Ecological and Evolutionary Consequences of Herbivory in the Perennial Herb *Lythrum salicaria*

LINA LEHNDAL
Abstract


In this thesis, I combined field, common-garden and greenhouse experiments to examine the ecological and evolutionary consequences of plant-herbivore interactions in the perennial herb *Lythrum salicaria*. More specifically I examined (1) whether resistance and tolerance to damage from herbivores vary with latitude and are positively related to the intensity of herbivory in natural populations, (2) whether effects of herbivory on plant fitness vary with latitude, (3) whether populations are locally adapted and whether herbivory influences the relative fitness of populations, and (4) whether the intensity and effects of insect herbivory on reproductive output vary locally along a disturbance gradient and are associated with differences in plant resistance.

A common-garden and a greenhouse experiment demonstrated that plant resistance decreased whereas plant tolerance increased with latitude of origin among populations sampled along a latitudinal gradient in Sweden. Oviposition and feeding preference in the greenhouse and leaf damage in the common-garden experiment were negatively related to natural damage in the source populations.

Experimental removal of insect herbivores in three populations sampled along the latitudinal gradient demonstrated that intensity of herbivory and its effects on plant fitness decreased towards the north. A reciprocal transplant experiment among the same three populations showed that herbivory affected the relative fitness of the three populations, but did not detect any evidence of local adaptation. Instead the southernmost population had the highest relative fitness at all three sites.

A herbivore-removal experiment conducted in nine populations in an archipelago in northern Sweden demonstrated that insect herbivory strongly influenced among-population variation in reproductive output. However, variation in resistance was not related to differences in intensity of herbivory at this spatial scale.

Taken together, the results demonstrate that resistance and tolerance to herbivory vary with latitude but in opposite directions, that intensity of herbivory is a major determinant of flowering and seed output, and that the strength of herbivore-mediated selection varies among populations in *Lythrum salicaria*. They further indicate that both physical disturbance regime and latitudinal variation in abiotic conditions may strongly influence the performance and abundance of perennial herbs because of their effects on interactions with specialized herbivores.

Keywords: Disturbance gradient, Female reproductive success, Galerucella calamiensis, Galerucella pusilla, Herbivore removal, Latitudinal gradient, Local adaptation, Nanophyes marmoratus, Plant-herbivore interactions, Plant size, Resistance to herbivory, Tolerance to damage

Lina Lehndal, Department of Ecology and Genetics, Plant Ecology and Evolution, Norbyvägen 18 D, Uppsala University, SE-752 36 Uppsala, Sweden.

© Lina Lehndal 2015

ISSN 1651-6214
urn:nbn:se:uu:diva-247088 (http://urn.kb.se/resolve?urn=urn:nbn:se:uu:diva-247088)
List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals.


II Lehndal, L., Ågren, J. Herbivory differentially affects plant fitness in three populations of the perennial herb *Lythrum salicaria* along a latitudinal gradient. (Manuscript)

III Lehndal, L., Ågren, J. Herbivory influences the relative fitness of three native *Lythrum salicaria* populations, but no evidence of local adaptation along a latitudinal gradient. (Manuscript)

IV Lehndal, L., Hambäck, P. A., Ericson, L., Ågren, J. Herbivory strongly influences among-population variation in reproductive output of *Lythrum salicaria* in its native range. (Submitted manuscript)

Paper I is reprinted with permission from the publisher.
Contents

Introduction ......................................................................................................................... 7
Consequences of herbivory ............................................................................................... 8
Resistance and tolerance to herbivory ............................................................................ 8
Variation along environmental gradients ....................................................................... 9
Local adaptation .............................................................................................................. 10
Aim of the thesis ............................................................................................................... 11

Materials and methods .................................................................................................. 12
Study plant ....................................................................................................................... 12
Specialist herbivores ....................................................................................................... 13
Study populations and experimental sites ..................................................................... 13
Among-population variation in resistance and tolerance to herbivory (I) ...................... 16
Effects of herbivory on plant fitness along a latitudinal gradient (II) ......................... 17
Reciprocal transplant experiment (III) .......................................................................... 17
Effects of herbivory on plant fitness along a disturbance gradient (IV) ...................... 18
Statistical analyses ......................................................................................................... 18

Results and discussion ................................................................................................... 20
Latitudinal variation in resistance and tolerance to herbivory (I) ................................. 20
Herbivory differentially affects plant fitness along a latitudinal gradient (II) ........... 22
No local adaptation, but herbivory influences relative fitness of three host plant populations (III) ................................................................. 22
Strong influence of herbivory on among-population variation in reproductive output along a disturbance gradient (IV) ............................................................... 25
Conclusions .................................................................................................................... 26

Summary in Swedish ....................................................................................................... 27

Acknowledgements ........................................................................................................ 32

References ...................................................................................................................... 33
Introduction

Plants are one of the most diverse life forms on the planet, and interactions with other organisms such as herbivores, pathogens and pollinators play an important role in shaping this diversity. Plant-herbivore interactions are among the most dominant species interactions on earth; herbivores feeding on leaves, flowers, seeds, roots and interior parts of plants remove 10-15% of the biomass produced annually (Carmona et al. 2011).

To avoid being consumed by herbivores, plants have evolved a diverse array of defences against the many insects, mammals and microorganisms that use them as a food source (Agrawal 2007, Walters 2011). The burning trichomes of a stinging nettle, the hot flavour of a chilli pepper and the thorns of a rose bush are well-known examples of the wide variation in defences against herbivore attack that can be found in plants. However, herbivores influence not only plant form and function, but also plant distribution and population dynamics because of their effects on plant performance and reproduction (Crawley 1989, Maron and Crone 2006). Thereby, herbivores often have strong effects on the structure and composition of many ecosystems and vegetation types. A striking example is the influence of grazing animals on the species-rich European cultural landscapes (Emanuelsson 2009).

The concept of biodiversity is usually associated with species-richness, but there is often a wide phenotypic and genetic variation also within species, both among and within populations. The genetic diversity within a population determines its ability to respond to selection and thus to changes in the environment. A central goal in evolutionary biology is to understand factors responsible for the origin and maintenance of diversity within and among populations, and such an understanding requires that the functional and adaptive significance of species interactions are examined.

