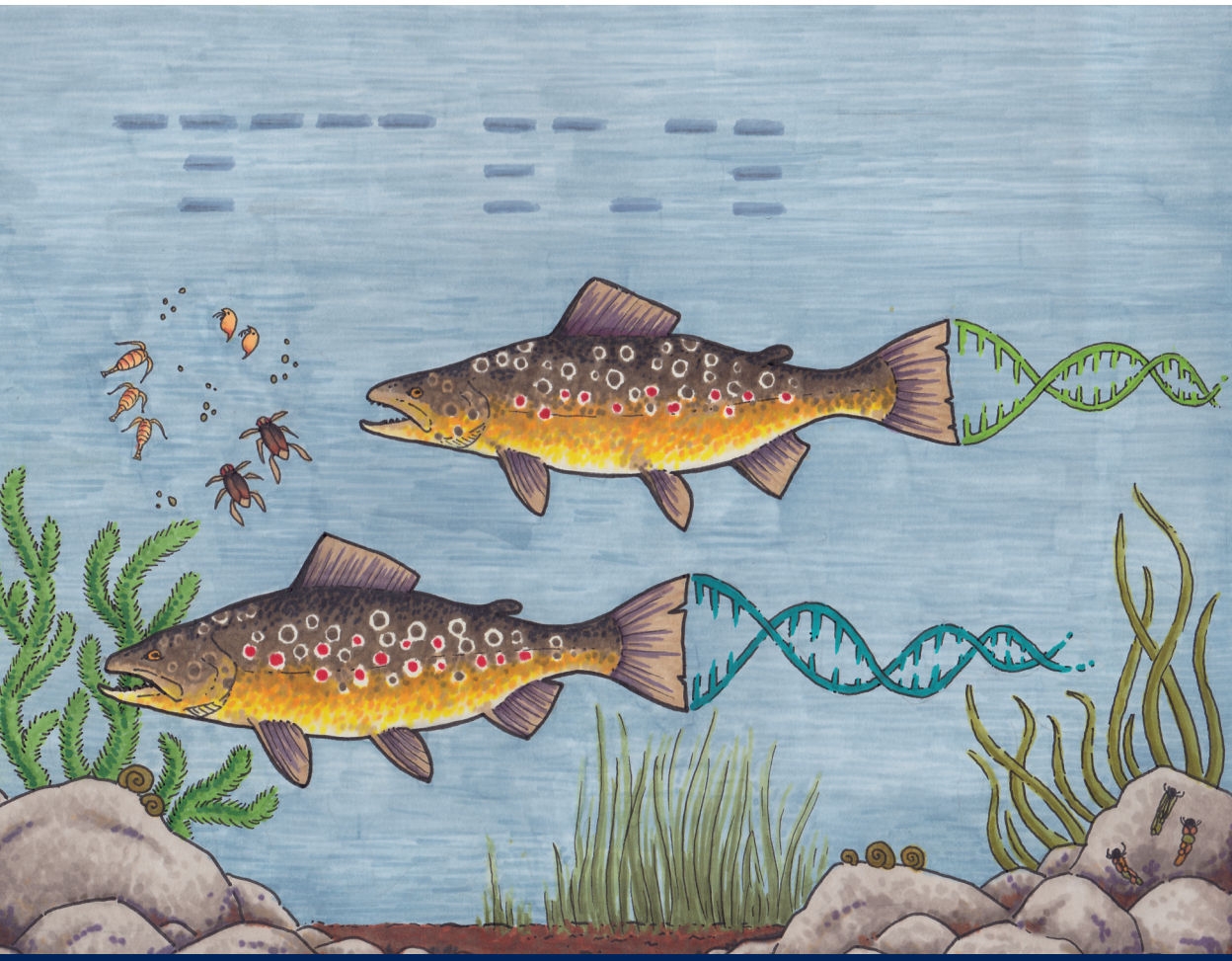
Stockholm University

Hidden biodiversity in an alpine freshwater top predator

Existence, characteristics, and temporal dynamics of cryptic, sympatric brown trout populations

Anastasia Andersson



Doctoral Thesis in Population Genetics at Stockholm University, Sweden 2021

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Anastasia Andersson

Academic dissertation for the Degree of Doctor of Philosophy in Population Genetics at Stockholm University to be publicly defended on Friday 24 September 2021 at 13.00 online via Zoom, public link is available at the department web site.

Abstract

Intraspecific genetic diversity is imperative to the survival of species in a changing environment, and it plays a vital role in ecosystem function. Since this type of diversity can be difficult to detect it is sometimes referred to as "hidden biodiversity". When separate and genetically distinct populations of the same species coexist within the same habitat, without apparent barriers to migration and obvious phenotypic divergence, this form of hidden biodiversity is called cryptic sympatry. Knowledge of cryptic sympatry is limited, however, and the aim of this thesis is to increase our understanding of this phenomenon by focusing on a species group where several cases of sympatry have been documented – the salmonids.

Using the brown trout (*Salmo trutta*) as a model, I characterized two previously reported cases of cryptic sympatry occurring in small Swedish alpine lakes with respect to both phenotypic and genetic characteristics. I explored the hypothesis that cryptic sympatry is more common than currently recognized by reviewing literature documenting sympatry, as well as by assessing the statistical power to detect sympatric populations with varying degrees of divergence using commonly applied sample sizes for loci and individuals. Further, I performed a large-scale search for sympatric populations in alpine lakes in central Sweden.

I found that cryptic, sympatric populations can coexist while apparently utilizing the same food resources and exhibiting the same adaptive plasticity to their shared environment (**Paper I**). In one of the empirical cases there were indications that the populations used different creeks for spawning, suggesting that segregation in spawning location contributes to the maintenance of sympatry (**Paper II**). Further, I found that differences between cryptic, sympatric populations of the same lake may be large with respect to levels of genetic diversity, inbreeding, and connectivity with populations in nearby lakes (**Papers II and III**).

I found support for the hypothesis that cryptic sympatry is more common than generally acknowledged (**Papers IV and V**). In the literature, cryptic sympatry is rarely reported and typically associated with higher divergence levels than between sympatric populations that differ phenotypically. My results suggest that this to a large extent may be due to limited statistical power when commonly used sample sizes in terms of individuals and loci are applied and the amount of divergence between populations is small (**Paper IV**). Cryptic sympatry was observed in over 40% of the screened localities (27 lakes), and was shown to be temporally stable over at least 40 years (**Paper V**).

Keywords: *cryptic sympatry, population genetic structure, sympatric populations, intraspecific biodiversity, genetic monitoring, conservation genetics, trophic polymorphism, genetic connectivity, temporal stability, Salmo trutta.*

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Stockholm University

Department of Zoology

Stockholm University, 106 91 Stockholm

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"There is grandeur in this view of life, with its several powers, having been originally breathed into a few forms or into one; and that, whilst this planet has gone cycling on according to the fixed law of gravity, from so simple a beginning endless forms most beautiful and most wonderful have been, and are being, evolved."

— Charles Darwin, The Origin of Species

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

I **Andersson, A.**, Johansson, F., Sundbom, M., Ryman, N., & Laikre, L. (2017). Lack of trophic polymorphism despite substantial genetic differentiation in sympatric brown trout (*Salmo trutta*) populations. Ecology of Freshwater Fish, 26, 643-652. doi:10.1111/eff.12308

II **Andersson, A.**, Jansson, E., Wennerström, L., Chiriboga, F., Arnyasi, M., Kent, M. P., Ryman, N., & Laikre, L. (2017). Complex genetic diversity patterns of cryptic, sympatric brown trout (*Salmo trutta*) populations in tiny mountain lakes. Conservation Genetics, 18, 1213-1227. doi:10.1007/s10592-017-0972-4

III Saha, A., **Andersson, A.**, Kurland, S., Keehnen, N. L. P., Kutschera, V. E., Hössjer, O., Ekman, D., Karlsson, S., Kardos, M., Ståhl, G., Allendorf, F. W., Ryman, N., & Laikre, L. Whole-genome resequencing confirms reproductive isolation between sympatric demes of brown trout (*Salmo trutta*) detected with allozymes. Manuscript.

IV Jorde, P. E., **Andersson, A.**, Ryman, N., & Laikre L. (2018). Are we underestimating the occurrence of sympatric populations? Molecular Ecology, 27, 4011-4025. doi:10.1111/mec.14846

V **Andersson, A.**, Karlsson, S., Ryman, N., & Laikre, L. Mapping and monitoring genetic diversity in brown trout population systems in alpine lakes by applying newly proposed indicators. Manuscript.

Candidate contributions to thesis articles*

	I	II	III	IV	V
Conceived the study	Substantial	Significant	Significant	Significant	Substantial
Designed the study	Substantial	Substantial	Minor	Significant	Substantial
Collected the data	Substantial	Substantial	Significant	Substantial	Significant
Analysed the data	Substantial	Substantial	Significant	Substantial	Substantial
Manuscript preparation	Substantial	Substantial	Significant	Significant	Substantial

*** Contribution Explanation**

Minor: contributed in some way, but contribution was limited.
Significant: provided a significant contribution to the work.
Substantial: took the lead role and performed the majority of the work.

