

**Oskarshamn and Forsmark
site investigation**

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bacteria in Laxemar and Forsmark
and biomass of Reed (*Phragmites
australis*) in Lake Frisksjön**

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Abstract

Biomass of benthic bacteria was investigated at one occasion in marine basins in the Laxemar area, the Forsmark area and in Lake Frisksjön. Bacterioplankton biomass was investigated once in marine basins in Laxemar and Forsmark and at two occasions in Lake Frisksjön. In addition, biomass of *Phragmites australis* (reed, both root and above ground biomass) in Lake Frisksjön was studied during the summer 2006.

Bacterioplankton biomass in the marine basins varied between 22 and 27 in Laxemar and between 15 and 36 mg C m⁻³ in Forsmark. In Lake Frisksjön bacterioplankton biomass varied was 18 mg C m⁻³ in June and 34 mg C m⁻³ in August. In the marine basins the benthic bacterial biomass varied between 3.4 and 12 g C m⁻² in Laxemar and between 1.4 and 5.0 g C m⁻² in Forsmark. In Lake Frisksjön the benthic bacterial biomass was 5.3 g C m⁻² (standard deviation 1.0).

There was a large variation between sampling points in root biomass as well as in above ground biomass of reed. The root biomass varied between 1 and 11 kg dw m⁻² (mean 5 kg dw m⁻²). The above ground biomass varied between 0.1 and 1.4 kg dw m⁻² (mean 0.7 kg dw m⁻²).

Sammanfattning

Under sommaren 2006 undersöktes biomassan av bentiska bakterier och bakterioplankton i några marina bassänger i Laxemar och Forsmark och i Frisksjön. I Frisksjön togs prover för bedömning av bakterioplanktonbiomassan även i augusti. Dessutom studerades biomassan av vass (*Phragmites australis*), såväl rot som ovanjordbiomassa, i Frisksjön.

För att undersöka bakterioplanktonbiomassan togs tre vattenprover från de marina bassängerna och två vattenprov från Frisksjön. En Willnerhämtare användes för att ta sedimentprover (8 från de marina bassängerna och 3 från Frisksjön) och de översta 5 cm användes för studier av bentiska bakterier. Bakterierproverna konserverades med formalin i fält och färgades in med acridineorange innan de filtrerades på ett polykarbonatfilter (0,22 μm porstorlek) och räknades i mikroskop. Bakterioplanktonbiomassan varierade mellan 22 och 27 mg C m^{-3} i de marina bassängerna i Laxemar och mellan 15 och 36 mg C m^{-3} i Forsmark. I Frisksjön var bakterioplanktonbiomassan 18 mg C m^{-3} i juni och 34 mg C m^{-3} i augusti. Biomassan av bentiska bakterier varierade mellan 3,4 och 12 g C m^{-2} i de marina bassängerna i Laxemar och mellan 1,4 och 5,0 g C m^{-2} i Forsmark. I Frisksjön varierade biomassan av bentiska bakterier mellan 4,2 och 6,1 g C m^{-2} (medel 5,3 g C m^{-2}).

10 prover av rotbiomassa och 20 prover av ovanjordbiomassa av vass undersöktes. Vid insamlandet av rotbiomassan av vass skars först ovanjordbiomassan bort. Sen grävdes rötterna upp med hjälp av en spade inom en känd area (0,25×0,25 m). Rötterna transporterades till laboratoriet där de rengjordes och vägdes. Delprov torkades över natt i 90 °C för att bestämma torrvikten. Ovanjordbiomassan av vass insamlades inom en känd area (0,25×0,25 m) och transporterades till laboratoriet och vägdes. Liksom för rotbiomassan torkades delprov i 90 °C för att kunna räkna ut torrvikten. Det var stor variation i rotbiomassa och ovanjordbiomassa mellan provpunkterna. Rotbiomassan varierade mellan 1 och 11 $\text{kg torrsvikt m}^{-2}$ och ovanjordbiomassan varierade mellan 0,1 och 1,4 $\text{kg torrsvikt m}^{-2}$.

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1 Introduction

This document reports the results gained by limnic and marine investigations during the summer 2006 (June 19–20, August 31, September 6–7), which is one of the activities performed within the site investigation at Oskarshamn and Forsmark. This activity is a part of the annual “field weeks” performed at the two sites, by Surface Net, a work group modelling the sites surface systems. The original results are stored in the database SICADA, and data is traceable by the P-report number.

In June 2006 bacterial biomass (both planktonic and benthic) was investigated in Lake Frisksjön and in marine basins in the coastal area situated in the site investigation area in Oskarshamn (Figure 1-1) and Forsmark (Figure 1-2). In Lake Frisksjön also the biomass of reed (*Phragmites australis*) was investigated. The root biomass was investigated in June and the above ground biomass was investigated in the end of the summer (31 August).

