INTERCORR 2014 ABRACO
Hotel Praia Centro – Fortaleza/CE
19 a 23 de Maio

Antifoulant compounds with copper for preventing bioadhesion. A case study.

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Abstract

Bioadhesion, biofilms formation and biocorrosion of metal surfaces are due to biological and bioelectrochemical processes in which microorganisms participate actively by adhering to surfaces through biofilms. It is very difficult to prevent bioadhesion, biofilms formation and the economic losses for the biocorrosion problems are estimated at billon USS therefore, the importance of finding an effective method to prevent it. The goal of this paper was to evaluate of performance of antifoulant compounds with copper applied on carbon steel structures. Bioassays were carried out in Mar del Plata harbor, Buenos Aires, Argentina (38º 02' S-57º 32W). Carbon steel panel's series with copper layer and without it (control) were vertically hung from a floating dock, below the water surface to provide the record of fouling organisms. Sample taking from the accumulative samplers each 4 months during one year. The abundance of sessile species was estimated by recording their occurrence on a grid marked in quadrates over the entire panel. After 1 year of immersion, panels covered by copper layer did not present macrofouling, but a significative bacterial adherence was present. On the other hand, on control panels a lot species were observed.

Keywords: bioadhesion, biocorrosion, carbon steel, copper layer, seawater.

Introduction

In the marine environment, all immersed surfaces (natural or artificial) are rapidly colonized by a succession of organisms, the outcome being known as 'biofouling'. Within the first seconds to minutes, surfaces are covered by conditioning films composed of organic and inorganic molecules (1). Given that biofouling has negative impacts on all aspects of marine industry including shipping (2, 3, 4), aquaculture (5), offshore platforms and piping (6) power stations (7), desalination plants (7) and geophysical exploration (8, 9), it is important to optimise the efficacy of surface-based technologie.

A wide range of chemicals are used as biocides, which have very different physico-chemical properties and therefore differing environmental fates, behaviour and effects. Copper has been used as an antifoulant for centuries and extensive research has been performed to understand how copper speciation influences bioavailability and toxicity.

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The goal of this work was to evaluate antifoulant compounds with copper for preventing bioadhesion A working plan was designed to systematically detect, quantify, isolate and determinate: bacteria, fungi, yeasts, algae, protozoans, larvae and macroorganisms such as polichaetes, molluscs, crustaceans, etc. Bioassays were carried out in Mar del Plata harbour-Bs. As. Argentina (38º 02'S-57º 32'W). Microscopic and scanning electron microscopic (SEM) observations of biological communities and studies of abiotic factors were made.

Materials and methods

Two polyurethane panels painted with commercial AMERCOAT 235 + ABC #3 and AMERCOAT + Cu, respectively and 2 polyurethane panels without coverings (control samples), 50 cm separated from each other were put vertically hung from a floating dock, about 0.3 m and 1.5 m below the water surface in Mar del Plata harbour Buenos Aires, Argentine (38º 02'S-57º 32'W) to provide the record of fouling organisms. Sample taking from the accumulative samplers each 4 months during one year. When immersion is performed and during sampling the following abiotic factors were measured: water temperature, air temperature and pH with a LUFTMAN p300 meter and conductivity with a Hanna conductivity meter.

a. Microbiological studies

The biofilms samples were refrigerated and sent to the laboratory in seawater. Biofilms were removed from surface by scraping and suspending it in physiological solution.

Once in laboratory, they were processed for microbiological studies. Samples were seeding in differential and selective culture media for viable heterotrophic aerobic bacteria, sulphate reducing bacteria (SRB) and acidifying activity bacteria, fungi and yeasts development. Viable microorganisms were enumerated by different techniques (10, 11, 12).

b. Microfouling and macrofouling studies

From the accumulative carbon steel panel's series, representative samples of 1 cm^2 were taken and preserved in physiological solution and in sea water and sent to laboratory in a container with ice.

The successional stages of community are determined from data on abundance and distribution of the accumulative samples obtained. Data obtained from control panels and those with copper coating are compared to determine the efficacy of the coating tested.

Once the sample is in laboratory, abundances are estimated and registered extrapolating the results to a percent scale with 5 defined categories (13).

R: rare, 1-25%
E: scarce, 25,1-50%
F: frequent, 50, 1-75%
A: abundant, 75,1-100%

Microfouling was systematically observed and determined using an optical microscope (X 100 y X 200).

