Master's thesis

Inferring fish abundance in lakes from subfossil midge (Diptera: Chironomidae) remains

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International Aquatic Master's Programme

15.05.2017

ABSTRACT

This study assessed the possibility of using subfossil remains of chironomid head capsules to reconstruct past fish abundances in lakes. Using data from 20 Swedish lakes, a weighted average (WA) model was created to predict the abundance of benthivorous fish (cyprinids and perch). Chironomid head capsules, identified to genus, served as the predictors to model already recorded (catch per unit effort) fish data. The relationship of chironomid species composition to lake morphometry, water chemistry and fish abundance were examined using ordination and variation partitioning methods. Moreover, the relationship between fish abundance and two chironomid species traits: body size and activity mode were examined to obtain information on the influential mechanisms behind these predator-prey interactions. Based on earlier studies, I hypothesized, first, that chironomid body size correlates negatively with fish abundance. Second, I expected that active and mobile species to dominate in the lakes without fish or with low fish abundance and inactive species to dominate in lakes with high fish abundance. The Chironomidae WA models explained 65 % and 60 % of the variation in fish density and biomass, respectively. When Chironomidae models were combined with previously published *Chaoborus*-based prediction models, both density and biomass models improved slightly. Spearman correlations between the body size and activity modes with fish abundance were significant only for smaller-sized chironomids. The smallest clingers showed a positive correlation with fish density, as did small crawlers with fish biomass. This may be due to the relief of predatory pressure from large-sized invertebrate predators, which are preyed on by fish. The burrowing taxa appeared to be more protected against fish predation, as large-sized burrowers correlated positively with fish biomass, and crawlers of the same size negatively correlated with fish biomass. Redundancy and variation partitioning analyses indicated that fish have a clear and independent effect on the chironomid assemblages.

TIIVISTELMÄ

Tässä tutkimuksessa arvioitiin mahdollisuutta käyttää surviaissääskijäänteitä järvien kalarunsauden takautuvaan arviointiin. Perustuen 20 ruotsalaisen järven aineistoon kehitettiin painotetun keskiarvon (WA) malli pohjaravintoa käyttävien kalojen (ahven- ja särkikalat) runsauden ennustamiseksi subfossiilisista surviaissääskijäänteistä. Suvulleen määritettyjen surviaissääskitoukkien pääkapselien runsaudet olivat ennustavina muuttujina kalojen runsaudelle (yksikkösaalis). Surviaissääskien taksonomisen koostumuksen yhteyttä järven morfometriaan, veden laatuun ja kalarunsauteen tutkittiin ordinaatio- ja varianssin osittamismenetelmin. Lisäksi tutkittiin kahden surviaissääskien lajipiirteen, koon ja aktiivisuuden, yhteyttä kalarunsauteen peto-saalis suhteen mekanismien selvittämiseksi. Teoriaan ja aiempiin tutkimuksiin perustuen oletin, että surviaissääskien koko korreloi negatiivisesti kalarunsauden kanssa. Lisäksi oletin, että aktiivisesti liikkuvat lajit ovat runsaimpia kalattomissa ja vähäkalaisissa järvissä ja paikallaan pysyvät lajit järvissä, joissa on runsaasti kaloja. WA mallin ennusteet selittivät 65 % havaitusta kalatiheydestä ja 60 % biomassasta. Yhdistämällä ennusteet aiemmin kehitetyn sulkasääskimallin antamiin ennusteisiin, ottamalla näiden kahden keskiarvo, sekä tiheyden että biomassan ennusteet paranivat hieman. Pienikokoisimpien, pintoihin kiinnittyvien surviaissääskitoukkien runsaus korreloi positiivisesti kalatiheyden kanssa ja pienten pohjalla ryömivien toukkien runsaus kalabiomassan kanssa. Tämä voi johtua vapautumisesta predaatiopaineesta, jonka aiheuttavat suurikokoiset selkärangatonpedot, joita kalat saalistavat. Myös kaivautuvat sääskentoukat näyttivät olevan paremmin suojassa kalapredaatiolta, sillä suurikokoisten kaivautujien runsaus korreloi positiivisesti ja samankokoisten ryömijöiden runsaus negatiivisesti kalabiomassan kanssa. Redundanssianalyysiin yhdistetty varianssien osittaminen viittasi siihen, että kaloilla on melko selkeä, muista tekijöistä riippumaton vaikutus järvien surviaissääskiyhteisöihin.

