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Effects of the copper-based antifouling paint "Fabi" on growth of the red alga *Ceramium tenuicorne*

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Abstract

The antifouling paint Fabi 3959 is painted on the hulls of vessels to avoid fouling caused by marine organisms attached to surfaces. The paint is registered for use on pleasure boats and other vessels weighing over 200 kg which are mainly running on the Swedish west coast (www.kemi.se).

Fabi 3959 contains copper as its active component, which is highly toxic to marine organisms and thus classified as a biocide.

Fabi antifouling paint was tested under laboratory conditions on the red macro alga *Ceramium tenuicorne*, in natural brackish water taken from the Baltic Sea. The *Ceramium* growth inhibition-test was performed using cloned algae exposed to leakage water with and without sediment. The samples containing only water held concentrations in the range of 0.11% of volume-18% of volume per liter, while the samples using sediment held doses measuring between 0.11% of volume-36% of volume leakage water per liter.

The study showed a growth inhibiting effect on the *Ceramium* in both water and sediment samples down to the lowest concentration used in the test. There was a difference between the water series and the sediment series in the EC₅₀ values of the leakage water. The mean EC₅₀ value was almost 10 times lower within the sediment series compared to the water series (0.114±0.10 and 1.024±0.75, respectively). This indicates that the sediment series are more toxic to *Ceramium* than the water series. However, if the mean values of EC₅₀ are expressed as copper-concentration, there is no clear difference between the two series (0.59 ± 0.13 µg/l for the sediment series and 0.62 ± 0.12 µg/l for the water series). Apparently, the test did not indicate that the sediment was absorbing the copper. Instead it cannot be excluded that another substance involved could have a growth inhibiting impact on *Ceramium*.

Keywords: *Ceramium*, *Ceramium tenuicorne*, antifouling paint, Fabi, growth inhibition-test

Sammanfattning

Båtbottenfärgen Fabi 3959 målas på fartygsskrov för att undvika påväxt av marina organismer. Färgen är registrerad att användas på fritidsbåtar och andra fartyg med en egenvikt på över 200 kg och med huvudsaklig fart på Västkusten (www.kemi.se). Den aktiva komponenten i Fabi 3959 är koppar, vilket är mycket giftigt för marina organismer och därför klassificeras den som en biocid.

Fabi båtbottenfärg testades i laborativ miljö, på den röda makroalgen *Ceramium tenuicorne* i naturligt brackvatten från Östersjön. Ett tillväxthämningstest utfördes på *Ceramium*-kloner vilka exponerades för lakvatten i bägare med och utan sediment. Proverna endast innehållande vatten bestod av koncentrationer i intervallen 0,11-18 volym% per liter medan proverna med sedimentvatten hade koncentrationer på 0,11-36 volym% per liter. Studien visade på en signifikant tillväxthämningseffekt på *Ceramium* i både vatten och sediment, ner till den lägsta använda koncentrationen. Det förelåg en tydlig skillnad mellan vattenserierna och sedimentserierna med avseende på resultaten av EC₅₀-värdena på lakvattnet; EC₅₀-värdena av den toxiska nivån för lakvattnet visade sig ligga tio gånger lägre i sedimentserierna än i vattenserierna (0,114 ± 0,10 i sediment och 1,024 ± 0,75 i vatten). Medelvärdena av EC₅₀ för koppar visar inte någon påtaglig skillnad mellan serierna (0,59 ± 0,13 µg/l för sediment 0,62 ± 0,12 µg/l för vatten). Testet indikerade därmed inte att sedimentet absorberade koppar. Istället föreligger en möjlighet att en annan substans orsakade tillväxthämning på *Ceramium*.