I am also a co-author of the following articles that were written during the course of my doctoral studies, but are not included in this thesis:

Kurland S., Wheat C. W., de la Paz Celorio Mancera M., Kutschera V. E., Hill J., **Andersson A.**, Rubin C-J., Andersson L., Ryman N., & Laikre L. (2019). Exploring a Pool-seq-only approach for gaining population genomic insights in nonmodel species. Ecology and Evolution 9, 11448-11463. doi:10.1002/ece3.5646

Andersson A., Laikre L., & Bergvall, U. A. (2014). Two shades of boldness: novel object and anti-predator behavior reflect different personality dimensions in domestic rabbits. Journal of Ethology 32, 123-136. doi:10.1007/s10164-014-0401-9

Contents

Introduction

Intraspecific genetic diversity has long been recognized as imperative to the adaptation and survival of species in changing environments (e.g. McGinnity et al. 2009; Allendorf 2013; Bitter et al. 2019; Lai et al. 2019). Such biodiversity also plays a vital role in ecosystem function and has been shown to affect ecosystem stability, resilience, and services in the same manner as species diversity does (Crutsinger et al. 2008; Cook-Patton et al. 2011; Yang et al. 2015; Des Roches et al. 2018; 2021). Therefore, intraspecific genetic diversity is of particular importance in species-poor environments, such as the boreal forest biome (Pastor 1996; Jetz and Fine 2012) and its aquatic systems (Laikre et al. 2008; Johannesson et al. 2011), because climate change is expected to severely affect freshwater habitats in the northern hemisphere (Chu et al. 2005; Prowse et al. 2006; Ficke et al. 2007). For these reasons, the identification, monitoring, and safeguarding of intraspecific genetic diversity is of utmost importance. This is recognized in international policy, including the United Nations Convention on Biological Diversity (CBD; www.cbd.int). Intraspecific diversity is sometimes referred to as "hidden biodiversity" (Des Roches et al. 2021) and this thesis focuses on a particular type of hidden biodiversity: genetically divergent populations that coexist in sympatry over restricted geographic areas without apparent barriers to migration, and without obvious phenotypic divergence (so-called cryptic sympatry).

Sympatric populations

Genetic diversity within species refers to differences within and between populations, with genetic structuring occurring at both larger and smaller spatial scales (Allendorf et al. 2013). Genetically distinct populations that coexist in the same area, at least during part of their lifecycle, are referred to as sympatric (Futuyama and Mayer 1980; Mallet et al. 2009). Such populations are of interest when studying microevolutionary processes and ecological interaction, as their existence may represent the very first steps of sympatric speciation (Maynard Smith, 1966; Via, 2001). They can reflect genetic adaptations to ecological niches and reveal reproductive isolation that occurs without detectible migration barriers (Kawecki, 1996, 1997; Turelli et al. 2001).

Sympatric populations have been documented in a variety of taxa, from insects to large mammals, in both terrestrial and aquatic ecosystems (Attard et al 2016; Guo et al., 2018; Knutsen et al., 2018; Orlov et al., 2012; Ravinet et al., 2016; Schönswetter et al., 2007; Verspoor et al. 2018; Kristensen et al. 2021). Sympatric populations often differ in appearance from one another,

and such differences are important factors in the detection of sympatry. When casual inspection does not reveal any clear differences in phenotype, sympatric populations can be described as cryptic (Bickford et al. 2007). Detecting such populations typically requires genetic data.

Regardless of whether populations are cryptic or not, it can be problematic to distinguish between sympatric populations and closely related sympatric sister species, since researchers who follow a strict interpretation of the biological species concept may classify reproductively isolated sympatric populations as separate species. Thus, it is likely that there are differences in the reports of cryptic sympatry between different research fields, taxa, and ecosystems and whether this type of diversity is considered to be below the species level (Bickford et al., 2007; Struck et al., 2018). Another issue is the definition of sympatry – at what spatial and temporal scale can populations be regarded as sympatric? Separate populations can coexist within the same area during relatively brief periods of time, such as during seasonal feeding migrations (Wood and Foote 1996; Hauser et al. 2014), or over much longer periods, such as the majority of the lifespan if individuals (Walker et al. 1988; Adams et al. 2008).

Sympatry in salmonid fishes

Salmonids are well-studied with respect to genetics, with frequent reports of populations exhibiting substantial levels of genetic divergence. This has been shown over large geographic distances (e.g. Ryman 1983; Crozier and Ferguson 1986; Bekkevold et al. 2019), as well as within water systems, and has even been demonstrated in geographically restricted areas, like the same body of water (i.e. sympatric populations; e.g. Prodöhl et al. 1992; Lu and Bernatchez 1999 Wilson et al. 2004; Gowell et al. 2012; Harris et al. 2015; Jacobs et al. 2018). The mechanism driving this structuring is thought to be the salmonids' homing behavior, which involves migration from feeding grounds to natal locations for spawning (Jonsson and Jonsson 2011; Ferguson et al. 2019). However, temporal segregation in spawning has also been shown to result in genetically divergent populations (Child 1984; Nielsen and Fountain 1999; Hendry et al. 2002; Fillatre et al. 2003; Schultz et al. 2006).

Sympatric salmonid populations may vary with respect to dietary niche use (Sandlund et al. 1992; Hirsch et al. 2013; Piggott et al. 2018), color (Sendek 2004; Lehnert et al. 2016), and morphology (Harris et al. 2015; Osinov et al. 2015), with some recent works coupling phenotypic divergence to genetics (Guðbrandsson et al. 2018; Jacobs et al. 2018).

Study species

The brown trout (*Salmo trutta;* Figure 1) is a member of the Salmonidae family, together with chars, Atlantic and Pacific salmon, whitefishes, and graylings. It occurs naturally throughout Europe and northern Africa, extending its distribution east towards the Ural Mountains and the northern parts of the Middle East, and it has been introduced to other countries in Africa, Asia, and South America, as well as to Australia (Elliott 1994; Jonsson and Jonsson 2011). The brown trout is an important species for many reasons. From a socio-economic perspective, it is a popular catch among sport fishermen and commercial fisheries alike (Laikre 1999; Marco-Rius et al. 2013). It also has a significant ecological role, especially in the mountains of Scandinavia, where it is often one of only a few fish species, or even the only species present (Laikre 1999; L'Abee-Lund et al. 2002; Frank et al. 2011; Jensen et al. 2012).

Genetic variation among populations accounts for the majority of biodiversity within this species (Ryman 1983), and it is often coupled to phenotypic differences, where populations can vary significantly in size, coloration, body shape, and diet. There are accounts of ecological niche separation occurring among sympatric brown trout populations (Ferguson and Mason 1981; Ferguson and Taggart 1991; Prodöhl et al. 1992; Duguid et al. 2006), as well as between brown trout and other fish species (Saksgård and Hesthagen 2004; Brodersen et al. 2012; Sanchez-Hernandez and Amundsen 2015).

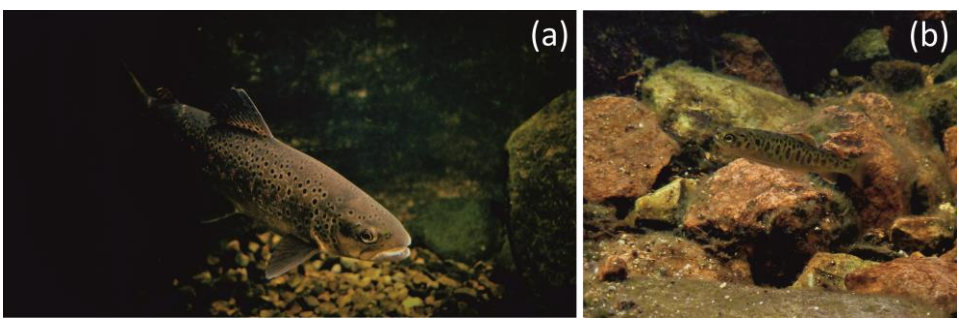

Figure 1 Brown trout (*Salmo trutta*). Panel (a) shows an adult and panel (b) a juvenile. Both pictures by Anastasia Andersson.

The species also shows a great variation in life histories, with several resident and migratory forms, as outlined in Ferguson et al. (2019). In short, the resident forms remain in lakes or rivers for both feeding and spawning, while the migratory forms move between their feeding grounds in lakes, rivers, and seas, and spawning locations in rivers and streams. Brown trout are iteroparous, which means they can reproduce several times during their lifetime.

Sexual maturity is reached at the age of about two to three years, with males maturing slightly earlier than females (Jonsson and Jonsson 2011).

