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1 **Bioaccumulation of the new antifoulant medetomidine in marine**
2 **organisms**

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11

Abstract

Biofouling is a huge problem globally and new alternative antifoulants are presently being investigated. One candidate is medetomidine, a commonly used sedative in veterinary medicine, which has been shown to effectively prevent settlement of barnacles. The purpose of this study was to measure uptake, elimination and bioconcentration of medetomidine in *Mytilus edulis*, *Abra nitida*, *Crangon crangon* and periphyton communities to evaluate the risk of bioaccumulation in the marine environment. Bioconcentration factors (BCF) and bioaccumulation factors (BAF) were used to assess the bioaccumulation. The calculations of these factors were based on the distribution of the radiolabelled medetomidine. BCF for *Crangon crangon* was 2.8 while *Mytilus edulis* had a BCF of 134 and the periphyton communities' BCF was 1195 l/kg fresh weight (FW). The concentration of medetomidine in the animals reached steady state after 24 to 48 hours for all test systems except for *Abra nitida*, which never stabilised enough to calculate a bioaccumulation factor (BAF). Elimination from the organism's tissues was rapid for three of the test systems with half-lives between 1 and 24 h. *Abra nitida* had a half-life of 96-120 h. The present study demonstrates that the bioconcentration and bioaccumulation of medetomidine differs between aquatic organisms and that microalgal communities in the form of periphyton have the highest bioconcentration factor of the organisms tested.

Keywords: medetomidine; antifouling substance; bioaccumulation; elimination; uptake; periphyton; benthos

1 1. Introduction

2 Medetomidine (4-[1-(2,3-dimethylphenyl)ethyl]-1H-imidazole) is commonly used as a
3 sedative in veterinary medicine and functions as an α -adrenoceptor agonist in mammals.

4 It is presently under development for use as an antifouling compound and has been
5 shown to successfully prevent settlement of barnacles at nanomolar concentrations
6 (Dahlström et al., 2000). Much is known about mammalian effects (MacDonald et al.,
7 1988; Scheinin et al., 1989; Vainio and Palmu, 1989; Kuusela et al., 2000) and
8 pharmacokinetics (Salonen, 1989; Kuusela et al., 2000; Ranheim et al., 2000) of
9 medetomidine. Effect studies have also been conducted on fish (Bellas et al., 2005;
10 Ruuskanen et al., 2005; Lennquist and Förlin, 2006; Hilvarsson et al., 2007; Lennquist et
11 al., 2008) and invertebrates (Bellas et al., 2006; Krång and Dahlström, 2006). However,
12 to our knowledge very little is known about bioaccumulation processes in aquatic
13 organisms.

14 Information on bioaccumulation and the risk of biomagnification of a chemical is
15 essential when conducting a risk assessment. Accumulation of a contaminant in the
16 aquatic environment is determined by its physical and chemical properties as well as
17 availability and persistence in water and the type of food chains exposed. The amount of
18 bioaccumulation is often correlated to the octanol-water partitioning coefficient (K_{ow})
19 with a linear relationship between bioaccumulation and K_{ow} for most substances below
20 $\log K_{ow}$ 6. Highly hydrophobic substances ($\log K_{ow} > 6$) do not display this correlation
21 since bioaccumulation is limited by molecule size.

22 The indicators traditionally used for bioaccumulation are the bioconcentration factor
23 (BCF) or the bioaccumulation factor (BAF). The factors are used as a measure of how

much of a certain contaminant an organism can absorb from the environment. BCF is defined as the ratio of the contaminant in the tissue of an organism to the concentration in the water, and BAF as the ratio of the contaminant in the tissue to the concentrations in any compartments (such as water, sediment, food) relevant for the main uptake routes from the environment. Both BCF and BAF are measured at steady-state conditions (Spacie et al., 1995) and most commonly presented as wet weight with the unit l/kg (Arnot and Gobas 2006) which is also used throughout this article. The criteria set by the European Chemicals Bureau for BCF states that a substance with a value above 2000 is bioaccumulative (Technical Guidance Document on Risk Assessment EUR 20418 EN/2). However, there are many more assessment endpoints from regulatory agencies such as the US Environmental Protection Agency, Environment Canada and the United Nations with BCF values ranging from 1000 to 5000 (Arnot and Gobas, 2006). These values do however only concern wet weight that causes some problems when working with systems like periphyton communities that are commonly measured as dry weight due to a high proportion of interstitial water. A BCF based on dry weight gives extremely high values and cannot be compared with fresh weight based BCFs. One way to normalise this difference is to express all values as lipid weight based concentrations since lipid content is one of the most important factors determining uptake of lipophilic compounds. Other factors like the amount of organic carbon in the organisms can however also influence accumulation, especially for primary producers. Normalisation to lipid content would then cause an overestimation of the bioaccumulation so to choose a normalisation method that suite all test species or systems can be difficult (Berglund et al., 2005). In this study no normalisations were performed.