Lake Frisksjön is a humic lake and the poor light condition in this kind of lake often suppresses phytoplankton. Instead, bacteria often play a significant role for the ecosystem as they can utilise dissolved organic carbon. Although bacterioplankton biomass has been studied in many humic lakes, the variation between lakes can be high, and it is therefore important to elucidate the size of the bacterioplankton pool in Lake Frisksjön to be able to make accurate estimation of their importance for the ecosystem. The biomass of sediment bacteria is often high but is less well studied than bacterioplankton and studies from humic lakes are practically absent.

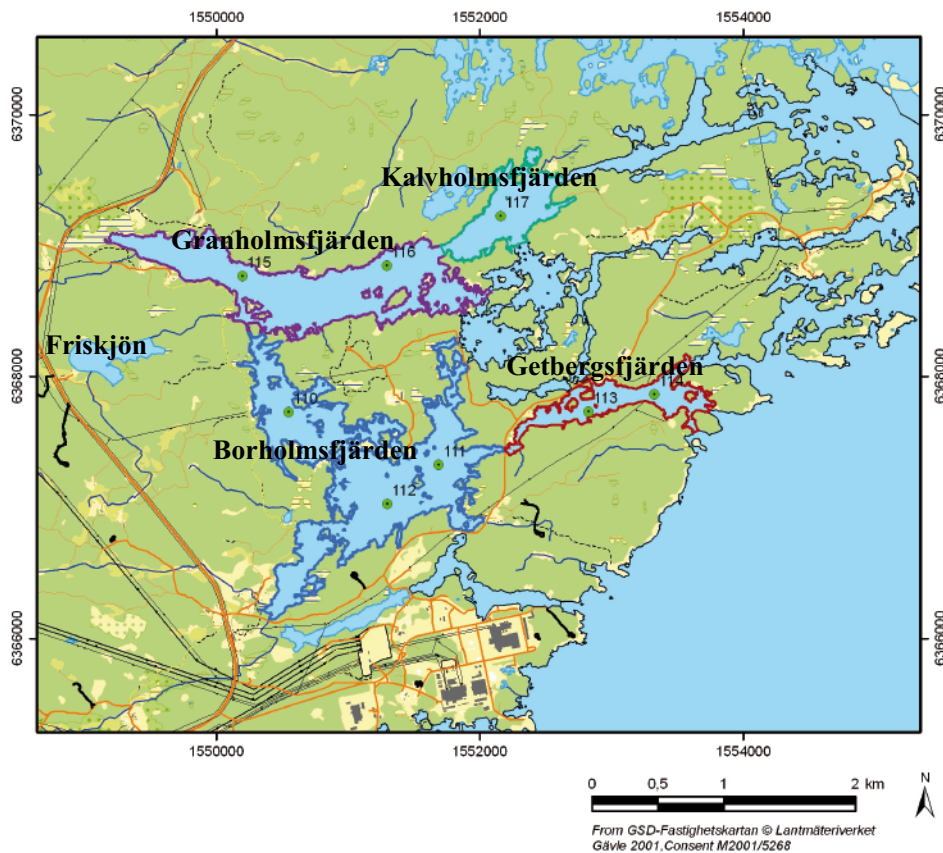


Figure 1-1. Lake Frisksjön, the sampled marine basins and the sample numbers.