Empirical case studies in this thesis

Two cases of cryptic sympatry have previously been described in Sweden and the present thesis focuses on expanding the knowledge of these two cases (Figure 2; **Papers I-III**). The first case of sympatry ever reported in brown trout was detected in Lakes Bunnersjöarna, which are two very small, interconnected lakes. Here, contrasting homozygosity at one allozyme locus (a lactate dehydrogenase coding locus, denoted *LDH-1*) indicated the existence of two sympatric populations (denoted deme I and II; Allendorf et al. 1976; Ryman et al.1979). Allele frequency differences in five additional allozyme loci supported this structuring and suggested that deme I was more genetically diverse than deme II. Stomach content analysis did not reveal any dietary divergence, and although a body size difference between the two demes was observed, this was not enough to assign individual fish to a population with any degree of acceptable accuracy (Ryman et al. 1979).

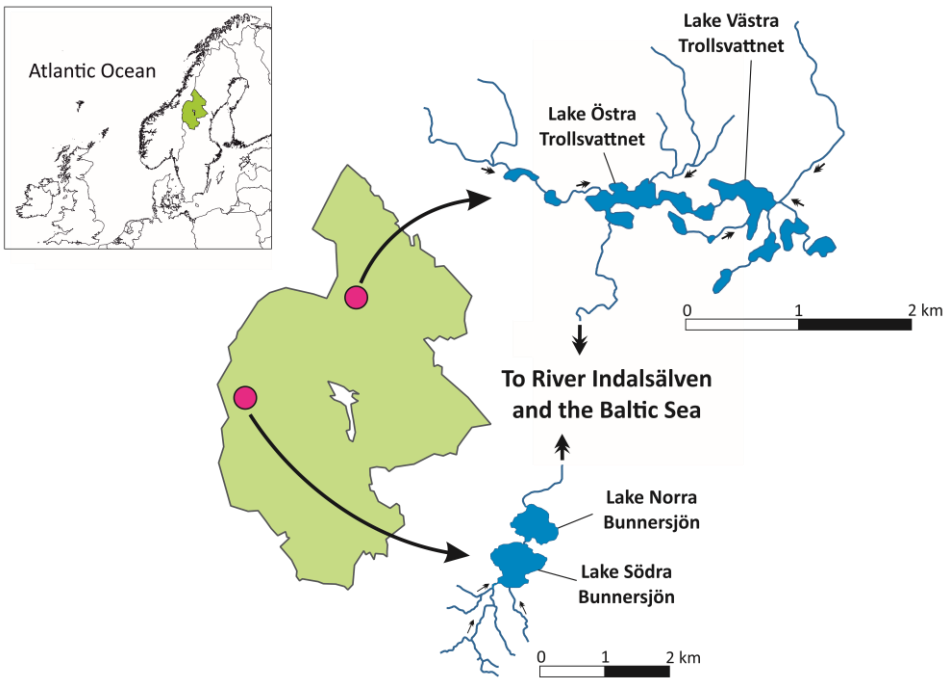

Figure 2 The location of the two twin Bunnersjöarna Lakes and the Trollsvattnen Lakes, where cryptic sympatric populations have previously been detected (Ryman et al. 1979; Palmé et al. 2013). Both localities are in the uppermost parts of their respective water systems, both of which drain into the Indalsälven River.

Lakes Trollsvattnen are also made up of two very small closely connected lakes, located north of the Bunnersjöarna Lakes. Population structure in

these lakes was discovered after several years of monitoring, initially through the deficiency of heterozygotes in several allozyme loci (Jorde and Ryman 1996) and was later confirmed by using the STRUCTURE software (Palmé 2013). After nearly two decades of monitoring, it was revealed that two populations (denoted A and B) occur at similar frequencies in both lakes, and that they have remained in stable sympatry for at least twenty years (Palmé et al. 2013). Like Lakes Bunnersjöarna, these populations do not have any apparent morphological differences. While they show substantial levels of differentiation with allozymes (F_{ST}≈0.1), only very minor body-size differences have been detected, in addition to differences in the proportion of sexually mature individuals, where population A is slightly larger and has a lower frequency of mature fish than population B (Palmé et al. 2013).

Objectives

The overall aim of this thesis is to expand the knowledge of cryptic sympatry, both genetically and phenotypically, with a particular focus on salmonids, by detailed characterization of two previously reported empirical cases of cryptic sympatric brown trout populations (Lakes Trollsvattnen and Lakes Bunnersjöarna, Jämtland County, central Sweden), as well as by exploring the hypothesis that cryptic sympatry in brown trout within restricted freshwater areas is more common than currently recognized.

Specifically, the objectives included:

- performing an extensive screening for trophic divergence between two sympatric populations, including diet, body size and shape, and gill raker metrics (**Paper I**),

- examining the genetic connectivity of sympatric populations to nearby lakes, potential differences in spawning characteristics, and confirming the genetic structures using SNP markers (**Papers I, II, and III**), and

- investigating the degree of genome-wide diversity and divergence of sympatric populations, while highlighting the evolutionary mechanisms governing this divergence (**Paper III**).

The objectives also included exploring the hypothesis that cryptic sympatry may be more common than generally appreciated, by:

- reviewing the accounts of sympatry in salmonid fishes in freshwater habitats to determine whether commonly used genetic markers might have resulted in under detection and under representation of cryptic sympatry in the literature, and assessing the statistical power of detecting cryptic sympatry when using genetic data (**Paper IV**), as well as

- conducting a screening for the potential existence and temporal genetic dynamics of sympatric populations in 27 small lakes located in the same geographic region as the two previously documented empirical cases in the alpine area of central Sweden (**Paper V**).

Methods

Sampling

Present day sampling was primarily performed as part of a long-term genetic monitoring research effort (The Lakes Bävervattnen Project; **Papers I, II, and V**). Muscle, eye, and liver tissue samples were collected for genotyping and stored on dry ice until they were placed in a tissue bank. When possible, length, weight, sex, breeding condition (mature to spawn or not; Figure 3), and age was recorded. This thesis also includes data collected prior to the research described here (**Papers I–III and V**). Allozyme genotypes, as well as ecological data, were obtained from a database, while tissue samples for genotyping with other markers were acquired from a frozen tissue bank, both maintained by the Department of Zoology at Stockholm University.

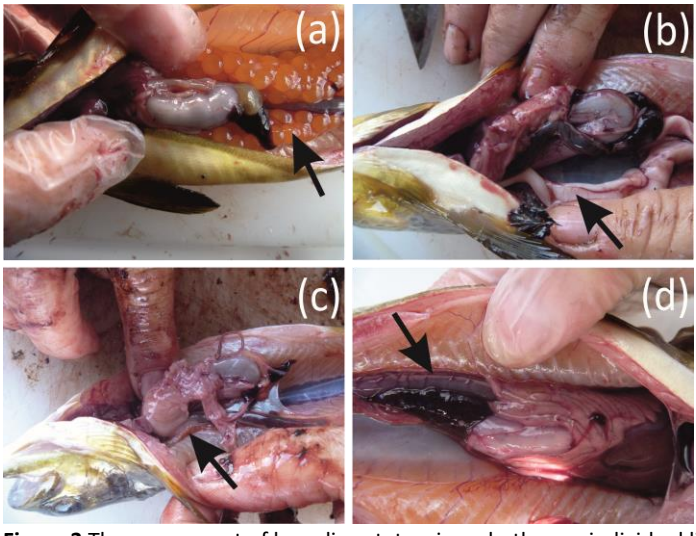

Figure 3 The assessment of breeding status, i.e. whether an individual likely would have spawned during the year of collection. The panels illustrate examples of a (a) mature female, (b) mature male, (c) immature female, and an (d) immature male. Black arrows indicate the gonads. All pictures by Anastasia Andersson.

Genotyping and sequencing

In this thesis, allozymes, a SNP array, and whole genome sequencing (WGS) were used. **Papers I and II** are based on allozyme markers, **Paper III** includes SNPs and WGS, which incorporates both pool sequencing and individual sequencing, while **Paper V** relies on SNPs only.

Allozyme genotyping

The genetic and phenotypic data used in **Papers I and II** were collected within The Lakes Bävervattnen Project which started in the 1970s. At that time, allozymes were the only genetic markers available for large-scale screenings. To be able to utilize already-existing genetic data representing several decades of annual sampling, allozymes were used in the first two papers. The fish were genotyped at a total of 14 polymorphic allozyme loci with co-dominant gene expression by horizontal starch gel electrophoresis (Allendorf et al.1977) at the Department of Zoology, Stockholm University.

DNA extraction, quality, and quantity control

Genomic DNA was extracted from approximately 50 mg of muscle tissue using the DNeasy Blood & Tissue Kit from Qiagen (Holden, Germany) for **Papers II and V** and the KingFisher Cell and Tissue DNA Kit from ThermoScientific (MA, USA) for **Paper III**, according to the manufacturer's instructions. DNA used for whole-genome resequencing received an additional RNase A treatment (**Paper III**). Evaluation of DNA quality was performed by electrophoresing an aliquot through a 1% agarose gel and subjectively assessing the proportion of high-molecular-weight DNA relative to degraded DNA. Double-stranded DNA was quantified using a Qubit fluorometer (ThermoScientific, MA, USA).