Understanding how herbivores drive the evolution of adaptive population differentiation in plants also has many practical implications. First, knowledge about how interactions between plants and herbivores will be altered with a changing climate and how that in turn will affect ecosystem function is of crucial importance to more accurately anticipate consequences of human-induced environmental change (Parmesan 2006). Second, elucidating how herbivores affect plant performance and population dynamics is highly relevant in an agricultural context, for example to be able to develop well-defended crops or successful biological control programs (Rausher 2001).
In this thesis, I explore the ecology and evolution of plant-herbivore interactions by evaluating how plant defence as well as intensity and fitness-consequences of herbivory vary along environmental gradients. As a model system I use the perennial herb *Lythrum salicaria*, which is particularly suitable to address these issues for several reasons: it has a wide distribution, it varies clinally in phenology and life-history traits (Olsson and Ågren 2002) and it is closely associated with a few, specialised insect herbivores.

**Consequences of herbivory**

Herbivory usually has a negative impact on plant performance by reducing plant fitness in terms of survival, growth, flowering propensity and seed production (Crawley 1989, Ehrlén 1995a, Maron 1998, Hunt-Joshi et al. 2004). An increasing number of studies have shown that herbivory also can influence plant population dynamics and distributions (e.g. Ehrlén 1995b, Maron and Crone 2006, Miller et al. 2009). Moreover, a few studies have shown that herbivores can drive evolutionary changes in plant populations (Agrawal et al. 2012, Ågren et al. 2013, Didiano et al. 2014).

The fitness-consequences of damage from herbivores can vary depending on several factors such as plant size (Horvitz and Schemske 2002), timing of damage relative to plant growth (Akiyama and Ågren 2012) and the amount of resources available to the plant (Wise and Abrahamson 2007). Because of variation in tolerance to damage, the effects of herbivory on plant fitness need not be proportional to damage levels (Wise and Abrahamson 2007, Fornoni 2011). Moreover, in perennial plants, damage from herbivores can have both short- and long-term effects (Knight 2003, Puentes and Ågren 2012).

**Resistance and tolerance to herbivory**

Plants have two general defence strategies against herbivores: resistance and tolerance (Núñez-Farfán et al. 2007). Resistance traits prevent or reduce damage by deterring herbivores, and can be either expressed constitutively (always expressed) or induced (expressed following herbivore damage) (Núñez-Farfán et al. 2007, Karban 2011). Resistance traits include various physical barriers such as waxes on the leaf surface, trichomes, thorns, secretory canals and leaf toughness, as well as chemical defences, often called secondary metabolites, such as terpenes, phenols, nitrogen-containing compounds and volatiles (Carmona et al. 2011, Walters 2011). Plant resistance can be operationally defined as one minus the proportion of leaf area removed by herbivores (Pilson 2000). Tolerance to herbivory is the ability of a plant to maintain fitness despite herbivory, and is usually defined as the reac-
tion norm of fitness across a damage gradient (Simms 2000). Tolerance traits reduce the negative fitness impacts of damage and enable compensation for damage, for example by regrowth or increased flower production (Strauss and Agrawal 1999) and are often related to traits associated with plant growth or life-history (Stowe et al. 2000, Núñez-Farfán et al. 2007).

If both resistance and tolerance have an allocation cost and resources are limited, resistance and tolerance may trade-off against each other (Fineblum and Rausher 1995). However, there is limited evidence for such trade-offs (Leimu and Koricheva 2006, Puentes and Ågren 2014, Więski and Pennings 2014). Instead, increasing evidence suggests that resistance and tolerance are not redundant strategies, but rather that plants commonly exhibit multiple defence strategies allocating resources to both resistance and tolerance (Mauricio et al. 1997, Leimu and Koricheva 2006, Carmona and Fornoni 2013).

Both resistance and tolerance include traits that vary genetically within plant species and are often, but not always, under selection exerted by herbivores (Fornoni 2011). Selection on traits that contribute to resistance and tolerance to herbivory may be driven also by factors other than herbivory. For example, phenology of plant growth and flowering can affect both the likelihood of herbivory (Feeny 1970, Bishop and Schemske 1998, Scheirs et al. 2002) and tolerance to herbivory (Lowenberg 1994, Del-Val and Crawley 2005, Akiyama and Ågren 2012).

Variation along environmental gradients

The strength and/or fitness-consequences of plant-herbivore interactions often vary spatially across habitats and along gradients of environmental conditions (Maron and Crone 2006, von Euler et al. 2014).

In plant species with a wide distribution, the intensity of herbivory may vary with latitude, and according to the Tropical Defense Hypothesis (Rasmann and Agrawal 2011), plant resistance should increase towards the equator as a result of increased herbivory. Several studies have reported latitudinal clines in intensity of plant-herbivore interactions (Dobzhansky 1950, Coley and Barone 1996, Pennings and Silliman 2005, Pennings et al. 2009, Scheemske et al. 2009), even though the presence, direction and magnitude of such clinal variation differ among species (Moles et al. 2011). Moreover, latitudinal clines in defence against herbivory have been documented in some systems and have been attributed to herbivore-mediated selection (Pemberton 1998, Scheemske et al. 2009, Rasmann and Agrawal 2011). The fitness-consequences of damage may vary with latitude because of differences in the magnitude of damage but also because of differences in the temporal overlap between herbivory and plant growth.
Spatial variation in plant-herbivore interactions occur not only on large geographic scales such as wide latitudinal gradients, but on local and regional scales as well. Variation in herbivore pressure and/or plant defence have been found along gradients of, e.g., altitude (Hodkinson 2005, Grassein et al. 2014) drought (Gutbrodt et al. 2011), sunlight (Louda and Rodman 1996), tidal height (Rand 2002), successional phase (Bishop 2002), population age (Stenberg et al. 2006), and disturbance (Knight and Holt 2005, Elderd 2006).

In this thesis I examine the effects of herbivory along two different kinds of environmental gradients; a latitudinal gradient on a large spatial scale (Papers I-III) and a disturbance gradient on a more local scale (Paper IV).

Local adaptation

Spatially variable selection due to heterogeneity in abiotic and biotic conditions may lead to the evolution of adaptive genetic differentiation and local adaptation, expressed as higher fitness of local compared with non-local populations in reciprocal transplant experiments (Kawecki and Ebert 2004, Ågren and Schemske 2012). Numerous studies have demonstrated adaptive differentiation among plant populations along environmental gradients (e.g. Joshi et al. 2001, Becker et al. 2006, Ågren and Schemske 2012, Toräng et al. 2015), but few have evaluated the extent to which interactions with natural enemies can explain the evolution and maintenance of local adaptation.

