RAPPORT
3 • 2008

Mercury Loading from forest to surface waters:

The effects of forest harvest and liming

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Papper
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Tryck
SJV, Jönköping

Upplaga
120 ex

ISSN 1100-0295
BEST NR 1790

Skogsstyrelsens förlag
551 83 Jönköping

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Foreword

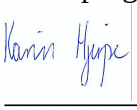
The Swedish Forest Agency has worked with measures to counteract soil acidification for more than 15 years. Based on knowledge and experience gained, an action programme was presented in 2001. The programme focused on countermeasures for acidification caused by pollution, compensation for nutrient removal with extraction of harvest residues, and management strategies adjusted to sustainable forestry. The implementation of the programme was proposed to contain a preparatory phase of three years followed by an action phase of ten years. The preparatory phase aimed at investigating and solving issues identified in the action programme. Further, large scale liming was suggested to develop tools for practical implementation.

In late 2004 the government permitted the Swedish Environmental Protection Agency to allocate SEK 10 million for measures to counteract acidification of forest soils according to the preparatory phase of the action programme. The Swedish Forest Agency developed, in co-operation with the Swedish Environmental Protection Agency, a project plan spanning from 2005 to 2007. In this plan, the study presented in this report was outlined.

Forest management has been found to affect the output of methyl mercury from soils to surface waters. In the present report, results from a study on the mercury effect of forest harvester tracks on the forest floor as well as on stream crossings, and forest liming are presented. Further, the report includes results from a study to develop an assay for net methylation potential in soils.

The authors are solely responsible for the report and the views in it. Consequently, it is not an expression of the views of the Swedish Forest Agency. The report has been reviewed by Kjell Johansson, the Swedish Environmental Protection Agency and the Department of Environmental Assessment, Swedish University of Agricultural Science.

Jönköping 2008



Karin Hjerpe
Project manager

Executive summary

Forest harvesting and other management/manipulation of forests in the boreal zone increase the loading of total mercury (Hgtot) and methylmercury (MeHg) exported from catchments as well as the amount of mercury found in biota downstream. Liming has also been indicated as a factor that can increase losses of Hg from forest catchments to streams. This raises serious questions about the potential role that forestry has played in the disastrous situation we now have, where mercury levels in pike and perch exceed health recommendations for human consumption in more than half of the Swedish lakes.

A paired catchment study is underway at Balsjö, in Västerbotten to improve the understanding of forest harvest impacts. The Swedish Forest Agency has supported a complementary project (2005-2007) to better address several issues about how forestry influences MeHg/Hgtot in surface waters. Those issues are the effects of 1) forestry tracks on shallow ground waters, 2) forest machinery stream crossings, 3) to explore the possibility of creating a measure of methylation potential in soils, and finally 4) the liming of forest soils where the riparian zone is not specifically excluded from liming. This report summarizes the results of these investigations as of October, 2007.

Tractors Tracks: The results from this study illustrate the variable nature of soil MeHg-concentrations. Before harvest soil MeHg-concentrations were similar throughout the entire study area. The tracks were created during the harvest in March 2006, when there was ca 1m of snow on the ground. No obvious disturbances from the tracks were noticeable at the soil surface, other than a slight compression of the soil. After harvest a tractor track and control points 3 meters to the side of the track had higher concentrations than control points elsewhere in the study area. If we assume that the 3 m control points were affected by compaction, then the harvest has increased the soil MeHg-concentration. However, if it is assumed that the 3 m control points were not affected by harvest, which cannot be proven, the harvest would not have affected the soil MeHg beneath the tractor tracks.

Stream Crossings: Whatever tractor tracks contribute to increased net methylation and total Hg concentrations in shallow groundwater under and close to the tracks, their impact on aquatic ecosystems is dependent on moving the increased concentrations of MeHg to streams. The places where tracks can be most effectively “connected” to surface waters are where tractors cross streams. The Balsjö study, as an example of good forest practice, had only one tractor crossing. Five of six sample pairs from above and below that crossing had no significant difference in MeHg and Hgtot levels. The single exception however points to the need to consider crossings as a potential “hot spot”. More analyses of this crossing, and crossing from logging operations in central Sweden are being investigated. There remains all reason to maintain the general recommendation to avoid driving across streams for the sake of mercury, as well as other water quality influences.

Methylation potential: The tractor crossing study illustrates the challenge of looking for the environmental influence of forestry when it may be express itself only intermittently. This is the background to the search for a measure of soil's potential to methylate Hg, as opposed to having to rely on instantaneous observation of MeHg concentrations, or even net methylation rates. A candidate methodology for measuring methylation potential in soils was created. The basis of this method is the use of additions of isotopically labeled total mercury and methyl mercury to be able to quantify both methylation and demethylation and consecutively also the rate of net methylation. To further access the controls of net methylation we also used additions of an energy source (carbon), and an electron acceptor (sulfate) in combination with labeled mercury species. While the concept was demonstrated, the resources required to apply this methodology were too high for routine use in the Balsjö project.

Forest Liming: A final component of this program has been to monitor the effects of forest liming on MeHg and Hgtot in runoff. Liming of forest soils is being studied as a measure to combat soil acidification in SW Sweden at several sites. Since this liming can extend to the near-stream areas that influence MeHg/Hgtot transfers from soils to surface waters, the effect of this liming on stream mercury levels was important to evaluate. The results to date from study streams in SW Sweden do not indicate any marked, consistent effect of liming on mercury outputs. Observation of the effects will continue until there is one full year of post-liming data on the sites that were limed.

Introduction

Alarming levels of mercury in the fish of many remote forest lakes have long been a concern in Sweden. The initial focus in the 1970's and 1980's was on the role of mercury (Hg) deposition and acidification. This has given way during the last decade to an awareness of the role that catchment processes play in methylating Hg and making a part of the atmospherically deposited Hg available for bioaccumulation in aquatic ecosystems. Swedish researchers have been instrumental in advancing the understanding of the Hg problem and defining a critical load for Hg deposition (Meili et al. 2003). These advances include localizing and identifying processes in the forest floor and near-stream wetland areas that increase the loading of bioavailable methylmercury (MeHg) to the aquatic ecosystem (1995), as well as showing the role of water table fluctuations in stimulating the sulfur reducing bacteria that are particularly effective in methylating mercury (Branfireun et al., 2001). This pointed to the potential role that forest management has in the Hg problem:

The great challenge for science and policy is to define and evaluate the prospective management alternatives, such as riparian management in forestry and reservoir regulation that might mitigate the enduring threat posed by Hg output from catchment soils to surface waters. (Bishop and Lee, 1997)

Several studies have since indicated that harvesting and other management/ manipulation of forests in the boreal zone increase the loading of methylmercury exported from catchments or the amount of mercury found in downstream fish (Garcia and Carignan, 2000; Porvari and Verta, 2003). Liming has also been indicated as a factor that can increase losses of Hg from forest catchments to streams (Meili, 1997). This raises serious questions about the potential role that forestry has played in the disastrous situation we now have, where mercury levels in pike and perch exceed health recommendations for human consumption in more than half of the Swedish lakes (Munthe and Hultberg, 2004). While existing research has raised questions, there is still far too little data to judge the actual contribution from forestry, and the possibility of mitigating these effects through measures such as riparian buffer zones (Munthe, 2003). To address these issues, the Swedish Forest Agency has supported this project to study:

1. The mercury effect of machine tracks on the forest floor.
2. The mercury effect of machine tracks at stream crossings.
3. Development of an assay for net methylation potential in soils.
4. The effect of forest liming on the near stream zone.

The first three studies are co-located at Balsjö, near Bjurholm, in Västerbotten, with the EU Life project: Forests for Water, in which forestry is put in relation to the Water Framework Directive. A FORMAS project (2005-2008) is specifically investigating the effects of forest harvest on mercury outputs in a paired catchment study at Balsjö. The final study was added to liming studies conducted by the Swedish Forest Agency itself and IVL, performed in different catchments in Southern Sweden.

Background

Previous studies have identified biogeochemical and hydrological conditions that can promote the transfer of more MeHg and Hgtot from soils to surface waters.

Biogeochemical conditions

The environment has a natural background level of mercury. But since the industrial revolution the combustion of fossil fuels and other anthropogenic activities have caused high deposition of inorganic Hg in the environment. The deposition is now decreasing, but there will be high levels of Hg present in the soils for centuries to come. The inorganic species of Hg is generally not a direct threat to organisms, though the threshold at which soil ecology is altered by Hg remains a subject of contention (Bringmark and Bringmark, 2001a; Bringmark and Bringmark, 2001b). Through methylation inorganic Hg can be transformed into the organic species methyl mercury (MeHg) which is a potent toxic compound that accumulates in the food web. Parallel to methylation, demethylation also occurs. So what one sees in the environment is the net balance of these antagonistic processes.

As a byproduct of acquiring energy, sulphur reducing bacteria (SRB) methylate Hg into MeHg. The general conditions required for these SRB are the presence of SRB, sulfur, Hg and a high quality carbon source, as well as anoxic conditions. Higher temperatures can also favour net methylation, given the availability of Hg^{2+}, energy and electron acceptors. Locations where net methylation is high due to synergies between these controlling factors are commonly referred to as methylation hotspots.

Hydrological conditions

Methylation can occur in surface waters (Xun et al., 1987), their sediments or in soils. When methylation takes place in soils, hydrology has an important influence on the redox conditions and it is responsible for transporting MeHg to surface water where it enters the food web. Consequently, to understand the MeHg production and the transport of MeHg from soils to surface waters also requires an understanding of the hydrology. Forest harvest reduces transpiration for a period of time, during which wetter soil conditions can promote methylation, and more rapid export of water from potential terrestrial methylation hotspots.

Specialized Harvest Disturbance Studies: Tractor tracks and Stream crossing

It is possible that much of the forest harvest effect on MeHg that has been observed in other studies derives from localized increases in the generation and output of methyl mercury on the tracks created by harvesters and forwarders. The compaction from heavy machinery can change the physical properties of the underlying soil. The high-porosity, sponge-like structure of the organic horizon is particularly susceptible to such disturbances.

In the Balsjö Research area stream chemistry concentration has been monitored bi-weekly since late 2004. In March 2006 one entire 37 ha catchment was clear cut without buffer zones, a nearby, 11 ha catchment was cut leaving buffer zones in the riparian zone (Figure 1 and 2), and a third 20 ha control catchment was not harvested (Appendix A1/A2).

Figure 1. View over the harvested area where a buffer zone is left along the stream. Photo: Lars Högbom, Skogforsk

Tractor tracks – Forest Floor

Figure 2. Logging just outside of the riparian buffer zone area. Photo: Lars Högbom, Skogforsk

To study the effect of tire tracks on the forest floor, two "Harvest:Tracked" transects were established on "Area North" between transect T12 and T13 (Figure 3 and 4). Each track was 30 m long and the harvester drove back and forth half a dozen times to make sure the soil was affected. In each of these tracks groundwater tubes were installed for ground water sampling (TractorTrack). As controls, groundwater tubes were installed 3 meters to the side of the track (TTrackRef). In addition other groundwater tubes throughout the Balsjö study area (5 within a similar soil type as the Tractor Track study site) form a secondary set of controls (Harvested). The groundwater was sampled in the fall of 2005 (prior to harvest), 2006 and 2007. The samples from 2007 have not yet been analyzed.

When sampled in the autumn of 2005, the tractor tracks and the 3-m controls had values of MeHg and Hg in shallow groundwater that did not diverge from the other shallow groundwater samples in organic soils in the catchment prior to harvest. However, TOC levels were significantly lower ($p=0.025$) in the Tractor tracks prior to harvest.

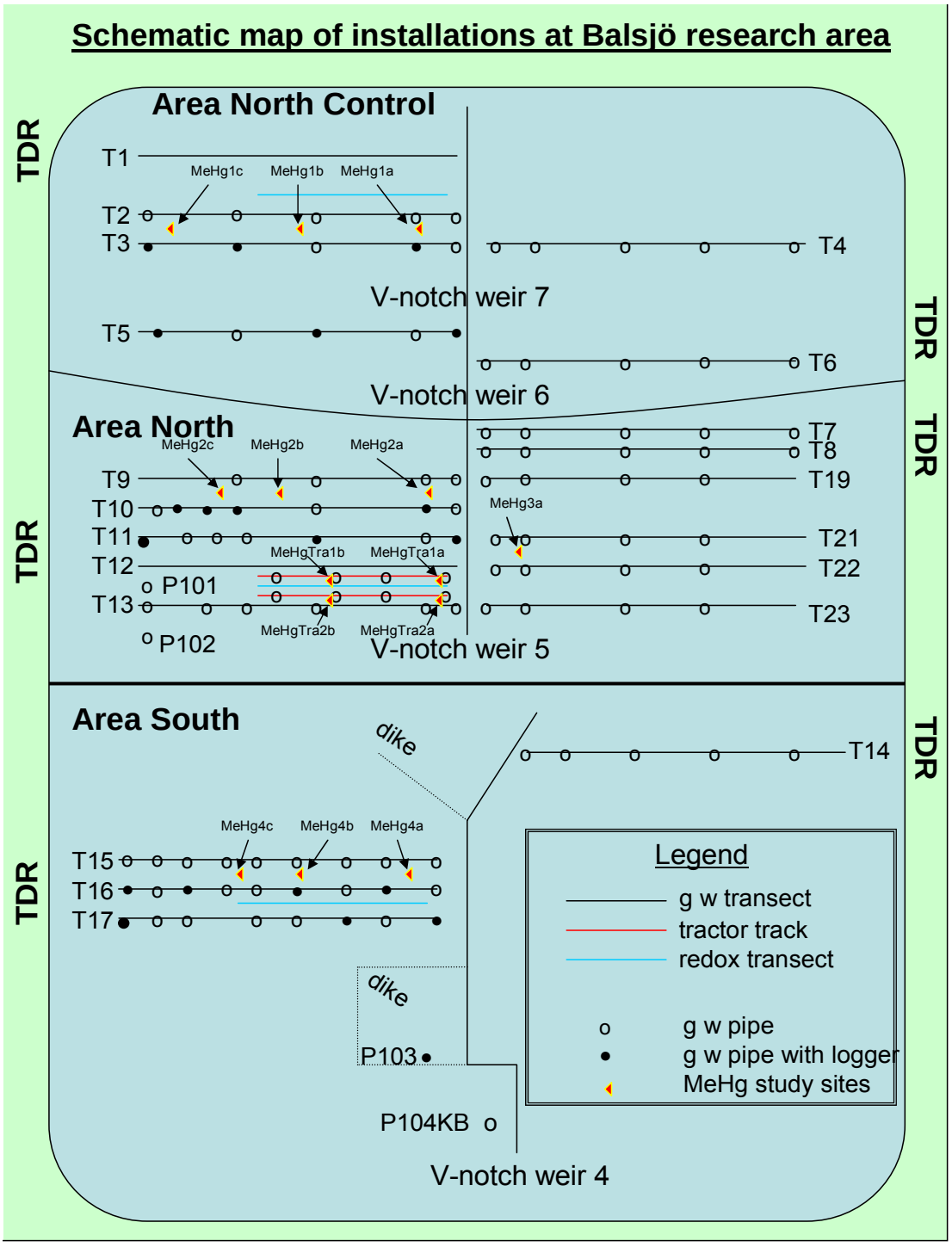


Figure 3. Schematic layout of the Balsjö Catchments Study, showing location of the tractor studies on the "Area North" catchment subjected to harvest with a riparian buffer zone.

