Sveriges lantbruksuniversitet
Swedish University of Agricultural Sciences

Faculty of Natural Resources and Agricultural Sciences

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– Use of *Fontinalis antipyretica* as an integrative sampler of nitrogen

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Department of Aquatic Sciences and Assessment
Independent project • 15 hec • First cycle, G2E
Biology and Environmental Science
stud.epsilon.slu.se 2015

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Credits: 15 hec
Level: First cycle, G2E
Course title: Independent project in biology
Course code: EX0689
Programme/education: Biology and Environmental Science

Place of publication: stud.epsilon.slu.se
Year of publication: 2015
Cover picture: Anders Johansson
Number of part of
Online publication: http://stud.epsilon.slu.se

Keywords: beaver, *Castor fiber*, colonization history, *Fontinalis antipyretica*, nitrogen retention, nitrogen, recolonization, Sweden

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Abstract

Beaver ponds affect the environment in many ways and beaver ponds have the potential to retain macronutrients. The retention of nitrogen in beaver ponds has mainly been suggested to be due to denitrification by bacteria. The aquatic moss *Fontinalis antipyretica* are frequently used as an integrative measure of water quality and the moss was hypothesized to retain nitrate, nitrite and ammonia in beaver system. The aim of the study is to investigate nitrogen retention in beaver ponds using *F. antipyretica* as an integrative sampler and to test if N-retention is dependent on the colonization history of the beaver pond. Twelve beaver systems were investigated in the study; Luleå (N=3), Sundsvall (N=3), Skinnskatteberg (N=2), Surahammar (N=2) and Örebro (N=2). Seven of the beaver systems were recolonization of abandoned ponds and five of them were colonized for the first time since extirpation by beavers, i.e. were reused and pioneer systems, respectively. *F. antipyretica* was picked in water systems with good ecological status, put in mesh bags and transplanted to sites up- and downstream the beaver ponds. The bags were left in the stream for 2, 4, 6 and 12 weeks. N content in *F. antipyretica* was higher upstream than downstream ($p<0.05$) and this result was stronger in reused than in pioneer beaver systems ($p<0.001$). The study suggests that *F. antipyretica* can be used as an integrative measure for retention of nitrogen in beaver systems. Especially, the results indicate that reused beaver dams have higher nitrogen retention potential than pioneer ones.

Keywords: beaver, *Castor fiber*, colonization history, *Fontinalis antipyretica*, nitrogen retention, nitrogen, recolonization, Sweden

Contents

Introduction

Beavers are referred as ecological engineers with the ability to profoundly change the environment and this has fascinated people since ancient times (Danilov et. al., 2010). For instance beavers have shown to increase the habitat heterogeneity in the riparian zone by increasing the number of herbaceous plats species at the landscapes scale (Wright et. al 2002). Furthermore, eutrophication of aquatic systems can be mitigated by the activity of beavers (Lazar et. al. 2013). Eutrophication of lakes is caused by agriculture, forestry, and wastewater from households and industry (Rosenberg et. al. 1990). Such potential retention of macronutrients (e.g. nitrogen and phosphorous) is an important ecosystem service provided by beaver ponds.

The Swedish beaver landscape today includes beaver ponds of different colonization history. In Fennoscandia beavers were almost hunted to extinction in the late 19^{th} century due to amongst others high fur prices (Hartman, 1994). In Sweden, the beaver population was probably extinct in 1871(Hartman, 1994). There was only a small population of beavers in southern Norway in the beginning of 20^{th} century (Hartman, 1994). Beavers from the Norwegian population were reintroduced to Sweden in 1922 and the population in Sweden probably exceeded 100 000 in the 90^{th} (Hartman, 1994, Hartman 1995).

The main nitrogen reduction processes in beaver dams are in order of decreasing importance denitrification, immobilization by soil microbes and plant uptake (Lazar et. al. 2013). Even though beaver dams have the potential to retain macronutrients our knowledge on the effect of colonization history and pond age on N-retention is limited.

The definition of a beaver system in this report is the combination of streams and ponds upstream and downstream the beaver settlement. Colonization history relates to the succession of beaver systems including colonization, abandonment and recolonization (Naiman et al. 1988). Pioneer beaver systems are here defined as those that are colonized by beavers for the first time after their reintroduction in the early 20^{th} century. Reused beaver systems are beaver dams that have been abandoned for some time and then been recolonized.

F. antipyretica is frequently used in biogeochemical investigations and environmental monitoring due to its ability to accumulate trace metals from the stream water (Lax & Selinus 2005, Pekka et. al. 2008). However, the use of the aquatic moss *Fontinalis antipyretica* as an integrative measure of N-concentration has to my knowledge not been investigated. The use of *F. antipyretica* as an integrative measure of nitrogen retention in beaver systems can be beneficial due to variation in concentrations of nitrite, nitrate and ammonium over the season. While, water samples only show the nitrogen concentration at a given time.

The aim of this thesis was twofold. In a first step, I studied if *F. antipyretica* can be used as an integrative sampler of N-retention in beaver ponds and finally, I evaluated if N-retention is dependent on the colonization history of beaver ponds.

Methods

Study areas

The study was conducted in twelve beaver systems in Sweden (figure 1). The systems in Lulea (N=3) and Sundsvall (N=3) represent the northern region in the study, while Skinnskatteberg (N=2), Surahammar (N=2) and Örebro (N=2) represent the southern region. For exact locations of the beaver systems see *Appendix 1*.

Luleå is located in middle boreal vegetation zone (Ahti et. Al. 1968). The climate is harsh with long winters and short summers, the annual average temperature and precipitation are 2^{0}C and 500mm, respectively (SMHI 2015). The dominating land use in the area is forestry. Sundsvall is located at the border between southern and middle boreal vegetation zone (Lönnqvist 2014). With an annual average temperature of 5^{0}C and precipitation of 700 mm (SMHI 2015). The dominating land use in the area is forestry. Skinnskatteberg, Surahammar and Örebro area are located in the border between the boreonemoral zone and southern boreal zone (Lönnqvist 2014). The annual average temperature and precipitation being 6-7^{0}C and 500-600mm, respectively. Agriculture is the dominating land use in the Örebro and Surahammar area. The dominating land use in Skinnskatteberg is forestry, and one locality was also affected by mining operation upstream upstream the studied beaver system. The concentrations of macronutrients varied among regions and location in the beaver system (table 1), but generally there were higher levels of macronutrients in in the southern beaver systems.

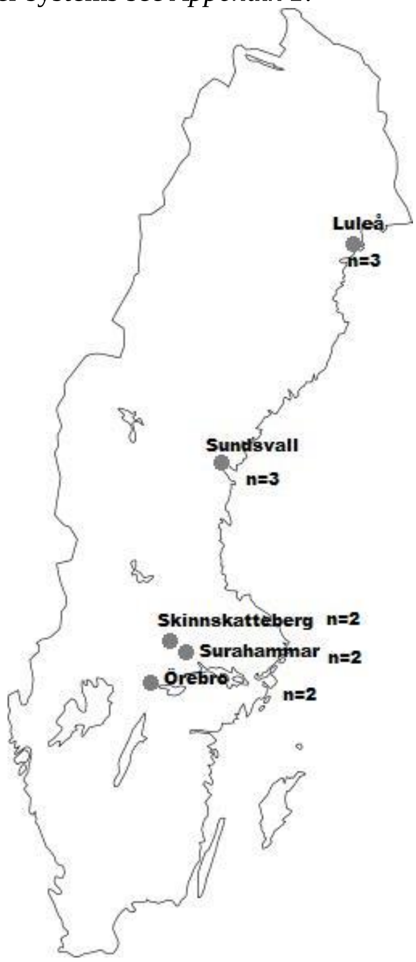

Figure 1: Locations where beaver systems were investigated and number of beaver systems (Johansson, 2014).

Table 1: Total nitrogen, NO_3+NO_2 and total phosphorus for the northern and southern region and for upstream and downstream the beaver pond. Maximum, minimum and mean values for the beaver pond in the specific region are presented (Lönnqvist 2014).

			Tot-N ug/l	NO_3+NO_2 ug/l	Tot-P ug/l
Northern region beaver systems	Upstream	Max	484	20	14
		Mean	335	12	11
		Min	191	6	6
	Downstream	Max	546	23	16
		Mean	374	15	13
		Min	226	7	7
Southern region beaver systems	Upstream	Max	1270	152	69
		Mean	615	70	26
		Min	382	21	14
	Downstream	Max	1690	611	612
		Mean	830	176	138
		Min	484	23	16

Choice of reference streams for collecting the aquatic moss

Reference streams with at least good ecological and chemical- status according to the Water Framework Directive (WFD; 2000/60/EC) were chosen for collecting the aquatic moss. Reference streams were chosen after contacting the respective county administration board (*see appendix 2*). The aquatic moss was collected and placed in the beaver systems between 22 of May to 5 of June 2013. Three moss samples per reference location were analyzed for tot-N, to approximate how much tot-N was at start in *F. antipyretica*.

Collection of the aquatic moss *F. antipyretica*

The aquatic moss *F. antipyretica* was collected by hand or by using a pair of scissors and placed in the tub containing some water so the moss would not be dried out. Only the yearly shoots characterized by bright green color were collected and the dark green / brown shoots were discarded. The moss was rinsed in running water to remove material that may be covering the moss. After picking a significant amount of moss (around 15min/bag/person) the moss was weighed and placed in mesh bags (see Appendix 3 for details on the mesh bags). A pilot study showed that 5g of moss are required per bag for the dry matter to be 1g (the minimum amount needed for chemical analyses). To reduce the amount of surplus water in the moss samples, the samples were squeezed with one hand 2-3 times medium-hard and then shaken. I removed the water from majority of the moss samples in the project to reduce between-observer errors related to moss weight. Then, I weighted the moss to 5.0 g with a field scale (accuracy 0.5g). A plastic-coted number was put in each bag to recognize the bags. The moss was added to the bags and I stapled the bags together so that they formed a tetra-pack shape that gave growing space for the moss. Then were the seam taped over the staples with duct tape and then stapled again. The bags with moss were stored in a larger tubs with water which had a lid for safe transportation in the car. A twine was attached in the eyelet of the mesh bag and the other end of the twine was connected together to the iron pipe. The length of the twine was determined from the water depth in the stream but usually 20-50cm. Four mesh bags were attached per iron rods and there were three iron rods for each location i.e. 12pcs upstream and 12pcs downstream per beaver system, 288 bags for the entire project. The bags' number and placement in beaver systems were recorded.