Nyckelord: *Ceramium*, *Ceramium tenuicorne*, båtbottenfärg, Fabi, tillväxthämningstest

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1. Introduction

1.1 Antifouling compounds and the Baltic Sea

Antifouling paints are used to protect the hulls of vessels, their underwater structures etc. from fouling caused by marine organisms attached to the surfaces (Debourg et al., 1993). When so happens, the new roughness of the vessel hulls increases turbulent flow and drag that reduces the vessel speed per unit of energy consumption (Milne, 1990). There are both biocide-based and biocide-free antifouling paints but the paint tested in this study, Fabi 3959, contains the active component copper. It is however registered only for use on pleasure boats and other vessels weighing over 200 kg which are mainly running on the Swedish west coast (www.kemi.se). Selling the products on one side of the Swedish coast brings the possibility for boat owners on the east coast to buy them (Fagergren, 2001). Today, boat owners that bought a copper-based antifouling paint before the prohibition in 2001 are allowed to continue using them until they are used up.

The Baltic Sea is one of the world's most polluted seas due to the 14 countries in the large drainage area that surrounds it, discharging nutrients and many different types of pollutants (Fagergren, 2001) out in the semi-enclosed basin (Figs 1-2). Another important contribution to the increasing concentration of contaminants is the long water residence time that is estimated to be 25-35 years (Jansson, 2003) combined with a limited inflow of sea water and a great supply of fresh water (Johansson, 2006). The Baltic salinity gradient consequently differs between 10 psu¹ in the southern Baltic Proper down to 2 psu in the northern Bothnian Bay (Kautsky and Kautsky, 2000). As most organisms in the Baltic Sea derive from either marine- or fresh water and living in brackish water, it is hard for them to adapt to these conditions. Instead, they are put under great physiological stress, which makes them even more sensitive to pollution. The average depth of the Baltic Sea is barely 60 meter and there is often a lack of oxygen on the bottom of the sea. The lack of oxygen depends on the formation of layers of saltier and heavier water below the halocline, that block oxygen-rich water to mix with the water poor in oxygen (Johansson, 2006). Lack of oxygen depends as well of a great consumption of oxygen when organic matter from e.g. algal blooms are decaying at the bottoms (Andrén, pers. comm.).

¹ Practical Salinity Unit

The fully marine conditions on the Swedish west coast along with a less polluted environment compared to the Baltic Sea, give the organisms an advantage to develop more diverse species communities on the west coast. Due to these two reasons, Fabi is allowed only on the west coast.



Figure 1. The Bothnian Bay and the Baltic Proper,

http://www.ab.lst.se/templates/Proj_Page_7587.asp



Figure 2. The Baltic Sea drainage basin

<http://www.grida.no/baltic/htmls/maps.htm>

The sediment bottoms of the Stockholm archipelago exhibit elevated concentrations of copper, with the highest levels closest to Stockholm (Kautsky et al., 2000). Organisms living in the Stockholm archipelago can hence absorb copper that is stored in the sediment and aquatic plants can take up copper by their roots and then transport it to the shoots (Greger and Kautsky, 1990; Greger and Kautsky, 1993). Further on, grazing molluscs can take up the copper and pass it on to organisms that are higher up in the food web. This occurs for example via fish to birds and humans. Birds that graze aquatic plants can take up copper directly by eating (Kautsky et al., 2000). Unfortunately, the launching of the newly painted pleasure boats coincides with the reproduction-period of many organisms that are important species for a well functioning ecosystem in the Baltic Sea.

Copper, which is an essential metal for aquatic organisms and virtually every living organism, is present in all sediment and natural waters (Hall et al., 1997). Even though copper is a minor nutrient for animals and plants at low concentrations, it becomes toxic at concentrations approximately 10-50 times higher than the concentration necessary to aquatic life (Hall et al., 1997). As copper is used with the intention to deter marine organisms, it is classified as a *biocide*. Biocides are defined as substances intended to have a negative effect on living organisms (Directive 98/8/EC, 1998). European saltwater environments have in past times experienced a high potential ecological risk from copper exposure (International Maritime Organization, 1997) and the main area of concern is the use of copper-based antifouling paints on both commercial and recreational watercraft. Multiple studies have shown the environmental copper input from the paints (Hall and Anderson, 1999).