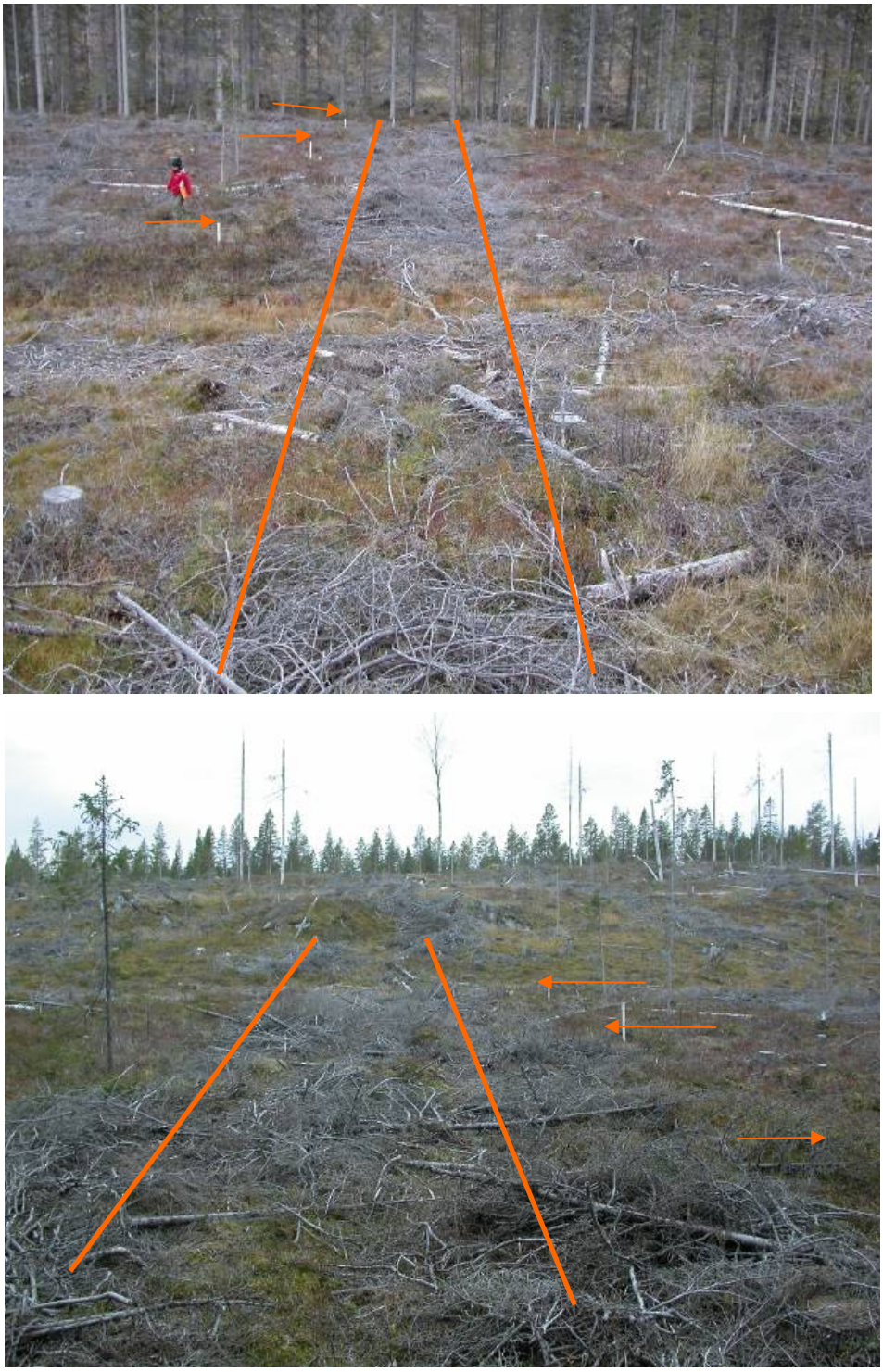


Figure 4. Site of the "Track studies". Above: looking down towards the stream. Below: looking uphill from the stream. The "control" groundwater tubes (TTrackRef) are noted with arrows. The groundwater tubes on the track itself (TraktorTrack) had been removed in late 2005 so they would not be destroyed by the tractor driving on the "track". These groundwater tubes were reinstalled shortly after the picture was taken in April, 2006.

After the harvest (Figure 5), in 2006, the shallow groundwater under the tracks together with the shallow groundwater parallel to the tracks had higher levels of MeHg than the shallow groundwater elsewhere in the harvested area. When

grouping the MeHg values from the Tractor Track and the TTrackRef these were significantly higher than the MeHg values from the groundwater samples from the rest of the sites (i.e. Harvested) to the 95th percentile. Considering the 90th percentile both the track itself and the "3m"-controls had higher MeHg concentrations than the reference points. For Hgtot and DOC no such difference was found.

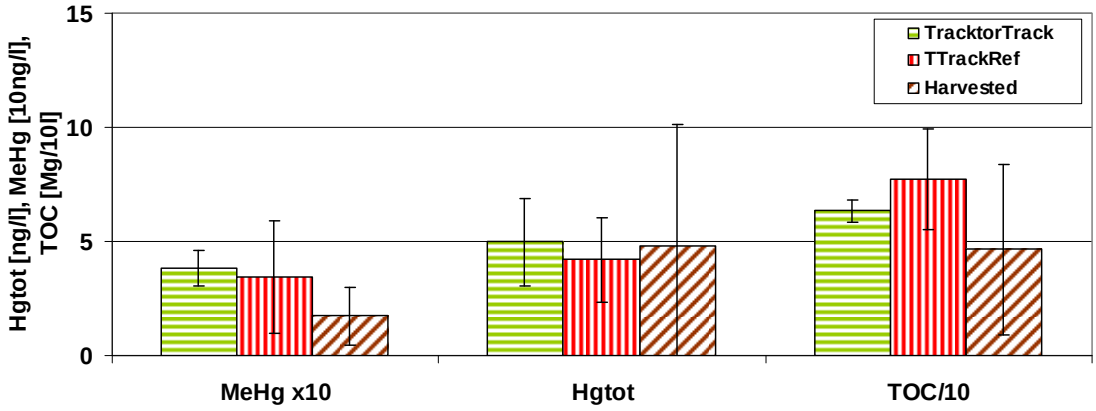


Figure 5. Mean concentrations of MeHg(multiplied by 10), Hgtot, and TOC(divided by 10) for the three treatment classes (columns). Bars display standard deviation.

Discussion of Track studies

Previous studies have shown that forestry can increase the output of Hg to surface waters. Garcia and Carignan (1999; Garcia and Carignan, 2000) observed elevated levels of MeHg in zooplankton and pike downstream from a clear-cut. Similarly, leakage of MeHg and Hgtot increased after clear-cutting a Finnish forest (Porvari et al., 2003). In Värmland, Gilles (2002) observed higher Hg-levels induced by forest activities, including rigorous soil damages from heavy machinery. In 1999 in the Gårdsjön research area in the vicinity of Göteborg a heavily impacted tractor track induced elevated leakage of MeHg to the nearby stream even though no forest was harvested in the catchment. A reasonable hypothesis is that the tractor track either created ponding with stagnant water on the hill slope resulting in anoxia and thus creating a methylation hotspot, or the track "connected" an existing upslope methylation hotspot to the stream (Munthe and Hultberg, 2004).

The results of the Balsjö track study indicate that both the tracks themselves, and the "3 m reference" points, have higher MeHg concentrations than the reference points elsewhere in the area (Harvested) (90th percentile). The size of the increase, essentially a doubling of MeHg, against the background reference points of considerable variability, is not as large as the effects seen after harvest on several other studies where there were manifold increases in MeHg outputs after harvest. Since the local controls (TTrackRef) as well as the tractor tracks increased after tracking, it cannot be determined whether the disturbance caused the increase or another aspect caused it, i.e. whether the "3m"-controls (TTrackRef) still act as controls even after the disturbance.

Given the large natural variability though, both within sites, and between catchments, we need more such studies to minimize uncertainty and definitely resolve this question.

A clear finding from the Balsjö study is that only little disturbance of the soil was visible on the surface. Whether the soil compaction caused increased MeHg output we cannot be sure, but even if the increase was human induced, the increase was not as large as seen in other disturbance studies. Thus this study suggests that good forest practise can minimize alterations in the soil chemistry.

Tractor tracks – Stream Crossings

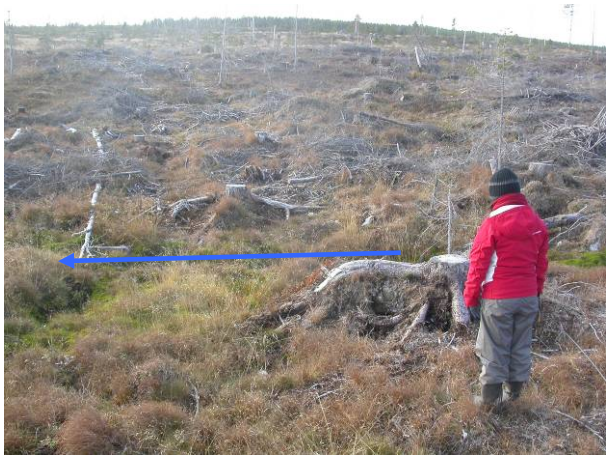


Figure 6. Stream crossing 18 months after disturbance. The stream is indicated by a blue arrow.

The effect of soil compression/disturbance in tracks becomes a problem for surface waters if and when the higher levels of mercury reach streams. Stream crossings for machinery are a location where the connections between tracks and surface waters can occur most readily. To study the effects of tractor tracks on stream crossings, one stream crossing located in the entirely clear cut area was sampled 6 times after the harvest (Figure 6). Water samples were taken 20 m upstream and 20 m downstream from the crossing, which is an example of poor practice with no efforts made to reduce crossing effects.

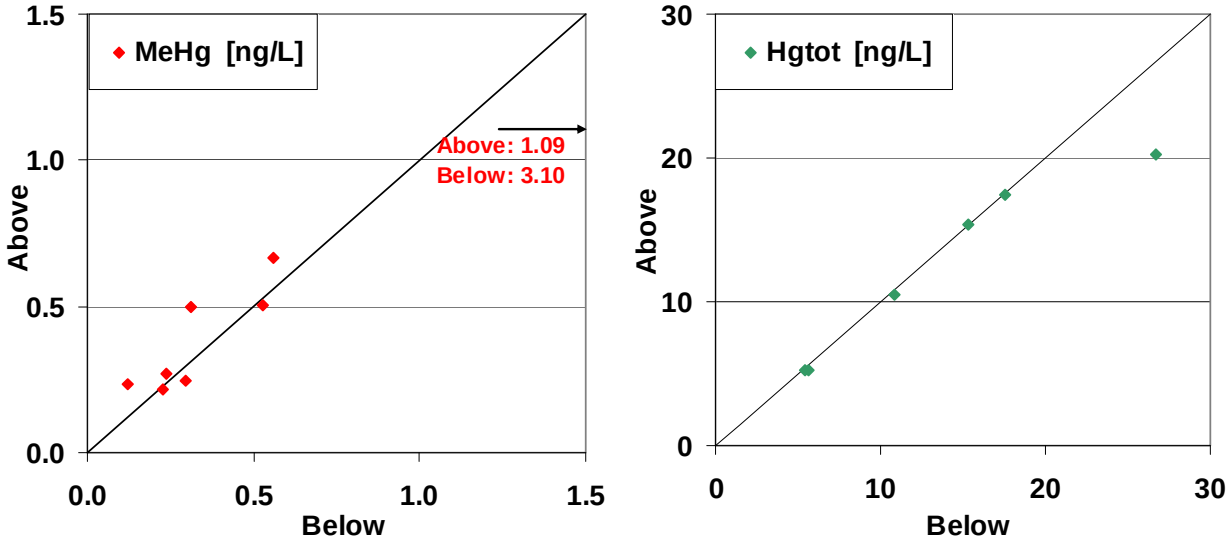


Figure 7. Stream concentration plot of MeHg (left) and Hgtot (right) from above and below the tractor stream crossing. One value from the MeHg-diagram is off scale, (below:3.10; above:1.09). The divergent MeHg value is not the same sample as the divergent Hgtot value visible in the right diagram.

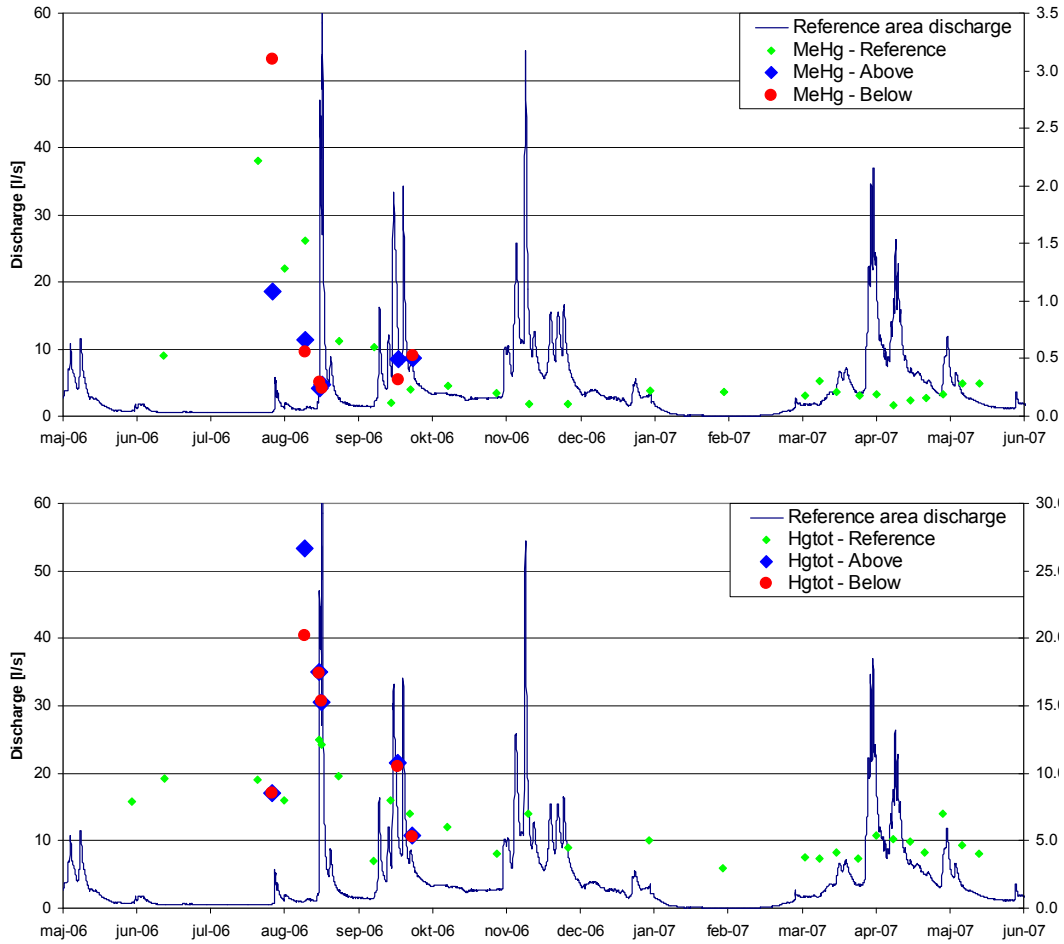


Figure 8. Timeseries of discharge and concentrations of MeHg (top) and Hgtot (bottom) from the Balsjö reference catchment together with concentrations of MeHg and Hgtot observed above (blue diamonds) and below (red dots) the stream crossing.

On five of six sampling occasions, no differences were observed above and below the stream crossing (Figure 7 and 8). On one occasion, in August 2006, after a long dry period, MeHg downstream from the crossing was much higher, though, than above the crossing. Two weeks later and after a moderate late summer rain, the Hgtot downstream was elevated, while the MeHg was not.

Discussion of Stream Crossings

The high value of MeHg concentration below the stream crossing (Figure 7) was observed after a long dry period in the summer. The reference site in the Balsjö research area (Figure 8) also had high concentrations under the same hydrological conditions. The highest MeHg concentrations are usually observed at this particular time of the year. The results suggest that this crossing has not become a consistent hotspot of mercury output, but it cannot be ruled out that it has this role occasionally.

Finding stream crossings was difficult since the harvest was done very carefully (according to NATURA2000 guidelines). Another adjacent area had been consid-

ered for a tractor stream crossing, but this had to be cancelled when it was discovered that it drained into one of the other treatment catchments. We are, however, collecting stream crossing data from an area in the vicinity of Örebro where we study the effects of stump-harvest on stream chemistry. The study of the crossing at Balsjö will also continue with more samples in order to investigate if there are any changes in a longer-term perspective.

Soil Methylation Potential

Inorganic mercury is derived from the atmosphere through dry and wet deposition and microbiologically transformed (methylated) into methylmercury within wetland areas.

Two counteracting processes control the presence of methylmercury within a given catchment: mercury methylation and methylmercury demethylation. Mercury methylation is primarily carried out by sulphate reducing bacteria (SRB) and the environmental factors controlling the SRB activity are as important as is the availability of inorganic mercury. Electron acceptors (sulfate) and electron donors (high quality carbon) ultimately controls SRB activity (Figure 9).

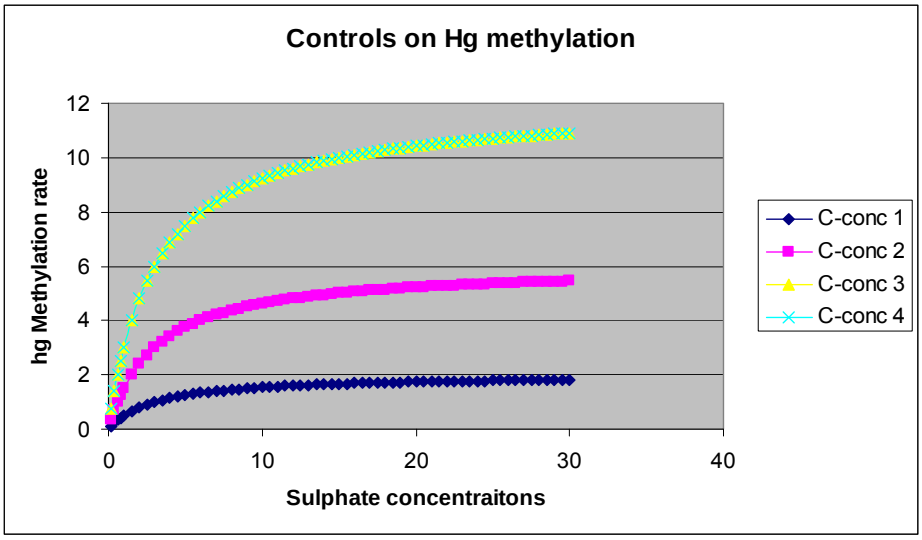


Figure 9. SRB methylation rate response to sulphate concentration at different concentrations of easily available carbon.

We have received support from FORMAS to use advanced isotope techniques to identify the methylation going on at a particular point in time (when we sample). Since methylation rates can vary during a season, from place to place, and in relation to the redox situation (e.g. sulfate availability) many such measurements would be needed to define the effect of forest harvest on methylation in the soil.

One way of getting a more robust indication of the effect of forestry on Hg methylation in the soil is to go from instantaneous measurements of methylation to defining the methylation potential of the soil. Essentially that is to measure the methylation under conditions where the soil microorganisms are supplied with an excess of nutrients and energy sources. There are relatively well-established procedures for developing such a measure of “methylation-potential” for different processes in microbiology. In this study, we experimented with the levels of carbon and sulfate needed to get a useful measure of soil methylation potential. This was conducted during the spring/summer of 2005.