The collection of bags and lab work

Bags with moss were retained from the beaver sites after 2, 4, 6 and 12 weeks, respectively. One bag per iron rod was taken on each occasion so I had three replicates upstream and three replicates downstream per beaver system occasion. In the lab, the moss was cleaned using a colander and small aluminum cups. A strong jet of water was sprayed on the moss in the cups to release sediment and other material. The water and the moss was then poured in and through the colander. The moss was brought back into the cup to repeat the process until all unwanted material was gone, usually 5-10 times. After the cleaning the moss was put in aluminum cups and dried in an oven at 60^0C overnight. The moss was weighed using a lab scale (accuracy 0.001g). The moss was then packed in plastic bags prior to analysis of total nitrogen content. A pilot analysis of one Sundsvall beaver system showed that the greatest difference in nitrogen content in *F. antipyretica* occurred during the last periods, i.e. weeks 6 and 12 (*see appendix 4*). Therefore, moss samples from the two last sampling occasions were analyzed for nitrogen in all beaver systems. The total nitrogen in *F.*

antipyretica were analyzed by a LECO CNS 2000 (LECO 1999) dry combustion analyser.

Data handling

The data and calculations were handled in Microsoft Excel 2013 and the statistical analyses were done in Minitab 17.

Calculations

The total nitrogen in *F. antipyretica* was corrected for the water content left in the samples (1). Then the values were transformed to g nitrogen / kg dry matter by multiply by 10 (2).

(1) Tot-N % / dry matter in *F. antipyretica* in % = Tot-N % of dry matter
(2) Tot-N % of dry matter * 10 = g nitogen / kg dry matter

To calculate the uptake of nitrogen in *F. antipyretica* per day the values were first corrected for the mean tot-N in *F. antipyretica* in the reference stream (were the moss were picked) to get the net nitrogen uptake (3). Then the net nitrogen uptake was corrected for the number of day the aquatic moss had been in the stream (4).

(3) Tot-N % of dry matter - Tot-N % of dry matter of the reference = net nitrogen uptake
(4) Net nitrogen uptake / NO. days in stream = uptake of nitrogen in *F. antipyretica* per day

Statistical analyses

One-way-ANOVA were preformed to investigate if there is a difference between tot-N in *F. antipyretica* upstream and downstream the beaver pond. The same analysis were done to investigate if there was a difference in nitrogen uptake per day in *F. antipyretica* between upstream and downstream the beaver pond. I compared means between upstream and downstream for region and pioneer/reused separately. Additional water chemistry data for DOC (dissolved organic carbon), pH, conductivity, sulfate, alkalinity (Lönnqvist 2014) and canopy cover (Levanoni, unpublished) were obtained to investigate possible correlations (Pearson product-moment correlation coefficient). The canopy cover were measured by a LAI-2200 plant canopy analyzer (LI-COR 2012).

Results

Weight of *F. antipyretica*

The theoretical dry weight (DW) at start was 1 ± 0.1 g. There was a weight increase in *F. antipyretica* in Luleå beaver systems (*figure 2, appendix 5*). However Sundsvall and the southern beaver systems did not show any weight increase. The weight did not differ significantly from upstream and downstream throughout all beaver systems (*figure 2*). The strongest correlation found to DW of the moss was canopy cover (*appendix 6*). The DW of the aquatic moss is higher when the canopy cover is lower (*appendix 7*) There was also negative correlation found with SO_4, pH, conductivity, tot-N, NO_2+NO_3, tot-P and alkalinity to the DW of the moss (*appendix 6*).

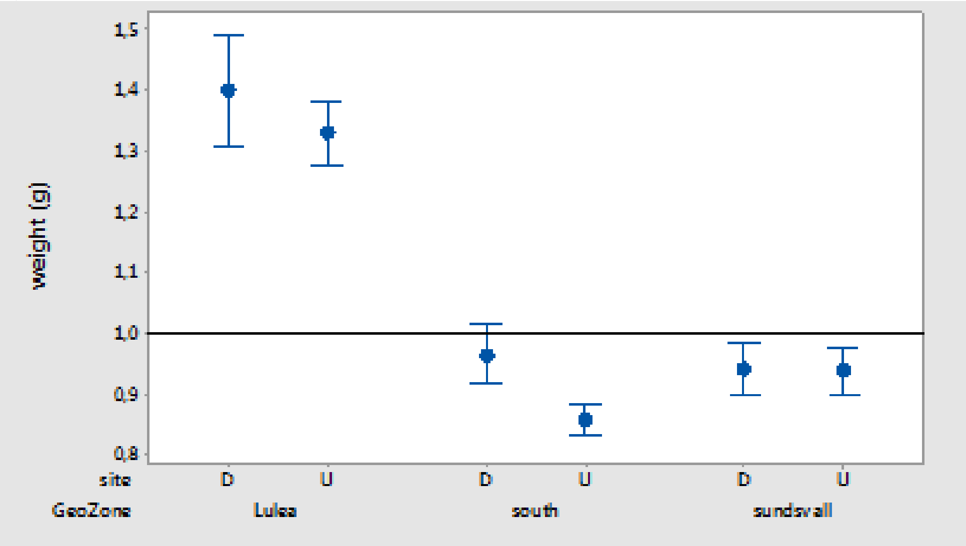

Figure 1**:** DW of *F. antipyretica* (mean ± 1 SE) in Lulea, Sundsvall and southern beaver systems. D denotes downstream and U upstream the beaver pond, respectively. The line indicates the theoretical starting value of 1 ± 0.1 g DW.

Nitrogen content in *F. antipyretica* upstream and downstream the beaver pond

The total nitrogen concentent in *F. antipyretica* for all the beaver systems was normally distributed (Anderson-Darling test, $p > 0.05$) (*appendix 8*). The mean total nitrogen content in *F. antipyretica* for all upstream locations was 25.6 g/kg DW and for downstream had an mean value of 24.6 g/kg DW (*figure 3*). There was a significant higher total nitrogen content upstream than downstream the beaver pond for all locations (p-value< 0,05) (table 2). There was a significantly higher nitrogen content in *F. antipyretica* upstream than downstream in the Sundsvall beaver systems (p-value < 0.001) (*figure 3, table 2*). In Luleå and southern Sweden, beaver systems showed no significance difference in nitrogen content in *F. antipyretica* between upstream and downstream. Individual figures for each beaver system on the total nitrogen in *F. antipyretica* are given in Appendix 3. There was a correlation

found on total nitrogen in *F. antipyretica* and conductivity. With higher conductivity lower nitrogen content in *F. antipyretica* (*appendix 9, 6*). There was also a positive correlation with weight and nitrogen content in *F. antipyretica* (*appendix 6*).
Most of the *F. antipyretica* lost nitrogen compared to the reference value (*figure 4*). There was a significantly higher uptake of nitrogen per day for upstream than downstream for all the beaver ponds combined (p-value < 0.01) and for the Sundsvall beaver ponds (p-value < 0.001) (*figure 4, table 3*). In Luleå and southern Sweden, beaver systems showed no significance difference in uptake of nitrogen per day between upstream and downstream (*figure 4, table 3*).

Table 2: One-way-ANOVA, comparison of total nitrogen in F. antipyretica between upstream and downstream for all the beaver systems in the study and separately for the different regions. The p-values, F-value and degrees of freedom are presented. The arrow show higher mean value upstream ↑ the beaver pond. Asterisks denote the level of significance (*, p<0.05 and ***, p<0.001).

	Degrees of freedom	F-value	P-value	Higher mean value
All in the study N=12	145	5.42	0.021 *	↑
Luleå =3	36	1.13	0.295	
Sundsvall =3	35	47.09	<0.001 ***	↑
Southern= 6	72	0.02	0.894	

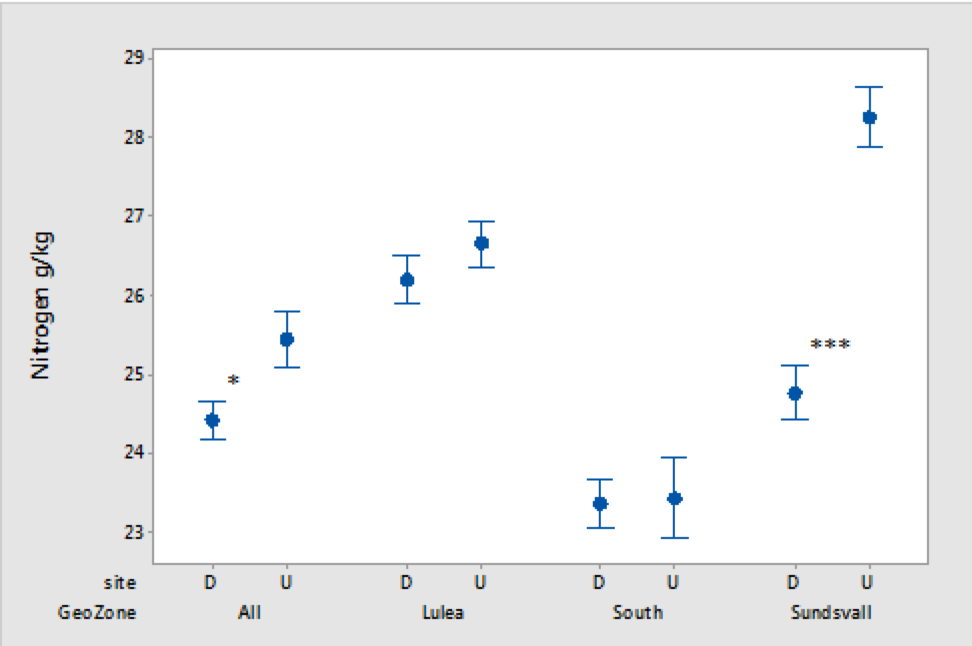

Figure 3: Total nitrogen content in *F. antipretica* (mean ± 1 SE) upstream and downstream the beaver pond in the three different regions and all systems combined (All). D denotes downstream the beaver pond and U upstream the beaver pond. Asterisks denote the level of significance (*, p<0.05 and ***, p<0.001).