Copper is effective against marine fauna, but unfortunately the compounds persist in the water and in sediments where it affects or even kills non-target organisms (Anonymous 1999). Copper can also be released into the environment in many other ways e.g. from copper roofs, sewage disposal plants, water mains, industries and pleasure boats.

When painted on to boats, the antifouling paints gradually leak copper into the water. The leakage results in a release of Cu^{2+} -ions and increases the bioavailable amount of copper near boats and marinas (Eriksson et al., 1998). Bioavailable copper is the fraction of copper that can be taken up and metabolized by an organism. The only stable form of copper in oxygenated environments, aside from metallic copper, is Cu^{2+} (Hall and Anderson, 1999; Landner and Lindström, 1998). The bioavailability and fate of Cu-ions is regulated by factors such as salinity, redox-potential (reduction-oxidation reaction; describes all chemical reactions in which atoms have their oxidation number changed), alkalinity, pH and the concentration of organic and inorganic particles in the water (Lindgren et al., 1998).

The effects that copper-based antifouling paints cause on marine organisms have been tested in many different studies e.g. by Eklund (2004) and Bruno and Eklund (2003). What this study contributes with to this research-area is the presence of natural sediment in the testing-series. Previous studies have focused on how the copper leaks into the water and what consequences it may cause on *Ceramium* and other marine organisms. The present study includes the influence of copper on water, but also what contributing effects natural sediment along with copper may have on the algae. The addition of sediment in the test made it

possible to simulate the natural environment of the Baltic Sea, which is important for a realistic comprehension of the environmental impact from the antifouling paints.

1.2. The study organism: Ceramium tenuicorne

The organism used in the test was the marine red macro alga *Ceramium tenuicorne*, which is frequently used in toxicity-tests. The alga is sensitive to the slightest change in its habitat which makes it a good indicator on the quality of the local environment (Eklund, 1998; Bergström et al., 2003). *C. tenuicorne* is a common and indigenous species in the Baltic Sea and other parts of the world and has a wide salinity range between 1 and 32 psu, which makes it possible to use in tests for relevant salinities in extended areas (Karlsson et al., 2006). Macro-algae are very important for the Baltic Sea ecosystem as they have many functions such as primary producers, food for primary consumers and as habitats for fishes and invertebrates (Tedengren, 1990).

The aim of this project was to investigate the effects of the copper-based antifouling paint “Fabi”. In order to do so, growth inhibition-test was performed on the red macro alga *Ceramium tenuicorne*.

2. Materials and methods

2.1. Fabi antifouling paint

The antifouling paint tested in this study, Fabi (registered as “Fabi 3959” at the Swedish National Chemicals Inspectorate), is suspected to be toxic to aquatic organisms as the active component is Copper(I) oxide 6% of weight. The Swedish National Chemicals Inspectorate only allows Fabi for boats mainly running on the Swedish west coast (from Trelleborg to the Norwegian border). The paint is manufactured by International Paint Ltd and represented by International Färg AB, Division Båtfärg (www.kemi.se).

Besides the biocide copper, Fabi consists of various substances, among them zinc and manganese. Both zinc and manganese are essential substances that can become harmful in high concentrations, and previous studies have shown that Fabi 3959 tends to leak substantial amounts of zinc (Ytreberg, pers. comm.). In aerobic (oxygen-based) environments, the presence of manganese oxides largely controls copper bioavailability (Hall and Anderson, 1999).

2.2. The sediment used in the test

The surface sediment (≈ 8 cm.) was collected by a sledge-scraper from a shallow bay close to St Uttervik, Södermanland county, Sweden (Latitude: N 58°51', Longitude: E 17°32') in September 2008. It was dry-sieved through a sieve with mesh size 5 mm (thus removing mussels, snails and other macro fauna). The sediment had a density of 1.15 g/ml (ww) and the organic content, estimated as loss on ignition, was 14% (von Seth, 2009).