An isotope dilution (ID) based method using gas chromatography with inductively coupled plasma mass spectroscopy (GC-ICP-MS) was used to determine the rate of methylation and demethylation simultaneously. By altering the concentrations of electron acceptors, electron donors and inorganic mercury in a peat sample

from the riparian zone, it was possible to determine their individual contribution to the rate of methylation and demethylation.

It was found that the rate of mercury methylation is limited by the availability of glucose, sulphate and inorganic mercury (Figure 10). Addition of only Hg^{2+} increases the rate of methylation slightly while further addition of sulfate results in a five-fold increase. Given excess of sulfate, addition of glucose further increased the rate of methylation. These results indicate the availability of sulfate to constitute the master control on methylation of mercury in this system.

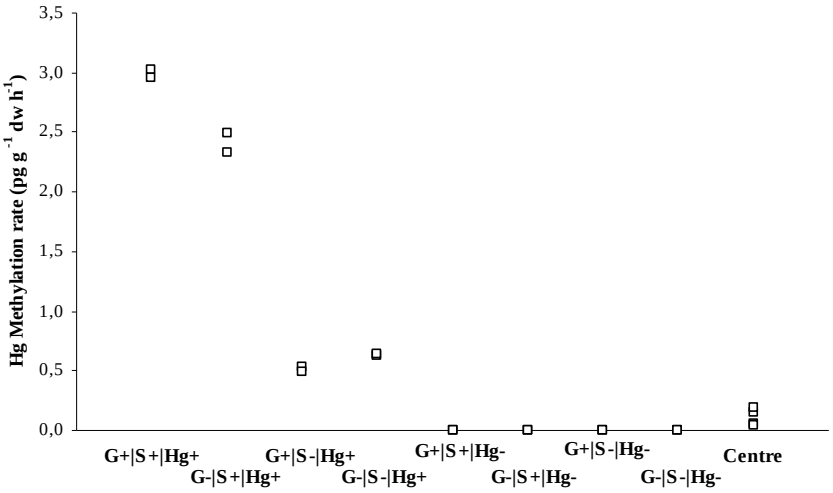


Figure 10. Response in rate of methylation to additions (+) or no addition (-) of mercury (Hg), glucose (G) and sulfate (S).

Discussion of Soil Methylation Potential

The findings presented in this study indicate that when mercury is in sufficient abundance, addition of sulfate and glucose increases the rate of mercury methylation further. The demethylation rate was not affected by addition of any of the three factors. Traditionally, the chemical behaviour of inorganic mercury has been the centre of attention whenever the mercury problem has been considered. This study both confirms the importance of biological induced mercury methylation but also stresses the importance of the complex control from available mercury, electron donors and electron acceptor on the rate of methylation. All these three controls are necessary to consider when addressing spatial and temporal variation in catchment distribution of methylmercury as well as the effect of environmental disturbance, i.e. forestry on aquatic bioaccumulation of methyl mercury.

This part of the study has demonstrated the feasibility of measuring a methylation potential.

While the concept was demonstrated, the resources required to apply this methodology were prohibitively high for routine use. Thus for the Balsjö paired catchment study of forestry effects, it was deemed more cost-effective to use the available resources to focus on more comprehensive “snapshots” of the mercury situation in shallow groundwater, rather than trying to systematically discern potential rates. For further information see Appendix B.

Forest Liming

Liming of forest soils is a way to remediate the linked problem of soil and surface water acidification created by acid deposition during the past century. There is some indication, however, that liming of the terrestrial environment can lead to increased outputs of mercury to surface waters (Meili, 1997). To test whether this is indeed the case with forest liming, the output of Hgtot and MeHg from catchments subjected to catchment liming of forest soils was investigated. During 2006 and 2007 a number of streams were sampled for mercury before and after liming in three areas in Southern Sweden, one in Götaland County, one in Halland County, and one in Jönköping County (table 1 and Figure 11). Samples were taken around 5-10 times per year before and after liming. At the present stage of writing this report post liming data was available for Götaland County (Figure 11). In addition to Hg-analysis our funding from the Swedish Forest Agency is even financing the analysis of general chemistry (performed by IVL) at some sites so that there will be a complete year of post liming data from all sites.

Table 1. Strems included in the evaluation of the liming project this far.

Götaland	Reference period begins	Number of samples before liming	Time of liming	Number of samples after liming
Alebäcken	20060404	14	Reference area	-
Habäcken	20060404	14	June 2007	5
Hulkebäcken	20060404	7	October 2006	12
Häbäcken	20060404	2	May 2006	17
Limbäcken	20060404	2	May 2006	17
Sågebäcken	20060404	7	October 2006	12

Figure 11. Location of the catchments included in the liming project. Map provided by the Swedish Forest Agency.

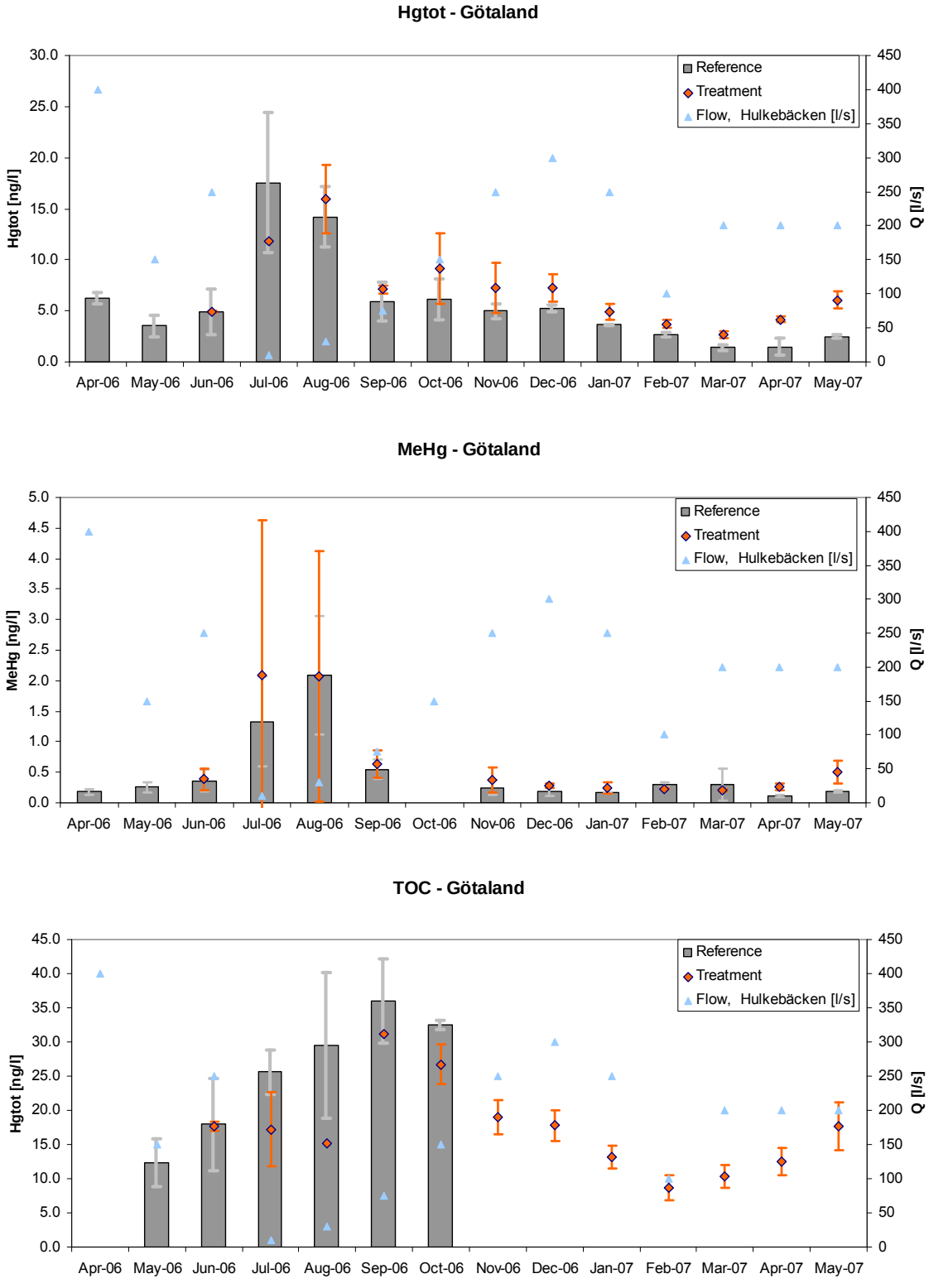


Figure 12. Mean values of Hgtot, MeHg, and TOC from the six catchment areas in Götaland County. Grey columns represent mean values of all reference and unlimed catchments. Orange diamonds represent mean values of all limed catchments. Grey and orange bars represent standard deviations.

Discussion of Forest Liming

Despite alterations in the general stream chemistry balance the results this far show no obvious effect of liming on the MeHg and Hgtot concentrations in the streams (Figure 12). Observation of the effects will continue until there is one full year of post-liming data on the sites that were limed, then a more comprehensive analysis will be made.

Timeseries from each individual stream are discussed in Appendix C, and contribute to the overall interpretation of the liming studies.

Future Reporting

All mercury analyses on samples taken during 2007 were not completed at the time of writing this report. Samples from the liming project are still being collected, and even so for the stream tractor crossing study. When sampling and analysis are finished we will summarize it to the Swedish Forest Agency.

References

Bishop, K., Lee, Y.H., Pettersson, C. and Allard, B., 1995. Terrestrial Sources of Methylmercury in Surface Waters - the Importance of the Riparian Zone on the Svartberget Catchment. Water Air and Soil Pollution, 80(1-4): 435-444.

Bishop, K.H. and Lee, Y.H., 1997. Catchments as a source of mercury/methylmercury in boreal surface waters. Metal Ions in Biological Systems, Vol 34, 34: 113-130.

Branfireun, B.A., Bishop, K., Roulet, N.T., Granberg, G. and Nilsson, M., 2001. Mercury cycling in boreal ecosystems: The long-term effect of acid rain constituents on peatland pore water methylmercury concentrations. Geophysical Research Letters, 28(7): 1227-1230.

Bringmark, L. and Bringmark, E., 2001a. Lowest Effect Levels of Lead and Mercury on Decomposition of Mor Layer Samples in a Long-Term Experiment Water Air and Soil Pollution, 1(3-4): 425-437.

Bringmark, L. and Bringmark, E., 2001b. Soil Respiration to Small-Scale Patterns of Lead and Mercury in Mor Layers of Southern Swedish Forest Sites. Water Air and Soil Pollution, 1(3-4): 395-408.

Garcia, E. and Carignan, R., 1999. Impact of wildfire and clear-cutting in the boreal forest on methyl mercury in zooplankton. Canadian Journal of Fisheries and Aquatic Sciences, 56(2): 339-345.

Garcia, E. and Carignan, R., 2000. Mercury concentrations in northern pike (Esox lucius) from boreal lakes with logged, burned, or undisturbed catchments. Canadian Journal of Fisheries and Aquatic Sciences, 57: 129-135.

Gilles, E., 2002. Den bäcknära zonen vid små skogliga vattendragi Värmland – Generell beskrivning, förekomstav traktorspår samt kvicksilverhalter ikörpåverkat ytvatten. Master Thesis from the Department of Forest Soils.

Meili, M., 1997. Mercury in lakes and rivers, Metal Ions in Biological Systems, Vol 34. Metal Ions in Biological Systems, pp. 21-51.

Munthe, J., 2003. Pathways and Processes of Mercury in the Environment. Selected papers presented at the Sixth International Conference on Mercury as Global Pollutant, Minamata, Japan, October 15-19, 2001 - Preface. Science of the Total Environment, 304(1-3): 1-1.

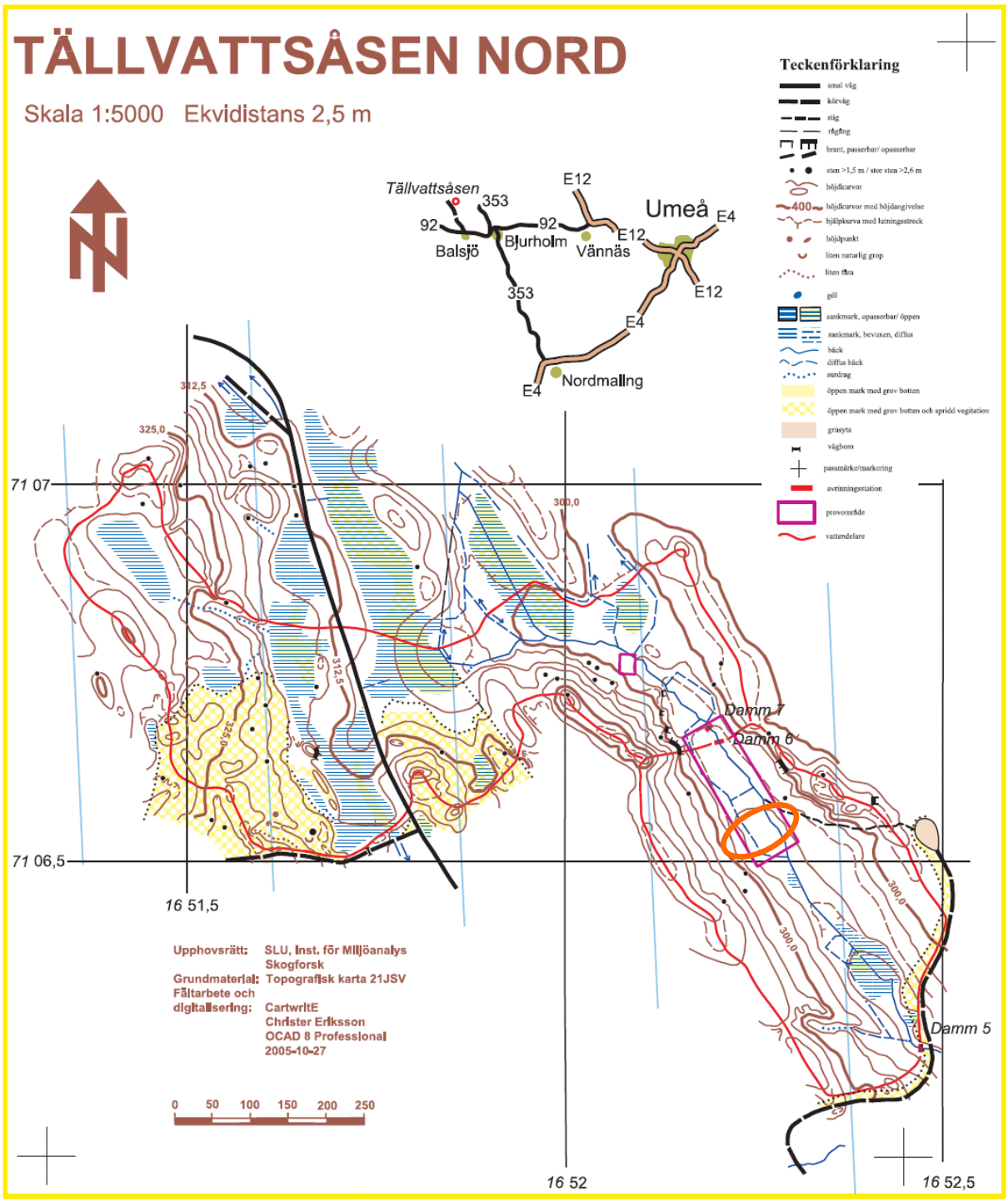
Munthe, J. and Hultberg, H., 2004. Mercury and methylmercury in runoff from a forested catchment - concentrations, fluxes, and their response to manipulations. Water, Air, and Soil Pollution, Focus: 4: 607-618.

Porvari, P. and Verta, M., 2003. Total and methyl mercury concentrations and fluxes from small boreal forest catchments in Finland. Environmental Pollution, 123(2): 181-191.