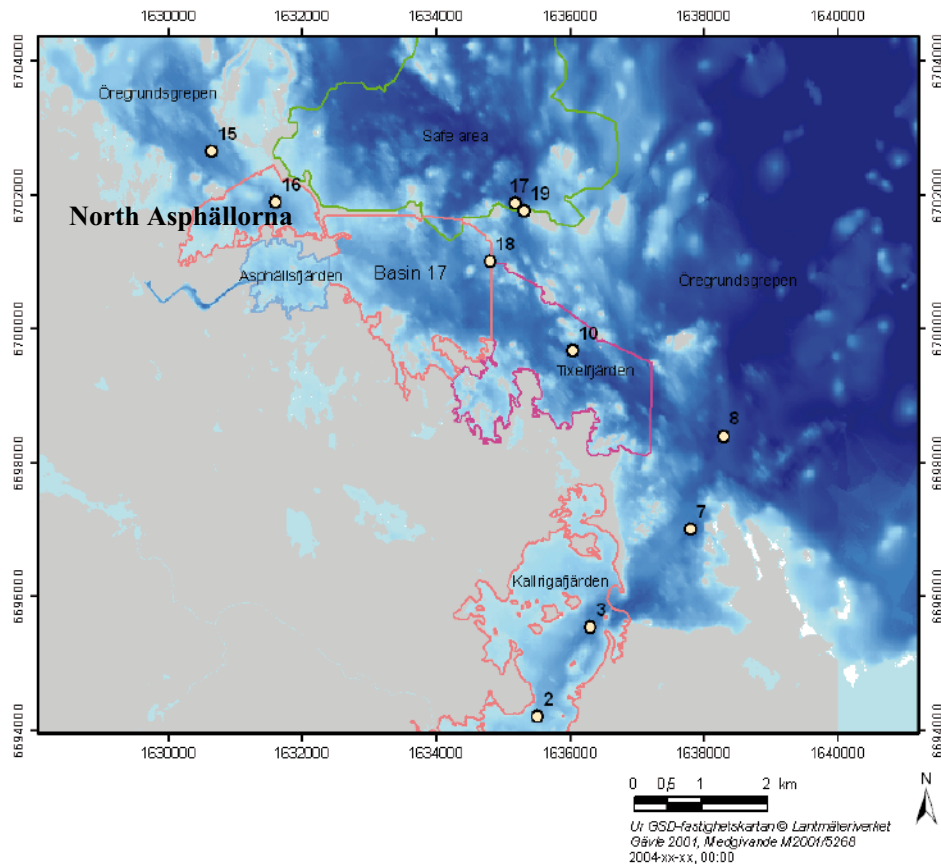


Figure 1-2. The sampled sites in Forsmark and the marine basin delimitations.

In the marine areas in the Laxemar area, the importance of benthic bacteria and bacterioplankton has been modelled in an ecosystem carbon budget /Wijnbladh et al. 2006/. The importance of benthic bacteria is large, but in the budget not balanced, either due to an underestimation of bacterial growth or an overestimation of them as energy source for detritivores. The data on bacteria used in the modelling was not from the Laxemar area, and because of the uncertainties of the model results it is important to estimate the amount of bacteria in the actual site. As both Laxemar and Forsmark is modelled quantitatively, site specific data from Forsmark is also needed.

According to a mass balance for Lake Frisksjön, reed contributes by far most to the total biomass of biota /Sobek et al. 2006/. Due to poor knowledge of the reed biomass in the lake, this calculation is to some extent based on data from the archipelago in the Simpevarp area and not from lake specific data. The shoot biomass of reed in Lake Frisksjön has been investigated once in 2004 /Aquilonius 2005/. However, the investigation was done in relatively thin reed stands and the root biomass was not included. Therefore additional data is needed to achieve reliable estimates of the reed biomass.

2 Objective and scope

Both bacteria and reed are assumed to be important components of humic lakes and marine ecosystems and the biomass estimates of these organisms are therefore important tools for the modelling of these ecosystems. Parameters measured in this study were:

1. Bacterioplankton and sediment bacterial biomass in coastal areas in Forsmark and Laxemar.
2. Bacterioplankton and sediment bacterial biomass in Lake Frisksjön.
3. Biomass of reed (both root and above-ground biomass) in Lake Frisksjön.

3 Equipment

3.1 Description of equipment tools

3.1.1 Bacterial biomass

A Ramberg tube (1.5 m) and a bucket were used to sample water from Lake Frisksjön. Sediment samples were taken with a Willner core sampler. Pipette and plastic jars were used to take sub-samples of bacteria. Filtered formaldehyde was used to preserve both bacterioplankton and sediment bacteria. Polycarbonate membrane filters (0.22 μm pore size), acridine orange and a Nikon Labophot microscope was used to count the bacteria.

3.1.2 Reed biomass

A wooden frame (size 0.25×0.25 m), a pair of pruning shears, a spade and plastic bags were used.

4 Execution

4.1 General

Bacterial biomass was estimated by sampling of water and sediments from Lake Frisksjön and from the coastal areas of Laxemar and Forsmark (see Figure 1-1 and 1-2). Known volume of water and sediment was preserved with formaldehyde and samples were brought back to the laboratory where bacteria were counted in a microscope.

Below ground biomass of reed was investigated in June and above ground biomass was investigated in August 2006.

4.2 Execution of field work

4.2.1 Bacterial biomass

Forsmark

Nine water samples were collected in Forsmark the 6th and 7th of September 2006. The sampled sites were 2, 3, 7, 8, 10, 15, 16, 17 and 18, see Figure 1-2. The samples were collected with a Ramberg tube. Sub-samples of 20 ml were preserved with formaldehyde, final concentration 4%.

Four sediment samples (site 2, 3, 16, 19, see Figure 1-2) were collected in Forsmark the 6th and 7th of September 2006. Sediment samples were collected with a Willner core sampler in site 2, 3 and 16 and the top 0–5 cm were transferred to plastic jars from which 1 ml sub samples were taken. The sub samples were preserved with formaldehyde, final concentration 4%. In site 19 the substrate did not allow this sampling technique and in this site two 5 cm cores were taken by SCUBA divers. The samples were treated in the same way as presented above.

Laxemar

Three pooled water samples (each consisting of two or three samples) were sampled from the marine basins on 19th of June. The water sample from Kalvholmsfjärden (see Figure 1-1) was included in the pooled sample for Granholmsfjärden. On 20 June and on August 31, pooled water samples were collected from Lake Frisksjön. The samples were collected with a Ramberg tube. Sub-samples of 20 ml were preserved with formaldehyde, final concentration 4%.