2.3. The leakage procedure

The paint was applied on the area of 10 cm² on small plastic boats at two occasions with an interval of 24 h. Before the leaking procedure started, the painted plastic boats were placed into beakers with 1 L natural brackish water for 1 h. This water was later discarded because it may contain residues of preservatives or solvents. The painted plastic boats were subsequently transferred in pre-cleaned autoclaved beakers. 3 beakers (W1, W2, W3) containing 1 L of filtered brackish water and 3 beakers (S1, S2, S3) containing 800 ml of filtered brackish water with 200 ml of surface sediment. Adding sediment into a few beakers gave the possibility to investigate if the sediment had any impact on the amount of contaminants in the water. In order to investigate the possibility of the sediment to absorb copper, the dose of copper was increased in the sediment tests to a concentration reaching 36% of leakage water, while the water tests only measure concentrations up to 18% of leakage water.

The beakers were covered with aluminum foil in order to prevent growth of any photosynthetic organisms and then placed on a shaking table for two weeks to stimulate water movement. The leaking was performed at 22 ± 2 °C. The leakage proceeded for 7 days with the volume proportion of copper used in the test corresponding to the allowed leakage of copper for the west coast of Sweden, i.e., 100 µg Cu/cm² in 7 days (Swedish Chemicals Inspectorate, 1998). After 24 h, 48 h, 96 h, and 1 week samples of 20 ml water were collected from each beaker in acid cleaned polyethylene bottles, to be analyzed for metal content. Water samples were then acidified (20 µl HNO₃ / 10 ml per bottle) and stored in plastic bags in a refrigerator before analysed for metals (Karlsson and Eklund, 2004). The leakage water bottles were stored in a refrigerator for later metal analyses on ITM (Department of Applied Environmental Science) at Stockholm University, while the rest of the leakage water was saved for the Ceramium growth inhibition-test. Due to a limited amount of time, only samples

from day 7 were included in the analyses. This is the water that was used for the growth inhibition study.

2.4. The growth inhibition-test

Growth inhibition was performed on *Ceramium tenuicorne*, according to the procedures described by Eklund (2004) and Bruno and Eklund (2003). The testing organisms were subsampled from a female generation since it, compared to the male generation, grows faster and more uniformly (Bergström et al., 2003; Eklund, 2005). Autoclaved artificial seawater added with nitrogen (3.46 mg/L), phosphorus (0.78 mg/L), iron (0.10 mg/L) and carbon (1.65 mg/L) was used as medium for growth-promotion of the algae. The water used in the *Ceramium* growth inhibition-test is the natural brackish water added into all the Petri dishes. All series therefore contain the same substances although the sediment series contain sediment as well. Four replicates of each test-concentration (from 0.11%-36%) and 6 replicates of the controls, containing only brackish water, were prepared for each test series. The concentrations were then transferred into sterile Petri dishes with covers and a diameter of 5 cm.

Every dish contained 2 algae-plants that had been randomly picked up with tweezers from the vessel in which they were gathered. Measurements were made on totally 120 algae.

Since the same tweezers were used to transfer all the algae from the vessel into the Petri dishes, there was a risk of increasing the concentration of toxic substances. In order to diminish the risk of lower concentrations to rise in toxicity, the algae were picked up from the control-concentration first and subsequently in rising concentration-order. To reduce the impact of external factors such as light, position and oxygen, the Petri dishes were randomly spread on a tray and put for cultivation in a culture chamber for 7 days at $22 \pm 2^\circ\text{C}$ and with a circadian rhythm consisting of 14 h light and 10 h darkness. To avoid any possible evaporation due to the ventilation-system, the trays were covered with plastic wrap (Eklund and Ek, 2004). The lengths of the algae were measured at day 0 and again at day 7 in order to estimate the growth inhibition. The measurements on the algae were carried out under a dissection microscope by placing the Petri dish on a millimeter-paper according to the method described by Eklund and Ek (2004). The length was measured from the first ramification to the tip of the longest branch (Fig.3).