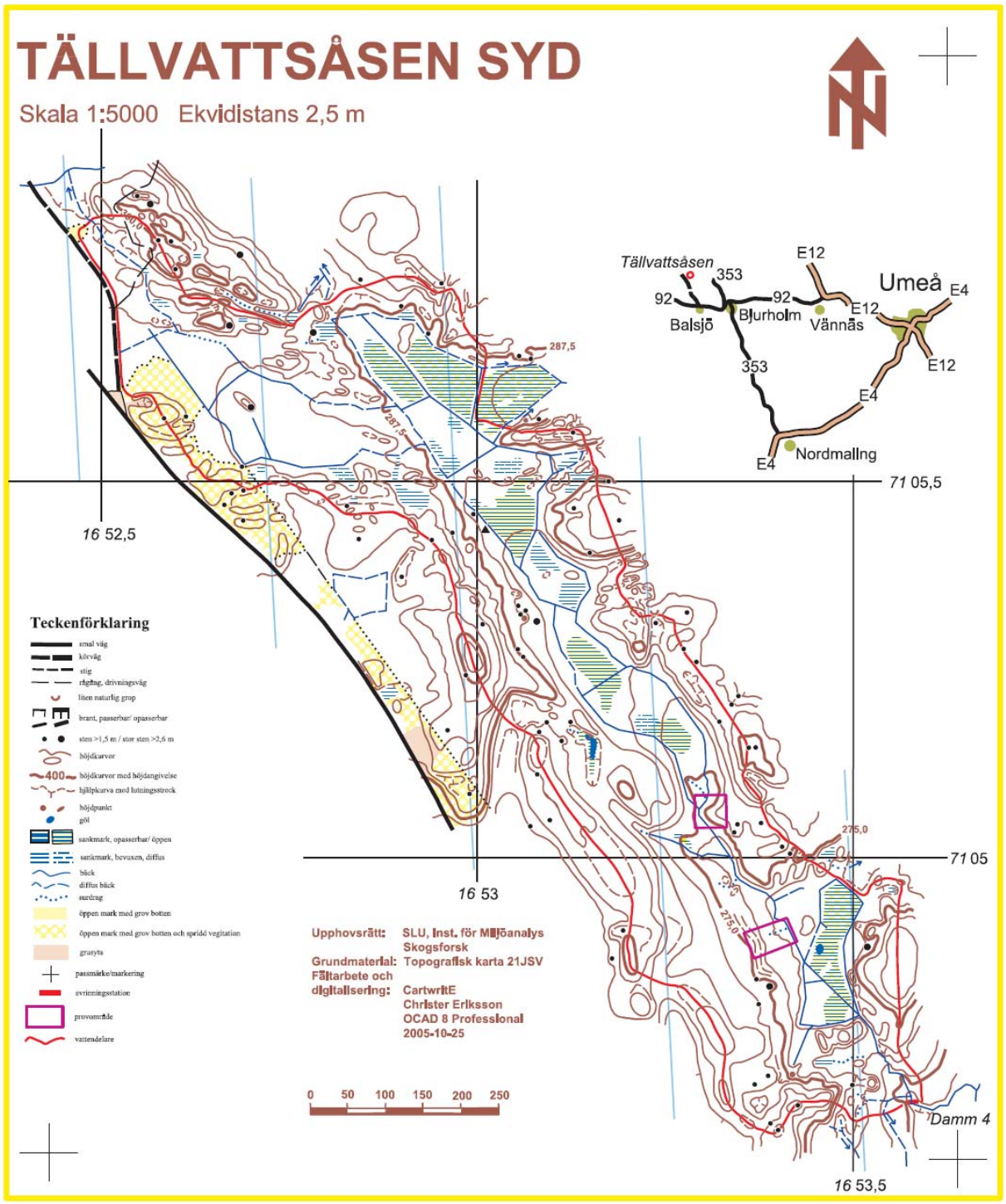
Porvari, P., Verta, M., Munthe, J. and Haapanen, M., 2003. Forestry practices increase mercury and methyl mercury output from boreal forest catchments. Environmental Science & Technology, 37(11): 2389-2393.

Xun, L., Campbell, N.E.R. and Rudd, J.W.M., 1987. Measurements of Specific Rates of Net Methyl Mercury Production in the Water Column and Surface Sediments of Acidified and Circumneutral Lakes. Canadian Journal of Fisheries and Aquatic Sciences, 44(4): 750-757.

Appendix A1. Area North Control - Reference area, and Area North - Buffer Zone Area



Appendix A2. Area South – Clear Cut Area



Appendix B. M.Sc.Thesis on soil methylation potential

Abstract

Methylmercury, one of the most toxic mercury species known to mankind, is able to accumulate in the aquatic food chain. This has resulted in fish consumption guidelines in Scandinavia and North America. Inorganic mercury is derived from the atmosphere through dry and wet deposition and microbiologically transformed (methylated) into methylmercury within wetland areas.

Two counteracting processes control the presence of methylmercury within a given catchment: mercury methylation and methylmercury demethylation. Mercury methylation is primarily carried out by sulphate reducing bacteria (SRB) and the environmental factors controlling the SRB activity are as important as is the availability of inorganic mercury. Electron acceptors (sulphate) and electron donors (high quality carbon) ultimately controls SRB activity.

In this study, an isotope dilution (ID) based method using gas chromatography with inductively coupled plasma mass spectroscopy (GC-ICP-MS) was used to determine the rate of methylation and demethylation simultaneously. By altering the concentrations of electron acceptors, electron donors and inorganic mercury in a peat sample from the riparian zone, it was possible to determine their individual contribution to the rate of methylation and demethylation.

It was found that the rate of mercury methylation is limited by the availability of glucose, sulphate and inorganic mercury. The findings presented in this thesis indicate that when mercury is in sufficient abundance, addition of sulphate and glucose increases the rate of mercury methylation further. The demethylation rate was not affected by addittion of any of the three factors.

Traditionally, the chemical behaviour of inorganic mercury has been the centre of attention whenever the mercury problem has been considered. This study both confirm the importance of biological induced mercury methylation but also stresses the importance of the complex control from available mercury, electron donors and electron acceptor on the rate of methylation. All these three controls are necessary to consider when addressing spatial and temporal variation in catchment distribution of methylmercury as well as the effect of environmental disturbance, i.e. forestry on aquatic bioaccumulation of methyl mercury.

Acknowledgements

I would like to thank **Mats Nilsson** at the Department of Forest Ecology, Swedish University of Agricultural Science, for your never-ending enthusiasm. I am especially grateful for the fact that you always make time for explanation and discussion, regardless of your workload.

None of the analyses would have been possible without the guidance from **Lars Lambertsson** at the Department of Analytical Chemistry, Umeå University. Thank you for your patience, Lars!

I would also like to thank Professors **Jim Harris** and **Karl Ritz** at Cranfield University at Silsoe for their critical review of and comments on my writing.

Nicklas Larsson, Umeå 24 August 2005

1. Introduction

Nearly all concern regarding mercury, (Hg), toxicity has been directed to its methylated form (monomethyl Hg; CH_3Hg^+; MeHg) within terrestrial and aquatic ecosystems (Hall & St. Louis 2004*a*). MeHg is a potent neurotoxin, which compared to inorganic Hg is highly lipophilic and able to penetrate both the placental and the blood-brain barrier (Grigal 2002; Hall et al. 2004*a*). MeHg is toxic by ingestion, inhalation and skin absorption with chronic exposure effects including the central nervous system and kidney damage. Chronic or longer term exposure includes memory disturbance, hypertension, vision problems, hallucinations, tremors and personality changes.

Because MeHg can cross the placental barrier, and because it can affect brain development, its effects are of special concern to pregnant or lactating women and young children. For this reason, together with the fact that MeHg accumulates in the aquatic food web, the Scandinavian countries have issued fish consumption guidelines, and a similar situation holds for parts of North America (Bishop and Lee 1997; Meili 1997; Hall et al. 2004*b*). For example, more than half of the 80,000 lakes in Sweden have pike (*Esox lucius*) with a total Hg concentration exceeding the guideline of 0.5 mg Hg kg^{-1} ww and in about 10,000 lakes pike Hg concentrations exceed the Swedish "blacklisting" limit of 1.0 mg Hg kg^{-1} ww (Håkanson 1996).

Although the MeHg situation no longer attracts much attention by mass media, the situation continues to pose a major threat to all species at the top of the food web.

1.1 Mercury is Deposited From the Atmosphere…

Hg occurs in two stable oxidation states in the atmosphere: Hg^0 (elemental) and Hg^{2+} (mercuric). Elemental Hg is the dominant species in the atmosphere and capable of being aerially transported for tens of thousands of kilometres followed by deposition to (even remote) terrestrial and aquatic ecosystems (Schroeder and Munthe 1998; Grigal 2002). This has led to its recognition as a toxic global pollutant. Although natural sources of Hg may be important on a local scale, they cannot explain geographic trends in soil Hg levels (Fitzgerald et al. 1998). This implies that anthropogenic emissions of Hg have led to deposition levels well in excess of natural levels throughout much of Europe and North America since pre-industrialised times (Johansson et al. 1991; Fitzgerald et al. 1998). Today, the major anthropogenic emission sources include combustion of fossil fuels, mining activities and waste incineration.

The significance of anthropogenic contribution of Hg from the atmosphere via terrestrial to aquatic ecosystems will outermost depend on biogeochemical processes in the watershed, including dry and wet deposition of atmospheric Hg, its storage in ecosystem components, its transfer among components and its loss as a gas or in solution (Grigal 2002).

1.2 … and Stored in Terrestrial Ecosystems

Hg is deposited to terrestrial systems via throughfall, litterfall and via open precipitation following oxidation of elemental to mercuric Hg. Deposited Hg may be lost from terrestrial ecosystems either through volatilisation back to the atmosphere or in solution via stream flow, and will then have great implications for aquatic ecosystems and for public health (Schroeder and Munthe 1998; Grigal 2002).

The biogeochemistry of mercuric Hg, hereafter referred to only as Hg, and MeHg is closely associated to sulphur (S), explaining why MeHg and Hg bind to reduced organic S, rather than to other functional groups in natural organic matter (Skyllberg et al. 2003). Xia et al. (1999) and Skyllberg et al. (2000) showed that the densities of reduced organic S functional groups are in sufficient abundance to bind all Hg in natural terrestrial systems. These findings agree with data from Aastrup et al. (1991), showing that much of the deposited Hg in Scandinavia has been stored and accumulated in the soils upon which it fell. This retention has historically protected aquatic systems from receiving the full potential load of atmospherically derived Hg, but has also led to a build up of Hg in soils and vegetation.

1.3 Mercury Methylation

Although the vast majority of all forms of Hg (total Hg; THg) in terrestrial ecosystems is in inorganic form, more than 90% of the Hg in fish comprises of MeHg, with nearly all of the fish-MeHg being acquired from ingestion of organisms containing MeHg (Bloom 1992; Bodaly et al. 1997). The direct deposition of MeHg is only about 1 - 2% of that of THg, which is insufficient to explain the amount found in aquatic biota (Gilmour and Henry 1991; Hall & St Louis 2004*b*). This implies that inorganic Hg is transformed to MeHg somewhere within the catchment.

1.3.1 The Role of Sulphate Reducing Bacteria

It is well documented that sulphate reducing bacteria (SRB) are primary Hg methylators; although not all SRB methylate Hg (King et al. 2000; Branfireun et al. 2001). The primary site of Hg methylation by SRB is just below the oxic/anoxic interface (Korthals & Winfrey 1987; Krabbenhoft et al. 1995; Gilmour et al. 1998) and MeHg production probably occurs in an accidental side reaction of a metabolic pathway (Choi et al. 1994). It is therefore not likely that SRB have acquired an active transport of Hg and diffusion of neutral Hg species ($[HgL_n^0]$) across the cell membrane is instead regarded as being the important uptake mechanism (Benoit et al. 1999; Benoit et al. 2001). Hence, the availability of Hg species is crucial for the bioavailability of Hg to SRB and for its future accumulation in aquatic food webs (Skyllberg et al. 2003).

However, the potential rate of methylation within any given catchment is not only dependent on the Hg speciation, but also on factors directly affecting the activity of the SRB community (Benoit et al. 2003). Environmental factors, such as temperature, pH, and dissolved oxygen concentration, the availability of electron donors, electron acceptors and carbon sources all influence the SRB activity (Korthals & Winfrey 1987; Gilmour & Henry 1991; Ramlal et al. 1993; Benoit et

al. 2003; Galloway & Branfireun 2004). Although all factors contribute, the availability of sulphate (electron acceptor) and high quality carbon (electron donor) are likely primary controls on SRB activity within any given boreal coniferous catchment (Gilmour & Henry 1991; Kelly et al. 1997; Branfireun et al. 1999; Branfireun et al. 2001; Branfireun et al. 2002; Benoit et al. 2003; Warner 2003).

Hence, semi-anoxic systems with high inputs of fresh organic matter and sulphur, such as peatlands, recently flooded reservoirs and periodically flooded riparian zones, are likely to exhibit high rates of Hg methylation (St Louis et al. 1994; Kelly et al. 1997; Branfireun et al. 1999; Heyes et al. 2000; Branfireun et al. 2001). Indeed, while upland forest soils act as sinks for atmospheric inputs of THg and MeHg because the majority of THg and MeHg binds to organic and mineral soil particles, several studies have shown that catchments containing water saturated areas are sources of both MeHg and Hg to downstream environments (St Louis et al. 1996; Bishop et al. 1995; Branfireun et al. 1996; St. Louis et al. 1996; Porvari et al. 2003*a*). This is probably not only a result of their higher MeHg production, but also because export of Hg and MeHg is largely controlled by the export of dissolved organic matter, which is very abundant in most water saturated areas within the boreal region (Aastrup et al. 1991; Krabbenhoft et al. 1995; Skyllberg 2003). In a typical boreal coniferous forest, water saturated areas are abundant naturally or may be induced by changes in land use, such as forest management or hydroelectric damming. The impacts of such activities have attracted more attention from research groups during the past five years (Garcia et al. 1999, 2000; Porvari et al 2003).

1.4 Methylmercury Demethylation

MeHg may be lost from the terrestrial environment either in association with organic matter, as outlined above, or become demethylated. MeHg can degrade by a number of abiotic and biotic pathways within the catchment. Abiotic pathways include photodegradation, as described by Sellers et al. (1996) and the reaction between MeHg and S2-, resulting in formation of dimethylmercury (Me2Hg) and H2S (Marvin-Dipasquale et al. 2000).

A number of bacterial strains, found under both anaerobic and aerobic conditions, are able to degrade MeHg (Weber et al. 1998). Demethylation is mainly carried out by Hg resistant bacteria as a resistance mechanism against organic Hg compounds (Marvin-Dipasquale et al. 2000). Although the specific environmental factors controlling the relative importance of these biotic pathways are largely unknown, MeHg and/or Hg concentrations are likely important (Marvin-Dipasquale et al. 2000).

The balance between Hg methylation and MeHg demethylation, i.e. the net methylation, will ultimately control MeHg concentrations within a catchment.

1.5. Study Aims

As an ancillary experiment to an ongoing EU Life project in Sweden (see Skogsvårdsstyrelsen 2003), this study will focus on the key factors that ultimately control the rate of Hg methylation and MeHg demethylation within boreal coniferous forest catchments.

This thesis aims at: (1) developing a suitable methodology for the future incubation of samples that will be conducted within later stages of the EU Life project in Balsjö; (2) finding the potential rate of net methylation in the riparian zone of a typical boreal coniferous forest catchment; and (3) determining if the availability of electron donors (high quality carbon) and electron acceptors (sulphate) are more important controls of Hg methylation than are the availability of Hg itself.

The current hypothesis is that the SRB community is limited primarily by (1) sulphate; and (2) glucose. Once the activity of the SRB is optimal, the availability of Hg will further limit the rate of methylation. This hypothesis will be tested in this study.

The activity of the bacterial strains responsible for MeHg demethylation is believed to increase with increased amounts of glucose. No response is expected at high concentrations of sulphate or Hg. This hypothesis will also be tested within this study.

2. Study methodology

2.1 Experimental Design – Initial Studies

Before the contribution of sulphate, glucose and Hg on the rate of Hg methylation and MeHg demethylation could be evaluated, the time required for the SRB to methylate Hg had to be determined. Further, the concentrations of sulphate, glucose and Hg generating the maximum methylation rate must be known. Initial studies like these are required in every new system that is about to be revised.

Therefore, the main study is preceded by; (1) an initial incubation time study and; (2) an optimal sulphate/glucose/Hg concentration study (Fig. 2.1; Table 2.1).

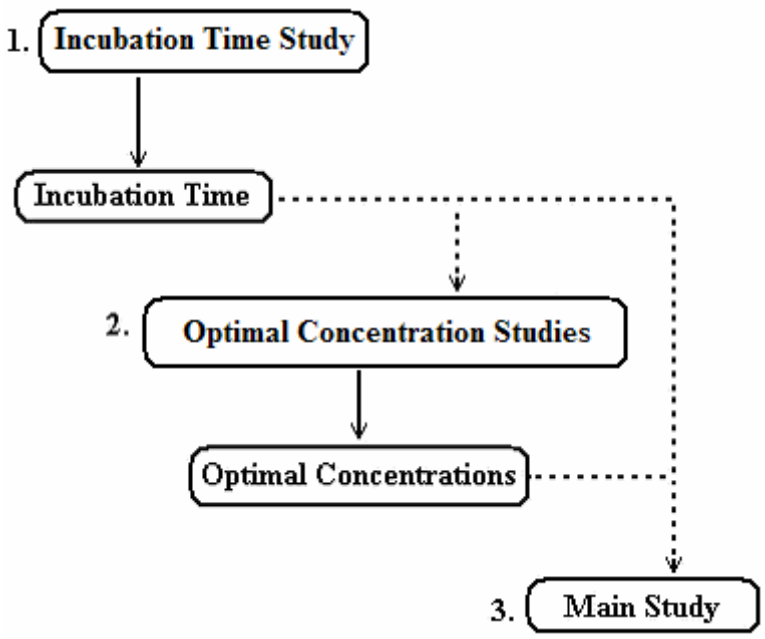


Figure 2.1 *The incubation time study will determine the time during which samples are incubated in the following studies. The optimal concentration study will determine what concentrations of sulphate, glucose and Hg to be used in the main study.*

Within the incubation time study, incubations are stopped after different times after which the amounts of produced MeHg are analysed. This makes it possible to determine what incubation time to use in the forthcoming studies. Following the time study, the SRB response to different concentrations of electron donors, electron acceptors and Hg will be evaluated. Concentrations resulting in maximum MeHg production will be used in the main study (Table 2.1).

Table 2.1 A comparison of the three studies conducted within this thesis. All treatments are duplicated.

