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# Surfactants Biodegradation in Sea Water

Primary Biodegradation and Bacterial Population Response in Sea Water

## 1. Introduction

The increasing production and use of surfactants has prompted a great number of studies in relation to their biodegradability. An important part of these studies has employed analytical techniques which assess surfactant disappearance [4, 6, 14, 15, 25]. The determination of the fate and ultimate biodegradability has lately been studied using surfactants labelled with  $^{14}\text{C}$  or  $^{3}\text{H}$  at different portions of the molecules in enriched or natural waters [11, 17, 18, 19]. However, to the date, little attention has been focused on the changes in the bacterial community of natural waters after surfactant addition.

Gledhill suggested that the rates and extent of  $\text{CO}_2$  production from a surfactant depend on factors such as number and types of microorganisms capable of metabolizing the tested compound, stability of the cells after growth, endogenous respiration, ecotoxicity and others [11]. It seems evident that more information on the evolution and changes of bacterial number and activity after surfactant discharge is desirable so as to reach a better understanding of surfactant biodegradation in sea waters.

The present paper describes the primary biodegradation of sodium dodecyl sulphate (SDS), sodium alkyl benzene sulphophate (LAS) and cetyl trimethyl ammonium bromide (CTAB) in sea water. Thymidine incorporation and epifluorescence counts were used to check the bacterial response to the surfactant addition.

## 2. Material and Methods

### 2.1 Surfactants

Sodium dodecyl sulphate (SDS) is a Merck analytical grade compound whose anionic active matter content was 99.2% according to the analysis performed in our laboratory.

Sodium alkyl benzene sulphophate (LAS): the raw alkylbenzene obtained from Petroquímica Espaçola S.A. (Algeciras, Spain), was submitted to a sulphonation process in our laboratory, and afterwards neutralized with  $\text{NaOH}$ . In the final product the anionic active matter content was 65%, water content was 33-34% and that of unsulphonated matter was 1-2%.

Cetyl trimethyl ammonium bromide (CTAB) is a Fluka analytical grade compound whose cationic active matter content was 99.6% according to the analysis performed in our laboratory.

### 2.2 Biodegradation assays

Sea water for all biodegradation assays was collected from Barceloneta, the natural beach of Barcelona, a Mediterranean city of 3 million inhabitants. Sea water samples were collected in 10 l plastic containers, and immediately transported to the laboratory. Biodegradation assays were carried out in 2 l-Ur-

lammeyer flasks provided with internal edges containing 1 l of sea water; 20 mg  $\text{l}^{-1}$  of surfactant was used in all experiments as active matter. Incubations were performed at 22 °C in a rotary shaker (120 oscillations per min.). Water samples received no exogenous inoculum, so that only the indigenous bacterial community was measured. Controls without surfactants were done in each biodegradation assay.

### 2.3 Bacterial counts

Enumeration of bacteria was carried out by epifluorescence, after acridine orange staining and filtration through 0.2  $\mu\text{m}$  filters of polycarbonate (Nuclepore), following the standard procedure of Hobbe et al. [13]. Observations were done through a Nikon Optiphot equipped with an epifluorescence system EP-D, mercury lamp Osram HBO 100 W/2, B and ND<sub>3</sub> filters (Nikon).

### 2.4 Bacterial activity

Bacterial heterotrophic activity was measured by the thymidine incorporation method of Johnman and Azam [9, 9]. Subsamples from biodegradation assays (10-20 ml) were incubated 30 min. at room temperature (22-23 °C) with 20 nM methyl- $^{3}\text{H}$  thymidine (80-90 Ci mmol $^{-1}$ , Amersham). At the end of the incubation period an equal volume of ice-cold 10% trichloroacetic acid was added and the mixture kept five minutes in an ice bath. The cold trichloroacetic acid insoluble material was collected by filtration through 25 mm acetate cellulose filters of 0.2  $\mu\text{m}$  pore size (Sartorius). Filters were dissolved in the scintillation vials with ethyl acetate (1 ml) before addition of scintillation fluid (10 ml). The radioactivity was assayed by liquid scintillation using a Tri-Carb 460 C, Packard Instruments Co. Efficiency was determined using internal  $^{3}\text{H}$ -n-hexadecane standard (Amersham).

### 2.5 Analytical procedures

The analysis of SDS and LAS anionic surfactants was carried out following the methylene blue spectrophotometric method according to the Longwell and Mamice method as modified by Alberer [1].

Determination of CTAB cationic surfactant was carried out by method based on the reaction of the surfactant with an anionic dyestuff (disulphine blue VN 150) followed by spectrophotometric measurement of the chloroform extract of the surfactant dyestuff complex [20].