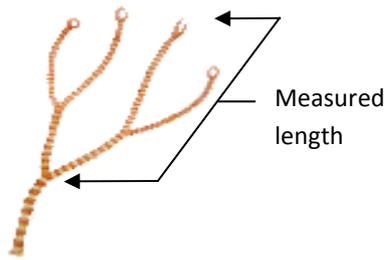


Figure 3. Piece of collected wild *Ceramium tenuicorne* with two branches and 4 initiated claws, length 3 mm. (adopted from von Seth 2009).

At the end of the test pH were measured in the control and in the lowest and highest test concentration (the pH-values were between 6.5 and 9.5).

2.5. Calculations

The algal growth was measured after 7 days and calculated as the percentage mean growth compared to the start length.

The Median Effective Concentration values (EC_{50} and EC_{10}), meaning the concentrations in which the algae express a 50% (EC_{50}) or 10% (EC_{10}) growth rate inhibition compared to the control, were calculated in REGTOX-EV6.xls (Regtox 6.3 software program.xls). This is a method that calculates EC-values for macro algae with corresponding 95% confidence limits by optimising the curve fit with successive iterations (Bruno and Eklund, 2003).

3. Results

3.1. Leakage water

The tests made during the leakage procedure do not show any large differences in the concentrations of copper among the different series. The mean values of copper (Cu) within the sediment-series are 16.5 (\pm SE 0.95) and 15.9 (\pm SE 0.24) within the water-series. The beakers containing sediment have a higher amount of manganese (Mn) compared to the beakers containing only water (see Table 1). The amounts of zinc (Zn) in the beakers with only water are much higher than the amounts in the beakers where water was added on to sediment (Table 1).

Table 1. Results from analyses performed on samples from day 7 of the leakage procedure where the amounts of manganese (Mn), copper (Cu) and zinc (Zn) in the water of the beakers were measured. CS = control with water containing sediment, CW = control with only water, S1, S2, S3 = sediment-tests 1, 2 and 3 (beakers containing both water and sediment), W1, W2, W3 = water-tests 1, 2 and 3 (beakers containing only water).

	Mn	Cu	Zn
	mg/l	mg/l	mg/l
CS 100%	6,7	2,8	7,1
CW 100%	0,1	0,5	2,0
S1 100%	4,5	16,8	444
S2 100%	3,8	14,8	377
S3 100%	5,2	18,0	522
W1 100%	0,3	16,4	1403
W2 100%	0,2	15,8	1440
W3 100%	0,2	15,6	1179

3.2. Results from the growth inhibition-test

There was a clear growth inhibition on *Ceramium* due to the Fabi antifouling paint for all tested series, at doses as low as 0.11% (Figs 4-6).

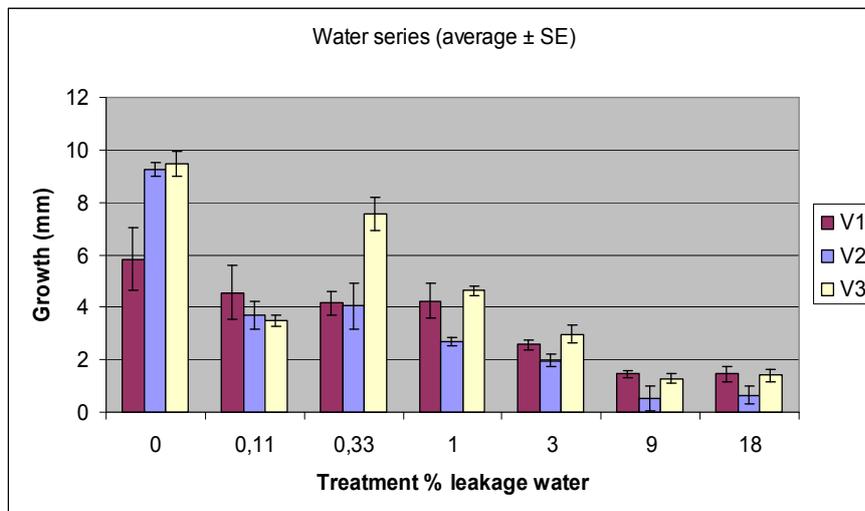


Figure 4. Effects of Fabi antifouling paint on *Ceramium tenuicorne* clones in the water series at the concentrations of 0 (control), 0.11, 0.33, 1, 3, 9 and 18% leakage water. Values are shown as mean relative growth ± SE (n=4; n=6 for controls)

