forming exudation is generally recognized. With this aim, in the present study, the use of fluorimetric techniques for the analysis of both the gel material taken in the field and the extracellular substances produced in laboratory cultures was considered. The technique chosen was the synchronous fluorescence spectroscopy, which involves the stepping of both the excitation and emission monochromators of the spectrofluorimeter at the same time, while monitoring the emission, keeping a constant wavelength difference between the monochromators (LLOYD, 1971). This approach, compared to conventional spectrofluorimetry, allows the narrowing of spectral bands and for this reason it is appropriately employed for the analysis of multicomponent mixtures containing fluorescent compounds, and in particular for the determination of dissolved organic matter (VO-DINH, 1978; VODACEK, 1989).

An aliquot of the gel material taken in July 1991 in surface waters offshore from Cesenatico was analyzed following filtration of the sample (0.22 µm). At the same time, a gel sample taken in July 1989 from the same area, using the dried mass on the extracts of which chemical analyses had already been undertaken (MARCHETTI *et al.*, 1989) was also analyzed. This mass was in turn extracted with distilled water for fluorimetric analysis following filtration. The synchronous ex-em fluorescence spectra of both samples are given in Fig.1, and show an evident similarity of the peaks around the wavelength of about 280 nm. Batch laboratory cultures of *Skeletonema costatum*, a marine diatom from the Northern Adriatic coastal waters, were gently filtered and the analyzed suspension medium showed fluorescence in the same region (Fig.1). Furthermore, the spectral responses of various carbohydrate solutions fell precisely in the same region. These results are in agreement with the chemical characterization of both gel (MARCHETTI *et al.*, 1989) and algal exudates (MYKLESTAD, 1989). Although the present study does not take into consideration the individuation of the single exudated compounds, which are species or strain specific (FOGG, 1990), the mentioned ex-em spectra reveal the presence of organic extracellular substances potentially capable of forming gels, in dissolved state. Thus, in order to verify the application to the study of the production trend in laboratory algal assays, aliquots of *S.costatum* cultures were taken at regular intervals along the algal growth curves for microscopic cell counts and fluorimetric analysis of the filtered medium. In replicated assays, results indicated an increase of organic matter dissolved in the medium during the log phase of the algal growth, with spectral characteristics identical to those in figure 1.

This series of preliminary verifications suggests that the synchronous fluorescence technique might be applied to study the factors which affect the production of extracellular organic substances in laboratory algal assays. A parallel field investigation monitoring the production trend is in progress. The proposed fluorimetric technique, new in the environmental studies, has important advantages: it is a simple method which can be used on a routine basis, without resorting to techniques which are more expensive and excessively time consuming (VO-DINH, 1978). Further investigations regarding both the quantitative and the analytical aspects, by parallel verifications with chemical analyses, are under way in order to confirm the validity of this method.



## REFERENCES

- FOGG G.E., 1990. - Water Pollution Research Report 16, EUR 12978. 207 p.  
 LLOYD J.B.F., 1971. - *Nature*, 231, p. 64.  
 MARCHETTI R., IACOMINI M., TORRI G., FOCHER B., 1989. - *Acqua-Aria*, 8, p. 883.  
 MYKLESTAD S., HOLM-HANSEN O., VARUM K.M., VOLCANI B.E., 1989. - *J. Plank. Res.*, 11, p. 763.  
 VODACEK A., 1989. - *Remote Sens. Environ.*, 30, p. 239.  
 VO-DINH T., 1978. - *Anal. Chem.*, 50, p. 396.

This work has been done during the 15th geochemical and geophysical Scientific Cruise (April-June 1984) aboard R/V "AKADEMIK PETROVSKI", and the participation of the author within the IOC/UNESCO programme "X.4 - Ocean and its resources". A total of 78 surface and bottom water samples were collected from the open mid-eastern Atlantic Ocean (35°-36°N and 12°-14°W), Ligurian, Tyrrhenian, Ionian and Maltese Seas (33°-44°N and 8°-28°E) with two stations in front of Genova and Monaco; Eight sampling stations from the Atlantic Ocean and twenty five stations from the Mediterranean Sea. The trace metals Cu, Zn, Pb, Cd, Ni, Co, Cr together with Mn, Fe, Li, Sr were determined in water samples.

Water samples were collected with pre-treated polycarbonate samplers. The sampling, processing and analysis were performed under strict clean conditions. The seawater samples were acidified with "suprapur" HNO<sub>3</sub> (1:1) and stored at 0°C in 0.5 - 2L acid-leached quartz bottles on board the ship, until measurement in onshore laboratories. No filtration of samples was performed because of the low amount of particles in open Mediterranean waters and due to the potential of contamination. A "Hitachi 180-70" double beam polarised Zeeman atomic absorption spectrophotometer was used to analyse the metals. All methods applied are described in detailed elsewhere (HANNA, 1985).

