Available online at www.sciencedirect.com
SCIENCE DIRECT®

Science of the Total Environment 353 (2005) 232–246

Science of the Total Environment
An International Journal for Scientific Research into the Environment and its Relationship with Humankind

www.elsevier.com/locate/scitotenv

Chemical characterization of different typologies of mucilaginous aggregates in the Northern Adriatic Sea

Michele Giani [a,*], Daniela Berto [a], Valentina Zangrando [a], Silvia Castelli [a], Paola Sist [b], Ranieri Urbani [b]

[a] *Istituto Centrale per la Ricerca Scientifica e Tecnologica Applicata al Mare, ICRAM, Brondolo, I-30015 Chioggia, Italy*
[b] *Dipartimento di Biochimica, Biofisica e Chimica delle macromolecole, Università di Trieste, I-34127 Trieste, Italy*

Abstract

The chemical composition of mucilage aggregates found during summer 2000, 2001 and 2002 in the North Adriatic Sea depends on the nature of the organic matter during aggregation, on the environmental conditions of the site of formation and on the transformations during ageing.

The mucilages were composed of organic matter, together with a significant inorganic fraction. Elemental analysis revealed 12.5–32.2% of organic carbon, 0–7.3% of inorganic carbon and 1.0–3.7% of nitrogen. The C_{org}/N ratios of most aggregates were between 7.5 and 12.6, values close to those found in the suspended matter; higher ratios were found in large-size (>5 m) aggregates which are probably older. The content of carbohydrates and proteins determined in the aggregates, respectively, $15.4 \pm 8.9\%$ and $7.9 \pm 4.8\%$, w/w, showed a prevalence of carbohydrates over proteins.

Neutral carbohydrate analysis of purified polysaccharides from mucilage samples showed very similar signatures with high relative abundance of galactose and glucose.

Humic, fulvic and humin substances extracted from the mucilages constitute an important fraction of the organic matter in the aggregates. The humin (a fraction insoluble in acidic and basic media) was present in all mucilage samples, indicating the refractory nature of a part of the organic matter in the mucilage.

The iron and calcium could play a role during the aggregation process to form a complex with polysaccharides and humic fractions. The C_{org}/N ratio 10 ± 2 found in the humic acids extracted from the Adriatic aggregates disclosed a marine origin.

The low phosphorus content and the high C_{org}/P ratio found in the aggregates might depend from high bacteria activity or from the aggregation of organic fractions depleted of phosphorus.

The principal inorganic species contained aluminium and silicon, part of which was of biogenic origin and was more significant in the offshore mucilage aggregates than in the coastal ones.

The Si_{biog}/C_{org} ratio showed that diatoms were always present in the aggregates, although it cannot be established whether these are the producers or these develop within the aggregates.

Keywords: Mucilage; Northern Adriatic Sea; Humic substances; Metals; Carbon; Nitrogen phosphorus; Sulphur; Carbohydrates; Proteins; Biogenic silicon

1. Introduction

Different microorganisms contribute to the organic content of the mucilaginous aggregates forming in late

---

* Corresponding author. Tel.: +39 041 5543933; fax: +39 041 5547897.
E-mail address: m.giani@icram.org (M. Giani).

0048-9697/$ - see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.scitotenv.2005.09.027

spring–summer in the Northern Adriatic Sea: diatoms (Revelante and Gilmartin, 1991; Najdek et al., 2002; Kovac et al., 2002), dinoflagellate (Pompei et al., 2003; Pistocchi et al., 2005a–this issue), cyanobacteria (Kaltenböck and Herndl, 1992) and bacteria (Herndl, 1992; Azam et al., 1999; Pistocchi et al., 2005b–this issue). However, it is very difficult to establish the relative importance of the different contributors. Independently of their origin, the aggregates form a microhabitat in which the enrichment of the dissolved organic matter and nutrients favours the growth of phytoplankton (Kaltenböck and Herndl, 1992) and bacteria (Herndl, 1992).