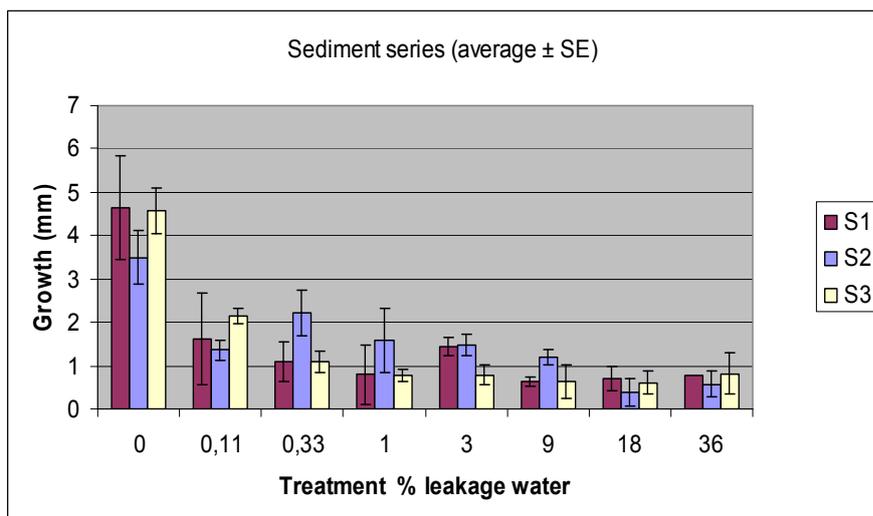


Figure 5. Effects of Fabi antifouling paint on *Ceramium tenuicorne* clones in the sediment series at the concentrations of 0 (control), 0.11, 0.33, 1, 3, 9, 18 and 36% leakage water. Values are shown as mean relative growth \pm SE (n=4; n=6 for controls).

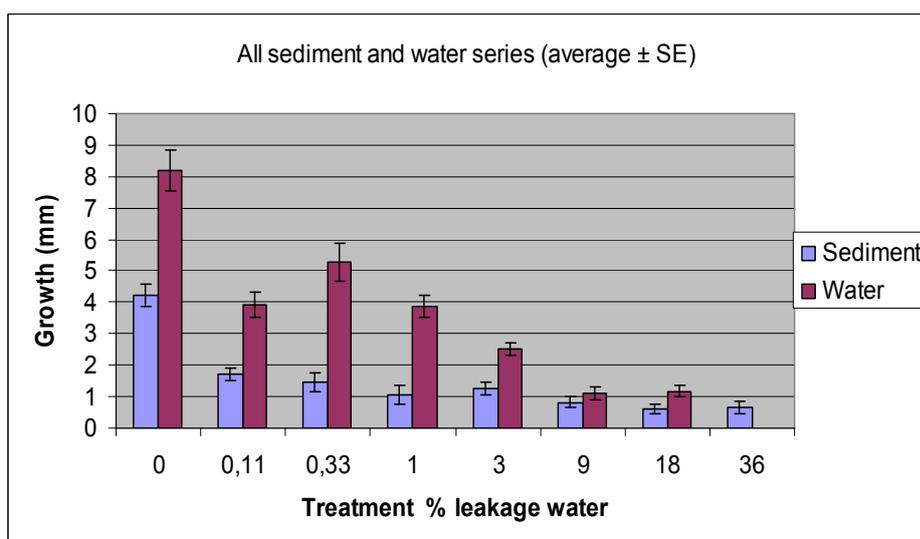


Figure 6. Effects of Fabi antifouling paint on *Ceramium tenuicorne* clones in both the sediment series and in the water series at the concentrations of 0 (control), 0.11, 0.33, 1, 3, 9, 18 and 36% (only sediment series). Values are shown as mean relative growth \pm SE (n=12; n=18 for controls).