SNP genotyping

SNP genotyping was carried out using two different platforms: the Illumina iSelect SNP array (Illumina, CA, USA) for **Paper II** and the EP1^{TM} 96.96 Dynamic array IFCs genotyping platform (Fluidigm, San Francisco, CA) for **Papers III and V**. The 3,093 SNP loci used in **Paper II** and the 96 SNPs used in **Papers III and V** were chosen from a 3,782 SNP panel identified in brown trout, which are evenly distributed across the 40 linkage groups (Bekkevold et al. 2020).

Whole genome sequencing (WGS)

For pool sequencing (Pool-seq), DNA from each fish (n=50) was pooled at equal concentrations to achieve a total of 3 µg for each population. For individual WGS, two fish per population were randomly chosen. Both Pool-seq and individual WGS samples were sent to the National Genomics Infrastructure (NGI) at the Science of Life Laboratory (SciLifeLab) in Stockholm, Sweden, for sequencing.

Exploration of genetic structure

Population genetic structure was primarily explored using the STRUCTURE software (Pritchard et al. 2000) as well as tests for deviation from Hardy-Weinberg expectations (**Papers I-V**). We also employed DAPC (Discriminant Analysis of Principal Components; Jombart et al. 2010) as a complement to explore genetic structuring (**Paper II**) as well as evaluate the statistical power to detect admixed populations based on genetic data (**Paper V**).

Moreover, we used phylogenetic trees to explore the genetic relationships between sympatric populations within their home lakes, as well as between these populations and others inhabiting different lakes, both within and outside of their respective water systems. The phylogenetic trees were based on allozyme, SNP, and pool-seq data, and were produced using the programs POPTREE2 (Tekezaki et al. 2010), MEGAX (Kumar et al. 2018), and TreeMix (v1.12; Pickrell and Pritchard 2012), with the latter used to simulate migration.

WGS data was used for estimating nucleotide diversity as divergence estimated using Nei's F_{ST} (Nei, 1973), with the program POPOOLATION v2 (Kofler et al., 2011). Inbreeding was measured as F_{ROH} and LnROH, which represent the fraction of the genome covered by "runs of homozygosity" (ROH) and their run length (LnROH; Gomez-Raya et al. 2015; Kardos et al. 2017; Magi et al., 2014; for a more detailed description of methods, see **Paper III**).

Phenotypic analyses

Body shape and gill raker morphology

Body shape was analyzed using the geometric morphometric method (Parsons et al. 2003; Rohlf and Marcus 1993), with the main steps outlined in Figure 4. In short, landmarks (i.e. points marking specific physical structures) were derived from photos, and shape components were derived from these landmarks. The shape components, which describe the general shape of the fish, were then used for statistical testing (detailed description in **Paper I**).

Gill raker metrics were assessed by raker count and raker length, as well as by measuring the distance between the five rakers closest to the elbow of the gill arch. These measurements were taken from the first gill arch on both the left and the right side of each fish, and the average was used in statistical analyses.

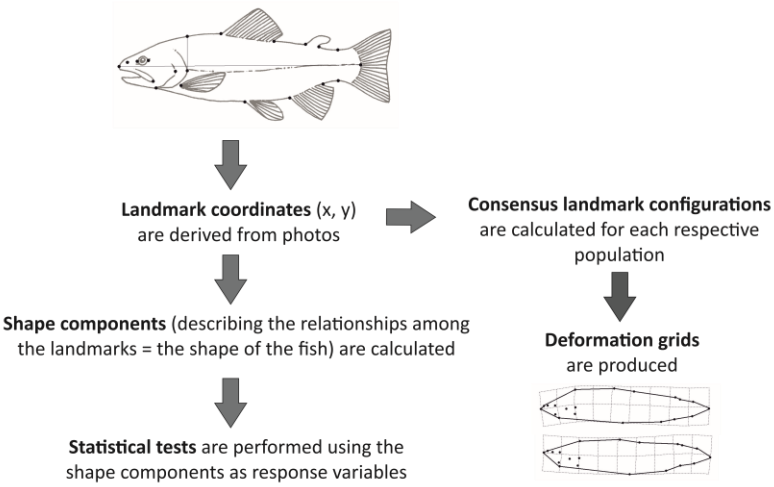

Figure 4 A flowchart of the major steps in the geometric morphometric approach used to investigate body shape differences. The diagram of the fish shows the actual placement of the landmarks in Paper I.

Dietary preference

Diet was examined by stomach content analysis and stable isotope analysis (**Paper I**). The stomach content analysis was carried out by dissecting stomachs and classifying the food items into eight broad categories: benthic prey, limnetic prey, surface insects, fish, mammal, plant material, non-food, and unidentified, using a stereo microscope (Figure 5). The proportion of each food category, ranging between 0–100%, was estimated by visual inspection.

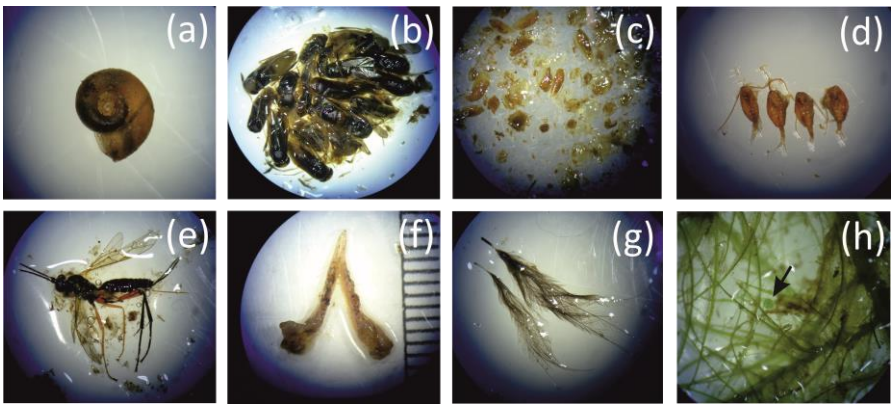

Figure 5. Food items from the stomachs of brown trout: (a) ramshorn snail, (b) *Corixidae spp.*, (c) *Eurycercus spp.*, (d) copepods, (e) *Apocrtia spp.*, (f) rodent jaw, (g) feathers, (h) plants, and a gelatinous sphere indicated by the black arrow. Panels (a-b) represent benthic prey, (c-d) limnetic prey, (e) surface insect, (f) mammal, (g) non-food, and (h) plant material and an unidentified object. All pictures by Anastasia Andersson.

Long-term feeding patterns were examined by measuring the stable isotope ratio of nitrogen (^{15}N to ^{14}N) and carbon (^{13}C to ^{12}C); a detailed description of these methods can be found in **Paper I**. The nitrogen isotope ratio is indicative of an individual's average trophic position, while the carbon isotope ratio provides information on primary carbon sources (Clarke et al. 2005).

Growth and breeding status

Differences in growth and breeding status between sympatric populations were examined by comparing body length and proportions of mature fish, respectively, from individuals that were between 2–8+ years of age (**Paper I**).

Literature review and computer simulations

The literature review of documented cases of sympatry in salmonids (**Paper IV**) was carried out using the Web of Science database. The keywords used when conducting the literature searches were "sympatric populations" in combination with the respective common name of five different salmonid species. Papers reporting occurrence of sympatric populations in freshwater habitats were included, while studies of sympatry in other environments, such as the sea, were excluded. Reported populations were classified as cryptic if they were detected based solely on genetic data, or non-cryptic, if the basis of detection was phenotypic differences. Furthermore, studies that reported cryptic population structure using the most common genetic markers (allozymes, microsatellites, and SNPs) were compared to those of non-cryptic populations, with respect to the number of loci, sample size, and among-population divergence measured as F_{ST}.

Computer simulations were used to evaluate the statistical power of detecting genetic structure from samples of individuals that lacked phenotypic cues indicating population membership (the methodology is described in detail in **Paper V**). In brief, datasets were first generated by random sampling from a simulated mix of two interconnected genetic populations with various levels of divergence, after which the power of detection was evaluated using Hardy-Weinberg tests (more specifically, the heterozygote deficiency test for detection of Wahlund effects; Raymond and Rousset 1995), STRUCTURE software (Pritchard et al. 2000), and DAPC (Jombart et al. 2010). Using these methods, I explored the effects of sample size, level of divergence between populations, and population representation in samples. An in-house computer program was used for the simulations (Jorde, unpublished).

Major Results and Discussion

Characterizing two empirical cases of cryptic sympatry

A lack of trophic polymorphism in the sympatric populations of Lakes Trollsvattnen

I found that the sympatric populations of the Trollsvattnen Lakes do not show any signs of trophic polymorphism. Instead, they demonstrate the same adaptive plasticity to the local food niches in the separate lakes, indicating that sympatric populations may coexist while apparently utilizing the same food resources. Only minor body-shape differences in the orientation of the head and tail were observed between the populations in one of the lakes (Lake Östra Trollsvattnet) (Figure 6; **Paper I**). When both lakes were pooled, no effect of population on body shape or gill raker morphology was found, and there were no long or short-term dietary differences.