The release of organic matter from phytoplankton by cell lysis, apoptosis or exudation and the successive bacterial transformations and photooxidation processes make the organic material even more complex. The adsorption of dissolved organic matter, the differential settling of particles, and the variation in buoyancy of the aggregates due to the hydrostatic effect of the gas produced by the biological activity facilitate the trapping of suspended matter, causing further changes to the stability and bacterial biotransformation (Decho and Herndl, 1995).

The aggregation of organic matter in seawater is determined by the physical and chemical properties of the aqueous medium, such as temperature (Thornton and Thake, 1998), turbulence, shear stress, differential settling, particle concentration (Simon et al., 2002), ionic strength, pH (Chin et al., 1998), concentrations of di- and trivalent cations (Thornton et al., 1999), and the chemical properties of the organic substances (molecular weight, functional groups) and particle surface properties (Žutić and Svetličić, 2000).

The chemical composition of the aggregates therefore depends on the nature of the organic substance at the moment of formation, in terms of size, age (biogeochemical transformations) and environmental conditions of the location in which the aggregate forms or is advected by currents. Previous studies evidenced the relevance of the polysaccharides (Marchetti et al., 1989) in the mucilaginous matrix as well as the presence of polymethylene chains (Kovac et al., 1998, 2002).

Aggregates very different in morphology and size have been observed during the mucilage events, both in the cluster of events of 1988–1990 (Stachowitsch et al., 1990) and in the more recent cluster of 2000, 2001 and 2002 (Precali et al., 2005–this issue). Information regarding the chemical composition of aggregates of different size and morphology is scarce (Posedel and Faganeli, 1991; Herndl, 1992), and almost no data exist for aggregates sampled far from the shallow coastal zone which is influenced by riverine discharge and bottom resuspension and can modify aggregate composition to a great extent.

Our objective was to determine the chemical and biochemical features of different aggregate typologies sampled both in the open sea and in coastal waters.

2. Materials and methods

2.1. Sampling

Mucilaginous aggregates were sampled by scuba divers during three different periods: June 9–July 6, 2000, July 3–6, 2001, June 3–July 31, 2002. The sampling data are reported in the Table 1, and the sampling sites are shown in Fig. 1.

Two L-polymethacrylate syringes or peristaltic pump with PVC tubing were used. The tubing was flushed with 50 l of seawater collected at the sample depth of the aggregates prior to each sampling. The aggregates were immediately frozen at −20 °C and thawed, dyalized (Cellusep, 3500 MWCO) with deionized MilliQ water, and then freeze dried.

Phytoplankton and zooplankton were sampled in the Po river prodelta using 30-μm and 200-μm nylon nets, respectively (Table 1). Aggregates comprising mainly the dinoflagellate *Noctiluca scintillans* were sampled at site K1 (Fig. 1). The plankton samples were washed with deionized MilliQ water and centrifuged (3000 rpm, 10 min), the washing were repeated 2–3 times until no chloride was detected by the addition of $AgNO_3$ solution. The plankton samples were frozen and freeze dried.

2.2. Analytical methods

Total carbon, organic carbon, nitrogen and sulphur were determined by a CHNS-O Fison analyser (Italy), mod. EA1108. Organic carbon was determined after removal of carbonates with concentrated HCl. The ash content was determined by ignition in a muffle at 450 °C for 4 h. Inorganic carbon was calculated as the difference between total and organic carbon.

Metals were determined by Atomic Emission of Inductively Coupled Plasma (ICP-AES), after digestion with a mixture of 69.5% HNO_3, 30% H_2O_2 and 48% HF (2:0.5:0.1 v/v). Biogenic silica was extracted with 0.2N NaOH (2 h, 95 °C) in accordance with Kamatani and Oku (2000) and determined using a spectrophotometer (ATI Unicam UV2) at 810 nm (Grasshoff et al., 1983). Biogenic silica was estimated by the regression line of silicon and aluminium (Kamatani and Oku, 2000). Humic fractions were separated as per the International Humic Substance Society methods (www.ihss.