	Incubation Time (h)	[Sulphate] (mM)	[Glucose] (mM)	[Hg] (ppb)
Study 1 Incubation Time Study	0, 23, 46, 64, 112	-	-	-
Study 2 Optimal Concentration Study	Optimal time from Study 1	0.1; 0.5; 2; 5	1; 5; 10; 20	1.7; 17; 170; 1700
Study 3 Main Study	Optimal time from Study 1	Optimal concentrations from Study 2. See experimental design in Fig. 2.2		

2.2 Experimental Design – Main Study

In the third and main study, the ambition is to reveal the contribution of sulphate, glucose and Hg to the rate of Hg methylation, respectively. This is possible through a factorial design at two levels of three factors, i.e. optimal (+) and low (-) concentrations of sulphate, glucose and Hg (Fig. 2.2). The design also contains four centre points, i.e. (+/2) concentrations of each factor.

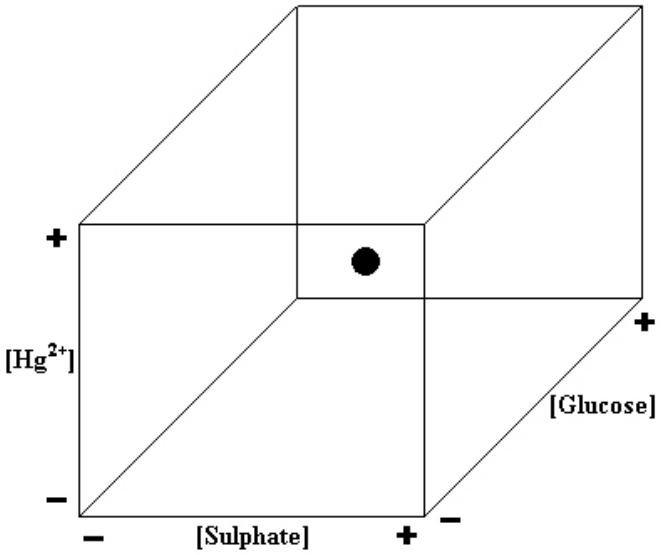


Figure 2.2 *The factorial design used in the main study illustrated in three dimensions. By using two levels for each concentration (+ and -) and four centre points (+/2) it is possible to verify the contribution of sulphate-, glucose- and Hg-concentration, respectively.*

2.3 Study Area

Samples were collected within a boreal coniferous catchment situated about 13 km NW of Balsjö (64°02’ N, 18°57’ E) in N Sweden. At a local scale, the study area has a distinct U-shaped topography and is drained by a small first order stream at the bottom of the depression. The riparian zone is mainly vegetated by Picea abies (L), Pinus sylvestris (L), Vaccinium myrtillus (L) and a number of

moss species, including Sphagnum spp. (L) and Polytrichum commune (L). See Fig. 1 to 3 in Appendix A for further portrayal of the study area.

2.4 Sampling, Storage and Incubation

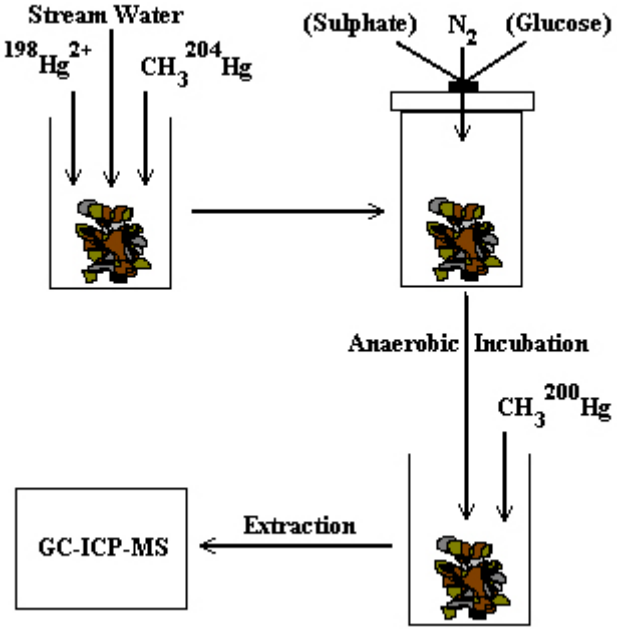


Figure 2.3 *Incubation protocol for studies 1 and 2. Approximately 20 g of peat was incubated along with 5 ml of stream water and Hg isotopes. Following gas exchange, 1 ml of sulphate and/or glucose was added to each sample as described earlier. After incubation, MeHg was extracted from a 5 g sub sample.*

Due to unexpected variance in the results from Study 1 and 2, the incubation protocol was slightly changed in the main study in order to improve the quality of the data (See Table 2, 3 and 4 in Appendix B for a comparison of standard errors). Instead of transferring the 20 g sub sample using tweezers, the fresh peat sample was homogenized with a rotary mixer before approximately 5 g of the homogenous slurry were transferred directly to polypropylene tubes (Fig. 2.4). The smaller mass of incubated peat allowed for extraction of the entire sample without any disturbing sub sampling following incubation. Addition of stream water, glucose, sulphate and Hg isotopes followed the procedure outlined above. All treatments and incubations were carried out in an anaerobic (N2) atmosphere.

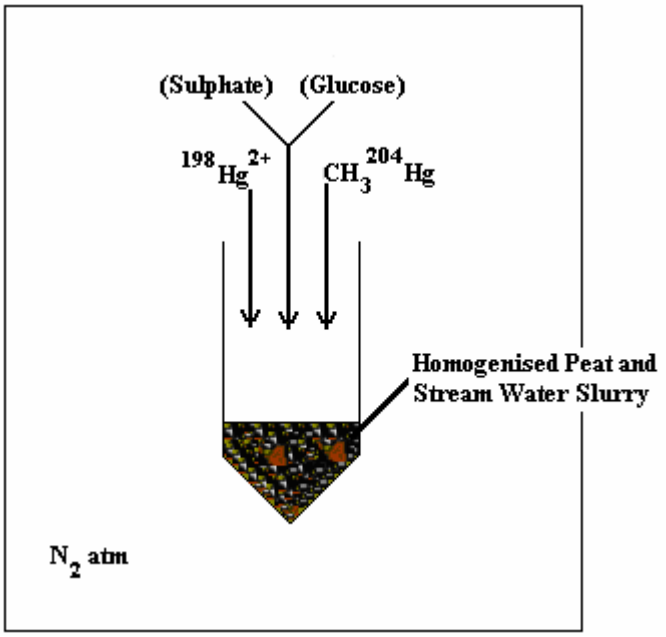


Figure 2.4 *A small part of the incubation protocol used in the main study. 5 g of the homogenous peat and stream water slurry were incubated along with Hg isotopes, sulphate and glucose according to the experimental design. Samples were incubated in an anaerobic box and gas exchange was therefore not required. Following incubation, MeHg could be extracted from the entire sample volume. See also Fig. 2.3.*

2.5 Analysis of Hg Methylation and MeHg Demethylation

2.5.1 MeHg Extraction

Extraction of MeHg from the peat followed the procedure developed by Qian et al. (2000) (see also Lambertsson et al. 2001 for a more detailed description). In essence, 0.5 ml of a 5 ppb $CH_3{}^{200}Hg^+$ solution was added to 5 g of each sample as a species-specific internal standard. After 15 minutes, 10 ml of 1.4 M KBr in 5% H_2SO_4, 10 ml CH_2Cl_2 and 2 ml 1 M $CuSO_4$ were added and samples were shaken for 1 h with at rotary mixer at 22 rev min^{-1}. After centrifugation at 3000 rev min^{-1} for 30 minutes, the organic phase was transferred into 50 ml polypropylene tubes and approximately 10 ml of Milli-Q water were added. The samples were then purged with N_2 until the organic solvent was completely removed. The remaining aquatic phase was transferred to a 10 ml glass tube along with 200 µl acetic acid/acetate buffer (pH 4.9), approximately 600 µl hexane and 100 µl 1% $NaBEt_4$. Samples were again shaken for 1 h at 22 rev min^{-1}, after which the remaining hexane phase was transferred to 2 ml GC-vials.

The isotope dilution based GC-MS-ICP method used to determine Hg methylation rates uses $Me^{200}Hg^+$ as an internal standard. The internal standard automatically corrects for variations in the solid-liquid as well as the liquid-liquid extraction efficiencies, the derivatisation yield and instrumental drift. The reported precision of measurements for MeHg determination using the identical method and similar substrates is 2.7% RSD (Lambertsson et al. 2001). For this reason, a duplicated treatment in all three studies is sufficient.

2.5.2 GC-ICP-MS

Ethylated Hg-species were automatically injected into an Agilent 6890N GC (Agilent Technologies, Palo Alto, CA, USA) fitted with a capillary column (internal diameter 0.32 mm; length 30 m; J&W Scientific, Folsom, CA, USA). The GC was coupled to an Agilent 7500 ICP-MS (Agilent Technologies, Palo Alto, CA, USA) operated at 600 W rf power with a carrier gas flow of 0.5 l min^{-1}. Data were collected by monitoring atomic masses of 198, 200, 202 and 204 at a 0.3 s integration time per point.

2.6 Calculation of Hg Methylation and MeHg Demethylation

Chromatographic signals were integrated using the Agilent Chromatographic Collector software. Based on the integrated areas, isotope dilution calibration calculations were performed using the following equation:

$$C_X = \left(\frac{C_S\,215.59}{200}\right)\left(\frac{A_S - R_m B_S}{R_m B_X - A_X}\right) \qquad (1)$$

Where; C_x = incipient $Me^{202}Hg^+$ concentration

C_s = spiked concentration of the added $Me^{200}Hg^+$ internal standard

A_s = atomic fraction of ^{200}Hg in the enriched isotope standard

B_s = atomic fraction of ^{202}Hg in the enriched isotope standard

A_x = atomic fraction of ^{200}Hg in the original sample

B_x = atomic fraction of ^{202}Hg in the original sample

R_m = the measured 200:202 ratio in the spiked sample

The level of the ^{202}Hg isotope was never manipulated during incubation or extraction and therefore used as a reference when determining methylated amounts of the added $^{198}Hg^{2+}$ tracer. Alterations in $^{198}Hg^{2+}$ concentration were determined using equation 2.

$$D_X = 0.2986\,C_X\left(R_s - \left(\frac{D_S}{B_X}\right)\right) \qquad (2)$$

Where; D_x = $Me^{198}Hg^+$ from methylation of the added tracer

C_x = incipient $Me^{202}Hg^+$ concentration from (1)

R_s = the measured 198:202 ratio in the spiked sample

D_s = atomic fraction of ^{198}Hg in the enriched isotope standard

B_x = atomic fraction of ^{202}Hg in the original sample

The ^{202}Hg isotope was also used as a reference when determining demethylated amounts of the added $^{204}MeHg^{2+}$ tracer. Alterations in $^{204}MeHg^{2+}$ concentration were determined using a modified version of equation 2, in which:

D_x = $^{204}Hg^{+}$ from methylation of the added tracer

C_x = incipient $Me^{202}Hg^{+}$ concentration from (1)

R_s = the measured 204:202 ratio in the spiked sample

D_s = atomic fraction of ^{204}Hg in the enriched isotope standard

B_x = atomic fraction of ^{202}Hg in the original sample

In order to determine the rate of Hg methylation and MeHg demethylation, D_x was divided by the effective incubation time, i.e. the entire incubation time minus an initial lag phase of 23 h that was identified in the incubation time study (see Fig. 3.1).

2.7 Statistical Analysis

Statistical differences in Hg methylation rates grouped by the factors glucose, sulphate and Hg were determined using GLM (General Linear Model) statistical analysis. In GLM, the Hg methylation rate was set as dependent variable with glucose, sulphate and Hg as category variables. All statistical analyses were completed using the MINITAB 14 software.

3. Results

3.1 Incubation Time Study

As predicted, the amount of methylated Hg increased over time (Fig. 3.1). After a lag phase of at least 23 h, the maximum MeHg production was reached somewhere in between t = 64 h and t = 112 h. The incubation time used in the following studies was therefore set to 64 h. See also Table 2 in Appendix B for further details.

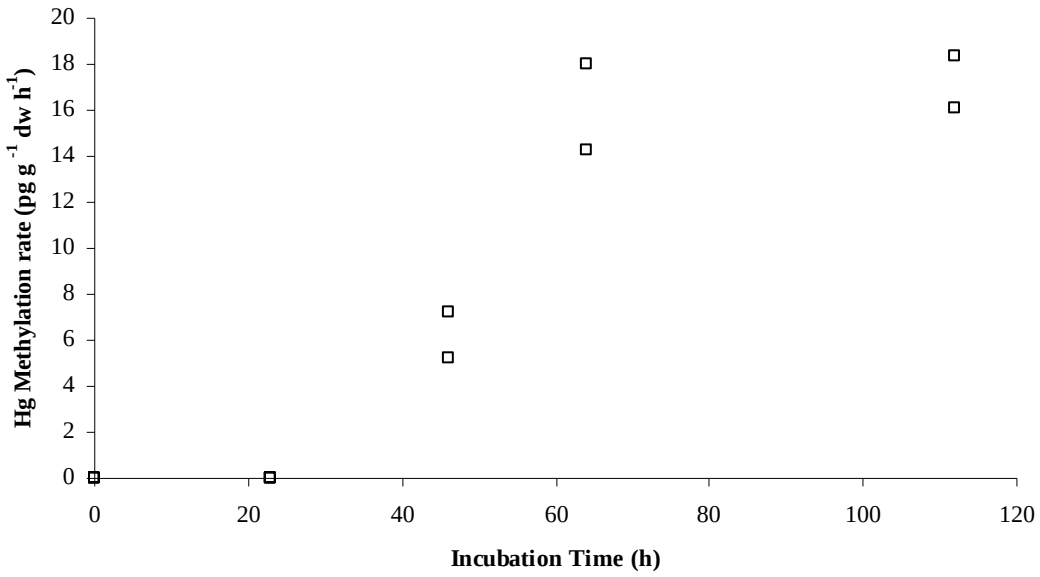


Figure 3.1 *Hg methylation rate after time = 0, 23, 46, 64 and 112 h following incubation. Based on these findings, the incubation time used in the following studies was set to 64 h.*

3.2 Optimal Concentration Study

The methylation rate increased with greater concentrations of sulphate and Hg until a certain point, after which the MeHg production declined (Fig. 3.2 and Fig. 3.3). Despite the great variation, the optimal concentrations of sulphate and Hg could be determined based on these findings. These were set to 1.5 mM (sulphate) and 17 ppb (Hg). Addition of glucose resulted in a decreased MeHg production (Fig. 3.4). This was likely due to disproportionate concentrations resulting in glucose having a toxic effect on the SRB community. Therefore, the optimal glucose concentration used in the main study had to be estimated (0.1 mM). In order to control the approximated glucose optimum, a second study using a lower concentration range was conducted parallel to the main study. The results indicate that the optimal glucose concentration is near the estimated value used in the main study (Fig. 3.5). More detailed results from the second study are presented in Table 3 (Appendix B). See also Table 4 in Appendix B for the complete experimental matrix used in the main study.

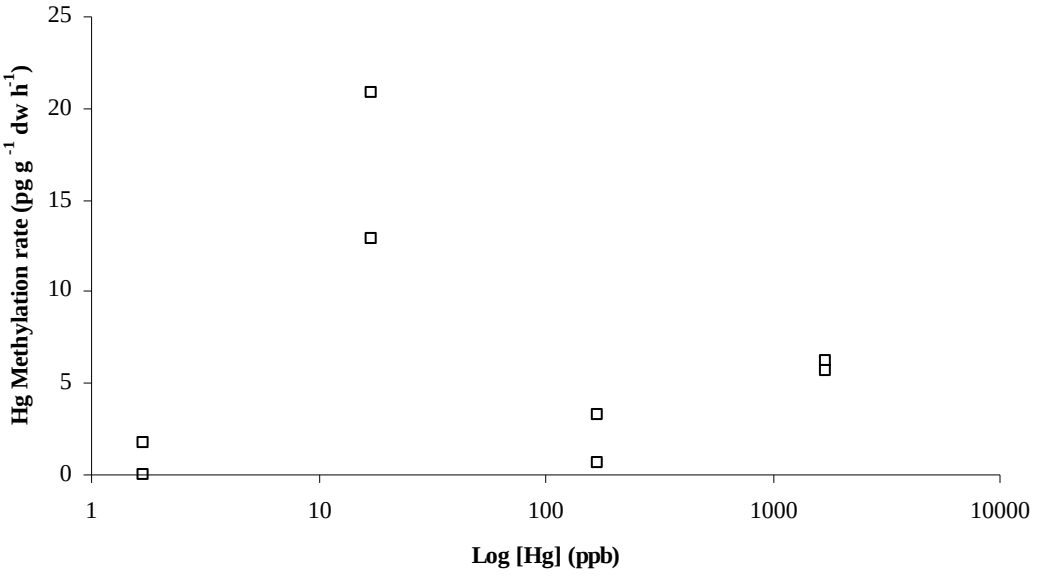


Figure 3.2 Hg methylation rates at different concentrations of Hg after 64 h of incubation. From these results, the optimal concentration of Hg was set to 17 ppb.