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1. INTRODUCTION

1.1. Maintaining lake ecosystems

Lake ecosystems provide freshwater resources that are essential for the humankind. Human communities are often dependent on ecosystem services provided by lakes including drinking water, irrigation water for farming, commercial and recreational fisheries and leisure activities. Human impacts on nature stemming from agriculture and urban activities have increased by about 8 times compared to 40–50 years ago (Jorgensen 2001). The nutrient loadings from agricultural practices, industrial effluents, untreated wastewater, and atmospheric deposition has greatly increased the amount of Nitrogen and Phosphorus in lakes and rivers (Smith et al. 1999). The consequences of additional nutrient inputs on aquatic ecosystems is seen across the globe in toxic algal blooms, loss of oxygen, fish kills, loss of biodiversity, and loss of aquatic plant beds (Carpenter et al. 1998). The United Nations (UN) refers to the global phenomenon of eutrophication as the most prevalent water quality problem (Jorgensen 2001). In addition to eutrophication, acidification with increased aluminum concentrations in water bodies is another major environmental issue. Lakes located in catchment systems with low concentrations of calcium to act as a buffering system are most prone to acidification, and may experience seasonal fish kills, or overall reduction in fish populations (EPA 2016).

The EU is working to improve the status of water bodies through its recent Water Framework Directive (WFD) initiative to restore all degraded water bodies to achieve a "good" ecological status. Lake status is determined based on the aquatic flora, fauna, and physical, chemical, and morphological quality elements. The good status allows only 'slight' deviation from the conditions prior to industrialization and major human use (EU 2000). Ensuring good water quality is also a priority by the United Nations, illustrated by Goal 6: Sustainable Water and Sanitation for all, of the Sustainable Development Goals (United Nations 2015). Water Goal 6 is one of the 17 goals to combat poverty and ensure prosperity of our planet within the sustainable development agenda adopted in September 2015.

1.2. The use of Chironomidae as paleoindicators of lake physicochemical conditions

In order to evaluate and measure restoration goals set by the WFD and UN, a clear way of evaluating the ecological status of a waterbody is required. This requires studying the changes in biota of an ecosystem. Chironomid midges (Diptera: Chironomidae) are commonly used to study human-induced changes in lakes, including eutrophication as well as hypolimnetic oxygen deficiencies in lakes (Brodersen & Quinlan 2006). Chironomids are common in these studies because certain species are sensitive to lake changes whereas others are tolerant (Brodersen & Quinlan 2006). Thienemann (1925) created the first basic model using profundal chironomid taxa to identify trophic status of temperate lakes and additional taxa have since been added to the model (Porinchu & MacDonald 2003). Profundal taxa are effectively used as lake indicators because the composition of species responds to changes in oxygen levels, reflecting the productivity of the lake. Therefore, profundal macroinvertebrates serve as indicators of lake status. The Benthic Quality Index (BQI, Wiederholm 1980) is a notable and widely used indicator metric, which is based on a small number of profundal chironomid taxa. BQI was later expanded to include 70 taxa for assessment of lake profundal macroinvertebrates (Jyväsjärvi & Hämäläinen 2014).

Chironomids are good indicators for paleolimnological studies because they occupy diverse aquatic habitats, their niche preferences are well known, and they leave behind high abundances of subfossilized head capsules in sediment. Chironomids are often the

most abundant macro-organisms in sediment samples due to their head capsules being composed of a durable chitin material that stay well preserved in sediment (Hofmann 1988). Identification of chironomid taxa within core samples could potentially serve as more reliable indicators of a lake's true history than other invertebrate fossil records.

Chironomid taxa are used as indicators for lake acidification and climate change. In cultural acidification, the overall amount of head capsules is expected to decrease, with an increase in *Phaenopsectra* and other members of the Chironomini tribe, accompanied by *Zalutschia zalutschicola*, from the Orthocladiinae subfamily (Brodin 1986, Brooks 2005). Long term patterns of chironomid head capsules has shown an increase of Chironomini tribe over the past 2500 years, which is thought to be associated with human induced warming of the planet. This shift in higher abundances of Chironomini in lakes triggered other studies to use chironomids as paleoclimatic indicators in temperature inference models (Porinchu et al. 2007, Larocque-Tobler et al. 2010). In a long-term study evaluating chironomid head capsules from an alpine lake spanning over 9000 years, Heiri & Lotter (2003) examined the effects of human activities in the catchment on lake communities.