Results and discussion

The abiotic factors of the sampling area varied between:

Water temperature: 12,1 ºC - 25 ºC
Air temperature: 12,4 ºC - 26 ºC
Conductivity: 45,9 mS - 48,9 mS
pH: 7,23 - 8,00

Bacillary bacteria and coco-bacillus were observed. Gram coloration revealed the presence of negative Gram bacillus and negative Gram coco-bacillus in all the samples.

Microbiological count can be read in Table N° 1.

Table No. 1: Microbial count values corresponding to the different microorganisms (365 days of immersion)

Samples/Microorganism	Heterotrophic aerobic bacteria CFU/cm^2	Acidifying bacteria mo/cm^2	SRB Mo/cm^2	Fungi and yeasts CFU/cm^2
AMERCOAT 235 + ABC #3	40 x 10^5	1000	1000	WD
Control	137 x 10^5	1000000	1000000	WD
AMERCOAT + Cu	145 x 10^6	WD	WD	WD
Control	120 x 10^6	1000000	1000000	WD

CFU/cm^2 :Colony forming unit by cm^2
mo: microorganism
WD: without development

The panel with **AMERCOAT 235 + ABC # 3** covering (Photographs n° 1) showed little fouling and the film cracked, maintains its typical reddish color but coating peeled off easily. The panel surface was mostly covered by a film of microorganisms and sediment. Unlike the sampling before, there was not macrofouling presence (Figure 1).

In this sampling there were not significant differences between fixation in the front and in the back.

Photograph No. 1: General aspect of the panels with **AMERCOAT 235 + ABC # 3** covering (upper panel). Control panel (lower panel).365 days of immersion.

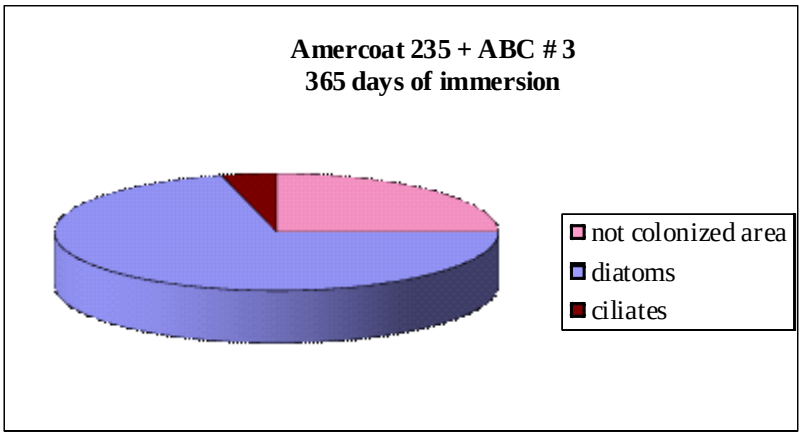

Figure 1.- Covering percent, 365 days of immersion.

The control panel showed an abundant fixation dominated by algae, calcareous poliquets (*Hydroides elegans*), cirripedia (*Balanus* spp.), ascidian (*Ciona intestinalis and Molgula* sp.) and colonies of bryozoans (Figure 2).

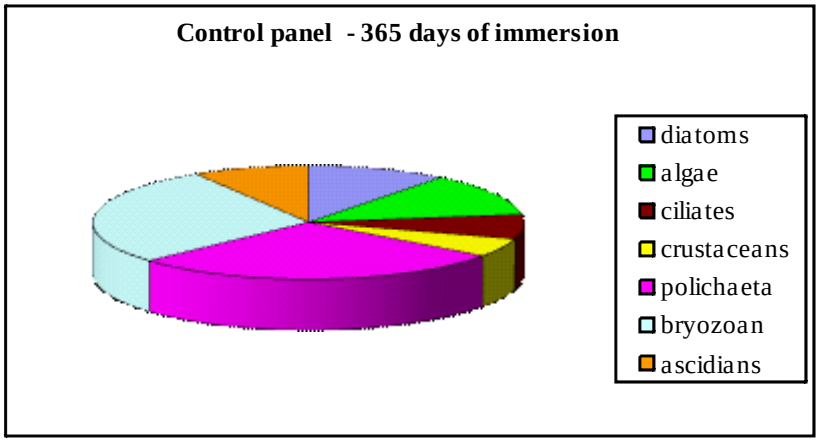

Figure 2.- Covering percent , 365 days of immersion

On the panel identified as **AMERCOAT 235 + CU** (Photographs n° 2), a film covering the whole surface was observed. The examination of this microfouling film under microscope revealed a great diversity and abundance of organisms and diatoms in particular. On the edges, well developed ascidians were registered (*Ciona intestinalis*), colonies of bryozoa *Bugula* sp. and green algae (*Enteromorpha intestinalis*) (Figure 3). The organisms with calcareous exoskeleton that had been registered in the samplings before peeled off; as it was explained before, this could be indicate that the covering interferes with the substrate cementation. Amphipods were also detected.