Table 3: One-way-ANOVA, comparison of nitrogen uptake per day in F. antipyretica between upstream and downstream for all the beaver systems in the study and separately for the different regions. The p-values are presented and the arrow show higher mean value upstream ↑ or downstream ↓ the beaver pond. Asterisks denote the level of significance (**, p<0.01 and ***, p<0.001).

	Degrees of freedom	F-value	P-value	Direction
All in the study N=12	145	7.66	0.006 **	↑
Luleå =3	36	1.24	0.274	
Sundsvall =3	35	40.70	<0.001 ***	↑
Southern =6	72	0.17	0.679	

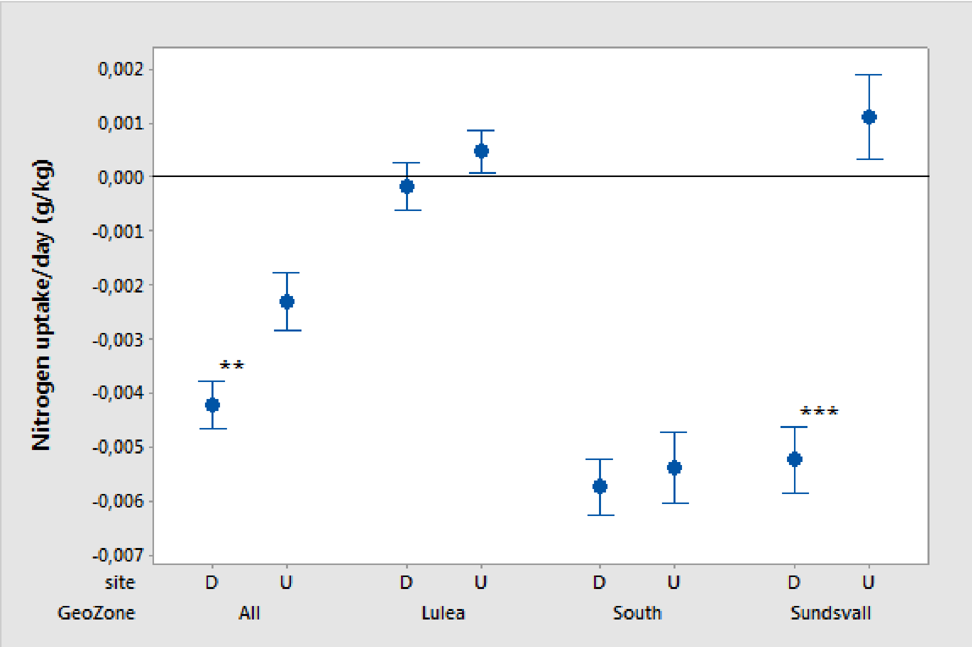

Figure 4: N-uptake per day in *F. antipyretica* (mean ± 1 SE) upstream and downstream the beaver dams in different regions in Sweden. D denotes downstream the beaver pond and U upstream the beaver pond. The line is the theoretical starting value. Asterisks denote the level of significance (**, p<0.01 and ***, p<0.001).

Effect of colonization history

Reused beaver systems

One-way-ANOVA comparison for the difference between upstream and downstream for all seven reused beaver dams revealed higher nitrogen content and N-uptake in *F. antipyretica* upstream (*table 4, 5, figure 5, 6*). The northern reused beaver systems showed higher values upstream. However, southern reused beaver systems (N=3) ones showed no difference in nitrogen content in *F. antipyretica* and N-uptake per day between upstream and downstream (*table 4, 5, figure 5, 6*).

Table 4: One-way-ANOVA, comparison of means between upstream and downstream for tot-N I F. antipyretica for reused beaver systems. The p-values, F-value and degrees of freedom are presented. The arrow show higher mean value upstream ↑. Asterisks denote the level of significance (***, p<0.001).

	Degrees of freedom	F-value	P-value	Direction
All reused N=7	83	17.29	<0.001 ***	↑
North reused N=4	47	37.86	<0.001 ***	↑
South reused N=3	35	3.28	0.079	

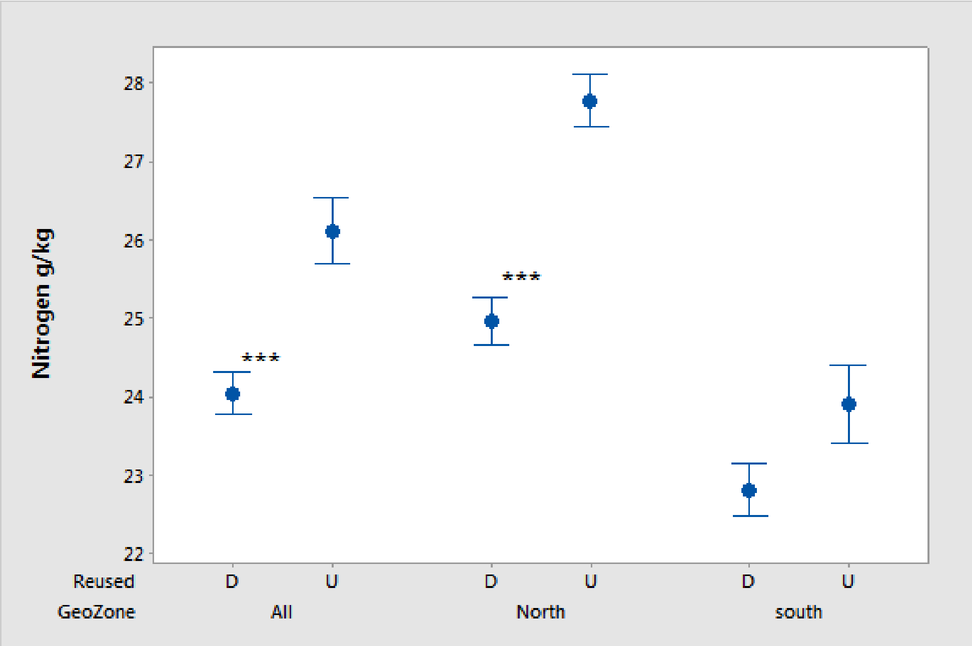

Figure 5. Total nitrogen content in *F. antipretica* (mean ± 1 SE) for reused beaver ponds, upstream and downstream. D denotes downstream the beaver pond and U upstream the beaver pond. All (N=7), north (N=4) and south (N=3) for reused beaver ponds are presented. Asterisks denote the level of significance (***, p<0.001).

Table 5: One-way-ANOVA, comparison of means between upstream and downstream for nitrogen uptake per day in F. antipyretica for reused beaver systems. The p-values are presented and the arrow show higher mean value upstream ↑ the beaver pond. Asterisks denote the level of significance (p<0.001).

	Degrees of freedom	F-value	P-value	Direction
All reused N=7	83	15.54	<0.001 ***	↑
North reused N=4	47	33.45	<0.001 ***	↑
South reused N=3	35	1.70	0.201	

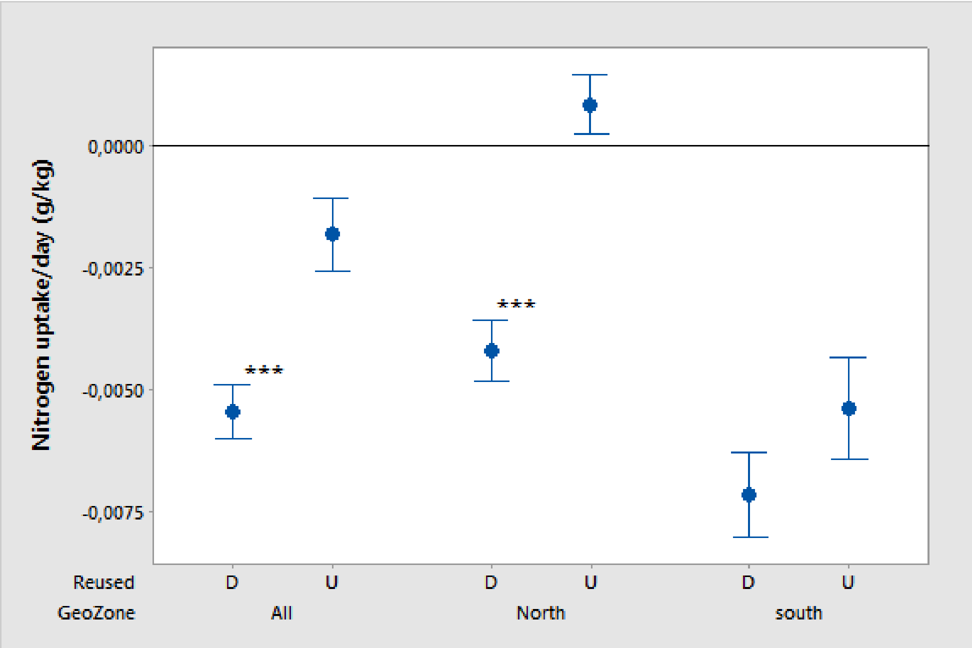

Figure 6 N-uptake per day in *F. antipyretica* (mean ± 1 SE) for reused beaver ponds, upstream and downstream. D denotes downstream the beaver pond and U upstream the beaver pond. All (N=7), north (N=4) and south (N=3) for reused beaver ponds are presented. Zero is the theoretical starting value. Asterisks denote the level of significance (***, p<0.001).