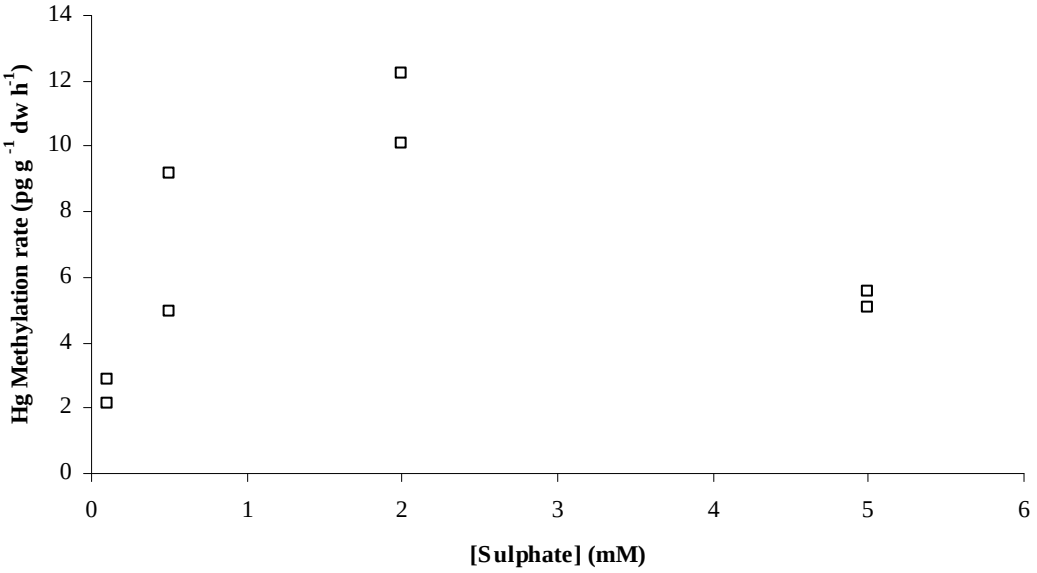


Figure 3.3 *Hg methylation rates at different concentrations of sulphate 64 h after incubation started. From these results, the optimal concentration of sulphate was determined to 1.5 mM.*

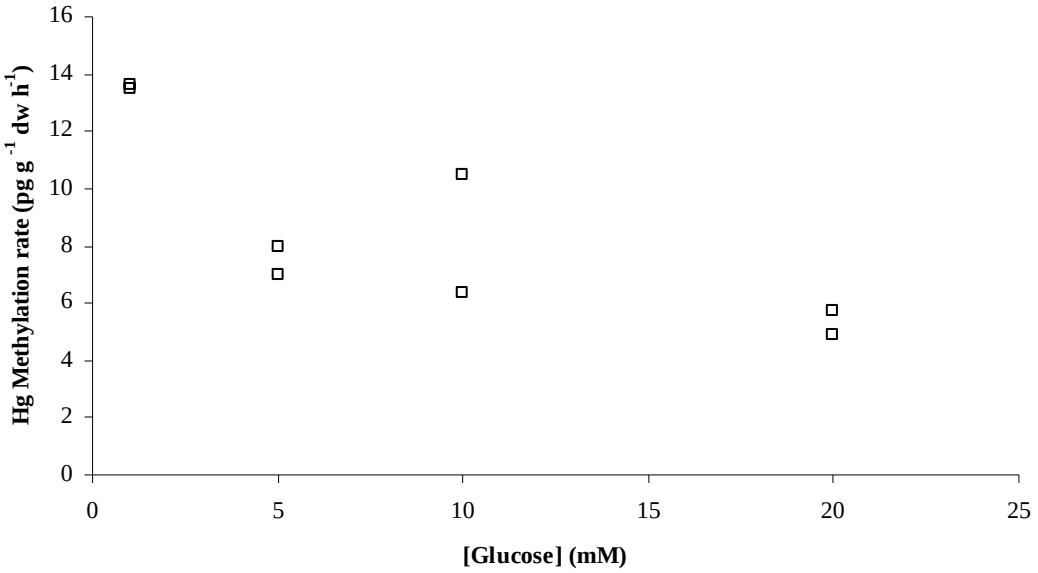


Figure 3.4 *Hg methylation rates at different concentrations of glucose after 64 h of incubation. Due to the constant decline in methylation rate with increased concentration of glucose, the optimal concentration had to be estimated (0.1 mM).*

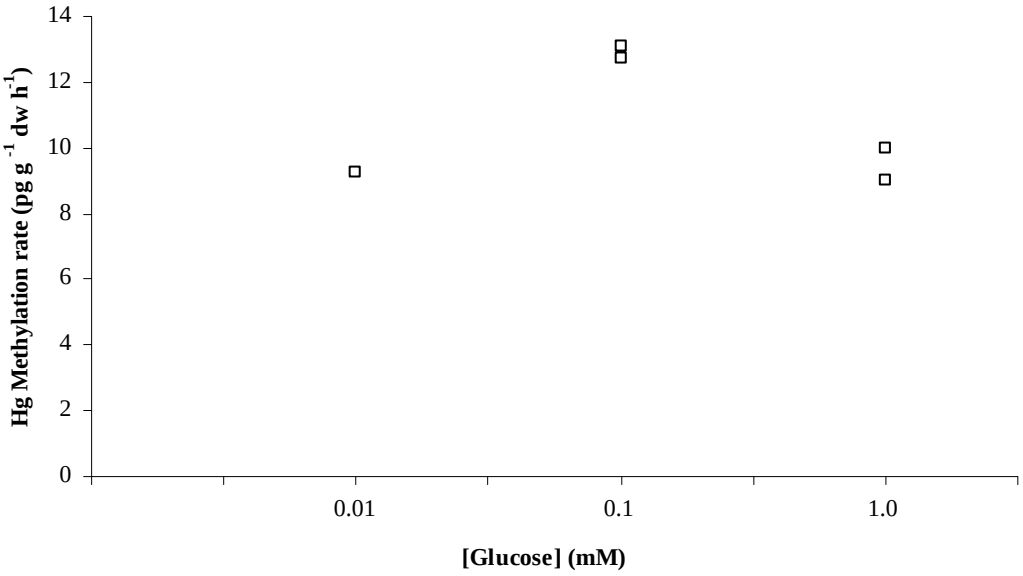


Figure 3.5 *Hg methylation rate at different glucose concentrations after 64 h of incubation, repeat experiment.*

The rate of MeHg demethylation was not significantly affected by addittion of sulphate, glucose or Hg (Table 3.1). However, the mean rate of MeHg demethylation was greater than was the mean rate of Hg methylation (compare Table 3.1 with Table 3 in Appendix B).

Table 3.1 Mean MeHg demethylation rate in samples incubated with different concentrations of sulphate, glucose and Hg.

[Sulphate] (mM)	[Glucose] (mM)	[Hg] (ppb)	N	Mean MeHg Demethylation Rate (pg g^{-1} dw h^{-1})	SE	Detection Limit (pg g^{-1} dw h^{-1})
0.1	-	-	2	12.4	0.4	3.5
0.5	-	-	2	11.3	0.2	3.2
2	-	-	2	11.6	1.5	3.1
5	-	-	2	12.0	1.2	3.4
	1	-	2	11.7	0.6	3.5
	5	-	2	11.3	0.3	3.2
	10	-	2	10.8	1.5	3.4
	20	-	2	10.5	0.6	3.3
		1.7	2	15.6	2.6	3.2
		17	2	10.3	1.9	3.5
		170	2	12.9	0.2	3.5
		1700	2	11.8	0.8	3.4

3.3 Main Study

The highest methylation rate was found in samples incubated with optimal concentrations of all factors (3.0 pg g^{-1} dw h^{-1}) and decreased as one or several factors were excluded (Fig. 3.6). No methylation could be detected in any sample incubated with low Hg concentrations. See Table 5 in Appendix B for additional details.

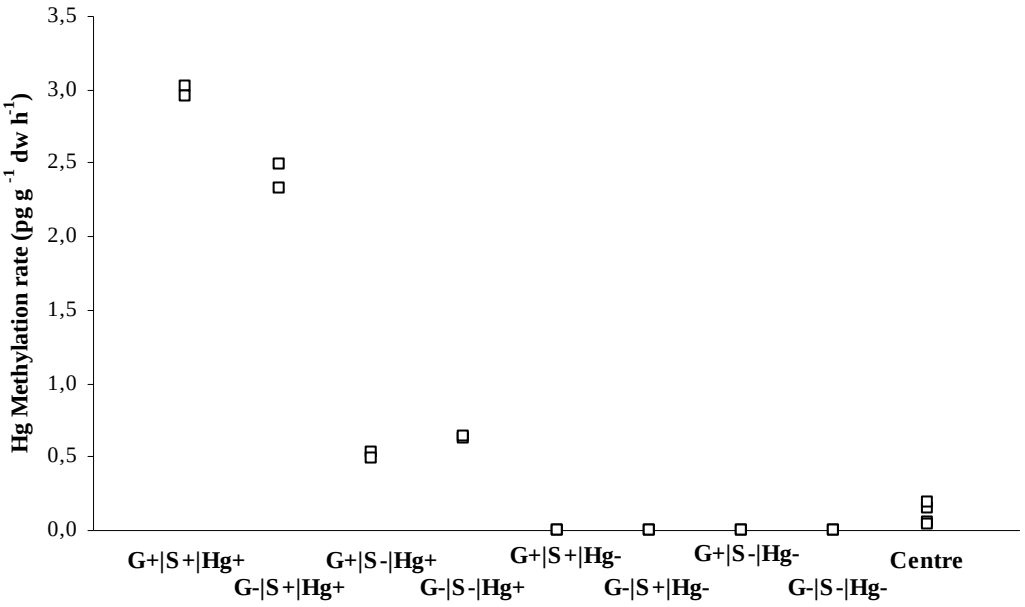


Figure 3.6 *The rate of Hg methylation at different concentrations of Hg, sulphate and glucose. The methylation rate was highest in samples incubated with high concentrations of all three factors, but dropped as one or several factors were removed. No methylation was detected in samples incubated with low Hg concentrations. Note: G = glucose; S = sulphate.*

Both addition of each single factor as well as their two- and three way interactions significantly increased the rate of methylation in the current sample (Table 3.2). High concentrations of glucose, sulphate and Hg all had a significant positive effect on the rate of Hg methylation. The treatment effect of Hg was greater than the effect of sulphate and glucose, respectively, and the effect of sulphate was greater than was the effect of glucose. The combined effect of high sulphate- and Hg concentration was greater than the combined effect of high glucose and Hg and also greater than the combined effect of all three factors.

Addittion of sulphate, glucose or Hg did not significantly affect the rate of MeHg demethylation (Table 6 in Appendix B). However, the mean rate of MeHg demethylation was greater than was the mean rate of Hg methylation.

Table 3.2 Analysis of variance for all factors used in the main study. All factors but Glucose and Glucose*Hg did significantly (P < 0.001) affect the Hg methylation rate.

Factor	DF	Seq SS[a]	Adj MS[b]	
Hg	1	3708	3708	5609
Sulphate*Hg	1	1572	1572	2377
Sulphate	1	1572	1572	2377
Glucose*Sulphate*Hg	1	44	44	66
Glucose*Sulphate	1	44	44	66
Glucose*Hg	1	19	19	29
Glucose	1	19	19	29
Error	8	5	1	-
Total	15	6983	-	-

R^2 (adj) = 99.9 %;
[a] - SS = sums of squares;
[b] - MS = mean squares

4. Discussion

4.1 Study Methodology

The current situation within the riparian zone in the study area is best described by the "low" Hg experiments without any addition of sulphate and glucose conducted in the main study. These experiments resulted in MeHg levels below the detection limit (Fig. 3.6). Due to two main reasons, this does not automatically mean that there is no ongoing methylation within the study area:

(1) All treatments in the main study resulted in much lower methylation rates compared to the initial studies (compare Table 2 and 3 with Table 5 in Appendix B). This was most likely a result of the great disturbance to the SRB caused by the mixing of the peat prior to incubation. Hence, the incubation time from the incubation time study was probably far from optimal for the mixed samples and a second time study would have been required in order to obtain comparable results.

(2) The importance of inorganic Hg is, of course, unquestionable for the formation of MeHg, but the results may partially be explained by experimental limitations. As mentioned earlier, Hg is strongly associated with thiol groups in natural organic matter (Skyllberg 2003) and thus hardly available to form neutral Hg complexes (Xia et al. 1999; Skyllberg et al. 2000) important to the SRB community. Hence, it is likely that the "low" addition of 1.7 ppb Hg (addition = 10 % of actual THg content) is adsorbed to thiol groups within the sample and therefore not available to SRB during the short time frame used in this study. Consequently, the concentration of Hg used in the "low" Hg treatments has been insufficient to allow for detetection using the actual method. As a result, potential treatment effects are not truly reflected in the statistical analysis (Table 3.2) since methylation rates in the "low" Hg experiments were set to zero.

If the Hg added in the "low" Hg experiments becomes unavailable through its association with thiol groups in the organic matter, the undetectable amounts of MeHg in the main study (Fig. 3.6) do not automatically imply that there is no current methylation in the riparian zone. Instead, as the organic matter is decomposed over time, this Hg will likely become more available to SRB. Hence, the zero methylation results presented in this thesis are true only for a short time frame within the study area.

As a consequence, few conclusions regarding the current situation in Balsjö should be drawn based on these results. An additional consequence is that the results from the main study do not correspond to the true potential for methylation that was to be revealed in this study.

4.1.1 Analytical Precision

The unexpected variance in the two initial studies was likely due to the rather heterogeneous nature of the peat (Fig 4 in Appendix A) with insufficient mixing of the added substrates and isotopes as a result (See Table 2, 3 and 5 in Appendix B for a comparison of standard errors). Further, the second subsampling prior to MeHg extraction may have generated additional mass bias error due to the usage of tweezers. Likely, the 5 g of sub sampled material did not correspond to the ac-

tual water: peat ratio of the incubation flasks since more peat than water was removed when using tweezers. The variance calls for great caution when drawing conclusion from the two studies.

These errors were corrected for by altering the incubation protocol slightly, as already outlined in section 1.2. The mean precision of measurements for MeHg determination in the main study was 2.4% RSD. This value consent with that of 2.7% RSD reported by Lambertsson et al. (2001) and further replicates were therefore not required.

Although resulting in some negative consequences for the results presented in this thesis, the trial and error approach has most likely generated a final methodology that is suitable for the future work that will be conducted within the EU Life project in Balsjö.

4.1.2 Evidence of Microbial Methylation

Back lying theories claim that the production of MeHg primarily is a by-product of a metabolic pathway within SRB (Choi et al. 1994). At the same time, the results presented in this thesis are based solely upon alterations of strictly chemical properties, i.e. differences in MeHg concentrations. Hence, one should raise the question whether the produced MeHg truly is a result of microbial conversion of Hg, or simply a result of chemical equilibrium during incubation.

The observed time lag (Fig. 3.1), together with the fact that glucose addition does affect the methylation rate (Fig. 3.2), indicates that the process is of microbial character. Further, previous studies using the same method on brackish water sediments have shown that incubation of sterile samples result in no formation of MeHg (Lambertsson, unpubl.). These findings offer sufficient evidence to verify that microbial conversions are the main source of produced MeHg.

4.2 MeHg Demethylation

The focus of many quantative studies has been on methylation. However, earlier studies have indicated that the build-up of MeHg may be due to suppressed demethylation rather than enhanced methylation (Meili 1997) and references therein.

Addittion of sulphate, glucose and Hg did not seem to significantly affect the rate of MeHg demethylation in this study (Table 3.1 and Table 6 in Appendix B). However, the rate of MeHg demethylation was somewhat greater than the rate of Hg methylation, i.e. the net methylation seem to be negative.

As already outlined, MeHg can degrade by a number of abiotic and biotic pathways (Sellers et al. 1996; Weber et al. 1998; Marvin-Dipasquale et al. 2000). Since there were no significant response to any of the added factors, no conclusions can be drawn whether the the main pathway of MeHg demethylation in this study is biological or chemical. As a consequence, the findings presented here can not be used to describe the current situation in Balsjö.

4.3 Hg Methylation

The results from the main study show that inorganic Hg, sulphate and glucose all limits the rate of MeHg production in the riparian zone of a typical boreal conifer-

ous catchment. These findings agree with the back lying theories and are also supported by previous work by others (see Branfireun et al. 2001; Bergman et al. 1997) and references therein.

However, based on the statistical analysis, the initial assumption that electron donors and electron acceptor may be more important than Hg itself, does not seem to be entirely valid. About 50 % of the total variation in the main study is explained by Hg (Table 3.2), but the zero methylation found in all "low" Hg treatments may not be true, due to experimental limitations (see discussion in section 4.1). If so, the statistical analysis will not be reliable.

Samples incubated only with optimal concentrations of Hg (Hg+|S-|G-) resulted in a methylation rate of 0.6 pg g^{-1} dw h^{-1}. Addittion of sulphate (Hg+|S+|G-) increased the methylation rate almost five times (2.5 pg g^{-1} dw h^{-1}) and the maximum methylation rate (3.0 pg g^{-1} dw h^{-1}) was found in samples incubated with optimal concentrations of all three factors (Hg+|S+|G+). Hence, once Hg is in sufficient abundance, addition of sulphate increases the MeHg production and when there is a surplus of both Hg and sulphate; glucose will boost the production even further. These findings confirm the importance of biological induced Hg methylation.

The methylation rates found in the Hg+|S-|G+ and Hg+|S-|G- treatments (Fig. 3.6) are determined by the initial sulphate concentration in the sample. Here, the activity of the SRB community is limited by the availability of sulphate which explains why no significant differences occurr between low and high glucose addittions. However, in the Hg+|S+|G- and Hg+|S+|G+ treatments, the entire SRB community becomes active and their potential to utilise electron donors is greater than the available amounts. Hence, glucose becomes the limiting factor, explaining why the methylation rate increases by about 20 % from low to high glucose addittion (Fig. 3.6).