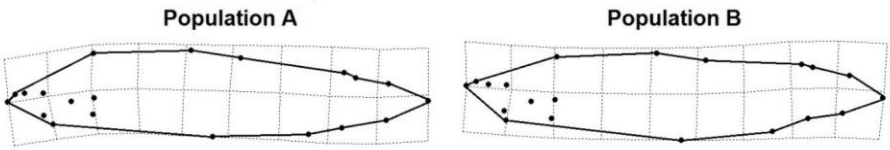

Figure 6 Deformation grids illustrating the shape differences between populations A and B in Lake Östra Trollsvattnet. The grids show the consensus shape of individuals assigned to the respective population based on genetic data, with grid deformations scaled up five times. The heads of the fish are to the left.

In contrast, body shape differed between lakes, with fish from Lake Östra Trollsvattnet being more shallow-bodied, with slightly more rakers in the upper part of the gill arch than fish from Lake Västra Trollsvattnet. These kinds of body shapes could suggest an adaptation to differential foraging strategies – slender bodies, upward-oriented heads, and dense gill rakers are often associated with limnetic feeders, while deep bodies, downward-oriented heads, and sparse gill rakers are linked to benthic foraging (Schluter, 1996). However, results from the stomach content analysis did not support this expectation: fish from Lake Östra Trollsvattnet fed primarily on benthic organisms (Figure 5, a-b), whereas individuals from Lake Västra Trollsvattnet preferred limnetic prey (Figure 5, c-d). This dietary divergence was also observed in long-term dietary preferences, where there was an effect of lake on both nitrogen and carbon isotopic signatures (Table S4 in **Paper I**). A divergence in carbon isotopic signatures ($p=0.008$) was observed, suggesting that the primary source of carbon in the diet is slightly different in the two lakes. There was also a small difference in nitrogen isotopic signatures which would be indicative of differential trophic positions, though it was not statistically significant ($p=0.07$; these results are not included in **Paper I**).

Comparison of body size and breeding status revealed that fish belonging to population A are slightly larger than fish belonging to B in all age classes and that population B had a higher proportion of mature individuals at the time of collection (0.51 vs. 0.33 for B and A, respectively), which confirms the previous findings of Palmé and colleagues (2013). There are indications that this may be due to fish from population B maturing at an earlier age than fish from population A; the average age of mature individuals is slightly lower for B which is also corroborated by mathematical modelling (Sjöström 2019).

Genetic relationships between sympatric populations and nearby lakes

The levels of divergence between sympatric populations and populations in nearby lakes vary greatly from case to case, and connectivity patterns can differ substantially, illustrated by the examples of the Trollsvattnen and Bunnersjöarna Lakes.

In Lakes Trollsvattnen, the two populations appear to coexist only within the lakes where they were originally discovered. Divergence between the sympatric populations (F_{ST}=0.09) was higher than the divergence between each respective population and the nearby lakes (F_{ST}=0.00–0.02 for population A and 0.03–0.08 for population B; **Paper II**). In addition, the populations exhibit disparate connectivity patterns to the populations in neighboring lakes. Population A shows high genetic similarities to nearby populations and thus appears to be a "common" form occurring throughout the water system, with some degree of differentiation among lakes. Population B, on the other hand, is the most unique group and does not seem to experience extensive genetic exchange with other populations in the system.

The two populations (demes) in Bunnersjöarna also show differential connectivity patterns to the closest lake (Lake Ånnsjön, located 8.5 km downstream; **Paper III**). Similar to the Trollsvattnen lakes, one of the demes (deme II) is genetically isolated while the other bears a closer resemblance to the fish inhabiting Lake Ånnsjön (F_{ST}=0.12 and 0.36 for demes I and II, respectively). The isolation of deme II is reflected in the nucleotide diversity, as well as inbreeding levels. Lake Ånnsjön has also been shown to harbor three sympatric populations (**Paper V**), and when this aspect was considered, pairwise F_{ST} values of 0.16, 0.08, and 0.10 were observed between the respective populations (Ånnsjön Population 1-3 in **Paper V**) and deme I. However, for deme II, the genetic dissimilarity to Lake Ånnsjön increased, with pairwise F_{ST} values estimated to 0.42, 0.42, and 0.47 for deme II versus each of the respective Lake Ånnsjön populations.

Spawning divergence of sympatric populations in Lakes Trollsvattnen

A spawning site segregation was revealed when the populations in the Trollsvattnen Lakes were compared to young-of-the-year parr collected from potential spawning sites in adjacent creeks. One of the populations (B) was found to be most genetically similar to parr from the northern inflow to Lake Västra Trollsvattnet, while the other (A) showed greater similarity to parr from the western inflow into Lake Västra Trollsvattnet, as well as parr from that lake's southern outflow. However, parr assigned to both populations were present at all sampled creek sites, which suggests that straying behavior may not be uncommon (as shown by Östergren et al. 2012). These findings contradict suggestions that spawning separation is often determined by preference for either inlet or outlet stream spawning (Ferguson et al. 2019).

On average, parr had slightly lower assignment probabilities, as well as lower levels of divergence, than adults. Divergence thus appears to increase over time, which may suggest the presence of diversifying selection. Though no allozyme loci appear to be under such selection pressure, the outlier analysis suggests that some of the SNPs may be.

Detection of cryptic sympatry using different genetic markers

In both empirical cases (the Trollsvattnen and Bunnersjöarna Lakes), cryptic sympatry has been detected using a few allozyme loci. To verify that these genetic structures are detectable with other markers, the possibility of identifying the populations using SNPs was explored (**Papers II and III**). My findings suggest that genetic structures representing cryptic sympatric populations are detectable using different genetic markers with relatively good consistency. This, in turn, indicates that such structuring likely reflects biologically relevant groupings and is not merely the result of using one particular marker system.

The STRUCTURE and DAPC analyses, which included a total of 60 fish from the Trollsvattnen Lakes genotyped at >3000 SNP loci, corresponded well between marker types; the two genetic clusters identified with SNPs mirrored the populations identified with allozymes to a great extent (Figure 7, a-b), with only a few fish having an inconsistent classification. When the 96 SNP panel was used, there was a slight increase in the number of fish that were classified to different populations with different markers, which may be due to the lower resolution of 96 loci compared to >3000 loci.

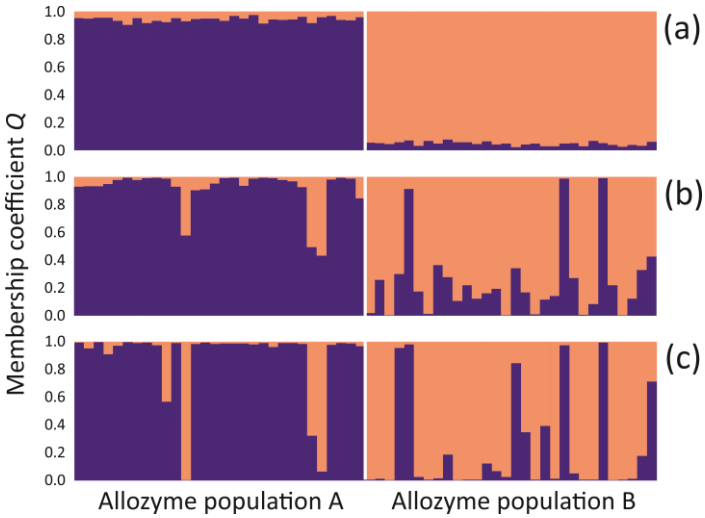

Figure 7 Comparison of the assignment of 60 individuals from Lakes Trollsvattnen representing the two sympatric populations A and B, which were scored for allozymes and SNPs. In all three cases, two populations were identified as the most likely number of upper-level structuring using STRUCTURE software. Panel (a) shows assignment based on 14 allozymes, panel (b) displays assignment based on >3000 SNPs, and panel (c) illustrates assignment based on the 96 SNP panel. Each fish is represented by one vertical bar.

For Lakes Bunnersjöarna, two genetic clusters were identified using the 96 SNPs. The structure analysis included a total of 140 fish, assigned to deme I (n=62) and deme II (n=78) based on their *LDH-1* genotype. The two populations reflected the groupings based on allozyme genotype almost perfectly; only one fish from allozyme deme I clustered together with fish from deme II (Figure 8).

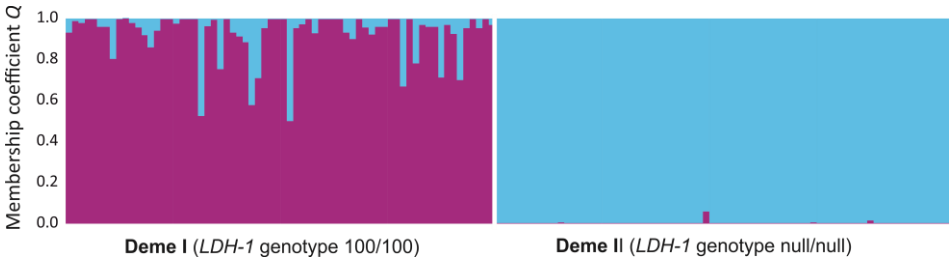

Figure 8 A barplot illustrating the assignment of individual fish belonging to the two demes of the Bunnersjöarna Lakes, with the 96 SNP panel, using STRUCTURE software. Each fish is represented by a vertical bar; colors indicate genetic clusters identified with SNPs.