The classical test of local adaptation is the reciprocal transplant, in which populations originating from different habitats are reciprocally planted at their sites of origin. If there is local adaptation, the native plants should outperform non-native plants at their home site (Kawecki and Ebert 2004). However, when studying adaptation to local herbivores, transplant experiments may not be enough on their own. Due to a lack of an undamaged control, it is difficult to disentangle if any detected fitness advantage of the local population is due to a better ability to cope with herbivory or with other environmental factors (Agrawal 2011). Therefore, to quantify the effects of herbivores on the direction and strength of selection observed in reciprocal transplant experiments, an experimental approach is required where the reciprocal transplant is combined with manipulation of herbivore presence, but this has rarely been done (but see Abdala-Roberts and Marquis 2007, Ortegón-Campos et al. 2011).
Aim of the thesis

The general aim of my thesis was to explore the ecological and evolutionary consequences of interactions between plants and their herbivores, and the mechanisms by which biotic interactions contribute to among-population variation in plant performance and defence. As a model system, I used the perennial herb *Lythrum salicaria* and its specialist insect herbivores.

I addressed the following questions:

1. Does the intensity of herbivory vary with latitude and disturbance regime? (I, II, III, IV)
2. Is plant fitness reduced by herbivory and, if so, which fitness components are affected? (II, IV)
3. Does among-population variation in resistance and tolerance to herbivory reflect differences in intensity of herbivory? (I, III, IV)
4. Are populations along a latitudinal gradient locally adapted, and are their relative fitness influenced by herbivory? (III)
5. Do the intensity and fitness-consequences of herbivory vary with plant size? (II)
Materials and methods

Study plant

The perennial herb *Lythrum salicaria* L., purple loosestrife, was used as the study system in this thesis. In Sweden, it is found in a variety of wetland habitats, such as lake- and seashores, riversides and fens. Purple loosestrife is native to Eurasia (Hultén and Fries 1986) while in North America and Australia it is an invasive species where it has become widespread during the last 100 years (Thompson et al. 1987).

One or several above-ground shoots are produced by each plant, developing from winter buds formed on the rootstock in the previous year. Flower buds are produced in leaf nodes in the upper part of flowering shoots. *L. salicaria* is tristylos and has pinkish-purple flowers. In Sweden, it flowers for six to eight weeks in July and August. The seeds mature six to eight weeks after flowering (Olsson and Ågren 2002).

In the study area, *L. salicaria* flowers are visited mainly by bumble bees, but also by solitary bees, honey bees, syrphid flies and butterflies (Waites and Ågren 2004; J. Ågren unpublished data).

Figure 1. A large, flowering individual of *Lythrum salicaria* growing on the seashore in one of the study populations (Långron).
Specialist herbivores

In Sweden, two specialist leaf beetles, *Galerucella calmariensis* L. and *Galerucella pusilla* L. (Coloptera: Chrysomelidae) are the main herbivores on *L. salicaria*. The monophagous leaf beetles overwinter in the soil as adults and emerge in the spring when the aboveground shoots of the host plant are developing. After emergence, the adult beetles mate and lay eggs on leaves and stems. After seven to ten days, the larvae hatch and feed for two to three weeks before pupating in the soil. Adult *G. calmariensis* and *G. pusilla* mainly feed on leaves, and larvae feed on leaves and flower buds (Hambäck et al. 2000). Herbivory of *G. calmariensis* and *G. pusilla* on purple loosestrife can cause extensive damage to the host plant, and the beetles are currently used as biological control agents in North America (Grevstad 2006).

The two *Galerucella* species have similar life histories, but adults of *G. calmariensis* are slightly larger than *G. pusilla* adults (Hambäck et al. 2003). The two species coexist in southern Sweden, but only *G. calmariensis* is present in northern Sweden (Fig. 2, study populations 9-12 and Skeppsvik 1-10). Moreover, the two northernmost populations (study populations 11 and 12) may have been colonized by *G. calmariensis* only recently. The two populations have been visited repeatedly from 1996 and onwards (1996, 2000, 2007, 2010-2014), and *G. calmariensis* was observed for the first time in 2011 (L. Lehndal and J. Ågren, personal observation).

The weevil *Nanophyes marmoratus* Goeze (Coleoptera: Curculionidae) is the main seed predator on *L. salicaria* in Sweden, and occurs across the study area. It feeds solely on *L. salicaria* and has a univoltine life cycle. The adults emerge in early summer on young *L. salicaria* shoots, and feed on young leaves and later also on flower buds. The females oviposit on young flower buds, and the emerging larva consumes the reproductive parts of the flower and pupates at the bottom of the bud (Blossey and Schroeder 1995).

Study populations and experimental sites

All study populations were located on shores of the Baltic Sea along the Swedish east coast (Table 1, Fig. 2). The study populations are found in open shore vegetation where interspecific competition is usually low because of disturbance from waves, ice and a fluctuating water table.

Study populations included in paper I were located along a latitudinal gradient along the Swedish east coast, separated by more than 1000 km and ranging from Karlshamn in the south to Kalix in the north (Fig. 2: populations 1-12). *L. salicaria* is rarely found at latitudes above 67° (Hultén and Fries 1986) and the northernmost populations were thus located near the northern range margin of the species. Both yearly mean temperature and the
length of the growing season vary clinally among the study populations (The Swedish Meterological and Hydrological Institute 2014).

The common-garden experiment (Paper I) was performed at Tomtasjön, Knivsta, Sweden (59.72°N, 17.76°E), in a semi-natural grassland where *L. salicaria* and its specialist herbivores *G. calmariensis*, *G. pusilla* and *N. marmoratus* are naturally present. The greenhouse experiment (Paper I) was conducted in the greenhouses of the Botanical Garden of Uppsala University (59.85°N, 17.63°E).

Study populations of paper II and III included three populations sampled along the latitudinal gradient, from mid to northern Sweden (Table 1, Fig. 2: populations 5, 10 and 11).

The study populations included in paper IV were located in the Skeppsvik archipelago, about 17 km east-southeast of Umeå in northern Sweden. The populations were separated by up to 5 km and were distributed from the inner part close to the mainland to the outer archipelago (Table 1, Fig. 2: populations Skeppsvik 1-10). In the outer part, populations are more exposed to disturbance from wave and ice action than are populations closer to the mainland and the study populations thus represented a disturbance gradient.