Photograph No. 2: General aspect of panels with **Amercoat 235 + Cu** covering (upper panel). Control panel (lower panel). 365 days of immersion.

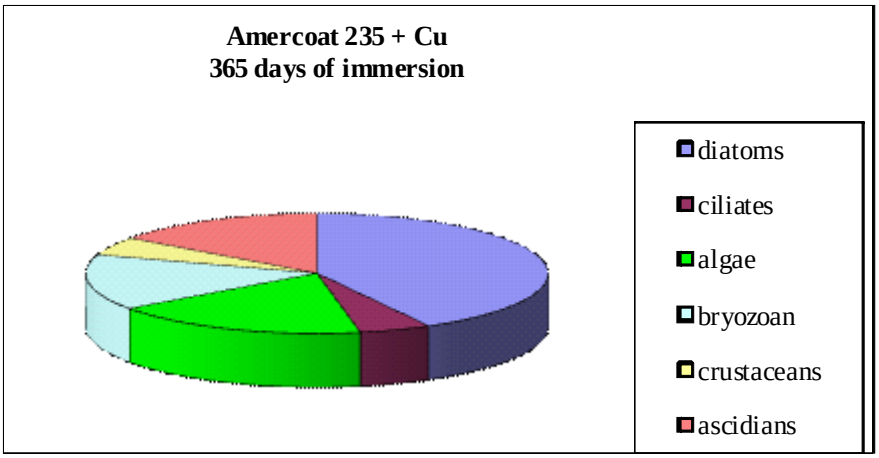

Figure 3.- Covering percent, 365 days of immersion.

In the control panel front a great amount of organisms was observed, of both macro and microfouling, while in the back there was a strong decrease of intensity and diversity of the populations established (Figure 4).

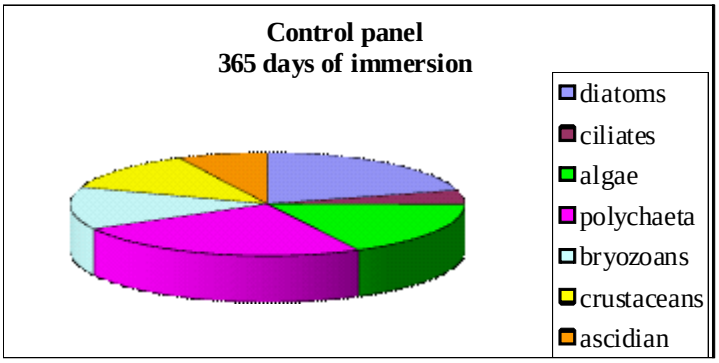

Figure 4.- Covering percent, 365 days of immersion

Conclusions

Microbiological results indicate that viable heterotrophic aerobic bacteria stuck on the panel surface without copper covering and with copper covering are of important values, higher during the hotter months. It is worth mentioning that microorganisms count was higher for panels with AMERCOAT + Cu covering.

In panels with AMERCOAT + Cu covering there were not acidifying bacteria and SRB, however, bacteria presence was variable on the AMERCOAT 235 + ABC#3 covering.
There were neither fungi nor yeast development in any case studied.

The AMERCOAT 235 + ABC#3 covering showed a better performance than the AMERCOAT 235 + Cu, but it also allowed some organisms to settle on.

In both cases it could be observed that coverings interfered with the mechanisms of cementation to substrate given that the organisms came easily off from the surfaces tested.
The panel with AMERCOAT 235 + ABC #3 covering showed cracking and peeled off easily.
For both kinds of coverings and taking into account that both faces of the panel undergo the same treatment, it is not possible to explain from a biological point of view the remarkable differences observed in the fouling settlement.
It would be advisable to go on with tests with the panel with AMERCOAT 235 + ABC #3 covering in order to determine the anti incrusting power in periods longer than a year as well as the quality of the film formed.

Acknowledgements: The authors acknowledge the financial support from CONICET (PIP 0200), CICPBA (Res. No. 243/13) and UNLP (11N 713 and 11X 632). To Lic. Patricia Battistoni for technical support.

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