Validating trophic status and indicating chemical or physical changes in lakes may be the most prevalent use of chironomids in paleolimnological studies so far, but in future studies they may also serve as strong indicators of biological conditions including the presence-absence and abundance of fish populations.

1.3. Potential use of Chironomidae as paleoindicators of past fish abundance

While studying how communities change in varied levels of eutrophication, Langdon (2006) found that chironomid communities are impacted by nutrients and lake productivity, but possibly equally affected by biotic factors, including macrophytes, fish populations and zooplankton. Fish predation effects have been inferred previously by Uutala (1986) when comparing trends of chironomid and *Chaoborus* remains to the time period of acidification and expiration of fish populations in the Adirondack Mountains. He observed that chironomid communities were greatly influenced by biotic factors, specifically predation.

Recent studies have identified fish populations as a major factor responsible for shifts in overall chironomid assemblages of lakes. Raposeiro et al. (2017) found that chironomid assemblages of Lake Azul (Azores Archipelago) were different before and after the introduction of fish populations over a 220-year time period. Lake Azul is a naturally fishless lake that experienced the introductions of several fish species at different times. With introductions of goldfish, carp and pike, the chironomid assemblage changed and chironomid feeding guild composition shifted from detritivores and predators to grazers. Milardi et al. (2016) observed that fish introduction in a boreal lake in Finnish Lapland decreased macroinvertebrate abundance in pelagic areas and increased in littoral areas. Within sediment samples, Chironomidae increased five-fold indicating that the introduction of Brown trout positively influenced the abundance of benthic Chironomidae, which was most likely linked to indirect top-down effects via predatory invertebrates depressed by fish.

More specifically, changes in chironomid genera have been associated with the expiration of fish populations. Heiri & Lotter (2003) noticed an absence of *Procladius* in recent years when examining chironomid trends over a 9000-year time span in an alpine lake. The absence of *Procladius* was hypothesized to be due to fish stocking, because they are considered a favored prey of salmonids (Serra-Tosio 1978, Heiri & Lotter 2003).

Additional studies have also observed that *Procladius* and other free-living chironomid taxa are favored prey of fish due to their large size and conspicuous pigmentation (Serra-Tossio 1978, Uutala 1990, Johnson et al. 1990, Armitage et al. 1995, Porinchu & MacDonald 2003). Also other free-living chironomids including *Corynoneura*, *Pentaneurini* and *Psectrocladius* are hypothesized to be preyed on more regularly than non-free-living chironomids by fish. On the other hand, smaller, free-living chironomids like *Corynoneura* and *Pentaneurini* are hypothesized to favor lakes with intermediate fish densities because fish tend to prey on larger invertebrate taxa, which are competing with and preying on the smaller taxa (Uutala 1986, Langdon et al. 2006).

The accurate identification of past fish abundances may be a useful tool for the assessment of present lake status and for setting restoration targets. Fish play a crucial role in stabilizing lake ecosystems, and benefit humans by providing us with food, recreation and employment. Human induced pressures on lakes, which include acidification and eutrophication, can cause natural fish populations to change and decline (Schofield 1976). Knowledge on past fish populations that once naturally existed is important background information for promoting success in lake restoration projects. For example, adding fish to a historically fishless lake can be costly and damaging to the natural ecosystem (Townsend 1996, Knapp et al. 2007). Difficulties can arise when trying to reconstruct past fish populations because of the poorly fossilized remains fish leave in sediments. Although subfossil fish scales have been used to estimate fish presence-absence in shallow eutrophic lakes, the scales were restricted to percid and cyprinid groups, and lacked representation of larger bodied fish species. Moreover, when attempting to accurately use fish scales for reconstruction purposes, a much larger amount of sediment than collected in standard coring practices is required (Davidson et al. 2003). This makes the direct use of fish remains a costly approach, and suggests the possible use of proxies i.e. prey taxa of fish as a better alternative to indirectly reconstruct past fish populations.

1.4. Objectives and hypotheses of study

The first objective of this study is to use Chironomidae subfossil remains to develop a model for fish abundance reconstruction. My hypothesis is that the chironomid species will respond differently to fish predation (directly or indirectly), making it possible to infer fish abundance from a lake's taxa composition. To evaluate this hypothesis, a weighted average model was developed to infer fish abundance using the chironomid head capsules from 21 study lakes.