![Figure 2. Maps of Sweden (left) and the Skeppsvik archipelago (right) showing the locations of the *Lythrum salicaria* populations used in the thesis. Populations 1-12 (grey circles) were used in Paper I, populations 5, 10 and 11 (grey circles) were used in papers II and III, and the ten populations in the Skeppsvik archipelago (black circles) were used in paper IV. See Table 1 for population characteristics.](image-url)
Table 1. Population names and coordinates of study populations used in the thesis and ranges of yearly mean temperature and length of growing season 1961-1990 (Climatic data from the Swedish Meteorological and hydrological Institute, 2014). Populations Skeppsvik 1-10 in the bottom of the table are located in the Skeppsvik archipelago, east-southeast of Umeå, Sweden.

<table>
<thead>
<tr>
<th>Population</th>
<th>Latitude (N)</th>
<th>Longitude (E)</th>
<th>Yearly mean temperature (°C)</th>
<th>Length of growing season* (days)</th>
<th>Used in thesis papers</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Stärnö</td>
<td>56.14°</td>
<td>14.83°</td>
<td>7-8</td>
<td>210-220</td>
<td>I</td>
</tr>
<tr>
<td>2. Kråkelund</td>
<td>57.45°</td>
<td>16.73°</td>
<td>6-7</td>
<td>190-200</td>
<td>I</td>
</tr>
<tr>
<td>3. Händelöp</td>
<td>57.67°</td>
<td>16.75°</td>
<td>6-7</td>
<td>190-200</td>
<td>I</td>
</tr>
<tr>
<td>4. Flatvarp</td>
<td>57.99°</td>
<td>16.81°</td>
<td>6-7</td>
<td>190-200</td>
<td>I</td>
</tr>
<tr>
<td>5. Forsmark</td>
<td>60.40°</td>
<td>18.22°</td>
<td>5-6</td>
<td>180-190</td>
<td>I, II, III</td>
</tr>
<tr>
<td>6. Eskön</td>
<td>60.79°</td>
<td>17.33°</td>
<td>4-5</td>
<td>170-180</td>
<td>I</td>
</tr>
<tr>
<td>7. Iggön</td>
<td>60.87°</td>
<td>17.33°</td>
<td>4-5</td>
<td>170-180</td>
<td>I</td>
</tr>
<tr>
<td>8. Lörudden</td>
<td>62.23°</td>
<td>17.66°</td>
<td>3-4</td>
<td>160-170</td>
<td>I</td>
</tr>
<tr>
<td>9. Långron</td>
<td>63.41°</td>
<td>19.50°</td>
<td>2-3</td>
<td>150-160</td>
<td>I</td>
</tr>
<tr>
<td>10. Vitskärsudden</td>
<td>63.65°</td>
<td>20.29°</td>
<td>2-3</td>
<td>150-160</td>
<td>I, II, III</td>
</tr>
<tr>
<td>11. Skagsudden</td>
<td>65.70°</td>
<td>23.10°</td>
<td>1-2</td>
<td>140-150</td>
<td>I, II, III</td>
</tr>
<tr>
<td>12. Pålänge</td>
<td>65.79°</td>
<td>22.89°</td>
<td>1-2</td>
<td>140-150</td>
<td>I</td>
</tr>
<tr>
<td>Skeppsvik 1-10</td>
<td>63.78°</td>
<td>20.62°</td>
<td>2-3</td>
<td>150-160</td>
<td>IV</td>
</tr>
</tbody>
</table>

*Number of days with an average temperature of > 5°C
Among-population variation in resistance and tolerance to herbivory (I)

To quantify among-population variation in resistance and tolerance to herbivory, I grew plants originating from 11 populations sampled along a latitudinal gradient through Sweden (Table 1, Fig. 2) in a common-garden experiment. The experiment was established in the summer of 2012, in a semi-natural grassland where herbivores are naturally present. Plants used in the experiment were grown from seeds collected in the source populations and had been growing in an experimental garden for three years prior to the experiment. Each population was represented by 100 plants except population 11, which contributed 72 plants. Half of the plants were randomly assigned to a herbivore-removal treatment and the other half were used as control plants. Plants in the experimental treatment were sprayed with an insecticide on five occasions during June and July when the herbivores are active, and control plants were sprayed with an equal amount of water. At the end of the summer, I quantified plant height and damage from herbivores. Plant resistance was estimated by calculating means of leaf and meristem damage in control plants, and tolerance to leaf damage was quantified as the ratio between population means of the difference in plant height and in leaf damage between controls and plants in the herbivore-removal treatment.

To quantify among-population variation in resistance to *Galerucella*, I performed an oviposition and feeding preference experiment in the greenhouse, in which I exposed plants to recently mated female beetles and used oviposition and proportion of leaf area removed as inverse measures of resistance. Plants used in the experiment were grown from seeds collected in 12 source populations along the latitudinal gradient (Table 1, Fig. 2). Seeds were sown in April 2011 and the experiment was carried out in June the same year. I used 27 mesh cages in each of which I placed 12 plants, one from each population, in a randomized order. I measured plant height and then released six *Galerucella* females into each cage and allowed them to freely feed and oviposit during six days. I then counted the number of eggs on each plant and quantified leaf damage.

I also performed a field survey where I documented plant size and damage in the 12 source populations. In August 2011, I recorded plant height and damage to 100 plants taller than 20 cm in each population. Damage in the natural populations was then related to among-population variation in resistance and tolerance to herbivory quantified in the common-garden experiment and in the greenhouse.
Effects of herbivory on plant fitness along a latitudinal gradient (II)

To examine among-population variation in effects of herbivory on plant reproductive output in three natural populations sampled along the latitudinal gradient (Table 1), I performed a herbivore-removal experiment. I reduced herbivore abundance on half of 200 plants in each population by spraying them with an insecticide and compared them with control plants that were sprayed with water. The insecticide was applied on five occasions during June and July 2013. Before the start of the experiment, in May 2013, I recorded plant height. At the end of the season, I recorded survival, height, number of floral shoots and leaf damage for each plant. I also collected one floral shoot from each plant to estimate flower, fruit and seed production in the laboratory.

Since *L. salicaria* is a perennial plant, the performance of all plants were assessed also in the next year, following the same procedure, but for logistic reasons I did not count the seeds but only estimated flower and fruit production in the lab.

Reciprocal transplant experiment (III)

To test the hypothesis that the three *L. salicaria* populations are locally adapted, and to examine whether herbivory influences the relative fitness of the study populations, I conducted a reciprocal transplant experiment that included a herbivore-removal treatment. Three hundred and sixty plants per population were reciprocally transplanted among three natural populations (120 plants per population planted at each site) along a latitudinal gradient separated by approximately 250 to 650 km (Table 1). Seeds were sown in May 2012. To reduce maternal effects, the seeds had gone through one generation of within-population random outcrossing in the greenhouse prior to the experiment. The plants where transplanted to the field sites in August 2012, and were planted individually along the seashore among naturally occurring *L. salicaria* plants.