### 2.6 Data analysis

Biodegradation of sodium dodecyl sulphate shows a first order decay which can be expressed as  $A = A_0 \times e^{-kt}$ , where  $A$  is the concentration at time  $t$ , and  $A_0$  is the initial concentra-

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## **Surfactant Biodegradation:**

**Surfactant Biodegradation, Second Edition**, Robert Donald Swisher, 1986-10-31      **Biodegradability of Surfactants** D.R. Karsa, M.R. Porter, 2012-12-06 The awareness and development of biodegradable surfactants pre dates current pressures by the environmental movement by nearly three decades wherein a responsible industry mutually agreed to replace hard non biodegradable components of household detergents by soft biodegradable alternatives without course to legislation. The only requirement at that time was for surfactants used in detergents to exhibit a primary biodegradability in excess of 80% this referring to the disappearance or removal from solution of the intact surface active material as detected by specified analytical techniques. This proved useful as observed environmental impacts of surfactants e.g. visible foam on rivers are associated with the intact molecule. Test methods for primary biodegradability were eventually enshrined in EU legislation for nonionic surfactants Directive 82/1242 EEC amended 73/404/EEC and for anionic surfactants Directive 82/1243/EEC amended 73/405/EEC. No approved test methods and resultant legislation have been developed for cationic and amphoteric surfactants to date. The environmental classification of chemical substances which of course includes surfactants and associated risk assessment utilises a second criterion ready biodegradability. This may be assessed by a number of methods which monitor oxygen uptake BOD carbon dioxide production or removal of dissolved organic carbon DOC. Some surfactants which comply with the above Detergents Directive are borderline when it comes to ready biodegradability.

*Surfactant Biodegradation* Robert Donald Swisher, 1970      **Surfactant Biodegradation** R. D. Swisher, 1987

*Bacterial Biodegradation of Polycyclic Aromatic Hydrocarbons (PAH) and Potential Effects of Surfactants on PAH*

*Bioavailability*, 1996      **Biodegradation of Phenanthrene in Soils in the Presence of Surfactants** Kauser Jahan, 1993

**Surfactants in Lipid Chemistry** John H. P. Tyman, 1992 The great majority of surface active compounds having Lipidic character are either semi synthetic or derived from fossil fuel sources. The contribution from lipid raw materials has increased over the past four decades and both sources are now recognized as essential. *Surfactants in Lipid Chemistry* Recent Synthetic Physical and Biodegradative Studies contains contributions dealing with a variety of natural product semi synthetic and purely synthetic materials. The topics covered include the place of lipid natural resources in the synthesis of surfactants both by organic chemical methodology and by enzymatic procedures aspects of the physical properties of non ionic and certain anionic surfactants and their involvement in detergency and current knowledge of the biodegradation of anionic surfactants in the environment. Recent progress in phenolic glycolipid and lipid amino acid chemistry is also described. This book will be of interest to all those concerned with modern surfactant chemistry.

**The Role of Biodegradation and Sorption in Determining the Fate of Nonionic Surfactants in the Environment** Kathleen A. Wight, 1997      *Biological Degradation of Organic Chemical Pollutants in Biofilm Systems* E. Arvin, 1995 There is no description available for this title

**Handbook of Detergents, Part B** Uri Zoller, 2004-08-30 The second installment of the multivolume Handbook of

Detergents deals with the potential environmental impact of detergents as a result of their production formulation usage consumption and disposal This volume forms a comprehensive treatise on the multidimensional issues involved and emphasizes the alignment of scientific knowledge with the relevant contemporary data and methodologies in toxicology ecotoxicology and environmental risk assessment With contributions from over 50 experts worldwide this volume discusses industry procedures involving surfactant and detergent treatments and explores global concerns centering on recent legislative and regulatory developments Environmental and Human Safety of Major Surfactants: Anionic surfactants. pt. 1. Linear alkylbenzene sulfonates. pt. 2. Alcohol ethoxy sulfates. pt. 3. Alkyl sulfates, 1991

**Novel Surfactants** Krister Holmberg,2003-07-03 Extensively revised and expanded this timely reference discusses the synthesis properties and potential applications of popular and emerging surfactant compounds and systems This reference reflects current research trends in green surfactants the production of surfactants using biotechnological methods and surfactants based on natural buildin Biodegradation Rolando Chamy,2013-06-14 This book contains a collection of different biodegradation research activities where biological processes take place The book has two main sections A Polymers and Surfactants Biodegradation and B Biodegradation Microbial Behaviour **Analysis of Surfactants** Thomas M. Schmitt,1992 Presents descriptions and practical examples of all techniques applied to the analysis of surfactants from the characterization of commercial compounds to their determination in formulated products and their detection at trace levels in the environment Literature is reviewed through 1990 with app **Biodeterioration and Biodegradation** H.W. Ross Moore,1991 Proceedings of the 8th International Symposium Windsor Ontario Canada 26 31 August 1990 **Environmental and Human Safety of Major Surfactants**,1991 Bioremediation of Surface and Subsurface Contamination Rakesh K. Bajpai,Mark E. Zappi,1997

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Surfactant-based Separations John F. Scamehorn,2000 Because they are biodegradable and work well in low energy separations surfactants are an active area of interest in separations science This book covers surfactant based separations

for the chemical and biochemical process industries and analytical chemistry. It includes discussion of widely used processes and novel techniques such as surfactant enhanced ultrafiltration ground water and soil remediation surfactant absorption and flotation processes extraction processes recycling of paper and plastics using surfactant and analytical separations using surfactants

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