Medetomidine has a QSPR-estimated log K_{ow} of 3.13 (pH 8, QSPR (Quantitative Structure Pharmacokinetic Relationship)). It is intermediately hydrophobic and may bioaccumulate (log $K_{ow} > 3$) according to the Technical Guidance Document on Risk Assessment (EUR 20418 EN/2). Because of the imidazole group in medetomidine, a substantial amount of the compound is likely to be adsorbed to sediment particles (Sundberg and Martin, 1974; Handa et al., 2006) and might therefore constitute a risk especially to sediment-living and sediment-eating organisms. However, a log K_{ow} below 4.5 is not enough to classify a substance to be bioaccumulative based on K_{ow} values alone (Technical Guidance Document on Risk Assessment EUR 20418 EN/2).

The present study aims to investigate the bioaccumulation of medetomidine in a selection of marine aquatic species to increase the knowledge of the substance behaviour. The different test organisms are chosen to get a broad understanding on the potential accumulation in the marine environment, from microbiota in periphyton communities, to filter- (blue mussel, *Mytilus edulis*) and sediment-feeders (*Abra nitida*). A crustacean, the brown shrimp (*Crangon crangon*), was also included since medetomidine is known to have effects on other crustaceans, i.e. barnacles. They were also selected because they are key species in their individual habitats and very abundant (Wikander 1980; Pihl and Rosenberg 1982; Gosling 1992).

This study is part of a larger risk assessment study of medetomidine conducted within the research programme Marine Paint funded by the Swedish Foundation for Environmental Research (MISTRA).

2. Materials & methods

2.1. Biological material

Periphyton growing on glass discs (1.5 cm²) mounted on polyethylene holders (Blanck and Wängberg, 1988) were sampled from the coastal environment. The glass discs were gently cleaned with paper tissues on all sides except the colonized one and sorted to achieve a homogenous set of glass discs for uptake and elimination studies.

Blue mussels, *Mytilus edulis*, with an average length of 30 mm (\pm 5 mm) were collected by hand at a depth of approximately 0.5-1 m. Brown shrimps, *Crangon crangon*, with an average length of 30 mm (\pm 5 mm) were collected using a push-net at a depth of approximately 1 m.

Periphyton, *Mytilus edulis* and *Crangon crangon* were collected from bays nearby Sven Lovén Centre for marine sciences (formerly Kristineberg Marine Research Station) at the Gullmarsfjord, on the west coast of Sweden. *Abra nitida* with an average length of 13 mm (\pm 1 mm) and sediment (silty clay >85% <4 μ m, TOC 4%) were collected using a benthic sledge (Warén) from a depth of approximately 100 m in the Gullmarsfjord.

2.2. Experimental solutions

Medetomidine was ¹⁴C-labelled in the methyl group directly bound to the chiral carbon. The specific activity was 0.51 GBq/mmol. It was synthesised using ¹⁴C-labelled alkyl substituted bromobenzene as starting material to prepare a Grignard reagent and supplied by the Marine Paint Formulation project. The medetomidine stock solution was dissolved in ethanol and experimental solutions were made by dissolving the stock solution in filtered seawater (GF/F for periphyton, 12 μ m for the other test species) to a nominal concentration of 20 μ g/l (100 nM). The concentration was chosen to achieve a ¹⁴C-level

1 above the detection limit but below a medetomidine concentration that cause
2 physiological effects in the test organisms. The concentration is not related to predicted
3 environmental concentrations (PEC), it is approximately 5000 times higher than PEC
4 values derived by modeling (unpublished data, Ohlauson C).

6 *2.3. Spiking of sediment*

7 Sediment was spiked by mixing 1.5 kg (wet weight) of sieved sediment (0.5 mm mesh
8 size) with 1.5 l of experimental medetomidine solution. The mixture was rotated in
9 darkness (15°C) for 24 h in a tumbler (Rotary tumbler Model QT66, Lortone, inc.,
10 Seattle, WA) before start of the experiment.

12 *2.4. Exposure and sampling*

13 Exposure of periphyton communities to ^{14}C -labelled medetomidine was performed in
14 glass containers (10x15x5 cm) with 300 ml of test medium placed on a shaker (60 rpm) at
15 $25 \mu\text{mol photons} \times \text{m}^{-2} \times \text{s}^{-1}$ with a 13:9 light dark regime and a constant temperature of
16 18°C). Five replicate containers were used each with 20 discs. After 48 hours of uptake
17 the discs were rinsed and transferred to clean containers with uncontaminated water for
18 another 48 hours to study elimination. To avoid reuptake of ^{14}C -labelled medetomidine
19 the water was changed again after 30 minutes. All solutions were changed every 24
20 hours. Samples were taken after 30 s, 1, 16 and 32 min (n=3) and at 1, 4, 8, 16, 24 and 48
21 hours (n=5), both during exposure to ^{14}C -medetomidine and uncontaminated water. The
22 sampled disc was immediately rinsed by quickly dipping it in water three times to
23 eliminate ^{14}C -medetomidine trapped in the interstitial water. The disc was then put into a

20 ml glass scintillation vial with the periphyton-colonised side upwards. Since fresh weight is difficult to estimate with good precision due to trapped interstitial water periphyton dry weight was measured at time 0 and after 48 hours in uncontaminated water (n=3). Discs were dried to constant weight ($\pm 1\%$) at 70°C and weighed. The periphyton biofilm was dissolved by incubating each disc in 0.5 ml HNO₃ at 100°C for 30 minutes. When the samples reached room temperature 0.5 ml H₂O₂ was added and the discs were incubated at 100°C for 30-60 minutes. The clean glass discs were rinsed in deionised water, dried in a fume hood and weighed again. When needed, periphyton dry weight was transformed to wet weight by multiplying with a factor of 12.5 (Sladecek and Sladeckova 1963; Sladecek and Sladeckova 1964).