In the herbivore-removal treatment, I reduced herbivore abundance on half of the transplanted plants in each population by spraying them with an insecticide on five occasions during June and July 2013. Control plants were sprayed with an equal amount of water. At the end of the season, I recorded height, number of floral shoots and leaf damage for each plant. I also collected one floral shoot from each plant to estimate fruit production in the laboratory. In the following year, I recorded the performance of all plants following the same procedure as in the first year.
Effects of herbivory on plant fitness along a disturbance gradient (IV)

To examine among-population variation in intensity of herbivory and the effects of herbivory on plant reproductive output along a disturbance gradient, a herbivore-removal experiment was conducted in an archipelago in northern Sweden. Nine populations were selected on different islands, ranging from the inner to the outer part of the archipelago (Fig. 2). In June 1997, between 68 and 99 *L. salicaria* were marked in each population. Thirty-three of the marked plants on each island were randomly assigned to the herbivore-removal treatment and the remaining plants served as controls. Plants in the herbivore-removal treatment were sprayed with an insecticide at night on four occasions in June and July 1997 and control plants were sprayed with an equal amount of water. In August when herbivory had ceased, leaf and meristem damage were recorded in each plant. At fruit maturation, one floral shoot was collected and brought to the laboratory where flower, fruit and seed production was estimated. To quantify among-year variation in intensity of herbivory, leaf damage to control plants was recorded also in the following summer, 1998.

To examine among-population variation in resistance against herbivory in the studied archipelago, plants originating from six populations were grown in a common-garden experiment in the same area. This experiment included five populations from the herbivore-removal experiment and one additional population (study population 10, Fig. 2), and a total of 1083 plants. Seeds were sown in a greenhouse in May 1993 and seedlings were transplanted to the field site in June the same year. To reduce maternal effects, seeds used in the common-garden experiment had gone through one generation of within-population random outcrossing in the greenhouse. Leaf damage to each plant was recorded in August 1997.

Statistical analyses

All statistical analyses were performed in R, a language and environment for statistical computing (R Core Team 2014).

In herbivore-removal experiments, ANOVA and generalized linear models were used to test the effects of plant population, herbivore-removal treatment (I-IV), site (III) and their interactions on different response variables using appropriate transformations and distributions. The effects of early-season plant height and herbivore-removal treatment on different response variables were examined with ANCOVA and generalized linear models (II).
Tukey tests and contrasts were used to determine which populations differed and in which populations there was a significant effect of herbivore-removal (I-IV).

In the greenhouse experiment, linear mixed models were used to examine the effects of beetle species (\textit{G. calmariensis} vs. \textit{G. pusilla}), plant population (fixed factors) and cage nested within beetle species (random factor) on the number of eggs per cm of plant and leaf damage (I).

Linear or quadratic regressions were used to test relationships between latitude of origin and plant height and resistance and tolerance to damage (I). Relationships between resistance in the common-garden and in the greenhouse and damage in the field were examined with linear regressions (I).

Relationships between mean leaf damage in the study populations in 1997 and 1998 and between damage in the common-garden experiment and in the source populations were quantified with Spearman rank correlation (IV).

The effect of block and population (fixed factors) and maternal family nested within population (random factor) on leaf damage in the common-garden experiment was examined with a linear mixed model (IV).
Results and discussion

Latitudinal variation in resistance and tolerance to herbivory (I)

Resistance and tolerance against insect herbivory varied among natural L. salicaria populations sampled along a latitudinal gradient across Sweden. Plant resistance to oviposition, leaf and meristem damage decreased (Figs. 3 and 4), whereas plant tolerance increased (Fig. 5), with latitude of origin. Oviposition and feeding preference in the greenhouse and leaf damage in the common-garden experiment were negatively related to damage in the source populations, which tended to decrease with latitude. The latitudinal variation in resistance was thus consistent with reduced selection from herbivores towards the northern range margin of L. salicaria. Variation in tolerance may be related to differences in the timing of damage in relation to the seasonal pattern of plant growth, as northern genotypes have developed further than southern have when herbivores emerge in early summer.

Figure 3. Relationships between latitude of origin and population mean ± SE proportion of leaf area removed in the control (filled circles) and in the herbivore-removal treatment (open circles) in the common-garden experiment (n = 11 populations). Standard errors are sometimes obscured by the symbols.
Figure 4. Relationships between latitude of origin and population means ± SE of (a) the number of eggs per cm of plant and (b) proportion of leaf area removed in the oviposition preference experiment, $n = 12$ populations.

Figure 5. Relationship between latitude of origin and tolerance to leaf damage quantified as the ratio of the difference in plant height ($\Delta$ height) and in proportion of leaf area removed ($\Delta$ leaf damage) between plants in the control treatment and in the herbivore-removal treatment in the common-garden experiment ($n = 11$ populations).
Herbivory differentially affects plant fitness along a latitudinal gradient (II)

The intensity of herbivory and the effects of herbivory on plant fitness varied among the three study populations, and were strongest in the southern population and intermediate in the central population. The mean proportion of leaf area removed ranged from 11% in the southern to 3% in the northern population (Fig. 6a). Herbivore removal increased plant height 1.5-fold in the southern and 1.2-fold in the central population (Fig. 6b), the proportion plants flowering 4-fold in the southern and 2-fold in the central population (Fig. 6c), and seed production per flower 1.6-fold in the southern and 1.2-fold in the central population (Fig. 6e), but did not affect plant fitness in the northern population. Herbivore removal thereby affected the relative fecundity of plants in the three populations: In the control, seed output per plant was 8.6 times higher in the northern population compared to the southern population, whereas after herbivore removal it was 2.5 times higher in the southern population compared to the northern (Fig. 6f). No effects of herbivore-removal on plant performance were detected in the year following the experimental treatment. Proportion of leaf area removed increased with plant size, but tolerance to damage did not vary with size.

The results demonstrate that native herbivores may strongly affect the demographic structure of *L. salicaria* populations, and thereby shape geographic patterns of seed production. They further suggest that the strength of herbivore-mediated selection varies among populations, and decreases towards the north.