Details of the analytical results will be presented in the oral and/or the poster sessions at the congress, together with the exact location and the total depth of the sampling stations. This study showed that the concentration (µg L<sup>-1</sup>) ranges and the grand averages from surface and bottom of all stations (together with ± standard deviation) were 0.08-0.05 (0.063 ± 0.1), 0.06-0.18 (0.086 ± 0.02) for Mn; 26-45 (0.33 ± 0.6); 25-1.44 (42 ± 12) for Fe; 0.11-0.14 (0.124 ± 0.01), 0.11-0.14 (0.124 ± 0.01) for Li; 3.7-4.8 (4.1 ± 2), 3.8-4.8 (4 ± 1.1) for Sr; 0.05-0.17 (0.093 ± 0.05), 0.05-0.17 (0.093 ± 0.05), 0.05-0.17 (0.11 ± 0.03) for Cu; 0.05-0.15 (0.08 ± 0.015), 0.04-0.11 (0.12 ± 0.06) for Zn; 0.3-0.6 (0.49 ± 0.1), 0.4-0.6 (0.5 ± 0.05) for Pb; 0.03-0.09 (0.075 ± 0.01), 0.07-0.1 (0.087 ± 0.01) for Cd; 0.26-0.36 (0.3 ± 0.02), 0.2-0.7 (0.36 ± 0.03) for Ni; 0.6-0.9 (0.76 ± 0.1), 0.6-0.9 (0.8 ± 0.04) for Co and 0.01-0.06 (0.06 ± 0.01), 0.04-0.1 (0.07 ± 0.01) for Cr (the values for surface and bottom respectively for each element).

The elements concentration of the open Mediterranean sea water samples are close to oceanic results gathered under similar conditions (except for Cd and Pb). These results indicate that a serious metal pollution problem does not exist in the open Mediterranean sea water so far at that time.

The elements Li, Sr, Pb, Co, Cr had bottom/surface ratio of the concentration values one and Mn, Fe, Zn, Ni had up to 1.4 values and Cu the only element which had a ratio of 1.9. This study shows that the shape of the Mediterranean profiles differ from other findings in the open ocean (e.g. Pacific and Indian Oceans) by two main features. Firstly, no increase with depth, secondly, in average, much lower concentrations have been found in bottom waters of the Mediterranean than in Pacific or Indian Ocean bottom waters. (BOYLE *et al.*, 1975, 76; DANIELSSON, 1980; WESTERLUND OHMAN, 1991). I believe that these characteristics are mainly caused by the special hydrochemical conditions and biogeochemical processes prevailing in the Mediterranean Sea, (low nutrients and vertical mixing). These results indicate that little biological oxidation occurs in Mediterranean deep waters. Moreover, due to the low primary production, the processes of thermalhaline vertical convections (STOMMEL, 1972) and the relatively high turnover rate of the deep water do not allow enrichment comparable to those measured in Pacific waters. This assumption does not imply that a biologically mediated transport from surface waters does not occur for these elements in Mediterranean. I suggest, and are supported by other authors (KREMLING & PETERSEN, 1981) and model calculations (TOPPING, 1974), that the overall analytical precision has to be much better than 5% to identify biologically mediated variations of trace metals in Mediterranean waters.

The overall mean concentrations of all elements determined in this study fit in very well with data reported by other authors (UNEP, 1985) for the world oceans. This supports the conclusions already made by BERNHARD (1978), KREMLING and PETERSEN (1981) that trace metal concentrations in seawater of the open Mediterranean do not seem to be much different from other oceans. I do hope that my data serve as a useful help for establishing the actual baseline concentrations in the Mediterranean sea environment.

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## REFERENCES

- BERNHARD M., 1978. - Heavy Metals and chlorinated hydrocarbons in the Mediterranean. *Ocean Management* 3: pp. 253-313.  
 BOYLE E.A., SCLATER F.X. and EDMOND J.M., 1976. - On the Marine Geochemistry of Cd. *Nature*, 263: pp. 42-44.  
 BOYLE E.A., SCLATER F.X. and EDMOND J.M., 1977. - The Distribution of Dissolved Cu in the Pacific. *Earth Plan. Sci. Lett.* 37: pp. 38-54.  
 DANIELSSON L.G., 1980. - Cd, Co, Cu, Fe, Pb, Ni and Zn in Indian Ocean water. *Mar. Chem.* 8: pp. 199-215.  
 HANNA R.G.M., 1985. - Report on the 15th Scientific Cruise of R/V "AKADEMIK PETROVSKI" in the Mediterranean Sea, UNESCO, Paris, 12 p.  
 KREMLING K.X. and PETERSEN H., 1981. - The Distribution of Zn, Cd, Cu, Mn and Fe in waters of the open Mediterranean Sea. *Meteor. Forsch.-Ergebnisse*, 23: pp. 5-14.  
 STOMMEL H., 1972. - Deep Winter-time Convection in the Western Mediterranean Sea. In: A.L. Gordon (Ed.), *Stud. Phys. Oceanogr. New York*, 2: pp. 207-218.  
 TOPPING G., 1974. - The movement of heavy metals in a marine ecosystem. *Int. Counc. Expl. Sea*, E31: pp. 1-7.  
 UNEP, 1985. - Report on the state of Pollution of the Mediterranean Sea. Athens: UNEP/IG. 56/Inf. 4.  
 WESTERLUND S.X. and OHMAN P., 1991. - Fe in the Water Column of Weddell Sea. *Mar. Chem.*, 35: pp. 199-217.