For the *Crangon*, *Mytilus* and *Abra* experiments, three replicates were used as well as controls without medetomidine to control for mortality throughout the experimental period. *Mytilus edulis* and *Crangon crangon* were exposed to ¹⁴C-labelled medetomidine for 8 days in glass or plexiglass aquaria containing 2.5 l of experimental solution. The shrimps were individually separated in order to avoid cannibalism. No sediment or bottom gravel was added. Following exposure, they were kept in uncontaminated filtered seawater for an elimination period of 5 days. Solutions were changed every 24 hours. Temperature was kept constant at 15°C and a 14:10 light dark regime was used. Samples were taken at 0, 1 day, 2, 4, 6, and 8 days of exposure and following 6 and 12 h, 1 day, 2, 3 and 5 days in uncontaminated water for the *Mytilus* experiment and at 0, 1 day, 2, 4, 6, and 8 days of exposure and following 6 h, 1 day, 2, 3 and 5 days in uncontaminated water for the *Crangon* experiment. *Abra nitida* was exposed to ¹⁴C-labelled medetomidine for 2

1 days in glass aquaria containing one third of the spiked water-sediment mixture (0.5 kg
2 sediment and 0.5 l experimental solution). Following exposure they were kept in
3 uncontaminated filtered seawater and sediment for an elimination period of 5 days.
4 Experimental solutions were not changed during the exposure period of 48 hours but
5 during the elimination phase water was changed every 24 hours. Temperature was kept
6 constant at 10°C and a 14:10 light dark regime was used. Samples were taken at 0, 3, 6
7 and 12 h, 1 and 2 days of exposure and following 3, 6 and 12 h, 1, 2, 3, 4 and 5 days in
8 uncontaminated water. One specimen was taken from each replicate at each sampling
9 occasion.

11 2.5. Tissue preparation

12 Periphyton samples were prepared for scintillation counting in a similar way as for
13 photosynthesis measurements (Blanck and Wängberg, 1988) i.e. dissolved with 1 ml of
14 dimethylsulfoxide and left for at least 10 min before adding 9 ml of scintillation cocktail
15 (Ready Safe™, Beckman Instruments). Vials were shaken vigorously to obtain
16 homogeneous samples prior to scintillation counting the next day. For brown shrimp and
17 *Abra nitida* whole animals, without exoskeleton or shells, were used. The soft tissue from
18 blue mussel was homogenised and a subsample of approximately 150 mg from each
19 mussel was used. The tissue of the experimental animals was placed in scintillation vials
20 and 2 ml of Soluene® (PerkinElmer) was added to each sample. All vials were placed in
21 an incubator (50°C, Heidolph incubator 1000 polymax 1040) and gently revolved until
22 the tissue was completely dissolved. 10 ml of scintillation cocktail (Hionic-Flour,

PerkinElmer) were added, samples were shaken and allowed to stand over night before scintillation counting.

2.6. Water and sediment samples

From the periphyton experiment water samples were taken for scintillation counting (0.5 ml) at 1, 16, 32 min and 1, 4, 8, 16, 24 and 48 hours (n=1), both during exposure to ^{14}C -medetomidine and uncontaminated water. 9 ml of scintillation cocktail (Ready Safe™, Beckman Instruments) was then added to each sample.

Water samples (2 ml) for later scintillation counting were also taken from the *Mytilus* and *Crangon* experiments at the start of the experiment and after 1 day (before solution change), 1 day (after solution change), 2, 3, 4, 5, 6, 7 and 8 days (n=3) and from the *Abra* experiment, at the start of the experiment and after 1 and 2 days (n=3). Sediment samples (100 mg) were taken at the start of the experiment and after 1 and 2 days (n=3). 10 ml of scintillation cocktail (Insta-gel Plus, Chemical Instruments) were added to water and sediment samples. All samples were shaken and allowed to stand over night before scintillation counting.

3. Results

The concentration of ^{14}C -labelled medetomidine reached steady state in the periphyton communities after 48 hours with a rapid uptake during the first 8 hours of exposure followed by a stabilisation after 24 hours (Figure 1). After the first 8 hours the concentration in periphyton was 15.5 $\mu\text{g/g}$ fresh weight (FW) (77.2 nmoles/g) and at steady state the concentration was 22.7 $\mu\text{g/g}$ FW (113 nmoles/g). Mean water

concentration was 19.0 $\mu\text{g/l}$ (94.6 nM) during the uptake phase of the experiment, giving a bioconcentration factor of 1195 and 14941 if presented as l/kg dry weight (Table 1 and Figure 5). Elimination was much faster than the uptake with a half-life of less than 1 hour. The steady state level reached was 2.80 $\mu\text{g/g}$ FW (14.0 nmoles/g) in periphyton giving a BCF of 722 at the end of the elimination period.