No local adaptation, but herbivory influences relative fitness of three host plant populations (III)

The reciprocal transplant experiment demonstrated that variation in intensity of herbivory contributes to differences in selection regimes among three populations sampled along the latitudinal gradient in Sweden. Herbivory significantly influenced the magnitude of fitness differences between populations at the southern site, where herbivory was most intense (Fig. 7). However, herbivory did not alter the direction of selection. Instead, the southernmost population had the highest relative fitness at all three sites both in the control and in the herbivore-removal treatment (Fig. 7). Moreover, the directions of between-population differences in resistance were consistent across sites, suggesting that the study populations were not adapted to specific behaviours or preferences of local herbivore populations. This study did thus not detect any evidence of local adaptation at the spatial scale examined.
Genetic drift, recent climatic warming and intermittent strong selection against southern genotypes at northern latitudes may all contribute to the documented patterns of among-population variation in fitness, while similarity in the behaviour and preferences of herbivore populations along the studied gradient may explain the consistent differences in resistance.

Figure 6. Herbivory and components of fitness of control plants and plants treated with insecticide in three *Lythrum salicaria* populations (Forsmark, Vitskärsudden and Skagsudden) in Sweden in 2013. Population means ± SE are given for continuous variables and proportions for flowering status (flowering or not flowering): (a) proportion of leaf area removed, (b) late-season plant height, (c) proportion of plants flowering, (d) number of flowers per reproductive plant, (e) number of seeds per flower and (f) female reproductive success (the number of seeds produced per plant including vegetative plants). Significant differences between control plants (open bars) and plants from which herbivores were removed (filled bars) are indicated. * P < 0.05, ** P < 0.01, *** P < 0.001.
Figure 7. Effects of population of origin (Fo, Forsmark; V, Vitskärsudden; Sk, Skagssudden) and herbivore removal (control vs. herbivore removal) on a) proportion of leaf area removed, b) late-season plant height, c) flowering status, and d) the number of fruits per reproductive plant of *Lythrum salicaria* in the year of the experimental herbivore removal at the Forsmark, Vitskärsudden and Skagsudden sites. The symbol for the local population is framed. Means ± SE are shown except for flowering status, which is illustrated as the proportion of plants flowering. Significant main and interaction effects in a three-way ANOVA are indicated (P = population, H = herbivore-removal treatment, S = site).
Strong influence of herbivory on among-population variation in reproductive output along a disturbance gradient (IV)

There was considerable variation in intensity of leaf herbivory and plant performance among nine study populations in an archipelago in northern Sweden. Leaf damage varied >500-fold and mean female reproductive success >400-fold among the study populations (Fig. 8). The intensity of herbivory was lowest in populations subject to strong disturbance from ice and wave action. Experimental removal of insect herbivores showed that the effect of herbivory on female reproductive success was correlated with the intensity of herbivory and that differences in insect herbivory could explain much of among-population variation in the proportion of plants flowering and seed production (Fig. 9). Population differentiation in resistance to herbivory was limited and not related to natural levels of herbivory in the source populations.

The results demonstrate that the intensity of herbivory is a major determinant of flowering and seed output in *L. salicaria*, but that differences in herbivory are not associated with differences in plant resistance at the spatial scale examined. They further suggest that the physical disturbance regime may strongly influence the performance and abundance of perennial herbs not only because of its effect on interspecific competition, but also because of effects on interactions with specialized herbivores.

Figure 8. Herbivory and performance of control plants and plants treated with insecticide in nine *Lythrum salicaria* populations in the Skeppsvik archipelago. Population mean ± SE (a) proportion of leaf area removed and (b) female reproductive success (the number of seeds produced per plant including vegetative plants). Significant differences between control plants (open bars) and plants from which herbivores were removed (filled bars) are indicated. * P < 0.01, ** P < 0.001, *** P < 0.0001.
Figure 9. Linear regression of increase in female reproductive success ($\Delta \log$ (female RS)) on (a) decrease in leaf damage ($\Delta$ proportion of leaf area removed), and (b) decrease in meristem damage ($\Delta$ proportion of plants with top meristem damage) after herbivore removal.

Conclusions

In this thesis I explored the ecological and evolutionary consequences of interactions between the perennial herb *Lythrum salicaria* and its specialist herbivores. I examined among-population variation in intensity of herbivory and in resistance and tolerance to herbivory along geographical gradients on different scales.

I have shown that herbivory is a major determinant of flowering and seed output in *L. salicaria* populations in the native range, that the intensity of herbivory varies both with latitude on a large spatial scale and with disturbance regime on a local scale, that resistance and tolerance vary with latitude but in opposite directions, and that among-population variation in resistance to herbivory reflects differences in intensity of herbivory. Further, I have shown that the relative fitness of three populations sampled along a latitudinal gradient was influenced by herbivory but that there was no evidence of local adaptation along the gradient. Finally, intensity of herbivory, but not plant tolerance to herbivory, varied with plant size.

Taken together, the results demonstrate and that the strength of herbivore-mediated selection varies among populations in *Lythrum salicaria*. They further indicate that both the physical disturbance regime and latitudinal variation in abiotic conditions may strongly influence the performance and abundance of perennial herbs because of their effects on interactions with specialized herbivores.
Samspelet mellan växter och växtätande insekter

Växter utgör en av de artrikaste livsformerna på vår jord, och denna mångfald har till stor del formats genom interaktioner med andra organismer, exempelvis herbivorer, pollinatörer och patogener. Samspelet mellan växter och herbivorer är en av de mest dominerande artinteraktionerna som finns; av den biomassa som produceras årligen konsumeras mellan 10 och 15 procent av olika typer av växtätare. För att undkomma att bli upptagna har växterna utvecklat olika försvarsmekanismer mot de insekter, däggdjur och mikroorganismer som använder dem som näringskälla. Några välkända exempel på växters anpassningar till växtätare är taggiga rosbuskar, brännande nässlor och kryddstarka chilifruktor. Den typ av försvar som på så vis avkräcker herbivorer kallas resistens, medan växternas förmåga att växa och reproduera sig trots att de blir skadade kallas tolerans.

Herbivorerna påverkar inte bara växternas form och funktion, utan även deras utbredning och populationsdynamik och på så vis har de stor effekt även på utformningen av många ekosystem. Ett slående exempel är hur artrika europeiska kulturlandskap till stor del formats av betande djur.

Begreppet biologisk mångfald förknippas oftast med artrikedom, medan vi lätt glömmer bort den fenotypiska och genetiska variation som finns inom och mellan olika populationer av en och samma art. Genetisk variation är avgörande för en populations förmåga att förändras till följd av naturlig selektion. Inom evolutionsbiologin vill man förstå vilka faktorer som formar och upprätthåller genetisk diversitet inom och mellan arter, och då behöver man undersöka genom vilka funktionella och adaptiva mekanismer arterna påverkar varandra.