Pioneer beaver systems

In pioneer beaver ponds nitrogen content and N-uptake per day in *F. antipyretica* did not differ between downstream and upstream in none of the regions or when combining all systems (*table 6, 7, figure 7, 8*).

Table 6: One-way-ANOVA, comparison of means in nitrogen in F. antipyretica between upstream and downstream for pioneer beaver systems. The p-values, F-value and degrees of freedom are presented.

	Degrees of freedom	F-value	P-value
All Pioneer N=5	61	0.29	0.594
North Pioneer N=2	24	0.33	0.569
South Pioneer N=3	36	0.84	0.367

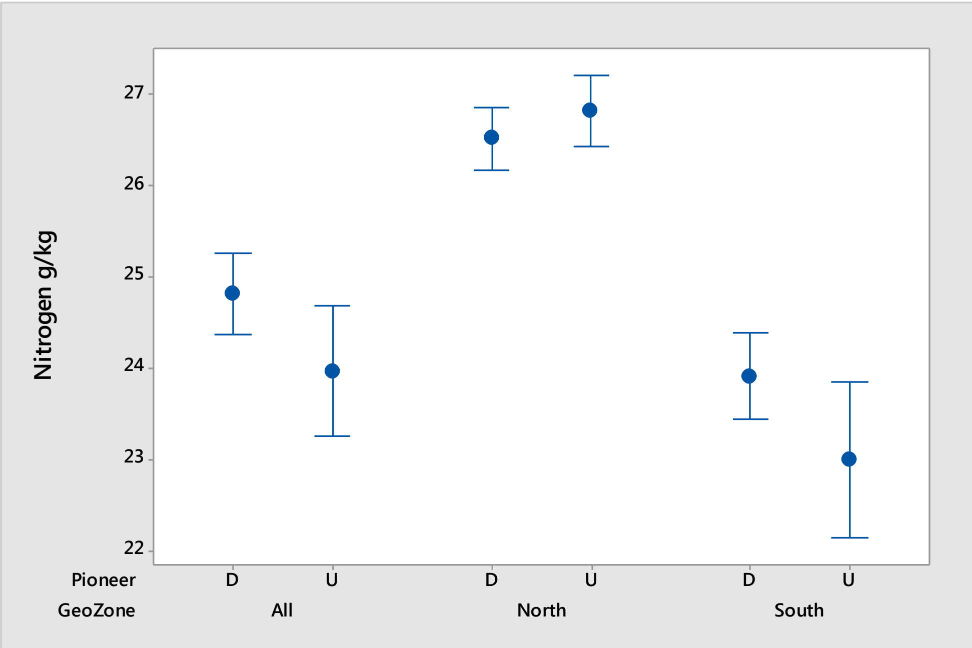

Figure 7 Total nitrogen content in *F. antipretica* (mean ± 1 SE) for pioneer beaver ponds, upstream and downstream. D denotes downstream the beaver pond and U upstream the beaver pond. All (N=5), north (N=2) and south (N=3) for pioneer beaver ponds are presented.

Table 7: One-way-ANOVA, comparison of means in uptake of nitrogen per day in F. antipyretica between upstream and downstream for pioneer beaver systems. The p-values, F-value and degrees of freedom are presented.

	Degrees of freedom	F-value	P-value
All pioneer N=5	61	0.23	0.633
North Pioneer N=2	24	0.33	0.574
South Pioneer N=3	36	1.26	0.269

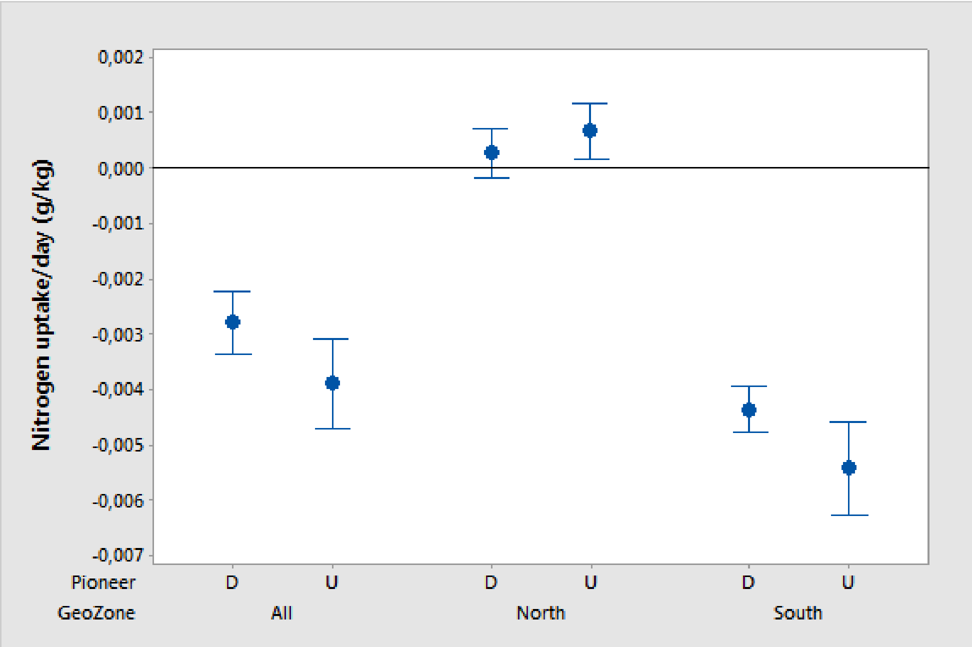

Figure 8 N-uptake per day in *F. antipyretica* (mean ± 1 SE) for pioneer beaver ponds, upstream and downstream. D denotes downstream the beaver pond and U upstream the beaver pond. All (N=5), north (N=2) and south (N=3) for pioneer beaver ponds are presented. The line is the theoretical starting value.

Discussion

Weight of *F. antipyretica*

The theoretical weight of *F. antipyretica* put into the beaver systems was 1 ± 0.1 g DW, but the only region where there was a weight increase was in the Luleå beaver systems. Luleå beaver systems showed the lowest phosphor, nitrate and nitrite concentration in the waters. So *F. antipyretica* cannot have been limited by those macro nutrients in the other beaver systems. However some samples of *F. antipyretica* in the southern regions were not as green and alive as those in Sundsvall and Luleå. In the southern regions the environment around the beaver ponds was many times affected by anthropogenic activity. Some were close to agricultural fields and one was in the same watershed as old mining operations. There is shown by (Davies 2006) that sulphate in high levels are toxic for *F. antipyretica.* Anthropogenic sources of sulphate are mine wastes, industrial waste water, agricultural runoff and domestic sewage (Davies 2006). Those activity might have affected the growth of *F. antipyretica* at some beaver systems in the southern region. But there was also healthy green *F. antipyretica* that showed no weight increase as those in Sundsvall region. There is a negative correlation with canopy cover and DW of *F. antipyretica* (appendix 8). Suggesting that the aquatic moss were suffering from reduced light conditions from the canopy. There can also be that the aquatic moss suffered from reduced light conditions in the bags itself thereby less growth. Many times the bags tracked sediment from the stream water and thereby reduced light conditions.

Nitrogen content in *F. antipyretica* upstream and downstream the beaver ponds

All beaver systems combined showed a significantly higher nitrogen content in *F. antipyretica* upstream than downstream, suggesting that there is a difference in nitrogen availability for *F. antipyretica* upstream- and downstream the beaver ponds. The low values of 20 g/kg and lower were all from the same beaver system (*appendix 6, BD_26*). There was something affected the nitrogen uptake in *F. atipyretica* at that beaver system (BD_26). In order to use *F. antipyretica* as integrative measure of N-concentration research is needed to know temporal variation, maximum and minimum levels of nitrogen content in *F. antipyretica*. The much lower nitrogen content in *F. antiypertica* in the southern region than in Sundsvall and Luleå also indicates that there are something affecting *F. antipyretica* ability to accumulate nitrogen (*figure 3*). There is suggested that with increasing ion activity in the water that non-halophytes also increases the uptake of ions to compensate for the osmotic imbalance (Davies 2006). This should explain why nitrogen is lower in *F. antipyretica* with higher conductivity in the water (appendix 9). However, there are 3 to 100 times more nitrate and nitrite in the southern regions than in the northern region. So the use of F. antipyretica as an intrusive sampler of nitrogen cannot be used as an absolute measure of nitrogen concentration in the water. However, There is shown by Sutter et al. (2001) that Cd, Pb and Zn inhibit the nitrogen assimilation in *F. antipyretica.* Anthropogenic factors might also affect the aquatic moss ability to assimilate nitrogen (see discussion weight). There is little understood of nitrogen assimilation of stream bryophytes (Stream bryophyte group 1999). However, current velocity are suggested to be important for nutrient uptake. There are shown to be an increased nutrient uptake with increased current velocity (stream bryophyte group 1999). This might affect the nitrogen assimilation in *F. antipyretica*.

In Sundsvall, beaver systems showed higher nitrogen content in *F. antipyretica* upstream and the NO_2^- and NO_3^- in the water showed higher values downstream. These results were unexpected and can indicate that there might be other factors controlling the nitrogen content in *F. antipyretica* such as ammonium or conductivity (see discussion above). The water samples of nitrite and nitrate were based on one replicate per location which is too low to say any think about the variation in nitrate and nitrite over the period of the experiment. This can mean that uses of *F. antipyretica* as an integrative measure of N-concentration gives a more comprehensive picture of the nitrogen in the system over long periods.