The second objective, related to the previous, is to analyze the differences in chironomid species compositions of lakes with varying fish abundance and fishless lakes. If species respond differently to fish predation, the species composition should vary predictably along the gradient of fish abundance. Third, certain species traits (body size and activity mode) were expected to respond predictably to fish abundance gradient, since chironomid species with certain characteristics were expected to be more susceptible to fish predation. Body size distribution of chironomids was hypothesized to shift towards small-sized species with increasing fish abundance. Furthermore, burrowing chironomid taxa were expected to dominate in lakes with high fish abundance, whereas lakes with low fish abundance or without fish are potentially characterized by greater abundances of free-living taxa.

2. MATERIAL AND METHODS

2.1. Study lakes and data collection

Chironomid head capsules were collected from the top sediment (0–1 cm) of 21 lakes in southern Sweden (Tolonen et al. 2012). The study lakes ranged in size from small to medium (3–383 ha) with varying levels of fish abundance. The study lakes have been subjected to varying degrees of acidification and differ in their morphometry and water chemistry characteristics (Table 1). Core samples were collected from the deepest point of each lake using a Kajak corer with 5.2 cm in inner diameter during April and May of 2008. Three sample replicates were pooled from each lake. The pooled sample of the top 1 cm of the sediment formed a single sample of 63 ml in each lake. Due to the low sedimentation rates in Swedish lakes (Guhrén et al. 2007), the (0–1 cm) surface sediment represented approximately the past eight years. Samples were preserved in 70 % ethanol in the field. In the laboratory, samples were homogenized and heated in 10 % KOH for 20 minutes. After cooling, samples were sieved through 93 μm mesh and preserved in 99 % ethanol. Chironomid head capsules were sorted under a dissecting microscope and mounted on Euparal$^{®}$ slides for identification.

Table 1. Information on the morphometry and water chemistry of the study lakes. The lakes are arranged in ascending order along the pH gradient. Liming status is given using the abbreviations N = not limed and L = limed lake. Starting year of liming is given in parentheses.

Lake	Liming status	Altitude (m a.s.l.)	Area (ha)	Max. depth (m)	Mean depth (m)	pH	P_{tot} (1 g 1^{-1})	TOC (mg 1^{-1})	Secchi depth (m)
Stavsjön	N	170	3	8.0	4.5	4.3	16	15.1	1.0
Liasjön	N	116	12	5.3	2.5	4.7	25	25.5	0.5
Gölasjön	N	166	6	4.2	1.3	4.8	23	32.0	0.3
Lillesjö	N	88	4	12.5	10.4	5.0	5	2.9	5.5
Stensjön	L (1992)	126	30	4.0	1.8	5.1	15	22.6	1.8
Hjärtsjön	N	274	128	6.0	3.4	5.3	5	4.2	5.4
Klintsjön	N	232	10	18.0	4.6	5.6	7	4.9	5.4
Stora Slätten	L (1980)	149	264	23.5	5.7	6.1	11	17.2	2.2
Bergsjön	N	174	326	7.0	3.4	6.1	19	11.7	2.2
Brändasjö	L	178	14	5.0	1.9	6.2	27	36.5	0.4
Ubbasjön	L (1983)	109	28	6.0	2.7	6.3	34	26.5	0.7
Römningen	L (1993)	121	383	4.0	1.1	6.4	20	19.2	1.3
Hultbren	L (1986)	240	193	3.0	1.0	6.4	26	20.0	2.2
Strönasjön	L (1983)	125	27	6.0	3.0	6.5	25	28.4	0.7
Gyslättäsjön	L (1985)	226	32	10.0	2.9	6.7	14	12.2	2.0
Humelsjön	L (1985)	130	34	4.0	2.1	6.8	30	13.7	1.3
Hacksjön	L	218	45	16.0	5.4	7.1	8	7.6	4.0
Blanksjön	L (1981)	39	20	17.0	4.9	7.2	8	11.1	4.4
Lunksjön	L (1987)	98	49	8.0	2.0	7.2	16	16.1	2.2
Frösjön	L (1989)	140	32	6.0	1.5	7.2	31	21.2	0.9
Levrasjön	N	7	292	19.0	10.6	8.3	9	5.4	4.6

2.2. Head capsule identification

Head capsules were identified to genera using mainly Wiederholm (1983) and when possible, to species level using an additional identification key (Brooks et al. 2007). Head capsules that could not be determined to species, either due to their positioning on the slide or their deteriorated state were classified to genus or subfamily. One specific-looking type of head capsules could not be confidently identified. These head capsules were reoccurring

throughout the core samples (in 9 of the 21 lakes) and when compared to one another appeared to be the same species. Based on the head capsule, this species is likely to be a 1st or 2nd instar of *Chironomus*, having similar mandibles and an underdeveloped mentum is labeled as small *Chironomus 1st Instar* (Appendix).