Striking differences in genome-wide diversity between the sympatric populations of Lakes Bunnersjöarna

The sympatric populations in the Bunnersjöarna Lakes are strongly differentiated (F_{ST}=0.24 using SNPs; and 0.13 using Pool-seq data), as illustrated in Figure 9, and display very different levels of genetic diversity throughout the whole genome. Deme II exhibited considerably lower levels of genetic diversity than deme I (**Paper III**). Estimates of expected and observed heterozygosity (H_E and H_O), allelic diversity measured as allelic richness (A_R), the number of alleles per locus (N_A), and the proportion of polymorphic loci (P_L) obtained from SNP data (not reported in **Paper III**) are summarized in Table 1, with all comparisons between deme I and II being statistically significant (all p<0.001).

Table 1 Genetic diversity estimates for deme I and II of the Bunnersjöarna Lakes, based on 96 SNP loci. To test for differences between the demes, t-tests and chi-square tests were used. H_E=expected heterozygosity H_O=observed heterozygosity, A_R=allelic allelic richness, N_A=the number of alleles per locus, and P_L=the proportion of polymorphic loci.

Diversity measure	Deme I	Deme II	Test statistic (df)	P value
H_E	0.27	0.08	t=7.51 (182)	<0.001
H_O	0.32	0.07	t=7.99 (157)	<0.001
A_R	1.79	1.25	t=9.15 (190)	<0.001
N_A	1.80	1.25	t=9.15 (190)	<0.001
P_L	0.80	0.25	χ^2=58.68 (1)	<0.001

Genome-wide diversity, measured as nucleotide diversity (π) and Watterson's theta (Θ), mirrored diversity patterns observed in SNPs, with deme II exhibiting much lower diversity levels. Additionally, individual WGS of two individuals from each deme revealed that deme II displayed considerable levels of inbreeding, measured as LnROH and F_{ROH}, on almost an order of magnitude higher than deme I.

Compared to other sympatric salmonid populations, genetic divergence between the demes is much higher than between other cryptic sympatric populations (**Papers I and II**;; Wilson et al. 2004; Adams et al. 2008; Palmé et al. 2013; Aykanat et al. 2015; Marin et al. 2016), making this a particularly special case. The high genome-wide divergence, together with relatively few SNPs/WGS read-windows, indicate, as outliers suggest, that most of the differentiation in this case is caused by genetic drift, though some genes could be under diversifying selection.

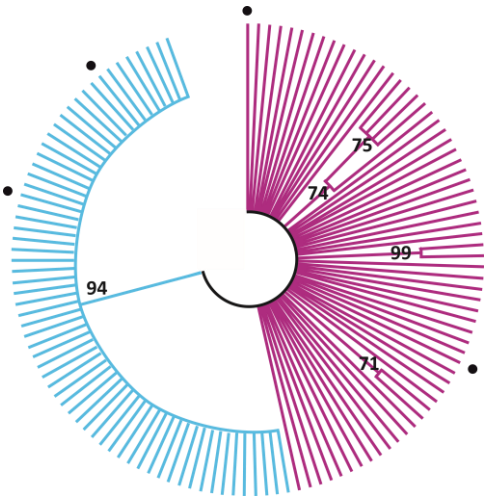

Figure 9 An individual-based, neighbor-joining tree, illustrating the genetic relationship between brown trout from Lakes Bunnersjöarna. Individuals in purple and blue are classified as belonging to deme I and deme II, respectively, based on the allozyme *LDH-I* genotype. The tree was constructed based on Nei's DA distance, and has been compressed to include branches with bootstrap values of at least 70%. Numbers along the branches indicate bootstrap values in percentages. The black dots mark the four individuals that were randomly selected for individual whole-genome sequencing.

Some of the outlier genes were involved in growth processes, which may relate to previously recorded body length differences between the demes (Ryman et al. 1979). Moreover, several of the presumably selected genes were linked to reproductive functions, which may support differences in reproductive characteristics. Spatial spawning divergence is a typical factor that separates brown trout populations (Ferguson et al. 2019), and segregation in spawning sites (inlet versus outlet) has been suggested to occur in Lakes Bunnersjöarna (Ryman et al. 1979). Altogether, these findings suggest that reproductive isolation between sympatric populations is possible even within extremely small bodies of water.

Exploring the hypothesis that cryptic sympatry may be more common than currently recognized

Detection and reports of cryptic sympatry in literature

When conducting the literature survey I identified 80 papers reporting cryptic sympatric populations in 9 localities and non-cryptic populations in 98 localities, encompassing at least 17 separate species, and confirming the suspicion that cryptic sympatry is reported less frequently than non-cryptic sympatry (**Paper IV**). Genetic markers used for assessing population structure were mostly microsatellites and allozymes, typically employing 5–22 loci

for microsatellites and 1–16 loci for allozymes. SNPs and sequencing data were used in 4 and 5 studies, respectively. In studies with SNPs, 94–3093 loci were used. Total individual sample sizes ranged from 48–4148 for studies based on allozymes, and from 20–468 for studies utilizing microsatellites. For SNP based studies, sample sizes varied between 24–744 individuals.

Comparisons of studies reporting cryptic versus non-cryptic sympatry indicated that divergence levels, as well as individual sample sizes, were higher in studies of cryptic populations. However, the number of loci used to detect population structure was essentially the same for both cryptic and non-cryptic populations.

On average, genetic differentiation between cryptic populations was higher than between non-cryptic ones (allozymes and microsatellites considered together averaged F_{ST}=0.13 and 0.10 for cryptic and non-cryptic populations, respectively). This is contrary to expectations, as phenotypically cryptic populations are thought to be evolutionarily young (Fišer et al. 2018), and therefore less genetically differentiated. The observation of higher divergence levels between cryptic populations indicates that the studies reporting cryptic sympatry might offer a biased view and primarily represent cases where statistical power of detection was sufficiently high. Our own observations from Lakes Trollsvattnen support this; these structures were only detected after extensive sampling – using several hundred individuals, and the 14 allozyme markers employed by the genetic monitoring program.

This underrepresentation of cryptic sympatry in literature could be due to several reasons. First, the statistical power to detect heterozygote deficiencies with genetic data alone may have been too low, especially if divergence was small. In this case, large sample sizes or large numbers of genetic markers would have been needed, which is not always possible to obtain. Further, population structuring may had been overlooked because heterozygote deficiencies could have been the result of various technical artefacts of microsatellites (a marker commonly used in genetic screenings; Band and Ron 1997; Waples 2015). Finally, there have been few statistical tools besides Hardy-Weinberg tests to detect population structure until the turn of the 21st century (i.e. STRUCTURE software; Pritchard et al. 2000).

Statistical power to detect cryptic sympatry

The statistical power of detecting cryptic structure was evaluated using a panel of 100 SNPs and 10 microsatellites. The SNP panel included 100 di-allelic loci, while the microsatellite panel consisted of 10 loci with 10 alleles each, which was designed to emulate the number of loci commonly used in actual studies, as indicated by the literature review. Simulated individual

sample sizes (n=20–400) were chosen to reflect the sizes typically used in empirical investigations of population structure.

Generally, the power of detection was very high if populations were well-differentiated and represented in equal proportions, regardless of method (STRUCTURE, DAPC or heterozygote deficiency tests) for both marker panels (Figure 10). STRUCTURE appeared to be superior to both DAPC and heterozygote deficiency tests when genetic divergence levels were at or above F_{ST}=0.10. However, when divergence was lower, power declined for all three methods, particularly for STRUCTURE and DAPC. DAPC was always inferior to the other methods, often not yielding meaningful results (power of either zero or unity, or no estimates at all), thus it was not considered further.

The power of STRUCTURE declined quite rapidly with decreasing genetic divergence levels. The rate of decline was dependent on sample size and the number of loci, with statistical power dropping dramatically at certain F_{ST} levels. These observations indicate the presence of a certain "threshold" level of divergence necessary for STRUCTURE to effectively detect genetic mixture. The power of the heterozygote deficiency test declined more slowly than STRUCTURE with reduced divergence (Figure 10) and this method became superior when divergence levels dropped below the threshold for STRUCTURE. However, the power to detect heterozygote deficiencies at such low divergence levels was poor in absolute terms, often below 20%, which suggests that weakly differentiated populations are likely to be undetected irrespective of method, even if sample sizes are as large as 400 individuals (Figure 10).