Eight sediment samples from the marine basins were collected on the 19th of June, and on June 20, three sediment samples were collected from Lake Frisksjön. Sediment samples were taken with a Willner core sampler and the top 0–5 cm were transferred to plastic jars from which 1 ml sub-samples were taken. The sub-samples were preserved with formaldehyde, final concentration 4%.

The multiple samples from each of the basins Borholms-, Granholms- and Getbergsfjärden were pooled according to Table 4-1. Sample 117 represented Kalvholmsfjärden.

Table 4-1. Pooling of multiple samples from the marine basins in Laxemar.

	Borholmsfjärden	Granholmsfjärden	Kalvholmsfjärden	Getbergsfjärden
Samples	110–112	115, 116	117	113, 114

4.2.2 Reed biomass

On June 19 and 20, root biomass of reed was investigated in Lake Frisksjön. A total of 10 samples were taken from three different stations in the lake (Table 4-2). A wooden frame (size 0.25×0.25 m) was placed in the reed belt and all roots within the frame were collected. The roots were brought back to the laboratory in plastic bags. In the laboratory the roots were rinsed, dead roots and any other species than reed was removed and wet weight of live reed roots was measured. Dry weight was analysed on 16 small sub samples which were dried in an oven (90°C) over night.

On August 31, above-ground samples of reed were investigated. 20 samples of reed shoots were sampled within a wooden frame (size 0.25×0.25 m), which was placed in the reed belt at 4 different stations (Table 4-2). The reed was brought back to the laboratory and weighted. Dry weight was analysed in 11 sub-samples which were dried in an oven (90°C) over night.

4.3 Data handling/post processing

4.3.1 Bacterial biomass

Bacterioplankton and sediment bacteria were counted and measured with an epifluorescence microscope (Nikon labophot). Sediment samples were first sonicated (1 min, 75 W); the other samples directly stained with acridine orange and filtered through 0.22 µm polycarbonate filters. A total of 400 cells were counted and at least 100 were measured for each sample.

Bacterial biomass was calculated according to /Loferer-Krössbacher et al. 1998/, using the formula $DW = N \times 435 \times V^{0.86}$ (where V is bacterial cell volume and N number of bacteria), and assuming that 50% of the dry weight was carbon.

4.3.2 Reed biomass

An average dry weight to wet weight ratio was calculated from sub-samples of dry weight for both root biomass and shot biomass. Dry weight per square meter was calculated by multiplying the wet weight per square meter with the average dry weight to wet weight ratio. Carbon biomass was calculated by multiplying the calculated dry weight with the conversion factor 0.395. This conversion factor is for *Scirpus acicularis* /Kautsky 1995/, but are assumed to be valid also for reed.

4.4 Nonconformities

No nonconformities was noted.

Table 4-2. Stations for reed sampling with x and y-coordinates and number of samples taken at each station. The coordinates for the actual sampling sites varies ± 20 m.

Station	x-coordinate	y-coordinate	No of root biomass samples	No of above-ground samples
1	6368179	1549655	5	10
2	6368129	1549404	2	2
3	6368118	1549323	3	5
4	6368092	1549178	–	3

5 Results

The original results are stored in the database SICADA and is traceable by P-report number P-06-232.

5.1 Bacterial biomass

The biomass of bacterioplankton and sediment bacteria is reported in Appendix 1 and in Figure 5-1 and 5-2 (Laxemar), 5-3 and 5-4 (Forsmark).

5.1.1 Laxemar

In Lake Frisksjön bacterioplankton biomass was 18 mg C m⁻³ in June and 34 mg C m⁻³ in August. The biomass of sediment bacteria in June varied between 4.2 and 6.1 g C m⁻² (mean 5.3 g C m⁻²). There was 1.3 10⁶ cells ml⁻¹ in the water and 5×10⁹ cells ml⁻¹ in the sediments. The total bacterial biomass in the lake was 4–7.6 kg C in bacterioplankton (calculated for the volume 257,000 m³) and 590 kg C in sediment bacteria (calculated for the lake area 0.13 km²).

The bacterioplankton biomass in the marine basins varied between 22 and 27 mg C m⁻³ (corresponding to 1.2 and 1.3×10⁶ cells ml⁻¹) (Figure 5-1).

The biomass of sediment bacteria varied between 3.4 and 12 g C m⁻² (corresponding to 3.0 and 7.3×10⁹ cells ml⁻¹). Comparably high biomasses were found in the basin Getbergsfjärden (mean 11 g C m⁻²) and similar and lower (mean 4.9, 4.9, 5.5 g C m⁻²) in the other basins (Figure 5-2).