The REGTOX-data shows that there is a difference in average effective concentration between the water series ($EC_{50} = 1.024$) and the sediment series ($EC_{50} = 0.114$). The EC_{50} values on the leakage water is 10 times lower within the sediment series compared to the water series, which indicates that the sediment series prove to be more toxic to *Ceramium* than the water series (Table 2).

The EC₅₀ values on the copper do not express a notable difference in toxicity between the series, meaning that the growth inhibition could depend on another substance.

Table 2. Median Effective Concentration values (EC₁₀ and EC₅₀) of growth rate inhibition on *Ceramium tenuicorne* (REGTOX-EV6.xls), expressed as percentage leakage water and as copper concentration (µg/l). W1-W3 represent test series with antifouling paint leakage into natural brackish water, S1-S3 represent test series with antifouling paint leakage into natural brackish water with sediment present (see text for details).

	% Leakage water		Expressed as Cu (µg/l)	
	EC ₁₀	EC ₅₀	EC ₁₀	EC ₅₀
W1	0.063828	2.498	0.470	0.858
W2	0.000410	0.084	0.460	0.473
W3	0.002007	0.490	0.460	0.534
<i>Average (W1,W2,W3)</i>	0.022082	1.024	0.464	0.622
S1	0.000000	0.001	0.460	0.460
S2	0.000094	0.313	0.823	0.847
S3	0.000031	0.026	0.460	0.465
<i>Average (S1,S2,S3)</i>	0.000042	0.114	0.581	0.591

4. Discussion

The results from this study showed a significant growth inhibiting effect on *Ceramium* caused by a copper based antifouling paint. The tests were made under controlled laboratory conditions and the method used in this study is the growth inhibition-test (Eklund, 2004).

According to the tests, the sediment series were approximately 10 times more harmful to *Ceramium* than the water series: the sediment-water had an EC₅₀ growth inhibition value 10 times higher compared to the water. Even though there is a significant difference in toxicity between the series, it cannot be explained by the copper since the series contain almost the same amounts of this metal.

The sediment did not appear to absorb or release copper. However, the amounts of zinc in the beakers with only water were much higher than the amounts in the beakers containing sediment. A possible explanation for this is that the sediment absorbed the zinc. There are previous studies showing that sediment has the ability to bind zinc in great quantities (Nicholas and Thomas, 1978).

The beakers containing sediment also had a higher amount of manganese compared to the beakers containing only water. This could be interpreted as a release of manganese from the sediment into the water. Manganese is expected to be more mobile in a low-oxygen environment than in one with high amounts (Voipio, 1981). The Baltic Sea is an area being rather low in oxygen, which could explain the mobility of the manganese in the tested sediment. However, the oxygen conditions in the sediment and water during the leakage process in the present study were not monitored.

As mentioned above, the beakers containing sediment did not contain higher amounts of copper than the ones with only water, yet the sediment series proved to give an EC₅₀ growth inhibition value approximately 10 times higher than the water series. This implies that the sediment from St Uttervik may have contained other toxic substances that were further released into the water. This is a very credible scenario since the sediment was taken from the Baltic Sea.

A notable observation in Figs 4-6 is that the tests “W2”, “S2” and “W3” show a lower growth inhibition in the 0.33%-concentration than in the 0.11%-concentration. Logically, the algae should suffer more from a higher dose of copper than from the lower dose. It is however, according to a theory called “hormesis” it is a common scenario that organisms respond positively to low exposures to toxicants and other stressors (Gustafsson, pers. comm.). As this study involved exposure to essential metals, low doses may consequently have had a healthy impact on *Ceramium*.

Sediment bottoms serve as an important function for the all-round condition of a sea and its impact on contaminants is therefore interesting to investigate further.