The more skewed the representation of the two populations was in a sample, the more the power of detecting population structure deteriorated. For minor deviations (such as 30/70) from equal representation (50/50), power was affected only slightly, whereas highly skewed representation (10/90 or more) appeared to limit power to a significant degree. Uneven representation affected STRUCTURE slightly more than the heterozygote deficiency test, though the power of detection remained fair (>50%) for highly differentiated populations (F_{ST}>0.1) for both methods, even if the representation was highly skewed, such as 5/95.

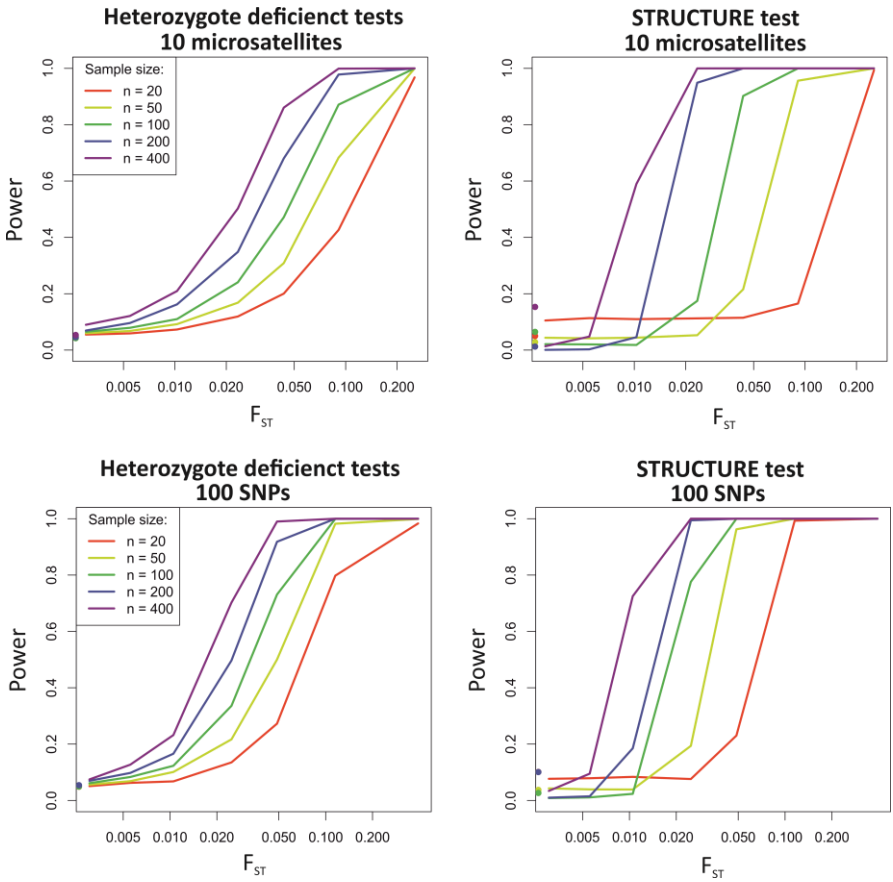

Figure 10 The power of detecting significant evidence for population mixture at different sample sizes (n individuals), genotyped for the microsatellite panel (top) and the SNP panel (bottom) and tested with a GENEPOP heterozygote-deficiency test (left) and a STRUCTURE test for K>1 (right). Dots on the left margins indicate proportions of significant runs from a single, panmictic population and represent the alpha errors of the tests (note that some dots overlap). Figure is reproduced from paper IV (Jorde et al. 2018).

Occurrence and temporal dynamics of cryptic sympatric populations in alpine water systems

My findings in **Paper V** suggest that the occurrence of cryptic sympatry is a relatively common phenomenon in the alpine lakes of central Sweden, as such structures were detected in over 40% of the lakes examined. The levels of divergence among sympatric populations within lakes were roughly the same at both points in time, indicating that these genetic structures have been temporally stable over 40 years which corresponds to about 6 generations.

Sympatric populations were detected in 12 of the 27 lakes, representing five lake systems (hereafter referred to as metapopulations). Sympatry occurred

throughout all but two metapopulations, with the same genetic clusters present at both points in time (Figure 11), which is in line with the findings of Bekkevold and colleagues (2019). The levels of genetic divergence (F_{ST}) among sympatric populations were in the same order of magnitude as those previously reported for sympatric populations using SNPs (**Paper II**; Renaut el al. 2011; Aykanat et al. 2015) and other marker systems (Hebert 2013; Lehnert et al. 2016). However, none of the sympatric populations differed as much as the two reproductively isolated brown trout demes in the Bunnersjöarna Lakes (**Paper III**).

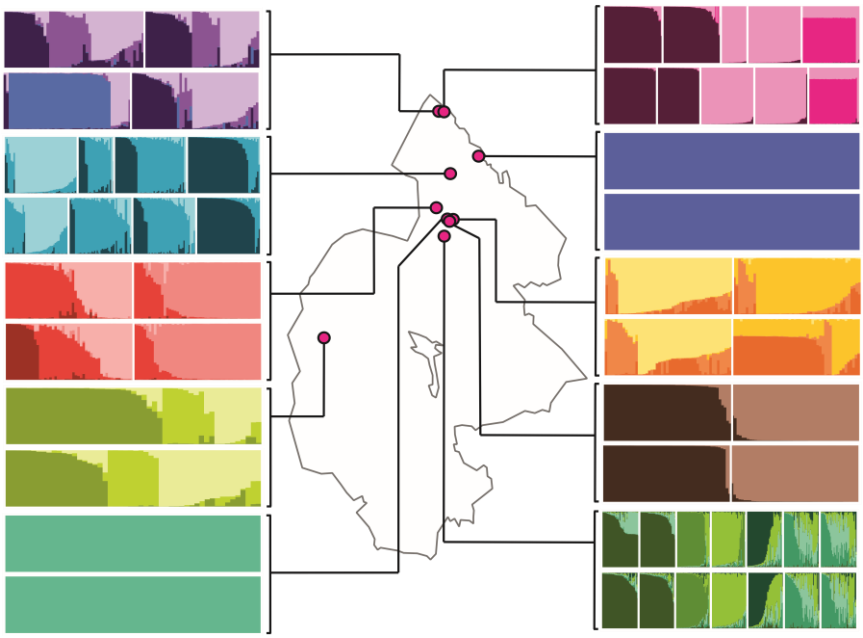

Figure 11 Barplots illustrating the results from STRUCTURE analyses exploring population genetic structure in 27 lakes, distributed over 7 lake systems, and 3 lakes without connection to any other locality included in the study, using a panel of 96 SNPs. Each fish is represented by one vertical bar and clusters are represented by different colors. Each metapopulation/single lake is shown in two panels representing the two time points: the past (1970–80s; top) and present (2010s; bottom). Lakes within metapopulations are separated by white vertical lines.

Both STRUCTURE and heterozygote deficiency tests were used to detect genetic structuring that could indicate the presence of sympatric populations. However, the convergence of the two methods for individual lakes was rather poor – the heterozygote deficiency tests failed to detect a heterozygote deficiency in many of the lakes where STRUCTURE identified more than one population. This can be explained by the higher power that STRUCTURE has to detect genetic structuring without any prior grouping of individuals when F_{ST} is fairly high between clusters (about 0.1), even if the representation of

clusters in the sample is skewed (**Paper IV**). STRUCTURE is expected to outperform the heterozygote deficiency tests because sample sizes (also affecting power) used in the tests were substantially smaller (maximum n=50) than those used in STRUCTURE (n=53–693). Pooling samples from different lakes is problematic for heterozygote deficiency tests; genetic differences between clusters in separate lakes make it difficult to distinguish the signal of potential genetic structuring within separate lakes from the genetic differences found between lakes. STRUCTURE is capable of doing this, however.

SNP data identifies additional genetic structuring and confirms spawning site divergence

Recently obtained SNP data for an additional >500 fish from the Trollsvattnen Lakes and adjacent creeks revealed further genetic structuring within one of the sympatric populations and supported the spawning locality segregation that had previously been detected with allozymes.

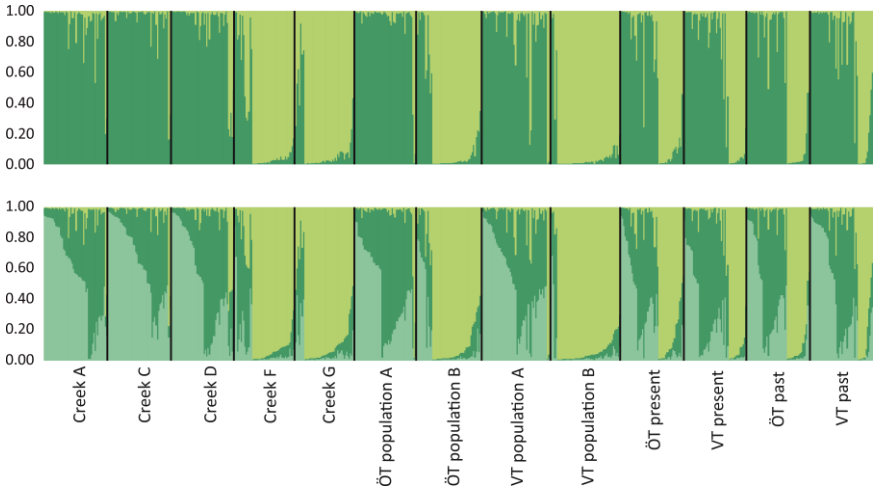

Figure 11 Barplots illustrating results from the recent SNP-based STRUCTURE analysis of >600 fish from the Trollsvattnen lakes, including parr from potential spawning locations (creeks A-G). ÖT=Lake Östra Trollsvattnet, VT=Lake Västra Trollsvattnet; A and B refer to the populations identified with allozymes, corresponding to the grayish-green and apple-green SNP clusters, respectively. The samples denoted "present" and "past" to the right in the plots are included in Paper V. The upper panel illustrates the segregation of the fish into two genetic clusters, while the lower panel shows three clusters, with the third cluster being nested within one of the original ones. Each fish is represented by a vertical bar.