Reused and pioneer beaver systems

All reused beaver systems showed significantly higher nitrogen content in *F. antipyretica* upstream than downstream the pond. Meanwhile all pioneer beaver systems did not show any difference in nitrogen content in *F antipyretica* between upstream and downstream the pond. This suggest that reused beaver systems have higher retentions of nitrogen than pioneer ones. This can partly be because of pioneer beaver dams have a higher amount of organic nitrogen from all the new dead trees around the beaver pond and in the sediment. While reused ones often are older beaver dams with lower amount of organic nitrogen. However, it can also be that the dam itself in reused beavers systems could be more solid than pioneer beaver systems. The solids might have higher active surface area thereby more active sites for the nitrogen to get stuck to the solids. The slightly higher values of nitrate and nitrite downstream in northern and southern region for reused beaver dams can be interpreted as mineralization of organic nitrogen is higher than in pioneer ones (*see appendix 10*). But this is disproving the results of higher nitrogen content in *F. antipyretica* upstream in reused beaver ponds than in pioneer ones (*see figure 6*). But the low amount of replicates of water samples must be beard in mind due to no information about the variance of nitrogen and there is statistically no difference between upstream and downstream for nitrate and nitrite.

Conclusions

F. antipyretica can presumably act as an integrative sampler of N-concentration in beaver systems and other water systems, despite the complexity of ion activity in the water. But to try quantifying the retention potential of nitrogen with the use of *F. antipyretica* requires more research about *F. antipyretica* ecophysiology and especially its properties of nitrogen uptake at different nutrient, conductivity, current velocity levels. The lack of DW increase of *F. antipyretica* are most likely due to reduced light conditions. Reused beaver ponds displayed a higher retention of nitrogen than the pioneer ones with this method.

Acknowledgments

Thanks are due to O. Levanoni, E. Arons, M. Grunander, S. Lindman, J. Dahl and J. Segersten with helping me with the fieldwork. Extra thanks to O. Levanoni for valuable discussion & chemical data. Thanks to A. Staniute, O. Levanoni and F. Ecke for critical reviewing. Thanks to F. Ecke for founding the nitrogen analysis and access with a car to the project.

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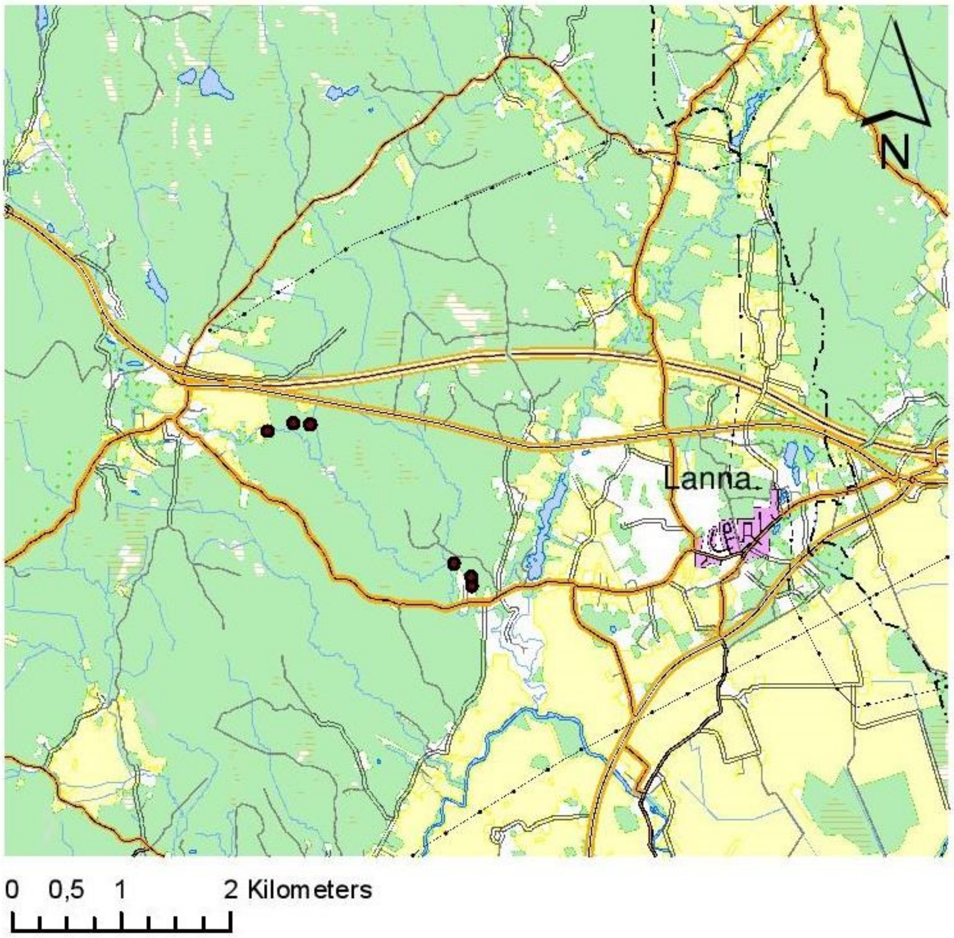
Appendix