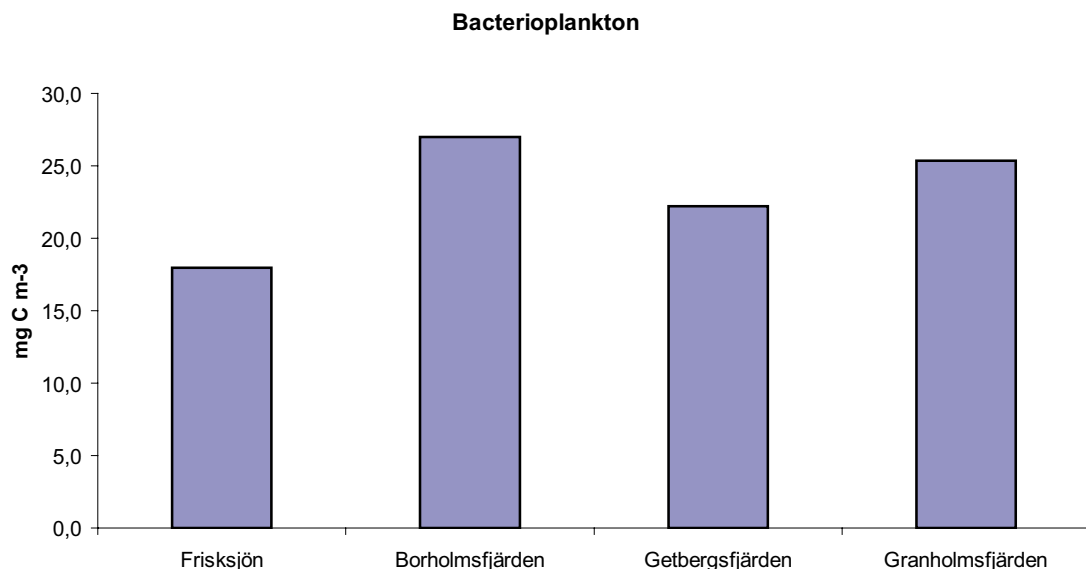


Figure 5-1. Bacterioplankton biomass in Lake Frisksjön and in the marine basins in June.

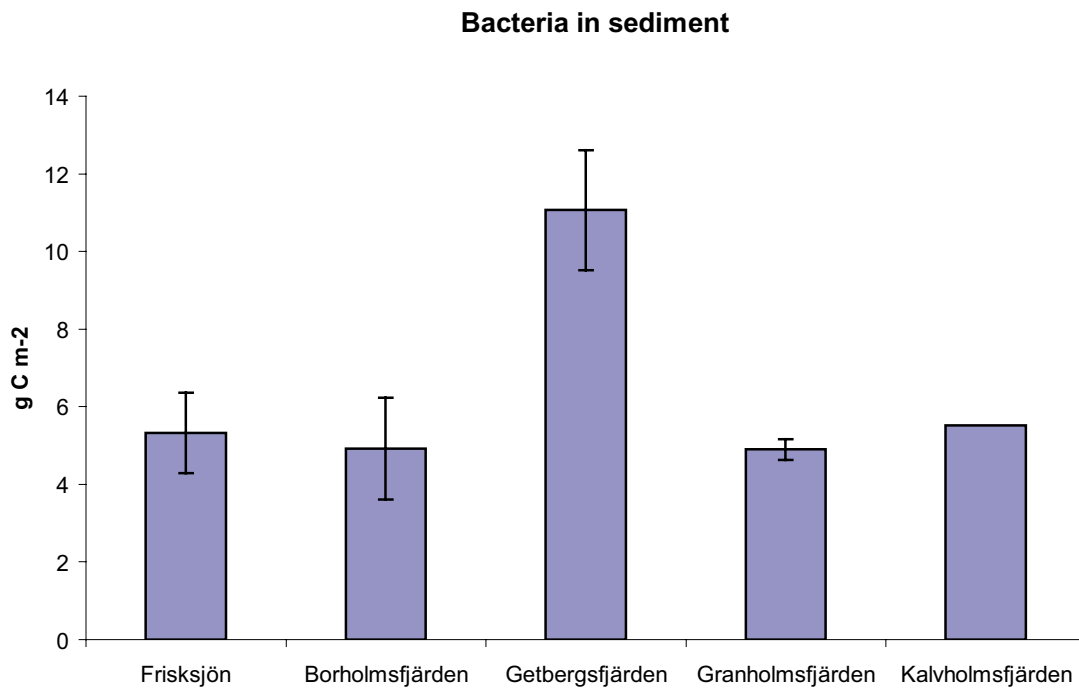


Figure 5-2. Biomass of benthic bacteria in Lake Frisksjön and in the marine basins.

5.1.2 Forsmark

Biomasses are presented in appendix 1 and means according to Table 5-1.

The bacterioplankton biomass in the marine basins varied between 15 and 36 mg C m⁻³ (corresponding to 1.2 and 2.8×10⁶ cells ml⁻¹). The mean for the three basins Kallrigafjärden, North Asphällorna and Öresundsgrepen were 25, 20 and 26 mg C m⁻³ respectively (Figure 5-3).