En ökad förståelse för hur herbivorer driver evolutionen av genetiska skillnader mellan populationer är viktigt inom evolutionsbiologin, men har också en praktisk betydelse som kanske är större nu än någonsin. För att förstå konsekvenserna av de accelererande klimatförändringar vi står inför behöver vi kunna förutsäga hur växt- och insektspopulationer reagerar på en förändrad miljö och hur detta i sin tur påverkar funktionen hos olika ekosystem. Dessutom är kunskapen om hur herbivorer påverkar växters funktion och populationsdynamik viktig inom jordbruket, exempelvis för att kunna ta fram grödor med ett bra försvar mot insektsangrepp eller för att utveckla nya metoder för biologisk kontroll av skadeinsekter.

**Resistens och tolerans mot herbivori varierade med latitud (I)**

I Sverige minskar växtsäsongens längd från söder till norr och det gör även antalet arter av insekter som äter på fackelblomster. Tidigare studier har visat att nordliga fackelblomsterpopulationer börjar växa tidigare på våren och slutar växa tidigare på hösten än vad sydliga populationer gör.

Mot bakgrund av detta ville jag undersöka om även resistens och tolerans mot herbivori varierar med latitud hos fackelblomster, och om dessa egenskaper är positivt relaterade till betestrycket i naturliga populationer. Jag odlade olika populationer i en gemensam miljö, en så kallad ”common garden”, och kvantifierade resistens genom att mäta hur mycket skador från insektsbete de olika populationerna fick. Toleransen kvantifierade jag genom att mäta skillnader i bladskador och storlek hos växter som utsätts för herbivori och växter där vi reducerat herbivorin med hjälp av en insekticid. För att få ytterligare resistensmått utförde jag även ett växthusförsök där honor av två arter av bladbaggar (*Galerucella pusilla* och *G. calmariensis*) som är specialiserade på fackelblomster fick välja vilka populationer de ville äta och lägga ägg på. De populationer som föredrogs av äggläggande honor och som utsattes för mest omfattande skador definierade vi som minst resistenta.

Resultaten visar att fackelblomsterpopulationer med sydligt ursprung var mer resistenta mot herbivori än populationer med nordligt ursprung, medan toleransen däremot ökade från söder till norr.

Jag genomförde även en fältstudie där jag dokumenterade skador från insektsbete i fält. Resultatet visar att skadorna tenderade att minska från söder till norr. Resistensen i både common-garden experimentet och växthusexperimentet var positivt relaterad till betestrycket i de naturliga populationerna. Detta kan tolkas som att de nordliga populationerna utsätts för mindre insektsbete och därför inte utvecklat lika hög resistens mot herbivori som de sydliga populationerna med ett högt betestryck. Den ökande toleransen norrut kan bero på att växterna från norra Sverige har hunnit komma längre i sin utveckling när insektarna kommer fram på våren och därför klarar angreppen bättre.

**Effekter av herbivori på växters framgång längs en latitudinell gradient (II)**

Herbivorer kan negativt påverka angripna växter genom att reducera deras tillväxt, överlevnad och reproduktiva förmåga, och på så vis kan de även påverka växters populationsdynamik och evolution. För att ta reda på hur
insektsbete påverkade växternas överlevnad och reproduktion utförde jag ett experiment där jag märkte upp plantor i tre naturliga fackelblomsterpopula-
tioner och uteslöt herbivori på hälften av dem genom att bespruta dem med 
en insekticid. De tre populationerna var belägna längs en latitudinell gradient 
från den mellersta till den nordligaste delen av en svenska ostkusten. Jag 
dokumenterade överlevnad, tillväxt och reproduktiv framgång hos växterna 
och kvantifierade effekterna av herbivori under två säsonger.
Både betestryck och effekter av herbivori på plantöhöjd, blommingsförmå-
ga och fröproduktion varierade mellan de tre populationerna och var krafti-
gast i den sydliga populationen. När vi uteslöt herbivorer påverkades den 
relativa fröproduktionen i de tre populationerna. I kontrollbehandlingen var 
fröproduktionen per planta 8.6 gånger högre i den nordliga populationen 
jämfört med den sydliga. När vi uteslöt herbivorer såg vi ett omvänt mönster 
där den sydliga populationen producerade 2.5 gånger fler frön per planta än 
den nordliga. Bladskadorna ökade med värdeväxtens storlek men toleransen 
mot herbivori varierade inte med växtens storlek.
Resultaten visar att herbivorer starkt kan påverka demografisk struktur 
hos fackelblomsterpopulationer och därigenom föra geografiska mönster i 
fröproduktion. Resultaten tyder också på att herbivorernas selektionstryck 
varierar mellan populationer och minskar norrut.

Inga lokala anpassningar men effekter av herbivori på populationers 
relativa framgång (III)
Växter som är lokalt anpassade klarar sig bättre i sin hemmiljö än vad växter 
av samma art men med ett annat ursprung gör. Detta brukar man testa genom 
att göra en så kallad reciprok transplantering. Då planterar man växter med 
olika ursprung och undersöker om populationer som planterats i sin hem-
mamiljö klarar sig bättre än populationer som flyttats dit från andra miljöer. 
Att populationer längs miljögradienter är lokal anpassade är relativt vanligt, 
men huruvida växtätande insekter bidrar till lokala anpassningar har sällan 
studerats.
I denna studie gjorde jag en reciprok transplantering mellan tre lokaler be-
lägna längs en latitudinell gradient på den svenska ostkusten. För att ta reda 
på herbivorins roll i hur de olika populationerna klarar sig lade jag till en 
experimentell behandling där jag med hjälp av en insekticid uteslöt insekter-
na på hälften av plantorna på varje lokal. Jag undersökte om de tre popula-
tionerna var lokalt anpassade till sina respektive miljöer, om skillnader i 
resistens och tolerans mellan populationer var lika på alla tre lokaler, samt 
om herbivori påverkade relativ framgång hos de tre populationerna.
Resultaten visade inga tecken på lokala anpassningar. Istället var den syd-
liga populationen mest framgångsrik på alla tre lokalerna och hade även 
mindre skador från herbivorer än de andra populationerna. När vi uteslöt 
växtätande insekter växte plantorna bättre och producerade mer frukter på de
två sydligaste lokalerna, men inte på den norra lokalen där herbivorin var väldigt begränsad.