A complete head capsule was counted as one, a partial head capsule as one half, and the sum represented the total raw number (*R*) of head capsules in each lake (Table 2). The sedimentation rates of the study lakes were unknown and likely differed due to varying productivity and sediment loads. Therefore the raw head capsule counts were transformed into proportional values, with each lake's total proportion of taxa equal to one. To compare the amount of head capsules between the lakes, the raw number of head capsules was multiplied by a density coefficient to determine density of head capsules (*D*) per gram of dry sediment (Table 2). The dry weight density coefficient was obtained by dividing the density coefficient per 100 mL of wet sediment by the sediment dry weight (g) per 100 mL of wet sediment.

Table 2. Chironomidae head capsule raw counts (*R*), densities per 1 g of dry sediment (*D*), and number of species and genera (S), and fish populations (mean catches ± S.D. during the years 2000–2007) of the study lakes. Number of test fishing years is given in parentheses after the name of the lake. Lakes are arranged in ascending order of fish biomass (CPUE g gill-net⁻¹).

Lake	Biomass (g gill-net⁻¹ day⁻¹)			Density (ind. gill-net⁻¹ day⁻¹)			Chironomidae		
	Perch	Roach	Total excl. pike	Perch	Roach	Total excl. pike	*R*	*D* (g)	*S*
Stavsjön (1)	0.0	0.0	0.0	0.0	0.0	0.0	74.5	528	17
Lillesjö (1)	0.0	0.0	0.0	0.0	0.0	0.0	110.0	1055	11
Gölasjön (3)	128.0	0.0	128.0 ± 33.2	1.6	0.0	1.6 ± 0.3	126.0	2,164	22
Liasjön (2)	178.8	0.0	178.8 ± 178.4	1.1	0.0	1.1 ± 1.1	63.5	546	20
Ubbasjön (3)	101.6	72.3	185.7 ± 81.7	4.1	4.6	9.2 ± 4.9	47.5	104	19
Brändasjö (1)	0.0	211.4	211.4	0.0	8.9	8.9	51.5	95	20
Gyslättäsjön (8)	137.4	240.6	418.6 ± 100.1	5.9	3.2	9.0 ± 2.9	87.5	281	18
Strönasjön (3)	45.0	58.7	424.2 ± 307.6	2.2	3.2	6.6 ± 4.7	60.5	95	22
Frösjön (1)	102.6	157.8	564.4	8.5	3.3	19.2	81.0	85	18
Klintsjön (3)	589.7	0.0	589.7 ± 173.7	26.0	0.0	26.0 ± 0.6	97.0	413	20
Stora Slätten (2)	805.0	40.8	974.6 ± 128.6	10.2	2.3	14.1 ± 2.7	84.0	77	25
Blanksjön (3)	571.8	411.2	983.0 ± 243.6	17.5	6.5	24.0 ± 4.8	98.5	308	24
Stensjön (1)	770.1	212.0	1,574.8	32.9	5.1	41.2	85.5	256	23
Römningen (1)	499.0	792.6	1,635.2	15.0	24.0	44.2	91.0	119	14
Levrasjön (1)	815.2	446.1	1,683.8	56.5	20.0	78.7	86.5	177	18
Lunksjön (1)	1,494.8	210.4	1,774.2	22.4	11.0	34.2	83.5	441	21
Hacksjön (1)	1,430.9	387.4	1,818.3	37.8	8.8	46.6	95.5	384	25
Hultbren (1)	1,370.0	488.8	1,858.8	67.1	11.6	78.8	125.0	179	28
Bergsjön (1)	1,295.8	452.7	1,932.0	38.0	26.1	68.9	82.5	109	18
Hjärtsjön (3)	2,611.2	0.0	178.8 ± 178.4	38.8	0.0	38.8 ± 7.2	122.5	2,328	13
Humelsjön (2)	1,319.8	2,130.0	178.8 ± 178.4	56.1	134.6	191.1 ± 76.2	105.0	576	25