The biomass of sediment bacteria varied between 1.4 and 5.0 g C m⁻² (corresponding to 1.2 and 4.3×10⁹ cells ml⁻¹) the high biomasses found in the more sheltered basin Kallrigafjärden and the area north of Asphällorna (see Figure 5-2 and Figure 5-3) and the low biomass in site 19 (see Figure 1-2 and 5-4).

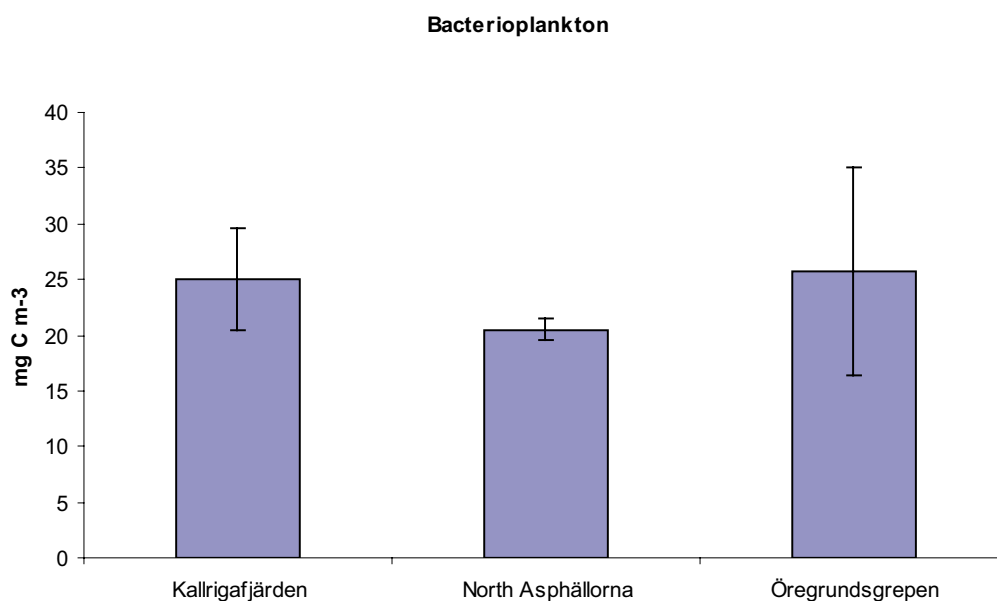


Figure 5-3. Bacterioplankton biomass in the marine basins in Forsmark.

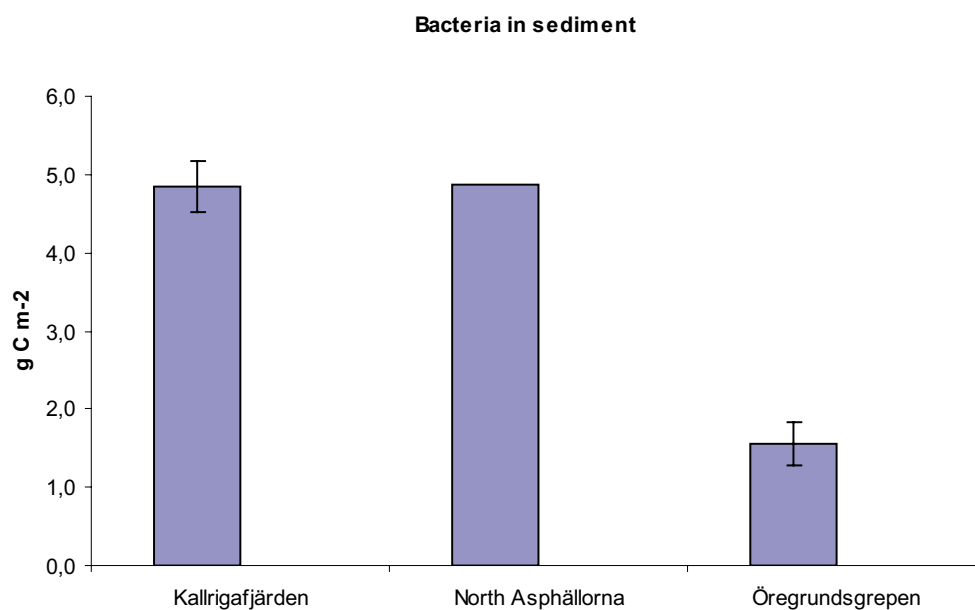


Figure 5-4. Biomass of benthic bacteria in the marine basins in Forsmark.

5.2 Reed biomass

There was a large variation in the root biomass of reed in Lake Frisksjön varying between 1 and 11 kg dry weight m⁻² (average 5, SD 4 kg m⁻²). The dry weight per wet weight ratio was relatively stable between the sub samples (average 0.12 ± 0.02). The carbon biomass of roots was 2.1 kg C m⁻² (SD 1.4 kg m⁻²).