This response is likely highly specific to every location within the study area and dependent on the geochemistry of each site. A site with high sulphate concentrations and, consequently, a greater active SRB population will likely suffer from faster depletion of electron donors. Spatial (and temporal) variations in geochemistry would explain the different methylation rates found in peat incubated with identical concentrations of Hg, glucose and sulphate but sampled from different locations and at different times along the riparian zone (see Fig. 3.1 to 3.6).

These results illustrate the importance of accounting for interactions between key factors when addressing the rate of Hg methylation in the riparian zone of a boreal coniferous catchment.

5. Conclusions

Of greater importance than finding a theoretical potential of net Hg methylation is the fact that the outcome of this study calls for a new paradigm explaining variations in aquatic bioaccumulation of Hg. For a long time the occurrence of MeHg in fish has been explained solely by Hg deposition and accumulation of THg in the environment, and factors controlling the availability of THg. As a replacement for this theory, the results presented in this thesis propose that biological transformation of inorganic Hg within a catchment is the utterly most important reason for Hg methylation and, consequently, the main cause of elevated MeHg levels in fish. The factors controlling methylation of Hg are therefore probably more central in explaining regional and landscape patterns in Hg accumulation in fish than are the occurrence of THg.

Future work should be focused on the effects of different land uses on the production and export of MeHg to downstream environments. Recently, the impacts of forestry have begun to attract attention from several research groups within the boreal region. It has been shown that clear cutting of forests increase not only the production of MeHg, but also the export of THg and MeHg to the surroundings under current management practices (Porvari 2003 *a*; *b*). Being the most widespread land use in the boreal region, finding how to reduce the effects of forestry is of unconditional importance. This thesis has hopefully assisted by laying one small piece of that puzzle.

6. References

Aastrup, M., Johnson, J., Bringmark, E., Bringmark, I. & Iverfeldt, Å. (1991) Occurrence and transport of mercury within a small catchment area. *Water Air and Soil Pollution* 56 55–167

Benoit J.M., Gilmour C.C., Mason R.P. & Heyes, A. (1999) Sulfide controls on mercury speciation and bioavailability to methylating bacteria in sediment pore waters. *Environmental Science & Technology* 33 951–957

Benoit, J.M., Gilmour, C.C. & Mason, R.P. (2001) The influence of sulfide on solid phase mercury bioavailability for methylation by pure cultures of Desulfobulbus propionicus. *Environmental Science & Technology* 35 (1) 127-132

Benoit, J.M., Gilmour, C.C. & Heyes A. (2003) Geochemical and biological controls over methylmercury production and degradation in aquatic ecosystems. *ACS Symposium Series* 835 262-297

Bergman, I., Svensson, B.H. & Nilsson, M. (1998) Regulation of methane production in a Swedish acid mire by pH, temperature and substrate. *Soil Biological Biochemistry* 6 729 - 741

Bishop, K., Lee, Y.H., Pettersson, C. & Allard, B. (1995) Terrestrial sources of methylmercury in surface waters – the importance of the riparian zone on the Svartberget catchment. *Water Air and Soil Pollution* 80 (1-4) 435-444

Bishop, K.H. & Lee, Y.H. (1997) Catchments as a source of mercury/ methylmercury in boreal surface waters. *Metal Ions in Biological Systems* 34 113-130

Bloom, N.S. (1992) On the chemical form of mercury in edible fish and marine invertebrate tissue. *Canadian Journal of Fisheries and Aquatic Sciences* 49 (5) 1010-1017

Bodaly, R.A., St Louis, V.L., Paterson, M.J., Fudge, R.J.P., Hall, B.D., Rosenberg, D.M. & Rudd, J.W.M. (1997) Bioaccumulation of mercury in the aquatic food chain in newly flooded areas. *Metal Ions In Biological Systems* 34 259-287

Branfireun, B.A., Heyes, A. & Roulet, N.T. (1996) The hydrology and methylmercury dynamics of a Precambrian Shield headwater peatland. *Water Resources Research* 32 (6) 1785-1794

Branfireun, B.A., Roulet, N.T., Kelly, C.A. & Rudd, J.W.M. (1999) In situ sulphate stimulation of mercury methylation in a boreal peatland: Toward a link between acid rain and methylmercury contamination in remote environments. *Global Biogeochemical Cycles* 13 (3) 743-750

Branfireun, B.A., Bishop, K., Roulet, N.T., Granberg, G. & Nilsson, M. (2001) Mercury cycling in boreal ecosystems: The long-term effect of acid rain constituents on peatland pore water methylmercury concentrations. *Geophysical Research Letters* 28 1227-1230

Branfireun, B.A. & Roulet, N.T. (2002) Controls on the fate and transport of methylmercury in a boreal headwater catchment, northwestern Ontario, Canada. *Hydrology and Earth System Sciences* 6 (4) 783-794

Choi, S.C., Chase, T. & Bartha, R. (1994) Metabolic pathways leading to mercury methylation in Desulfovibrio desulfuricans LS. *Applied And Environmental Microbiology* 60 (11) 4072-4077

Fitzgerald, W.F., Engstrom, D.R., Mason, R.P. & Nater, E.A. (1998) The case for atmospheric mercury contamination in remote areas. *Environmental Science & Technology* 32 (1) 1-7

Galloway, M.E. & Branfireun, B.A. (2004) Mercury dynamics of a temperate forested wetland. *Science of the Total Environment* 325 239–254

Garcia, E. & Carignan, R. (1999) Impact of wildfire and clear-cutting in the boreal forest on methyl mercury in zooplankton. *Canadian Journal of Fisheries and Aquatic Sciences* 56 339-345

Garcia, E. & Carignan, R. (2000) Mercury concentrations in northern pike (Esox lucius) from boreal lakes with logged, burned, or undisturbed catchments. *Canadian Journal of Fisheries and Aquatic Sciences* 57 (2) 129-135

Gilmour, C.C. & Henry, E.A. (1991) Mercury methylation in aquatic systems affected by acid deposition. *Environmental Pollution* 71 (2-4) 131-169

Gilmour, C.C., Riedel, G.S., Ederington, M.C., Bell, J.T., Benoit, J.M., Gill, G.A. & Stordal, M.C. (1998) Methylmercury concentrations and production rates across a trophic gradient in the northern Everglades. *Biogeochemistry* 40 327–345

Grigal, D.F. (2002) Inputs and outputs of mercury from terrestrial watersheds: a review *Environmental Reviews* (10) 1-39

Hall, B.D., St Louis, V.L. & Bodaly, R.A. (2004*a*) The stimulation of methylmercury production by decomposition of flooded birch leaves and jack pine needles. *Biogeochemistry* 68 (1) 107-129

Hall, B.D & St Louis, V.L. (2004*b*) Methylmercury and total mercury in plant litter decomposing in upland forests and flooded landscapes. *Environmental Science & Technology* 38 (19) 5010-5021

Heyes, A., Moore, T.R., Rudd, J.W.M. & Dugoua, J.J. (2000) Methyl mercury in pristine and impounded boreal peatlands, experimental Lakes Area, Ontario. *Canadian Journal of Fisheries and Aquatic Sciences* 57 (11) 2211-2222

Håkanson, L. (1996) A simple model to predict the duration of the mercury problem in Sweden. *Ecological Modelling* 93 (1-3) 251-262

Johansson, K., Aastrup, M., Andersson, A., Bringmark, L. & Iverfeldt, A (1991) Mercuty in Swedish forest soils and waters – assessment of critical load. *Water Air and Soil Pollution* 56 267-281

Kelly, C.A., Rudd, J.W.M., Bodaly, R.A., Roulet, N.P., St Louis, S.T., Heyes, A., Moore, T.R., Schiff, S., Aravena, R., Scott, K.J., Dyck, B., Harris, R., Warner, B. & Edwards, G. (1997) Increases in fluxes of greenhouse gases and methyl mercury following flooding of an experimental reservoir. *Environmental Science & Technology* 31 1334-1344

King, J.K., Kostka, J.E. & Frischer, M.E. (2000) Sulfate-reducing bacteria methylate mercury at variable rates in pure culture and in marine sediments. *Applied Environmental Microbiology* 66 (6) 2430-2437

Korthals, E.T. & Winfrey, M.R. (1987) Seasonal and spatial variations in mercury methylation and demethylation in an oligotrophic lake. *Applied Environmental Microbiology* 53(10) 2397–2404

Krabbenhoft, D.P., Benoit, J.M., Babiarz, C.L., Hurley, J.P, Andren, A.W. (1995) Mercury cycling in the Allequash Creek watershed, northern Wisconsin. Water Air and Soil Pollution 80 (1-4) 425-433

Lambertsson, L., Lundberg, E. & Nilsson M. (2001) Applications of enriched stable isotope tracers in combination with isotope dilution GC-ICP-MS to study mercury species transformation in sea sediments during in situ ethylation and determination. *Journal of Analytical Atomic Spectrometry* 16 (11) 1296-1301

Marvin-DiPasquale, M., Agee, J., McGowan, C., Oremland, R.S., Thomas, M., Krabbenhoft, D. & Gilmour, C.C. (2000) Methyl-mercury degradation pathways: A comparison among three mercury-impacted ecosystems. *Environmental Science & Technology* 34 (23) 4908-4916

Meili, M. (1997) Mercury in lakes and rivers. *Metal Ions in Biological Systems* 34 21-51

O'Driscoll, N.J., Lean, D.R.S. & Loseto, L.L. (2004) Effect of dissolved organic carbon on the photoproduction of dissolved gaseous mercury in lakes: Potential impacts of forestry. *Environmental Science & Technology* 38 (9) 2664-2672

Porvari, P. & Verta, M. (2003*a*) Total and methyl mercury concentrations and fluxes

from small boreal forest catchments in Finland. *Environmental Pollution* 123 181–191

Porvari, P., Verta, M. & Munthe J. (2003*b*) Forestry practices increase mercury and methyl mercury output from boreal forest catchments *Environmental Science & Technology* 37 (11) 2389-2393

Qian, J., Skyllberg, U., Tu, Q., Bleam, F. & Frech, W. (2000) Efficiency of solvent extraction methods for the determination of methyl mercury in forest soils. *Fresenius' journal of analytical chemistry* 367 (5) 467 - 473

Ramlal, P.S., Kelly, C.A., Rudd, J.V.M. & Furutani, A. (1993) Sites of methyl mercury production in remote Canadian Shield lakes. *Canadian Journal of Fisheries and Aquatic Sciences* 50 (5) 972-979

Schroeder, W.H. & Munthe, J. (1998) Atmospheric mercury - An overview. *Atmospheric Environment* 32 (5) 809-822

Sellers, P., Kelly, C.A., Rudd, J.W.M. & MacHutchon, A.R. (1996) Photodegradation of methylmercury in lakes. *Nature* 380 (6576) 694-697

Skogsvårdsstyrelsen (Swedish National Board of Forestry) (2003) Forests for Water – Demonstration of opportunities on forest land to support the implementation of the Water Framework Directive. Source:*http://www.svo.se/dokument/sks/projekt/life/forests_for_water/ Publications%20and%20information%20material/plansch%20engelsk.pdf* [Online]

Skyllberg, U., Xia, K., Bloom, P.R., Nater, E.A. & Bleam, W.F. (2000) Binding of mercury(II) to reduced sulfur in soil organic matter along upland-peat soil transects. *Journal of Environmental Quality* 29 (3) 855-865

Skyllberg, U., Qian, J., Frech, W., Xia, K. & Bleam, W.F. (2003) Distribution of mercury, methyl mercury and organic sulphur species in soil, soil solution and stream of a boreal forest catchment. *Biogeochemistry* 64 53–76

St Louis, V.L., Rudd, J.W.M., Kelly, C.A., Beaty, K.G., Bloom, N.S. & Flett, R.J. (1994) Importance of wetlands as sources of methyl mercury to boreal forest ecosystems. *Canadian Journal of Fisheries and Aquatic Sciences* 51 (5) 1065-1076

St Louis, V.L., Rudd, J.V.M., Kelly, C.A., Beaty, K.G., Flett, R.J. & Roulet, N.T. (1996) Production and loss of methylmercury and loss of total mercury from boreal forest catchments containing different types of wetlands. *Environmental Science & Technology* 30 (9) 2719-2729

Warner, K.A., Roden, E.E. & Bonzongo, J.C. (2003) Microbial mercury transformation in anoxic freshwater sediments under iron-reducing and other electron-accepting conditions. *Environmental Science & Technology* 37 (10) 2159-2165

Weber, J.H., Evans, R., Jones, S.H. & Hines, M.E. (1998) Conversion of mercury(II) into mercury(0), monomethylmercury cation, and dimethylmercury in saltmarsh sediment slurries. *Chemosphere* 36 (7) 1669-1687

Xia, K., Skyllberg, U.L., Bleam, W.F., Bloom, P.R., Nater, E.A. & Helmke, P.A. 1999. X-ray absorption spectroscopic evidence for the complexation of Hg(II) by reduced sulfur in soil humic substances. *Environmental Science & Technology* 33 257–261.

Appendix

A – Study Area

Figure 1 Photo taken on top of the N facing slope approximately 100 m away from the small first order stream. The slopes are dominated by Pinus sylvestris and Picea abies, with a field layer nursing Vaccinium myrtillus and several moss species.

Figure 2 *The small first order stream runs at the very bottom of the study area. The sphagnum dominated riparian zone has an apparent wetland character that differs from the more obvious upland environment in Fig. 1.*

Figure 3 *Samples were collected at the stream/peat interface in June 2005.*

Figure 4 *A close up on the material overlying one of the collected peat samples reveals the network of sphagnum- and vascular plant roots providing the SRB community with fresh carbon compounds. The heterogeneity of the material becomes obvious.*

B – Analytical Results

Table 1 Total Hg and organic matter content in the collected peat samples from the stream/peat interface. Sampling was conducted in the same area and under identical conditions.

Sample	Sampling Date	Total Hg content (ppb)	Organic Matter Content (%)
1	June 15	17.3	-
2	″	17.3	-
3	″	17.3	-
4	July 3	-	5.3
5	″	-	5.2
6	″	-	5.3
7	″	-	5.3
8	″	-	5.3

Table 2 Mean Hg methylation rate in samples incubated during 0, 23, 46, 64 and 112 h. (Incubation Time study)

Incubation Time (h)	N	Mean Hg Methylation Rate (pg g^{-1} dw h^{-1})	SE	Detection Limit (pg g^{-1} dw h^{-1})
0	2	0*	-	0.1
23	2	0*	-	0.1
46	2	6.2	0.9	0.1
64	2	16.1	1.9	0.1
112	2	17.2	1.1	0.1

* undetectable in GC-ICP-MS analysis

Table 3 Mean MeHg production in samples incubated with different concentrations of sulphate, glucose and Hg. (Optimal concentration study)

[Sulphate] (mM)	[Glucose] (mM)	[Hg] (ppb)	N	Mean Hg Methylation Rate (pg g^{-1} dw h^{-1})	SE	Detection Limit (pg g^{-1} dw h^{-1})
0.1	-	-	2	2.5	0.4	0.1
0.5	-	-	2	7.0	2.1	0.1
2	-	-	2	11.6	0.6	0.1
5	-	-	2	5.3	0.2	0.1
	1	-	2	13.5	0.0	0.1
	5	-	2	7.5	0.5	0.1
	10	-	2	8.4	2.0	0.1
	20	-	2	5.3	0.4	0.1
		1.7	2	0.9	0.9	0.1
		17	2	16.9	3.9	0.1
		170	2	2.0	1.3	0.1
		1700	2	5.9	0.3	0.1

Table 4 The full experimental matrix used in the main study. The incubation time and optimal (+) concentrations were derived from the initial studies. (-) concentration of sulphate and glucose equals no addition. Due to analytic reasons, the (-) Hg treatments must be added a small amount of Hg. Centre point (C) concentrations equals (+/2).