The future effects on usage of toxic antifouling paints on the Swedish west coast could initiate problems on the Swedish east coast as well. A potential risk for future negative effects on the environment are sediment-bottoms serving as end stations for heavy metals used in toxic antifouling compounds (Landner and Lindeström, 1998). One basic factor regarding the chemistry of sea water is the interaction between sediments and water. There are three main aspects in this certain interaction: bacterial activity, chemical equilibria and effects of the redox potential (Voipio, 1981). Focus is not being put on the chemistry of sea water in this article, yet it is important to understand that several aspects have to be taken into account concerning the presence and impact of metals in sediment and water.

As mentioned in the beginning of this article, the main problem faced in the Baltic Sea from the use of copper-based antifouling paints, is that the launching of the newly painted pleasure boats which coincides with the reproduction-period of many marine organisms. According to an article by Fagergren (2001), pleasure boats are the major single source of copper in lakes and in coastal waters, which shows a clear relation between increased copper content and the launching of newly painted pleasure boats.

The most troublesome fouling species in the Baltic Sea, barnacle, is easily removed from the boat hull in a very environmentally friendly way. In order to escape the fouling-problem, one method that can be used is to brush off the young barnacle from the boat hull. By using this method within a couple of weeks after settling of the larvae, the barnacles are still small and therefore easily removed (Fagergren, 2001). Another way to avoid fouling is to take the boat to a freshwater area e.g. Lake Mälaren, for only a few days. The young barnacles cannot survive under these freshwater-conditions, and the problem will consequently be solved in an effortless way (Fagergren and Kautsky, 2000). There are though many variables that can affect fouling: salinity, temperature and light, water currents, time for launching and hauling ashore, geographical situation and how much the boat is in use during the summer (The County Administrative Board in Stockholm, 2000).

Copper-based antifouling paints are forbidden since 2001 on boats along the Swedish east coast, but the paint-company “International” has recently reintroduced them to the market. The company claims that the rules have been changed, but according to the Swedish Chemicals Inspectorate the rules are the same as before. “International” continues selling the products, referring to “individual responsibility” among the customers: boat owners should be reading the markings saying that it is only legal to use the copper-based antifouling paints on the west coast. Since the law only forbids *using* these paints on the east coast, “International” has interpreted that there is no prohibition for companies to *sell* them (DN, 2009). As the law speaks today, boat owners are allowed to paint their boats with copper-based antifouling paints, as long as they mainly run their boats on the west coast. With these instructions, boat owners are not prohibited to launch their newly painted boats on east coast-marinas, neither are they forbidden to take their boats to the east coast as long as they mainly run on the west coast.

As the main copper-leakage occurs when the boat is newly painted, the law would probably be more effective if it would put focus on where the boats are put into marinas instead of where they generally run.

5. Conclusion

The antifouling paint Fabi 3959 is very harmful to the red macro alga *Ceramium tenuicorne* and it is consequently direct and indirect dangerous to other marine organisms as well. It can cause a direct harmful reaction on many organisms but also indirectly by damaging the ecosystem of the Baltic Sea. By using the growth inhibition-test, it became evident that concentrations as low as 0.11% of the leakage-water containing Fabi antifouling paint, was clearly harmful towards *Ceramium*. This indicates the fact that Fabi is destructive to the environment.

According to this study, the toxicity of the tested paint does not necessarily have to be explained by the copper: the metal analysis of the waters used in the test showed that the beakers containing sediment gave an EC₅₀ growth inhibition value 10 times higher (i.e. they were 10 times more toxic) than the beakers containing water. The analysis of the amounts of copper did however not show a significant difference between the water- and sediment series. This implies that the active component of the paint does not have to be the main reason to the growth inhibition; instead there could be other substances of the antifouling paint causing the observed growth inhibition.

In this study, the water containing sediment showed to be more harmful to the algae than the tests that were performed on plain water. By adding sediment into the test, the results showed that sediment absorbs zinc and releases manganese, but the theory about sediment absorbing copper does not seem to apply for the tested system.

Considering today's insufficient law against using antifouling paints on the east coast, a possibility to create better rules for boat owners, would be to focus on where the boats are put into marinas, instead of focusing on where the boat mainly will be running during the summer.

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