The above-ground biomass of reed varied between 0.1 and 1.4 kg dry weight m⁻² (average 0.70 kg dw m⁻²). The dry weight per wet weight factor was relatively stable and varied between 0.33 and 0.49 (average 0.43). The carbon biomass was 0.28 (SD 0.15) kg C m⁻².

Table 5-1. Marine basins and samples representing different basins in Forsmark.

	Kallrigafjärden	North Asphällorna	Öregrundsgrepen
Bacterioplankton	2, 3	15, 16	7, 8, 10, 16, 17, 18
Benthic bacteria	2, 3	16	19 (two samples)

6 Summary and discussions

The concentration of bacterioplankton found in the marine basins in Laxemar (22.2–27.0 mg C m³) and in Forsmark (19.8–36.6 mg C m³) was similar to those annual averages found by /Kuparinen 1987/ in Tvärminne, Finland (11–36 mg C m³). In another study, /Mohammedi et al. 1993/ found 0.6–1.2×10⁹ cells ml⁻¹ in benthic sediment, which is low compared to those in Laxemar (3.0–7.2×10⁹ cells ml⁻¹) and Forsmark (1.2–4.3×10⁹ cells ml⁻¹). In the study by Mohammedi, however, the sediment was in much deeper water (> 100 m) which can explain the low number of cells in that study. The lower concentrations (1.2 and 1.5×10⁹ cells ml⁻¹) found in Forsmark is likely due to that those sites are exposed areas with low carbon content in the sediment.

The number of bacterioplankton in Lake Frisksjön (1.3×10⁶ cells ml⁻¹) was within reported values for humic lakes but in the lower range /Nürnberg 1999/. The bacterioplankton biomass is often high in humic lakes due to high amounts of dissolved organic carbon. However, there are several parameters besides access to carbon that can limit the bacteria, e.g. access to nitrogen or phosphorus, grazing pressure by zooplankton, virus infections and temperature /Elser et al. 1995, Vrede et al. 1999, White et al. 1991, Fuhman 1999/. Number of sediment bacteria can vary within a large span, but typically range between 10⁸ and 10¹⁰ cells per ml sediment /Schallenberg 1999/. The benthic bacterial number in this study was 10⁹, and is thus within the reported range.

The above-ground reed biomass (0.70 kg dw m⁻²) was almost 10 times higher than reported in a previous report from Lake Frisksjön (0.05 kg dw m⁻²) /Aquilonius 2005/. The earlier reed investigation was performed in thin stands of macrophytes which may explain this difference. The above ground reed biomass in our study was not exceptionally high and was for example lower than in the marine area in Laxemar, where the mean biomass for 6 stations was 1.25 kg dw m⁻² /Alling et al. 2004/.

In contrast to the above ground biomass, the rhizome biomass in Lake Frisksjön was higher in Lake Frisksjön than in the marine areas in Laxemar /Alling et al. 2004/. This may be a true difference, but may also be a result of the relative few replicates in the marine areas.

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Bacteria

Bacterioplankton numbers, bio-volumes and biomasses in the marine basins in Laxemar and in Lake Frisksjön. MCV = mean cell volume.

Sample	Bacterial number 10^6 cells ml ⁻¹	MCV	Biovolym $10^6 \mu\text{m}^3$ ml ⁻¹	Dry weight $\mu\text{g dw L}^{-1}$	Carbon weight $\mu\text{g C L}^{-1}$
110–112	1.32	0.064	0.084	54.0	27.0
113–114	1.20	0.057	0.068	44.4	22.2
115–117	1.51	0.051	0.077	50.7	25.4
Frisksjön June	1.26	0.042	0.053	36.0	18.0
Frisksjön August	1.95	0.053	0.104	67.9	34.0

Benthic bacterial numbers, bio-volume and biomasses in the marine basins in Laxemar and in Lake Frisksjön.

Sample	Bacterial number 10^9 cells ml ⁻¹	MCV	Biovolume $10^6 \mu\text{m}^3$ ml ⁻¹	Dry weight mgDW m ⁻²	Carbon weight mgC m ⁻²
110	4.72	0.080	378	11,699	5,850
111	3.03	0.072	218	6,847	3,424
112	4.34	0.082	356	10,991	5,495
113	7.29	0.113	824	24,317	12,159
114	6.16	0.109	672	19,933	9,967
115	3.34	0.093	310	9,417	4,708
116	4.39	0.074	325	10,176	5,088
117	4.94	0.071	351	11,048	5,524
Frisksjön	4.29	0.060	257	8,302	4,151
Frisksjön	4.86	0.081	393	12,166	6,083
Frisksjön	5.61	0.064	359	11,480	5,740

Bacterioplankton numbers, bio-volumes and biomasses in the marine basins Forsmark.