Uteslutningen av herbivorer påverkade populationernas relativa framgång på den sydligaste lokalen där betestrycket var högst. Men den relativa rangningen i skademått mellan populationerna ändrades inte då vi tog bort herbivorer, vilket tyder på att herbivorin påverkar selektionens styrka men inte dess riktning.

Man kan tänka sig att genetisk drift, sentida uppvärmning av klimatet, och oregelbundet förekommande selektion mot sydliga genotyper på nordliga breddgrader är faktorer som bidrar till att de sydliga populationerna klarar sig bäst både i norr och i söder. De konsistenta skillnaderna mellan populationer i resistens mot insektsbete skulle kunna bero på att herbivorerna på de olika lokalerna har likartade beteenden och födopreferenser.

Herbivorer har stark påverkan på mellanpopulationsvariation i reproduktiv framgång längs en störningsgradient (IV)

I denna studie undersökte jag hur mycket variation det fanns mellan populationer i betestryck och dess påverkan på växternas reproduktiva förmåga längs en störningsgradient i en skärgård i norra Sverige. Detta gjorde jag genom att analysera data från ett experiment som utfördes av mina medförfattare under somrarna 1997-1998.

Nio populationer på olika öar från inre till yttre skärgård valdes ut. I den yttre skärgården utsätts öarna för kraftiga störningar i samband med höst- och vinterstormar då de ofta blir helt överspolade av vågor eller skrapade av is, medan de inre öarna i stor utsträckning är skyddade från sådana störningar. Ett antal fackelblomsterplantor på varje ö märktes upp, varav en del besprutades med en insekticid några gånger under försommaren. På sensommaren dokumenterades skador från insektsbete, blomning och fröproduktion. För att ta reda på hur mycket insektsbetet varierade från år till år dokumenterades bladskador hos obesprutade kontrollplantor även året efter.

Resultaten visar att både betestrycket och växternas reproduktiva förmåga varierade mycket mellan populationer, och att populationerna närmast land utsattes för mest herbivori. När herbivorerna uteslöts såg vi att skillnaderna i skador från insektsbete till stor del förklarade skillnaderna i blomning och fröproduktion.

För att ta reda på om det fanns genetisk variation i resistens mellan populationer gjordes även ett common-garden experiment med plantor från sex populationer i skärgården. Resultaten visar att skillnaderna mellan populationer i resistens var ytterst begränsade.

Sammantaget visar resultaten att hur mycket herbivori de olika fackelblomsterpopulationerna utsatts för till stor del avgör hur många plantor som blommar och hur många från de producerar, men att dessa skillnader inte verkar ha resulterat i skillnader i resistens mot herbivori på denna rumsliga skala. Dessutom tyder resultaten på att fysisk störning kan påverka framgång
och populationsdynamik hos perenna växter inte bara på grund av dess effekt på mellanartskonkurrens utan i stor utsträckning även på grund av effekter på interaktioner med specialiserade herbivorer.

**Slutsatser**
I den här avhandlingen har jag undersökt de ekologiska och evolutionära konsekvenserna av interaktioner mellan den perenna örtens fackelblomster och dess specialiserade herbivorer. Jag har studerat hur betesträck och resistens och tolerans mot herbivori varierar längs miljögradienter på olika geografiska skalor.

Dessa studier har visat att herbivori i stor utsträckning avgör blomningsförmåga och fröproduktion hos fackelblomster och att betesträck och resistens och tolerans mot herbivori varierar både med breddgrad över en vid geografisk skala och med störning på en lokal skala. De har även visat att både resistens och tolerans mot herbivori varierar med latitud men i olika riktning, samt att mellanpopulationsvariation i resistens mot herbivori längs en latitudinell gradient motsvarades av skillnad i hur kraftig herbivori populationerna utsattes för. Dessutom har studierna visat att relativ framgång hos tre populationer längs samma latitudinella gradient påverkades av herbivori men att dessa populationer inte var lokalt anpassade. Till sist har de visat att insektsbete ökade med värdväxtens storlek, medan toleransen mot herbivori inte visade tecken på att variera med växtens storlek.

Sammanfattningsvis visar resultaten att styrkan på den selektion som herbivorer utsätter fackelblomster för varierar mellan populationer. Resultaten tyder även på att både fysisk störning och latitudinell variation i miljöfaktorer kan ha stor inverkan på reproduktion och populationsdynamik hos perenna växter på grund av effekter på interaktioner med specialiserade herbivorer.
Acknowledgements

Först och främst vill jag tack min handledare Jon Ågren för inspiration, idéer, konstruktiv kritik och ovärderlig textredigering och min biträdande handledare Peter Hambäck för diskussioner, idéer samt hjälp i fält och med statistik. Jag vill också tacka Lars Ericson för gott medförfattarskap och experträd gällande transplantering av fackelblomster.


Jag är tacksam för ekonomiskt bidrag från Svenska Växtgeografiska Sällskapet, Stiftelsen Extensus, Helge Ax:son Johnsons stiftelse, Lars Hiertas minnesfond, Tullbergs fond för biologisk forskning samt Uddeholms, Bjurzons och Regnells resestipendier.

Tack Ulf Gärdebo för att jag fick hålla till på din mark och för hjälp med transport av plantor. Tack Stefan Björklund för tillverkning av burar.


Tack alla doktorandkollegor och nuvarande och före detta avdelningskollegor för fikor, diskussioner, seminarier, fester, filmkvällar, Teneriffa-ekursion och mycket mer. Jag kommer att sakna er!

Ett speciellt tack till Camille Madec och Maria Usecka-Perzanowska för kontors-sällskap, vänuskap och stöd genom åren. Ett extra-spezialt tack till Camille för att du alltid tog dig tid för till mina tusen statistikfrågor!

Tack Markus och trollen för att ni hjälper mig att komma ihåg vad som är viktigt, och för all praktisk hjälp och moraliskt stöd. Utan er hade inget funkat, ni är bäst i världen!
References


The Swedish Meteorological and Hydrological Institute. 2014.


Acta Universitatis Upsaliensis

Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology 1238

Editor: The Dean of the Faculty of Science and Technology

A doctoral dissertation from the Faculty of Science and Technology, Uppsala University, is usually a summary of a number of papers. A few copies of the complete dissertation are kept at major Swedish research libraries, while the summary alone is distributed internationally through the series Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology. (Prior to January, 2005, the series was published under the title “Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology”.)

Distribution: publications.uu.se
urn:nbn:se:uu:diva-247088