Appendix 1

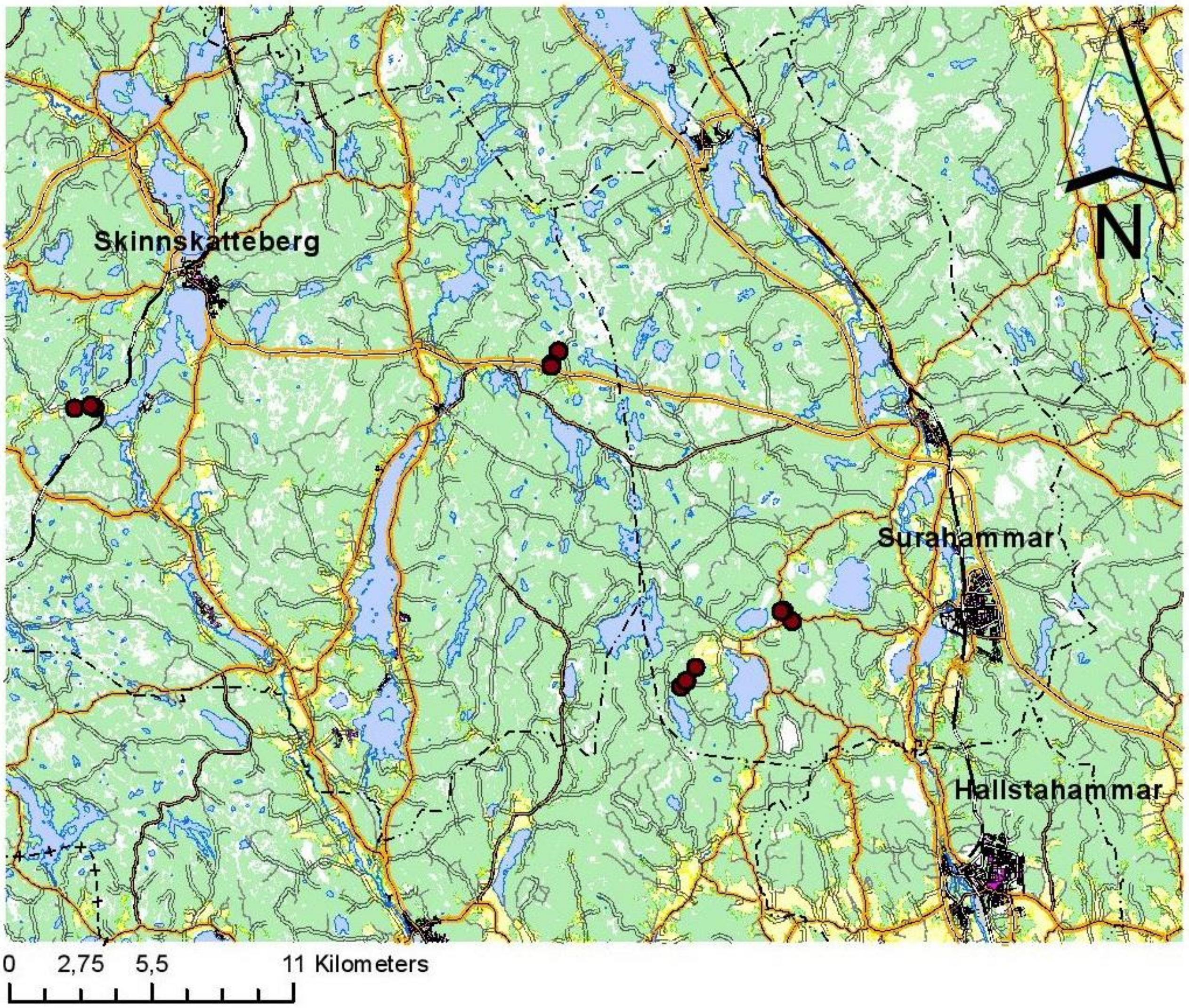
Beaver systems locations

Beaver system west of Örebro

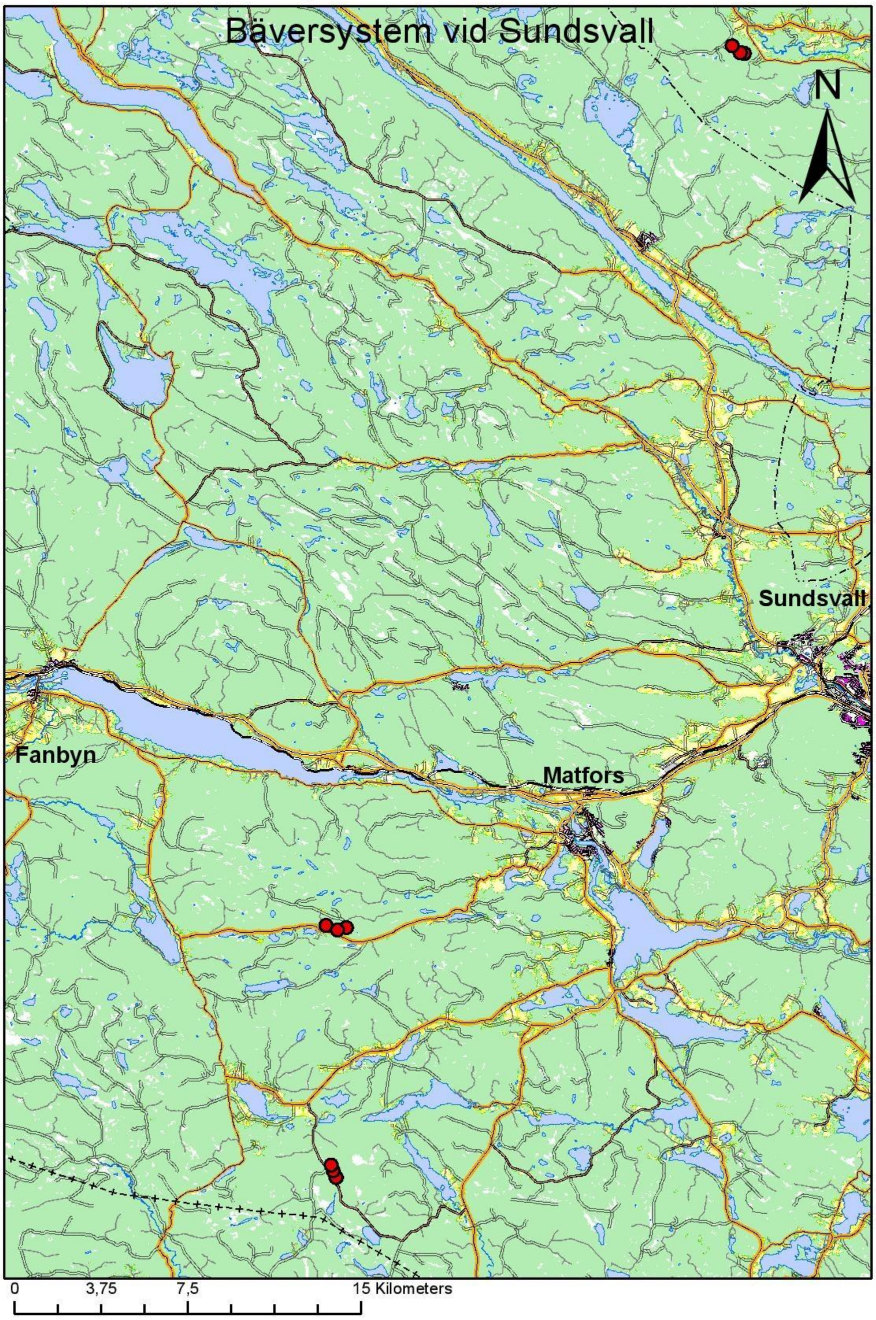


Beaver system north of Luleå

Beaver systems west of Västerås



Beaver system west of Sundsvall



Appendix 2

Table 1: Locations where the aquatic moss was picked in the reference streams and their coordinate, number of moss bags prepared from the different locations and how many beaver systems that were investigated in that area.

Location	Reference stream	Coordinate SWEREF99 TM	NO moss bags tot.	NO beaver syst. investi-gated
Luleå	Kvarnbäcken	7342233, 818146	72	3
Sundsvall	Lindsjöån	6906225, 598393	72	3
Örebro/Ves-terås	Venabäcken/ Årsbäcken	6612460, 553146 6621404, 547766	144	2 & 4

Appendix 3

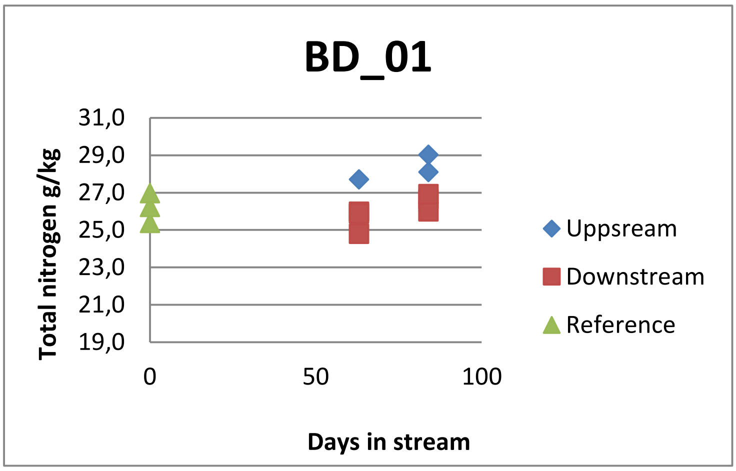
Making mesh bags

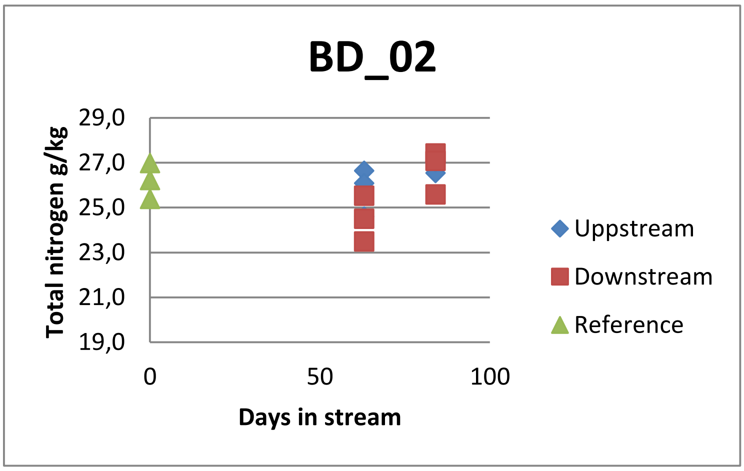
Material: polyester filter cloth, scissors, sewing machine, soldering iron, eyelets

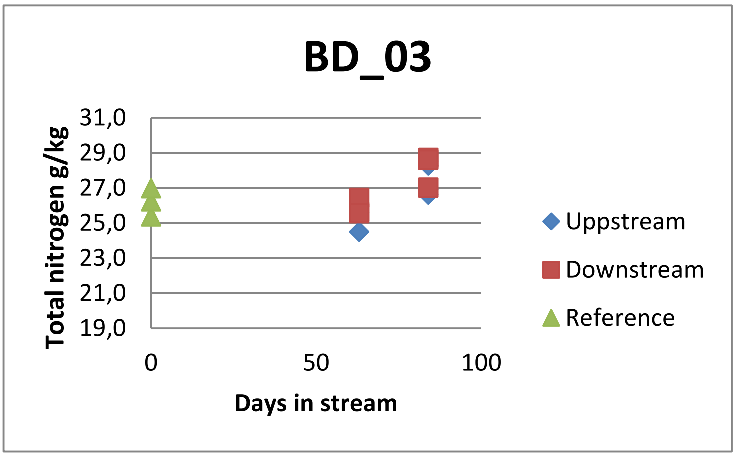
Rectangular pieces 30x20 cm polyester filter cloth was cut out which have 0.5mm sized meshes. Thereafter, turn it double and sews one of the long sides and one of the short sides together with a sewing machine. A hole in one corner of the bag was made using a soldering iron to prevent it to be chipped up. Threading then in an eyelets of aluminum in the hole so the hole will not be torn open by the twine that will keep the bag in the stream. For this project it needed about 300pcs of bags.

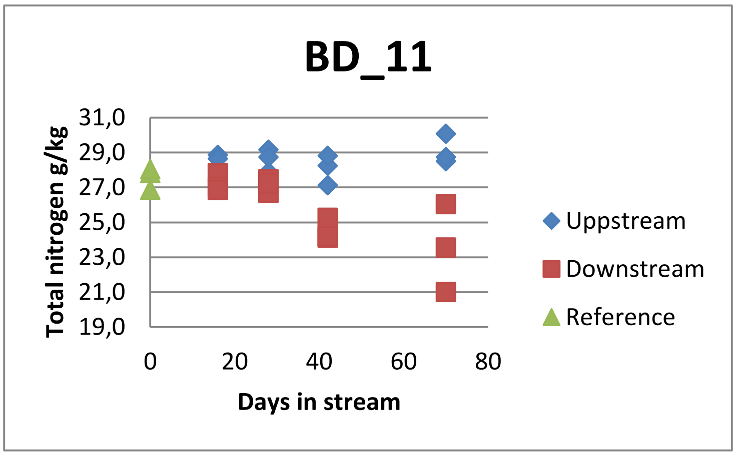
Appendix 4

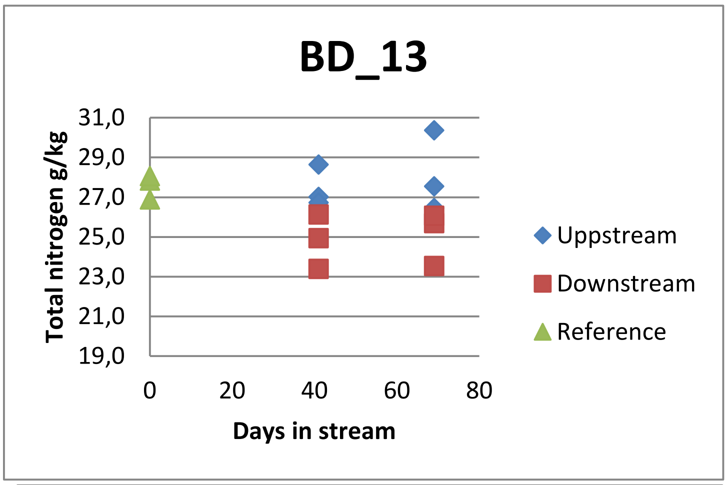
Nitrogen content in *F. antipyretica* in different beaver systems BD 01-03 is in Luleå region, BD 11, 13, 14 is in Sundsvall region and BD 21-26 is in the southern region.