In **Paper V**, Lakes Trollsvattnen appear to harbor three populations, whereas two were identified using allozymes. This is because the SNP data identifies additional structuring within one of the allozyme populations (population A), as revealed in a new structure analysis (unpublished results; methods de-

scribed in **Paper V**). When comparing the two-population and three-population scenarios (Figure 11), the consistency of assignment is excellent, with only 17 of 653 fish classified inconsistently.

The SNP data supports the divergence in spawning locality between the sympatric populations in the Trollsvattnen Lakes, as reported in **Paper II** (Figure 11). The observation that parr, classified as corresponding to population A using SNPs, are found predominantly in the western inflow and the southern outflow, while parr corresponding to population B occur in the northern inflow, is in line with the findings in **Paper II**.

Conclusions

This thesis expands our knowledge on the characteristics, spatial and temporal dynamics, and occurrence of cryptic sympatry – i.e. the occurrence of genetically distinct populations coexisting in the same habitat with no apparent barriers to gene flow and without obvious phenotypic differences. The main findings are:

- Cryptic, sympatric populations can coexist without apparent trophic divergence, utilizing the same food resources and displaying the same adaptive plasticity to their shared environment (**Paper I**).

- Patterns and levels of genetic connectivity of sympatric populations to nearby lakes can be very different, both between the coexisting populations as well as between different cases of sympatry (**Papers II and III**).

- Spawning segregation in space appears to be a mechanism that enables cryptic sympatric populations to exist, and the frequency of spawners can differ markedly between populations (**Papers I and II**).

- The genetic structures representing the cryptic sympatric populations in the two empirical cases (Lakes Trollsvattnen and Lakes Bunnersjöarna), originally detected with allozymes, can be identified with SNPs with a high consistency. This suggests that even a few allozymes in some cases can identify genetic structuring that reflects genome wide patterns (**Papers II and III**).

- Sympatric populations can exhibit strikingly different levels of genome-wide diversity, inbreeding, and divergence, suggesting that reproductive isolation can be strong (**Paper III**).

- Cryptic sympatry in salmonids is rarely reported in the literature. For reported cases, the divergence between sympatric populations is considerably higher for cryptic than for non-cryptic sympatry. This may be due the generally poor statistical power for detecting population structure without prior grouping of individuals when applying commonly used sample sizes (in terms of individual and loci), especially if population representation in the sample is skewed (**Paper IV**).

- When actively searching for cryptic, sympatric populations they appear to be common and stable over time; such hidden structures were detected in over 40% of the small alpine lakes included in the screening effort (**Paper V**).

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Svensk sammanfattning

Genetisk variation är biologisk variation inom arter som är nödvändig för att de ska kunna anpassa sig till förändrade miljöförhållanden och överleva i ett långsiktigt perspektiv. Denna variation kallas ibland även för dold biodiversitet eftersom den inte syns för blotta ögat, utan kräver laborativa analyser för att upptäckas. Genetisk variation har i vissa studier visats kunna ha liknande effekt som artdiversitet för ekosystemens funktion och återhämtningsförmåga.

En speciell typ av genetisk variation som verkligen kan kallas för dold är genetiskt åtskilda bestånd som samexisterar över begränsade geografiska områden. Det handlar alltså om bestånd av samma art, som trots att de existerar i samma miljö inte parar sig med varandra alls eller gör det väldigt sällan. Detta är en typ av biologisk mångfald som är föga känd och den kallas på fackspråk för ”kryptisk sympatri”. Samexisterande bestånd kan även förekomma i en form som inte är dold, det vill säga de samexisterande bestånden skiljer sig åt i till exempel utseende och/eller beteende. Sådana synliga samexistenser har dokumenterats i flera olika organismer, däribland laxartade fiskar.

Två fall av dolda samexisterande bestånd har tidigare upptäckts hos öring i mycket små fjällsjöar i Jämtlands län. Det ena fallet finns i Bunnersjöarna i Vålådalens naturreservat och det andra i Trollsvattnen i Hotagens naturreservat. Fallet i Bunnersjöarna upptäcktes redan på 1970-talet då de första kartläggningarna av öringens genetiska variation gjordes, och det andra upptäcktes på 1990-talet i samband med ett långsiktigt projekt som övervakar genetisk variation i utvalda vatten i Hotagenreservatet.

Syftet med denna avhandling har varit att öka kunskapen om kryptiskt samexisterande bestånd. Med öring som modell var målet först att i mer detalj studera de två rapporterade fallen i Trollsvattnen och Bunnersjöarna med avseende på deras arvsmassa, utseende och beteende. I ett andra steg undersöktes om det skulle kunna vara så att dolt samexisterande bestånd är vanligare än vi hittills vetat. Detta gjordes genom att granska litteratur som dokumenterar samlevande bestånd hos laxartade fiskar, utvärdera den statistiska säkerheten att upptäcka sådana bestånd med ofta använda materialstorlekar (både vad gäller antalet undersökta individer och antalet undersökta gener), samt att genomföra en storskalig genetisk kartläggning av förekomsten av kryptisk sympatri i små svenska fjällsjöar.

Bestånden i Trollsvattnen undersöktes med avseende på födoval och kroppsform men visade inga tecken på att nyttja olika födoresurser. Detta var förvånande då det enligt ekologisk teori inte ska vara möjligt för två bestånd att använda samma resurs utan att det ena beståndet konkurrerar ut det andra.

De enda skillnaderna som upptäcktes var i kroppslängd och andelen lekmogna individer vid insamlingstidpunkten, där det ena beståndet var något längre och hade en lägre förekomst av lekmogna individer. Bestånden ser också ut att använda olika bäckar för lek, vilket tyder på att skilda lekplatser är en mekanism för upprätthållandet av samexistens. I Bunnersjöarna upptäcktes slående skillnader i många gener fördelade över hela arvsmassan. Det ena beståndet var markant mer inavlat än det andra och hade mycket lägre grad av genetisk variation vilket tyder på att det var starkt isolerat. Motsvarande skillnader upptäcktes inte i Trollsvattnen. Även i Trollsvattnen hade det ena beståndet mer genetiskt utbyte med bestånd i närliggande vatten än det andra, men skillnaderna var inte alls så stora som i Bunnersjöarna.

Resultaten visar att samexisterande bestånd kan förekomma inom mycket begränsade områden och uppvisa mycket olika nivåer av genetisk variation, samt ha olika grad av genetiskt utbyte med bestånden i närliggande sjöar.

Det fanns även tecken på att dold samexistens är vanligare än vad vi tidigare har förstått. Litteraturgranskningen visade att kryptisk samexistens sällan har rapporterats i vetenskapliga artiklar. Betydligt vanligare är rapporter om samexisterande bestånd som kan särskiljas utseendemässigt (icke-kryptisk sympatri). De genetiska skillnader som rapporteras mellan samexisterande bestånd är högre för kryptiska bestånd än för icke-kryptiska. Detta beror på att möjligheten att upptäcka kryptiskt samexisterande bestånd är ganska låg om de inte skiljer sig starkt genetiskt. När vi sedan aktivt sökte efter dolda samexisterande bestånd i ett större antal fjällvatten visade det sig att sådana bestånd är ganska vanliga – de förekom i över 40 procent av de undersökta sjöarna. Resultaten visade också att dessa samexisterande bestånd är stabila över tid.

Sammanfattningsvis visar fynden i denna avhandling att kryptiskt samexisterande öringbestånd kan nyttja samma födoresurser utan att konkurrera ut varandra, och att val av olika lekplats över mycket begränsade områden kan vara en faktor som upprätthåller samexistens. Sådana bestånd kan även uppvisa stora genetiska skillnader både mellan varandra och med närliggande bestånd, trots till synes goda möjligheter till genetiskt utbyte. Vidare tyder resultaten på att kryptisk samexistens är ett relativt vanligt fenomen, som är stabilt över tid, men att det kan vara svårt att upptäcka. Många individer och gener behöver undersökas för att dolda samexisterande bestånd ska upptäckas med hög säkerhet.

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