Steady state was reached within 24 hours of exposure for both *Mytilus edulis* (Figure 2) and *Crangon crangon* (Figure 3). The bioconcentration factor at the end of exposure is calculated to 134 and 2.8 respectively (Table 1). BCF did not change significantly over time (24 hours till 8 days) for either *Mytilus* or *Crangon* (Figure 6). Elimination of medetomidine was also rapid with a half-life of less than 6 h in *Mytilus* and 6-24 h in *Crangon*. The mean water concentration was 19.2 $\mu\text{g/l}$ (95.8 nM) in the *Mytilus* experiment and 16.0 $\mu\text{g/l}$ (80.0 nM) in the *Crangon* experiment. The tissue concentration in *Crangon* is lower on a few sampling occasions (48 and 96 h) compared to after 24 h exposure because of a lower water concentration than expected, but increased as soon as the water concentration increased (Figure 3). However, BCF values were stable between 1.5 and 2.8 (Figure 6). The concentration of medetomidine in the tissue of *Abra nitida* has probably not reached steady state after the exposure period of 48 hours (Figure 4 and 5), thus a correct BAF cannot be calculated. However, a ratio of the concentration in tissue after 48 hours and the concentration in all compartments (sediment and water) gives a factor of 2.6. This might be an underestimation compared to a true BAF for *Abra nitida*. The elimination half-life of medetomidine is approximately 96-120 h in *Abra* (Table 1). The mean water concentration in the *Abra* experiment was 0.55 $\mu\text{g/l}$ (2.75 nM)

1 and the mean sediment concentration 18.1 $\mu\text{g/kg}$ (90.6 nmoles/g) demonstrating
2 medetomidine's ability to adsorb to sediment particles.
3 No significant mortality ($< 5\%$) and no difference between controls and treatments were
4 observed in any of the experiments.

6 **4. Discussion**

7 Medetomidine is shown to have a maximal bioconcentration factor of 1195, which
8 occurred in periphyton at steady state. The rate of uptake is quite high in periphyton
9 communities compared to other test species. This is probably due to periphyton's large
10 surface area and high lipid content (Wang et al., 1999). Whether medetomidine is rapidly
11 mobile in and out of the essentially unicellular periphyton organisms, or only adsorbed to
12 the surface cannot be conclusively clarified in the present study. However, the fit to a
13 two-compartment bioconcentration model (Newman, 1995) is consistent with a surface
14 adsorption with a rapid first phase followed by a secondary slower accumulation (data not
15 shown). The rapid elimination ($t_{1/2} < 1$ h) implies as well that most of the medetomidine is
16 adsorbed and only a small fraction is taken up by the algal cells since the concentration
17 decreased immediately when exposure ended. The medetomidine remaining in
18 periphyton after 48 hours is 79 nmoles/g dry weight corresponding to 2.3 pmoles per
19 square centimeter.

20 For brown shrimp a considerably lower bioconcentration factor (2.8) is found compared
21 to blue mussel (BCF 134). A similar result has previously been observed in these two
22 species when exposed to nonylphenol (Ekelund et al., 1990), demonstrating again that
23 bioaccumulation is species dependent, with a BCF of 100 for brown shrimp and 3400 for

1 blue mussel. As a consequence, bioaccumulation should always be studied in several
2 species to get the full range of possible bioconcentration and bioaccumulation factors.
3 The experiment with *Abra nitida* suggests that it may take longer time to reach a steady-
4 state condition in sediment systems and the elimination of medetomidine also seems to be
5 slower. One possible explanation for the slower uptake of medetomidine in *A. nitida*,
6 compared to the other species tested, might be because of medetomidine's ability to
7 interact with surrounding molecules, e.g. metals in the sediment (Sundberg and Martin,
8 1974), and might therefore be transported through the gut of the organisms without being
9 readily absorbed by the tissues. Further experiments with longer exposure periods and
10 other sediment-eating organisms are needed to validate the results from this study which
11 indicates that only minor uptake of medetomidine is through the sediment. However, the
12 concentration used in the present study (20 µg/l, 100 nM) may have been high enough to
13 impair the bivalves feeding behaviour thus decreasing the uptake of medetomidine.
14 Earlier studies have shown a decreased sediment reworking activity already at 10 nM but
15 the amount of faeces produced were however not affected up to 375 nM indicating that
16 the bivalves fed normally, but at a different layer in the sediment, when exposed to
17 medetomidine (Bellas et al., 2006). The animals used in the present study buried into the
18 sediment and stayed buried throughout the experiment. However, feeding activity was not
19 measured and the sediment was not removed from the gut prior to analysis.
20 Our results show, in agreement with mammalian data, a rapid uptake and elimination of
21 the ¹⁴C-labelled molecule in all test organisms, except for *Abra*. Medetomidine is
22 biotransformed in the hepatic tissue in mammals and not more than 5 % is excreted as
23 unchanged medetomidine (Salonen, 1989). *In vitro* studies in rat show that medetomidine