2.3. Fish data

Fish abundance data (Table 2) were obtained from the National Register of Survey (NORS) test fishing database of the Swedish Board of Fisheries by Tolonen et al. (2012). Due to the low sedimentation rates in Swedish lakes (Guhrén et al. 2007), fish data from the past eight years was thought to correspond to surface sediment (0–1 cm) accumulation. Using the available fish data, lake-specific catches per unit of effort (CPUE) were calculated and used as proxies for fish density (number of individuals gillnet⁻¹ day⁻¹) and

biomass (g gillnet^{-1} day^{-1}). Fish data for the lakes were obtained by the European standard test fishing methodology, using Nordic multi-mesh gillnets with mesh-sizes 5, 6.25, 8, 10, 12.5, 15.5, 19.5, 24, 29, 35, 43 and 55 mm in randomized order in each net (CEN 2005). The fishing procedure was based on stratified random sampling, where each lake was divided into depth strata, within which gillnets were placed randomly. Piscivorous fish species, pike (*Esox lucius*) and pikeperch (*Sander lucioperca*), were not included in the fish biomass and density because these fish do not feed directly on chironomids.

2.4. Chironomidae weighted averaging model

A weighted averaging regression model was constructed following the steps of Jeppesen et al (2001) transfer function to test the possibility of using Chironomidae midge fauna remains to predict fish populations in lakes. The WA model uses the identified head capsules from the study lakes, selected to range varying abundances of fish density and fish biomass. A WA model was appropriate for this study because it assumes that taxa demonstrate a unimodal response to environmental variables, and suggests that species peak abundance is at the optimum level of the specific variable gradient (ter Braak & Juggins 1993). This modeling approach works well with noisy, species-rich data (ter Braak & Juggins 1993), making it highly suitable for chironomid taxa, which are known to inhabit a wide range of ecosystem conditions. The transfer function used is a series of four calculations including estimation of the taxa optima and tolerances to determine the initial inferred estimate, which is then regressed on the observed variable to obtain the coefficients *a* and *b* or the deshrinking parameters, leading to the final inferred estimate (Jeppesen et al. 2001). This model uses the estimated optima and tolerances of taxa for fish abundances from the 21 study lakes as the independent predictor variables, and the known fish abundance data as the dependent variable.

The first two equations used to develop the model are the estimators of the optimum (u) and tolerance (t) of each taxon (k) with regard to fish abundance (x). The abundance-weighted average is used to estimate the taxa optima (Eq. 1), and the abundance-weighted standard deviations are used to determine the taxa tolerances (Eq. 2).

$$\hat{U}_k = \sum_{i=1}^{m} y_{ik}\, x_i \Big/ \sum_{i=1}^{n} y_{ik} \quad [1]$$

$$\hat{t}_k = \left[\sum_{i=1}^{n} y_{ik\,(x_i - \hat{u}_k)^2} \Big/ \sum_{i=1}^{n} y_{ik} \right]^{1/2} \quad [2]$$

Where y_{ik} is the relative abundance of the taxon k in each lake i, and x_i is the corresponding fish abundance.

The abundance-weighted average of optima across all taxa present in a site was calculated to determine the initial inferred value for the x variable (Eq. 3).

$$\hat{x}_i = \sum_{k=1}^{m} y_{ik}\, \hat{u}_k \Big/ \sum_{i=1}^{m} y_{ik} \quad [3]$$

The tolerances were used as additional weights to give more emphasis to smaller niche gradients, opposed to larger, broader gradients, thereby improving the estimate (ter Braak & Juggins 1993).

The next step was to regress the initial inferred estimate against the observed variable (actual fish biomass or density) in a linear regression to determine the *a* and *b* coefficients used for the deshrinking regression. Using the deshrinking coefficients (*a* and *b*), the final inferred abundance was calculated using the classical deshrinking equation (Eq. 4).

$$\hat{x}_{if} = \frac{(\hat{x}_i - a)}{b} \quad [4]$$

In order to compare and evaluate the Chironomidae WA model results, the model was combined with another model using mandibular remains of phantom midge larvae (Diptera: *Chaoborus*) as the predictor for fish abundance (Tolonen et al. 2012). The data for both models were collected from the same study lakes therefore the models could easily be combined by taking the average of the inferred abundance estimates. The observed fish biomass and density were regressed against the model estimates and also the Mean Absolute Error (MAE) was calculated to evaluate model performances.