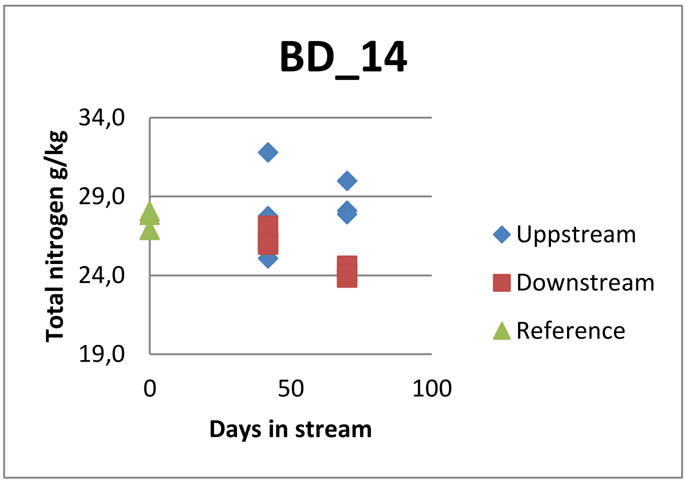


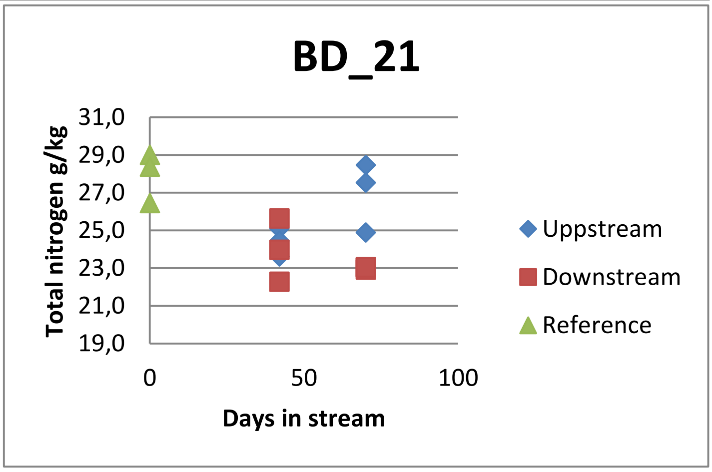


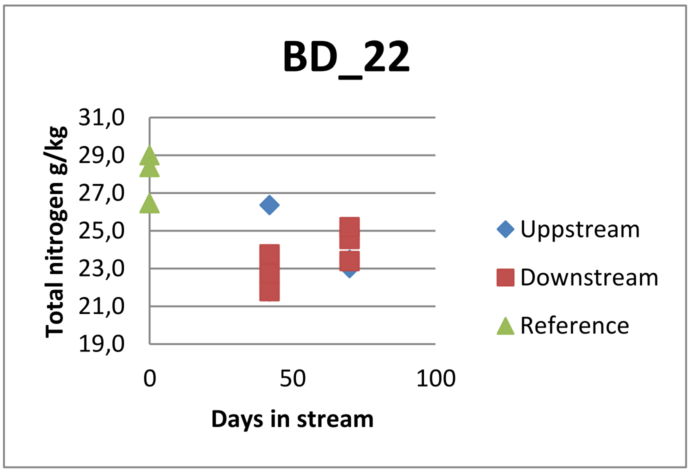


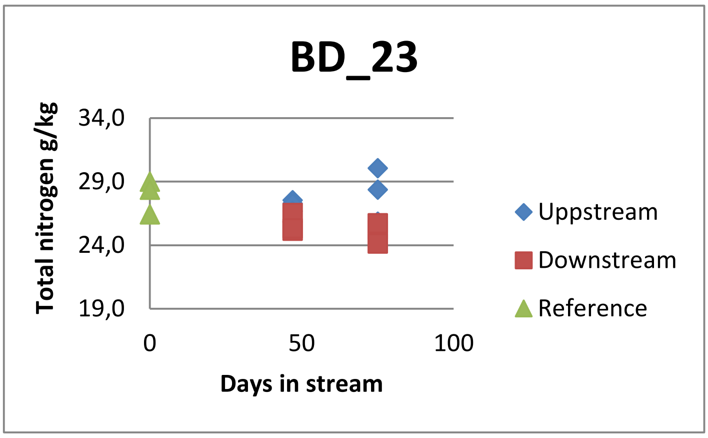


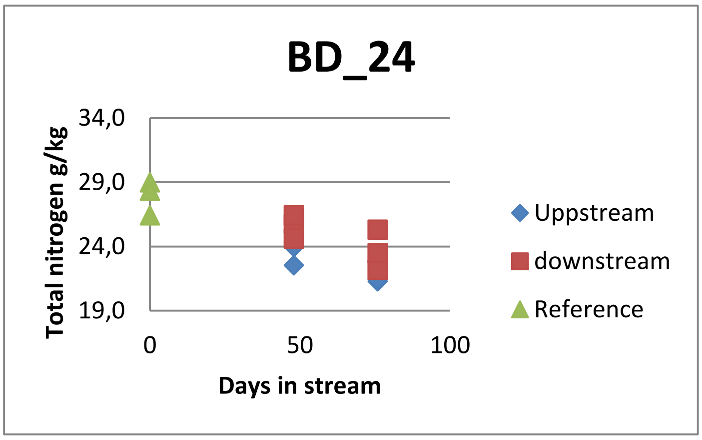

BD_13
Total nitrogen g/kg
31,0
29,0
27,0
25,0
23,0
21,0
19,0
0
20
40
60
80
Days in stream
Uppstream
Downstream
Reference

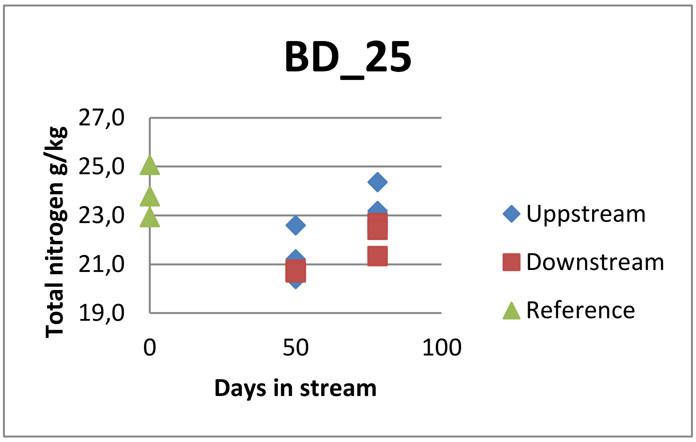

BD_14
Total nitrogen g/kg
34,0
29,0
24,0
19,0
0
50
100
Days in stream
Uppstream
Downstream
Reference

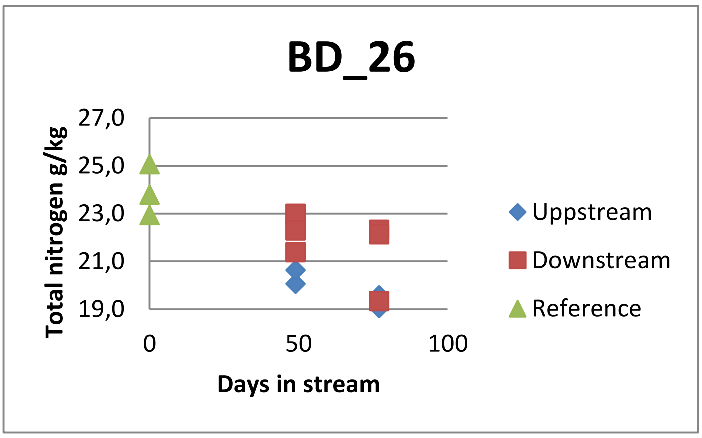

BD_21
Total nitrogen g/kg
31,0
29,0
27,0
25,0
23,0
21,0
19,0
0
50
100
Days in stream
Uppstream
Downstream
Reference


BD_22
Total nitrogen g/kg
31,0
29,0
27,0
25,0
23,0
21,0
19,0
0
50
100
Days in stream
Uppstream
Downstream
Reference

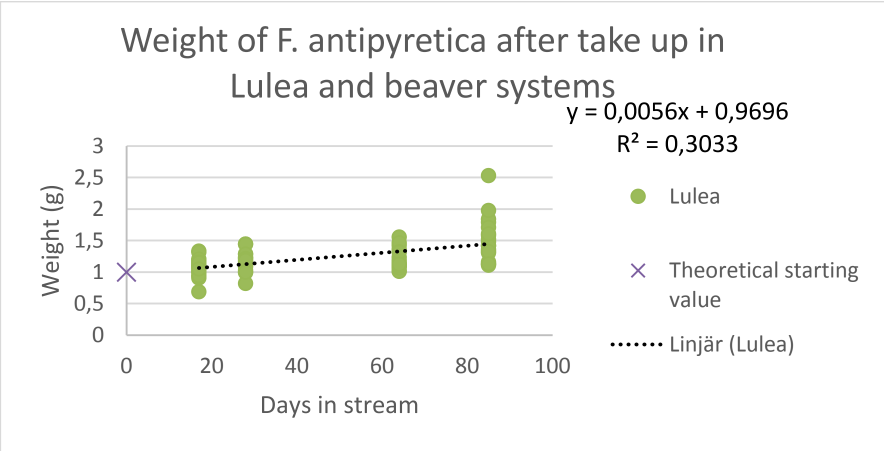

BD_23
Total nitrogen g/kg
34,0
29,0
24,0
19,0
0
50
100
Days in stream
Uppstream
Downstream
Reference


BD_24
Total nitrogen g/kg
34,0
29,0
24,0
19,0
0
50
100
Days in stream
Uppstream
downstream
Reference


BD_25
Total nitrogen g/kg
27,0
25,0
23,0
21,0
19,0
0
50
100
Days in stream
Uppstream
Downstream
Reference


BD_26
Total nitrogen g/kg
27,0
25,0
23,0
21,0
19,0
0
50
100
Days in stream
Uppstream
Downstream
Reference