only have one major metabolite, hydroxymedetomidine, while three major metabolites and some minor metabolites were found *in vivo*. Two different pathways are suggested after hydroxylation and they depend on the concentration of medetomidine. None of the major metabolites have α -receptor activity, thus this transformation removes the active molecule (Salonen and Eloranta, 1990). However, the study indicates that methylation might occur to a very minor extent which could produce a metabolite that would be slowly eliminated and thus accumulate in fatty tissues during long time exposure (Salonen and Eloranta, 1990). Metabolites are not analysed in the present study and it cannot be entirely excluded that a small amount of the N-methylated conjugate is formed. Further studies with collection of excretion products and analysis of metabolites are needed to investigate whether this potentially bioaccumulating metabolite is produced in invertebrates as well. The medetomidine used in this experiment was also only ^{14}C -labelled in one position, which makes it impossible to distinguish between intact medetomidine and metabolites containing the labelled carbon. For further studies medetomidine with several ^{14}C -labels will be used.

A comparison of medetomidine to other antifouling substances regarding bioaccumulative potential displayed varying results. Tributyltin, TBT, which was prohibited for use in 2008 due to the environmental damages caused by the substance (Champ, 2003), has a octanol-water partitioning coefficient in the same range as medetomidine with $\log K_{ow}$ of 3.74 (Laughlin et al., 1986). BCF for TBT was also in the same range as for medetomidine regarding algae with values above 1000. For filtrating molluscs the BCF for TBT was however much higher, with values above 100 000 reported (Ranke and Jastorff 2000 and references therein). Studies have also

demonstrated that $\log K_{ow}$ is not a good predictor of bioconcentration or bioaccumulation for TBT with experiments resulting in much higher BCF or BAF than predicted (Huang and Wang, 1995). Another antifouling biocide, Irgarol 1051, have $\log K_{ow}$ values between 2.8 and 3.95 recorded. Like TBT and medetomidine Irgarol also has a rather high affinity for algae and water plants with BCFs of at least 1000 published. Other organism groups have lower BCFs for Irgarol, with values around 200 reported for fish (Ranke and Jastorff, 2000 and references therein). Both TBT and Irgarol are good examples that $\log K_{ow}$ does not display the full risk of bioaccumulation of a substance. Experimental studies of several organism groups are necessary to separate substances with a high potential to accumulate from the other non-accumulative substances with similar octanol-water partitioning coefficients.

5. Conclusion

Medetomidine seems to be a new antifouling compound with rapid elimination and a BCF below the bioaccumulative classification level ($BCF < 2000$) set by the European Chemicals Bureau (Technical Guidance Document on Risk Assessment EUR 20418 EN/2). However, further studies on degradation and metabolites are necessary before a complete risk assessment can be produced.

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4

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1 Figure captions

2 Figure 1

3 Bioaccumulation in periphyton communities exposed to a nominal concentration of 20
4 $\mu\text{g/l}$ (100 nM) of medetomidine. ● tissue concentration of medetomidine during exposure
5 Δ tissue concentration of medetomidine during elimination \square water concentration of
6 medetomidine.

7
8 Figure 2

9 Bioaccumulation in *Mytilus edulis* exposed to a nominal concentration of 20 $\mu\text{g/l}$ (100
10 nM) of medetomidine. ● tissue concentration of medetomidine during exposure Δ tissue
11 concentration of medetomidine during elimination \square water concentration of
12 medetomidine during exposure. Data show the median value \pm range (n=3).

13
14 Figure 3

15 Bioaccumulation in *Crangon crangon* exposed to a nominal concentration of 20 $\mu\text{g/l}$ (100
16 nM) of medetomidine. ● tissue concentration of medetomidine during exposure Δ tissue
17 concentration of medetomidine during elimination \square water concentration of
18 medetomidine during exposure. Data show the median value \pm range (n=3).

19
20 Figure 4

21 Bioaccumulation in *Abra nitida* exposed to a nominal concentration of 20 $\mu\text{g/l}$ (100 nM)
22 of medetomidine. ● tissue concentration of medetomidine during exposure Δ tissue

concentration of medetomidine during elimination ■ total concentration of medetomidine in the sediment and water during exposure. Data show the median value +/- range (n=3).

Figure 5

Bioaccumulation in periphyton and *Abra nitida* exposed to a nominal concentration of 20 µg/l (100 nM) of medetomidine. ○ Periphyton BCF during exposure ■ a ratio of the concentration in tissue and the concentration in all compartments (sediment and water) during exposure for *Abra nitida*. Data show the median value +/- range (*Abra* n=3, periphyton n=5).

Figure 6

Bioaccumulation in *Crangon crangon* and *Mytilus edulis* exposed to a nominal concentration of 20 µg/l (100 nM) of medetomidine. ○ *Crangon crangon* BCF during exposure ■ *Mytilus edulis* BCF during exposure. Data show the median value +/- range (n=3).

Table 1

Bioconcentration factors in organisms exposed to medetomidine.