Incubation Time (h)	N	[Sulphate] (mM)	[Glucose] (mM)	[Hg] (ppb)
64	2	1.5	0.1	17
64	2	1.5	0	17
64	2	0	0.1	17
64	2	0	0	17
64	2	1.5	0.1	1.7
64	2	1.5	0	1.7
64	2	0	0.1	1.7
64	2	0	0	1.7
64	4	0.75	0.05	8.5

Table 5 The result of different concentrations of glucose, sulphate and Hg on the mean rate of Hg methylation. (Main study)

[Sulphate]	[Glucose]	[Hg]	N	Mean Hg Methylation Rate ($pg\ g^{-1}\ dw\ h^{-1}$)	SE	Detection Limit ($pg\ g^{-1}\ dw\ h^{-1}$)
+	+	+	2	3.0	0.04	0.46
+	-	+	2	2.4	0.08	0.24
-	+	+	2	0.5	0.03	0.26
-	-	+	2	0.6	0.01	0.29
+	+	-	2	0*	-	0.31
+	-	-	2	0*	-	0.25
-	+	-	2	0*	-	0.20
-	-	-	2	0*	-	0.24
C	C	C	4	0.05	0.03	0.06

Note: (+), (-) and (C) concentrations according to Table 4 above.
* undetectable in GC-ICP-MS analysis

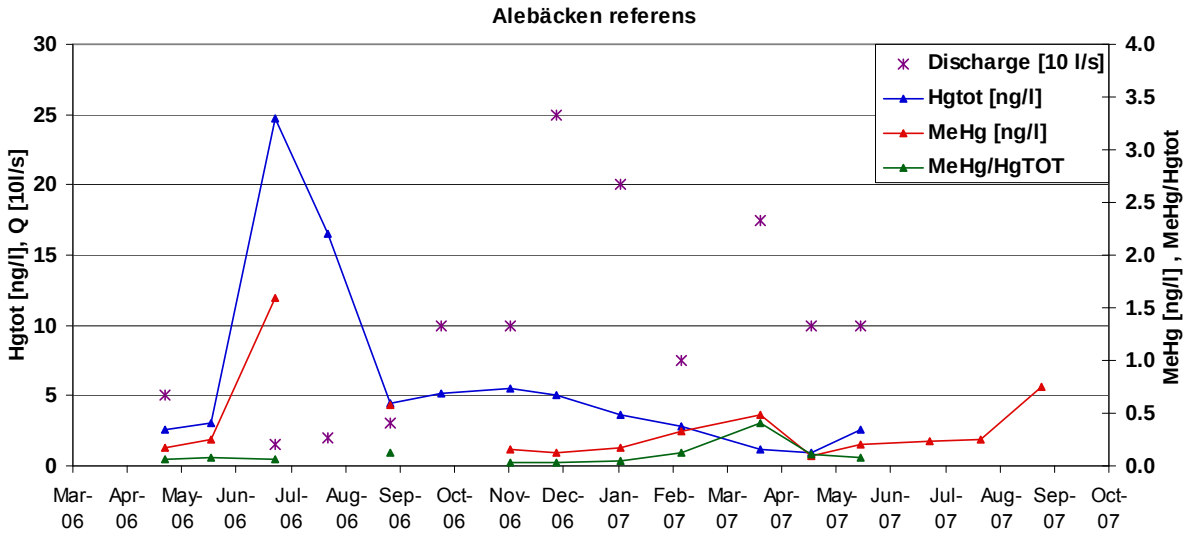
Table 6 The result of different concentrations of glucose, sulphate and Hg on the mean rate of MeHg demethylation. (Main study)

[Sulphate]	[Glucose]	[Hg]	N	Mean MeHg Demethylation Rate ($ng\ g^{-1}\ dw\ h^{-1}$)	SE
+	+	+	2	13.5	0.1
+	-	+	2	13.7	<0.1
-	+	+	2	13.7	<0.1
-	-	+	2	13.5	<0.1
+	+	-	2	13.6	<0.1
+	-	-	2	13.6	<0.1
-	+	-	2	13.7	<0.1
-	-	-	2	13.6	<0.1
C	C	C	4	13.6	<0.1

Note: (+), (-) and (C) concentrations according to Table 4 above.

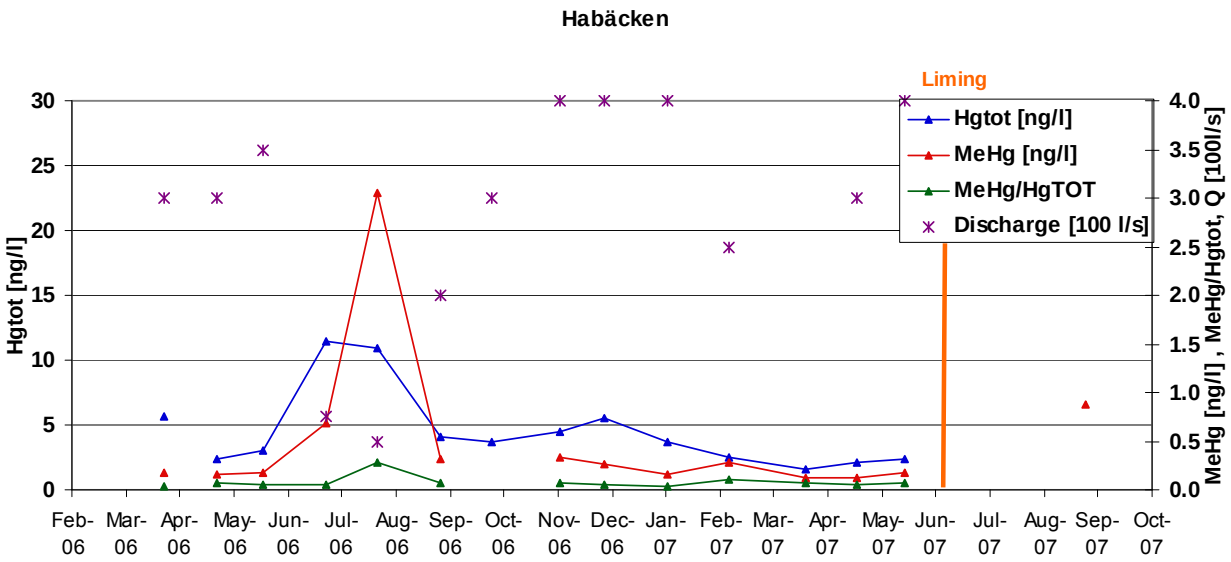
Appendix C. Timeseries of MeHg/Hgtot-ratio and its components

Flow from Hulkebäcken added for information.



Alebäcken (no liming):

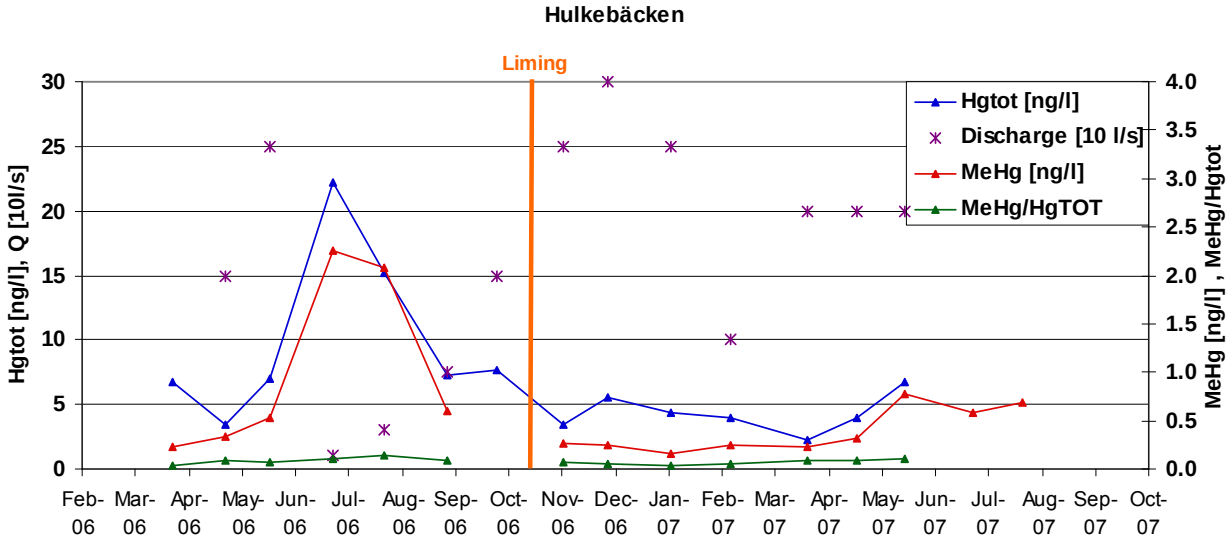
Sampling started in the spring 2006. Both MeHg and Hgtot peaked in June 2006. A small MeHg increase in August was noted which is the latest sampling we have the analysis result for. The ratio MeHg/Hgtot increased in early spring 2007.



Habäcken (liming summer 2007, i.e. later date than analysis has reached):

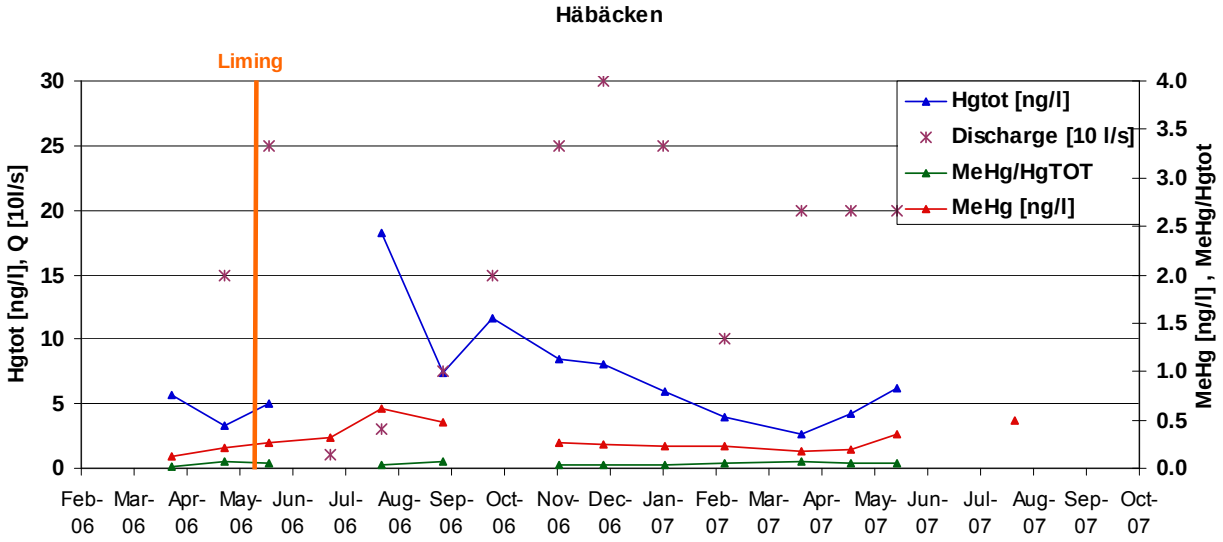
MeHg concentrations from summer 2006 peaked in July and increased again in august 2007, same pattern as for Alebäcken.

Since the Hgtot also increased during summer 2006 the ratio between the components increased only moderately.



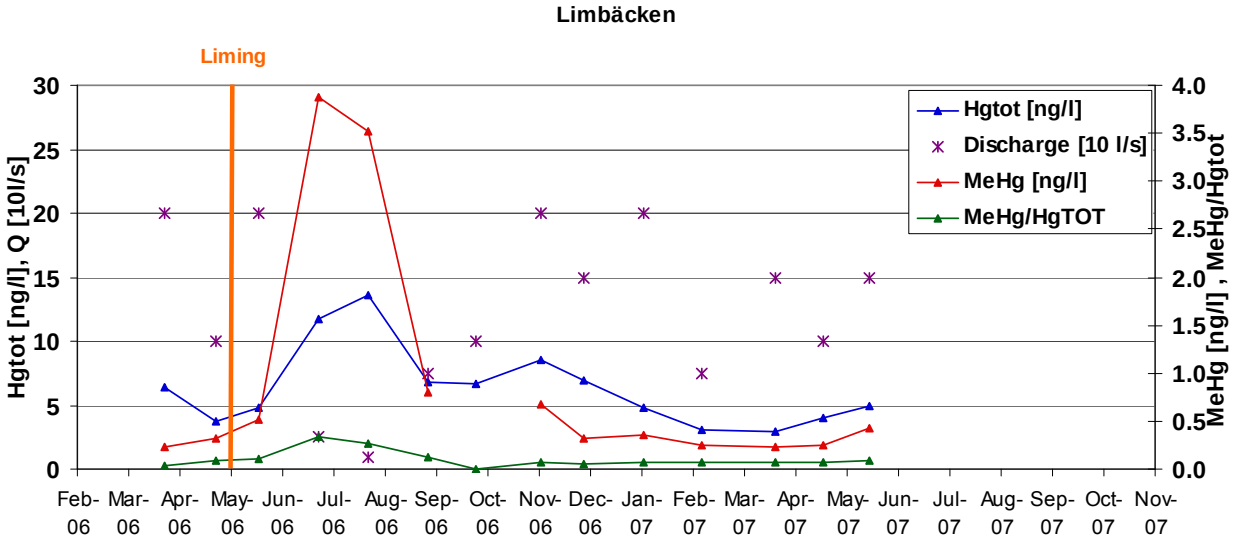
Hulkebäcken (liming October 2006): The ratio MeHg/Hgtot hardly changes at all during summer base flow compared to Habäcken. Otherwise the patterns for the individual components are similar to those for Habäcken with increase in MeHg

in June/July 2006. The post-liming concentrations of MeHg do not seem to be affected by liming.



Häbäcken (liming May 2006):

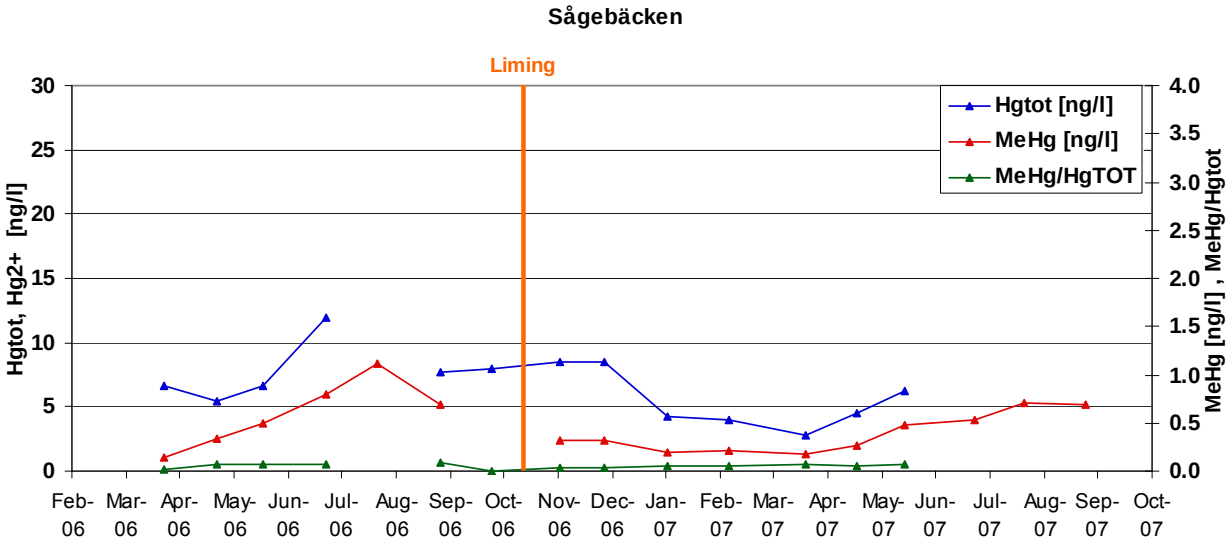
The summer MeHg concentrations did not increase as much as for the unlimed streams.



Limbäcken (liming May 2006):

Limbäcken had MeHg concentrations that were extremely high shortly after liming.

Both MeHg and Hgtot concentrations seemed to generally vary more in this stream compared to its neighbouring streams. The peak in MeHg and Hgtot for June/July fits in time with the unlimed streams.



Sågebäcken (liming October 2006):

Sågebäcken seemed to have more moderate variations in MeHg concentrations which were generally lower, a very similar patter to Häbäcken, though limed 5 months later. No obvious effect of liming on MeHg or Hgtot.

Av Skogsstyrelsen publicerade Rapporter:

1988:1 Mallar för ståndortsbonitering; Lathund för 18 län i södra Sverige
1988:2 Grusanalys i fält
1990:1 Teknik vid skogsmarkskalkning
1991:1 Tätortsnära skogsbruk
1991:2 ÖSI; utvärdering av effekter mm
1991:3 Utboträffar; utvärdering
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1991:5 Contortarapporten
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1993:3 Skogsskador i Sverige 1992
1993:5 Nyckelbiotoper i skogarna vid våra sydligaste fjäll
1993:6 Skogsmarkskalkning – *Resultat från en fyraårig försöksperiod samt förslag till åtgärdsprogram*
1993:7 Betespräglad äldre bondeskog – *från naturvårdssynpunkt*
1993:8 Seminarier om Naturhänsyn i gallring i januari 1993
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1994:5 Historiska kartor - *underlag för natur- och kulturmiljövård i skogen*
1994:6 Skogsskador i Sverige 1993
1994:7 Skogsskador i Sverige – *nuläge och förslag till åtgärder*
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1995:2 SUMPSKOG – ekologi och skötsel
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1997:2 Naturvårdsutbildning (20 poäng) Hur gick det?
1997:3 IR-95 – Flygbildsbaserad inventering av skogsskador i sydvästra Sverige 1995
1997:5 Miljeu96 Rådgivning. Rapport från utvärdering av miljeurådgivningen
1997:6 Effekter av skogsbränsleuttag och askåterföring – *en litteraturstudie*
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1997:9 GIS–metodik för kartläggning av markförsurning – *En pilotstudie i Jönköpings län*
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1998:2 Studier över skogsbruksåtgärdernas inverkan på snäckfaunans diversitet *(with English summary: Studies on the impact by forestry on the mollusc fauna in commercially uses forests in Central Sweden*
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1998:6 Övervakning av biologisk mångfald i det brukade skogslandskapet. *With a summary in English: Monitoring of biodiversity in managed forests.*
1998:7 Marksvampar i kalkbarrskogar och skogsbeten i Gotländska nyckelbiotoper
1998:8 Omgivande skog och skogsbrukets betydelse för fiskfaunan i små skogsbäckar
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1999:2 Internationella konventioner och andra instrument som behandlar internationella skogsfrågor
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Liming may affect flora and fauna. Ectomycorrhizal fungi have been found to be particularly sensitive. In the present report, a study aiming at 1) a review of results from previous studies of lime and ash addition effects and 2) an additional field study to monitor if three tonnes of lime per hectare have caused any significant effects 17 years after liming is presented

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