Appendix 5



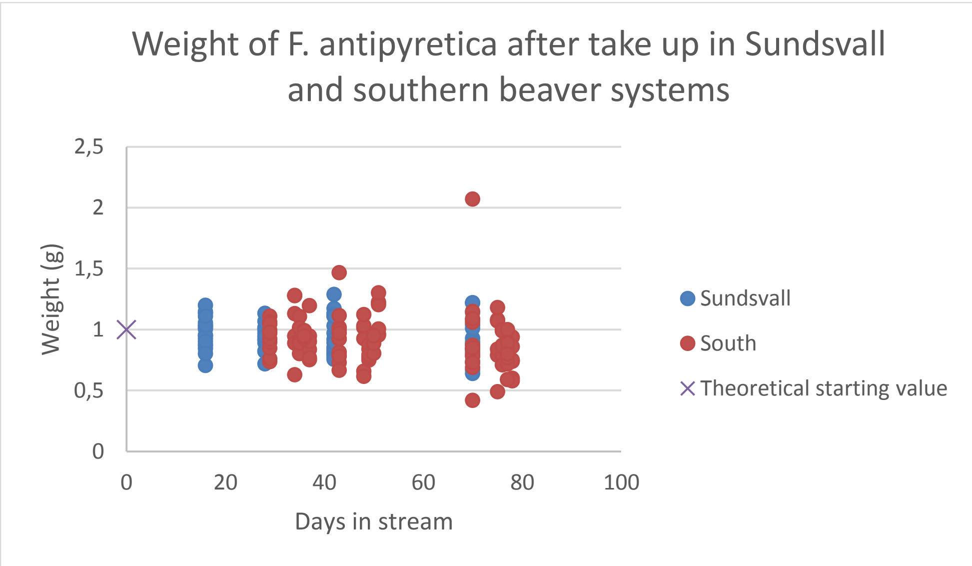


Figure 1 & 2: DW of *F. antipyretica* after have been taken up from beaver systems in Sundsvall, Lulea and southern Sweden. Both upstream and down streams are included. Only Lulea show significant weight increase.

Appendix 6

Table 1 Correlations with nitrogen content in *F. antipyretica*.

Parameter	P-value	Pearson product-moment correlation coefficient
Canopy cover	0,728	0,029
DOC	0,331	0,081
pH	0,534	-0,051
Kond. mS/m25	<0,001 ***	-0,315
Tot-N ug/l	0,847	-0,016
NO2+ NO3	0,082	-0,144
Tot-P	0,763	-0,025
SO4 mekv/l	0,062 [a]	-0,361
Alk./Acid mekv/l	0,413	-0,068
Weight	0,004 **	0,236

[a] one beaver system showed high values of SO_4 those were considerd outliers and were removed from the test. The p-value was initially <0,001.

Table 2 Correlations with DW in F. antipyretica.

Parameter	P-value	Pearson product-moment correlation coefficient
SO4 mekv/l	0,212	-0,104
Canopy cover	<0,001	-0,385
DOC	0,077	-0,147

pH	**0,006**	**-0,227**
Kond. mS/m25	**0,001**	**-0,266**
Tot-N	**0,008**	**-0,220**
NO2+ NO3	**0,001**	**-0,279**
Tot-P	**0,019**	**-0,194**
Alk./Acid mekv/l	**0,022**	**-0,189**

Appendix 7

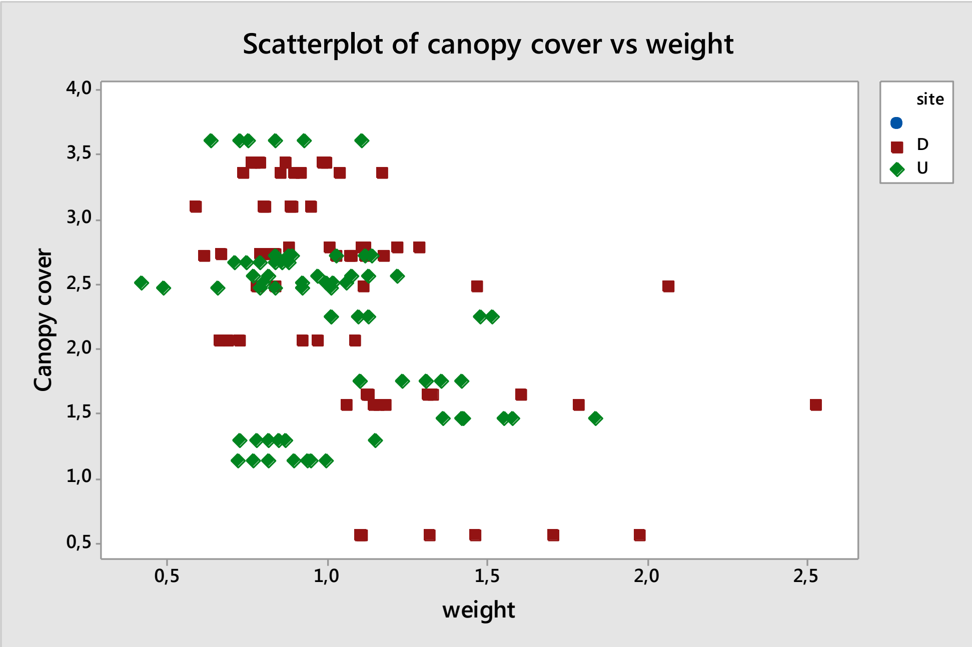
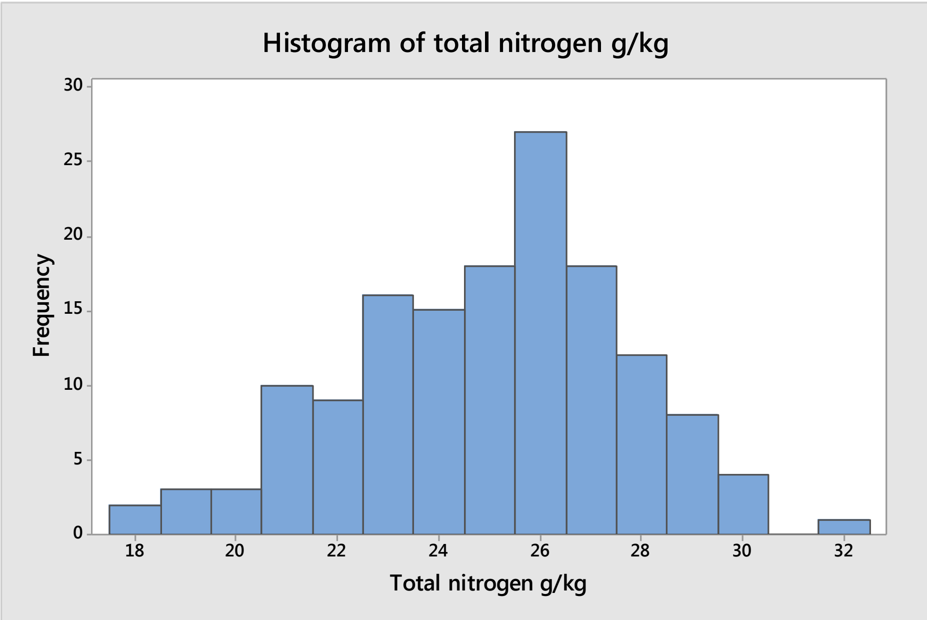


Figure 1: Canopy cover and DW of the aquatic moss *F. antipyretica.* Pearson product-moment correlation coefficient is -0.385. There is a significant increace of DW when canopy cover is lower (p<0,001).

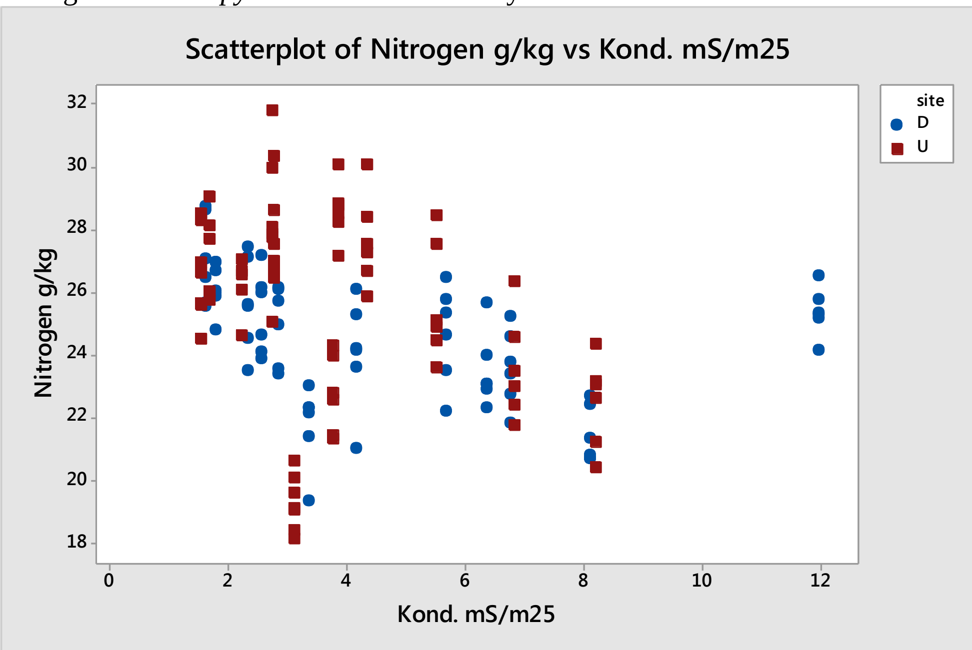
Appendix 8



Figur 1: Histogram for total nitrogen for all the *F. antipyretica* in the study showing normal distribution.

Appendix 9

Nitrogen in *F. antipyretica* and conductivity in the water correlation.



Appendix 10

Nitrate and nitrite for reused (fig.2) and pioneer (fig. 1) beaver systems.