Parameter	Community/Species			
	Periphyton	<i>Mytilus edulis</i>	<i>Abra nitida</i>	<i>Crangon crangon</i>
Mean medetomidine conc. in water (µg/l)	19.0	19.2	0.55	16.0
conc. in water (nM)	95	96	2.75	80
Mean medetomidine conc. in sediment (µg/kg)	-	-	18.1	-
Conc. of medetomidine in tissue at end of exposure ^a (µg/g FW)	22.66	2.54	0.049	0.044
Bioconcentration factor (l/kg FW)	1195 (14941) ^b	134	2.6 ^c	2.8
Estimated half-life $t_{1/2}$ (h)	<1	<6	96-120	6-24

^afor periphyton communities, *Mytilus edulis* and *Crangon crangon* steady-state was reached within the exposure period but probably not for *Abra nitida*

^bbased on dry weight

^cbioaccumulation factor calculated without steady-state reached

Figure 1

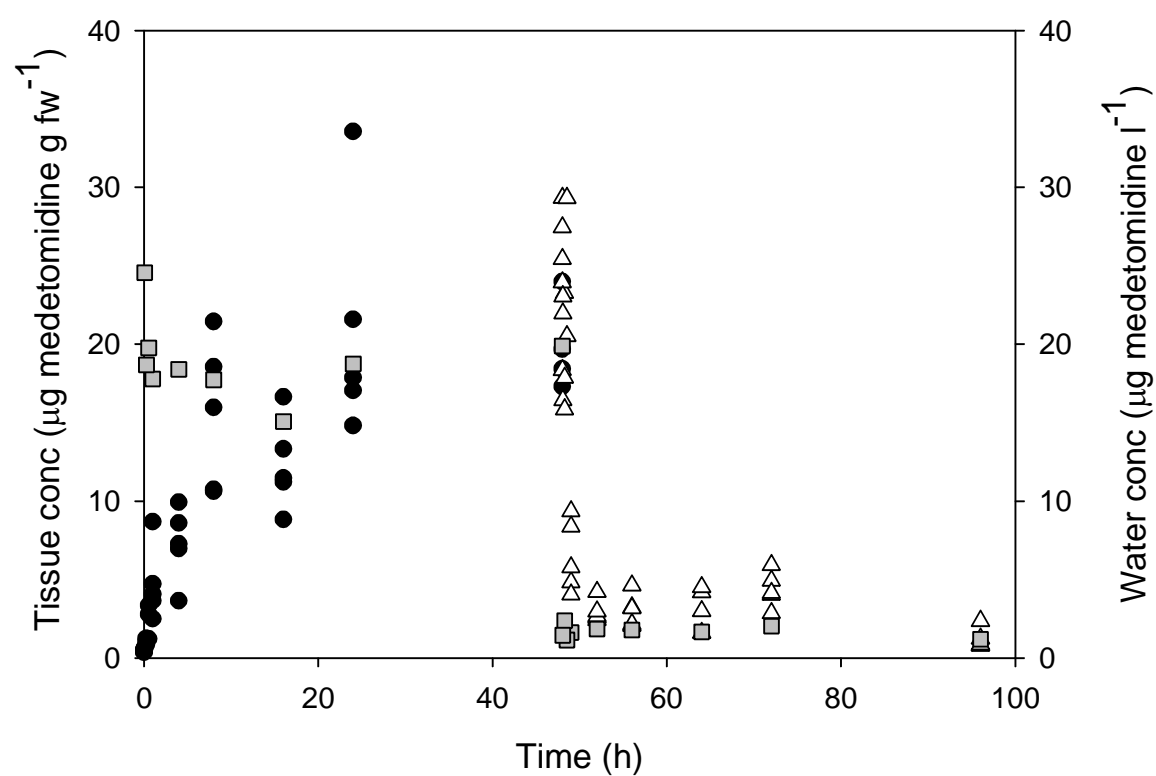


Figure 2

Figure 2

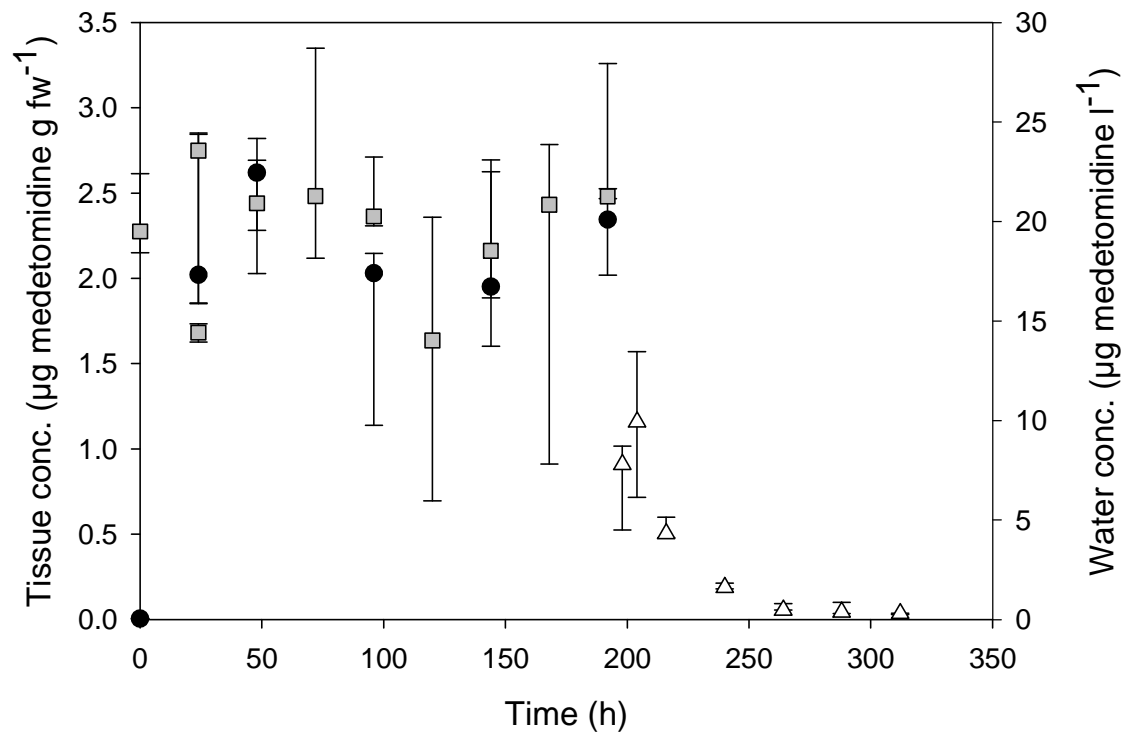


Figure 3

Figure 3

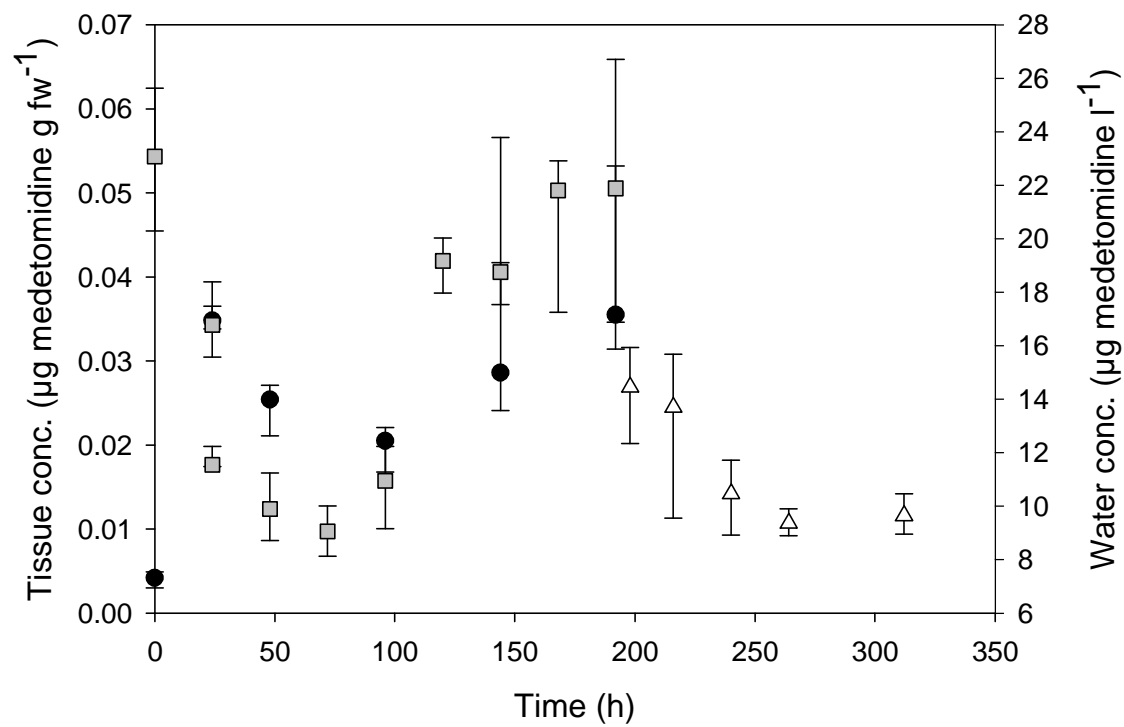


Figure 4

Figure 4

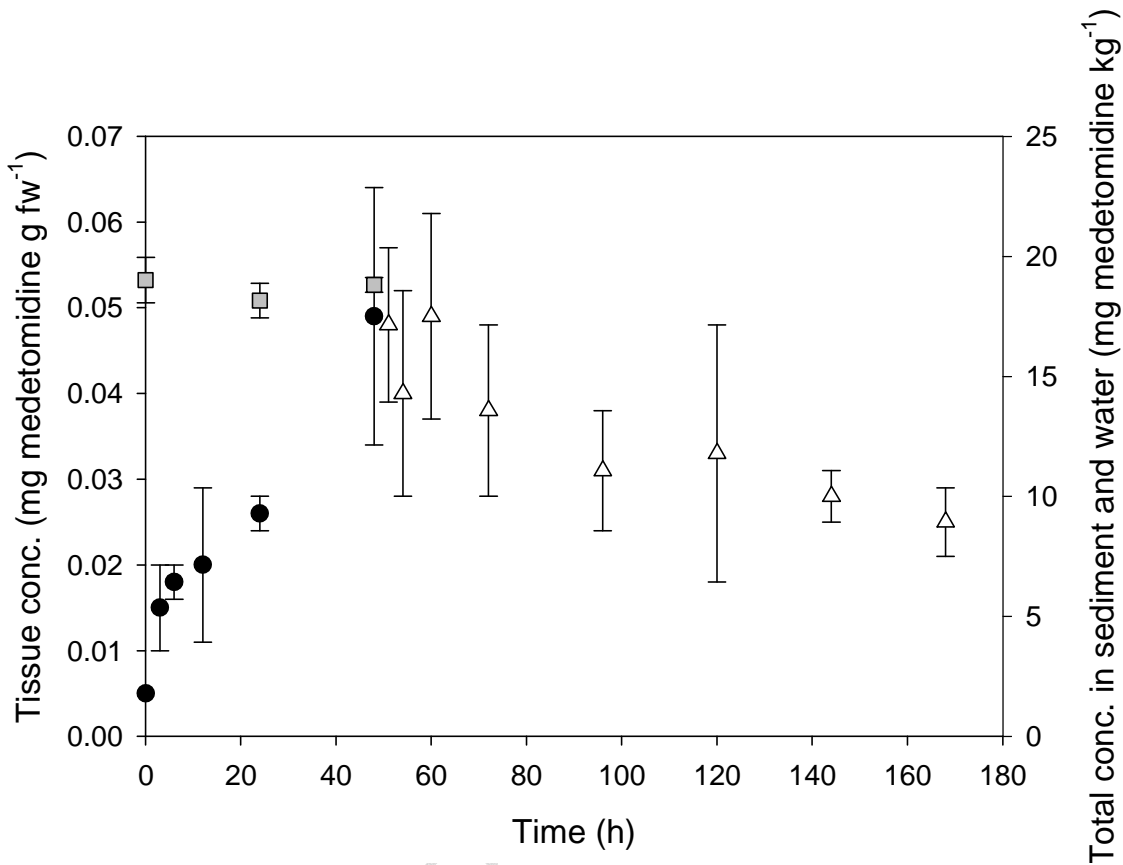


Figure 5

Figure 5

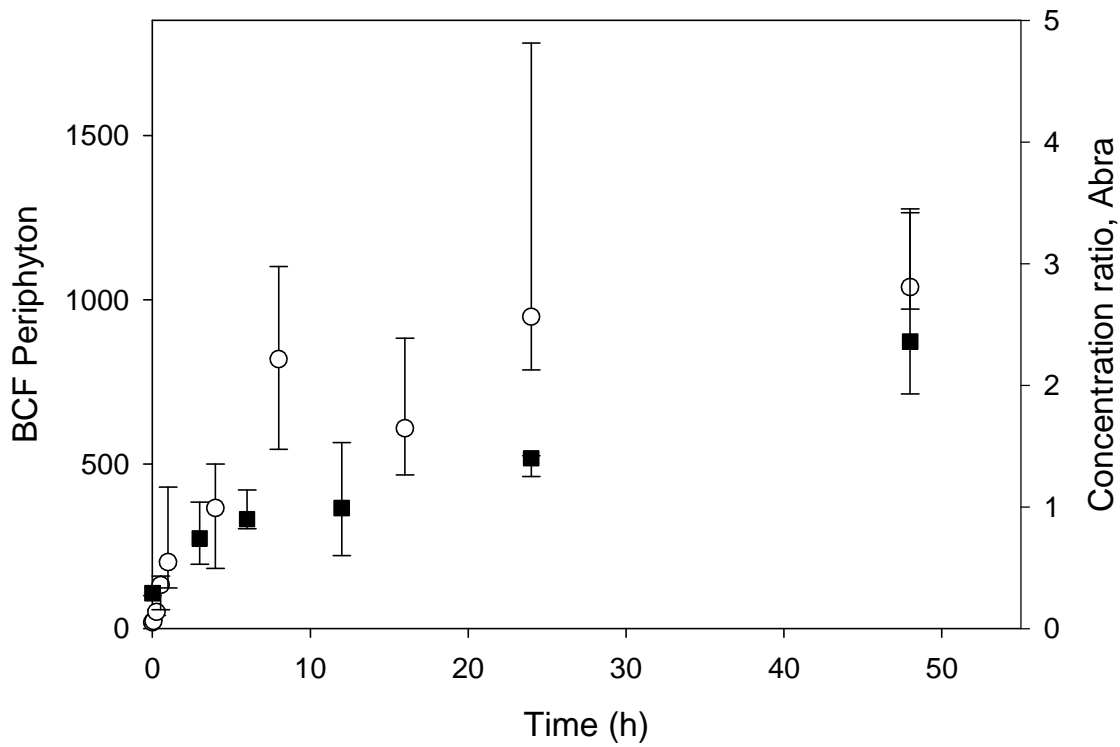


Figure 6

Figure 6