Sample	Bacterial number 10^6 cells ml ⁻¹	MCV μm^3	Biovolym $10^6 \mu\text{m}^3$ ml ⁻¹	Dry weight $\mu\text{g dw L}^{-1}$	Carbon weight $\mu\text{g C L}^{-1}$
2	2.07	0,040	0.083	56.7	28.3
3	1.27	0.052	0.066	43.6	21.8
7	1.21	0.036	0.044	30.3	15.1
8	1.40	0.038	0.053	36.6	18.3
10	1.81	0.040	0.072	49.4	24.7
15	2.17	0.027	0.059	42.3	21.1
16	1.31	0.045	0.059	39.6	19.8
17	2.24	0.045	0.101	67.6	33.8
18	2.80	0.038	0.107	73.3	36.6

Benthic bacterial numbers, bio-volume and biomasses in the marine basins in Forsmark.

Sample	Bacterial number 10^9 cells ml ⁻¹	MCV μm^3	Biovolume $10^6 \mu\text{m}^3$ ml ⁻¹	Dry weight mgDW m ⁻²	Carbon weight mgC m ⁻²
2	4.28	0.08	325.02	10,140.30	5,070.15
3	3.80	0.08	296.19	9,207.29	4,603.64
16	2.99	0.11	328.48	9,731.41	4,865.71
19	1.46	0.077	112	3,494	1,747
19	1.15	0.076	88	2,738	1,369
19				Average 19:	1,558

Reed

Dry weight per wet weight calculations for above-ground biomass of reed (*Phragmites australis*) in Lake Frisksjön.

Sample	Sampling station	Number of straws	Wet weight g	Dry weight g	Dry weight per wet weight
1	1	1	17.08	6.99	0.41
2	1	1	14.74	7.03	0.48
3	1	1	2.98	1.47	0.49
4	1	1	28.79	12.57	0.44
5	1	1	2.77	0.92	0.33
6	1	1	16.73	7.44	0.44
7	2	1	18.04	7.88	0.44
8	3	1	12.22	4.92	0.40
9	4	1	25.36	11.41	0.45
10	4	1	30.9	13.44	0.43
11	4	1	24.67	10.94	0.44
Mean					0.43
Standard deviation					0.04
Min					0.33
Max					0.49

Above ground biomass of reed (*Phragmites australis*) in Lake Frisksjön.

Sample	Sampling station	Number of straws	Wet weight g sample ⁻¹	Dry weight g sample ⁻¹	Dry weight g dw m ⁻²	Carbon weight g C m ⁻²
1	1	5	59	25	407	161
2	1	8	123	53	851	336
3	1	10	149	65	1,034	409
4	1	6	61	26	420	166
5	1	5	72	31	499	197
6	1	6	70	30	481	190
7	1	12	83	36	577	228
8	1	4	54	23	372	147
9	1	7	71	31	492	194
10	1	5	103	45	715	282
11	2	6	96	41	664	262
12	2	1	18	8	125	49
13	3	5	210	91	1,454	574
14	3	7	169	73	1,168	461
15	3	6	152	66	1,052	416
16	3	6	49	21	338	133
17	3	7	172	75	1,194	472
18	4	1	31	13	214	85
19	4	4	85	37	589	233
20	4	6	187	81	1,298	513
Mean			101	44	697	275
Standard deviation			55	24	382	151
Min			18	8	125	49
Max			210	91	1,454	574

Dry weight per wet weight calculations for root biomass of reed (*Phragmites australis*) in Lake Frisksjön.

Sample	Sampling station	Wet weight g	Dry weight g	Dry weight per wet weight
1	1	39.85	4.72	0.12
2	1	30	3.66	0.12
3	1	34.03	3.76	0.11
4	1	37.31	4.85	0.13
5	1	36.63	3.89	0.11
6	1	40.19	6.16	0.15
7	1	32.25	3.84	0.12
9	1	39.67	5.15	0.13
10	1	41.94	5.4	0.13
11	1	42.82	4.31	0.10
12	1	37.03	5.8	0.16
8	2	28.69	3.69	0.13
13	3	33.28	3.53	0.11
14	3	45.84	5.12	0.11
15	3	37.61	4.27	0.11
16	3	35.78	4.58	0.13
Mean				0.12
Standard deviation				0.02
Min				0.10
Max				0.16

Root biomass of reed (*Phragmites australis*) in Lake Frisksjön.

Sample	Sampling station	Wet weight g sample ⁻¹	Dry weight g sample ⁻¹	Dry weight g dw m ⁻²	Carbon weight g C m ⁻²
1	1	596	73	1	462
2	1	2,389	293	5	1,853
3	1	1,372	168	3	1,064
4	1	3,016	370	6	2,339
5	1	4,554	559	9	3,532
6	2	274	34	1	212
7	2	1,070	131	2	830
8	3	4,224	518	8	3,275
9	3	5,627	690	11	4,363
10	3	3,624	445	7	2,810
Medel		2,675	328	5	2,074
Stdav		1,829	224	4	1,418
Min			34	1	212
Max			690	11	4,363