2.5. Correlation of chironomid species and traits with fish abundance

Spearman correlation analyses were carried out to determine the association of individual chironomid taxa with fish density and biomass. Spearman correlation analyses were also used to examine correlations between species traits of chironomids, i.e. potential body size and substrate association (activity mode), with fish abundance (predation pressure). Potential maximum dry weights (mg) of the larval stage of each chironomid species were based on the literature length-weight regression equations (Tolonen et al. 2016). The taxa were divided by their size into one of four size divisions: < 0.3, 0.3–3, 3–10 and > 10 mg, following the size classification in Tolonen et al. (2003). The sum of the relative proportions of each taxa belonging to a certain size class represented the total relative proportion of that size class. Then, the taxa were grouped by their activity modes: crawler, clinger, and burrower. Lastly, the sizes and activity modes were combined into 12 pairings and correlated with fish biomass and density. The substrate attachments were classified according to Tolonen et al. (2016).

2.6. Redundancy analyses and variation partitioning

A redundancy analysis (RDA) was run using R software to visualize the differences in chironomid assemblages of the study lakes in comparison to their fish abundance. An RDA ordination was chosen because it uses scores that are constrained by linear combinations of the explanatory variables (ter Braak & Smilauer 2014). Variance partitioning was used to evaluate unique and shared effects of three variable groups: i) water chemistry, ii) lake morphometry/landscape positioning, and iii) fish biomass/density on the chironomid assemblages. First, to avoid problems of multicollinearity among explanatory variables, one variable considered to be the most important was selected for subsequent analyses among the correlated variable sets (r > 0.6) within each explanatory parameter group (e.g. water chemistry). Second, using statistical package vegan in R (Oksanen et al. 2013), RDA with a forward selection of explanatory variables was run (Blanchet et al. 2008) with an ordiR2step function to select significant variables within each of the three factor groups. Third, RDA with variation partitioning was run using varpart function in R package vegan to comprise fractions uniquely explained by each explanatory variable group, shared proportions explained by different variable groups together, and unexplained variation. The analyses were completed using the Arcsin-Sqrt-transformed proportional chironomid taxa abundances.

3. RESULTS

3.1. Correlations between individual chironomid taxa and fish abundance

The average number of taxa identified in the study lakes was 20, whereas taxon richness ranged from 11 to 28 (Table 1). There were 24 common taxa, which occurred in at least four lakes, and their proportions were correlated with fish density and/or biomass (Table 3). Of these taxa, four showed a positive significant correlation with fish abundance. *Corynocera oliveri* showed a positive correlation ($r_s = 0.62$) with fish density, and occurred in six lakes, which had a fish density ranging from 46.6 to 100 individuals gill-net^{-1} day^{-1} (Fig. 1 A). *Lauterborniella*, a small (< 0.3 mg) crawler, correlated positively with fish density ($r_s = 0.45$), and appeared to prefer lakes with a medium fish abundance, abundant in the deep lakes of Hacksjön, Klintsjön, and Blanksjön (≥ 16 m maximum depth), and in Hultben (3 m maximum depth) (Fig. 1 B). *Protanypus* correlated positively with fish density $(r_s = 0.50)$, and was only found in Lunksjön, Klintsjön, Hultbren, Humelsjön, and Hacksjön, which ranged in fish density from 26.0 to 191.1 individuals gill-net^{-1} day^{-1} (Fig. 1 C). *Chironomus plumosus* correlated positively with fish biomass $(r_s = 0.46)$, and was abundant in Bergsjön, Lunksjön, Humlesjön, and Stora Slätten (Fig. 1 D).

Two chironomid species, *Z. zalutschicola* and *Sergentia coracina,* showed significant negative correlations with fish abundance. *Z. zalutschicola* showed strongest negative correlation with fish density, ($r_s = -0.61$) and had the highest abundances in five of the six lakes with lowest fish abundances (Fig. 1 E). Brändasjö, Stavsjön, Liasjön, Gölasjön, Ubbasjön, Gyslattäsjön are all relatively small (area 3–32 ha) and shallow, with a maximum depth between 4–10 m, a secchi depth less than 2, and a pH ranging from 4 to 6. *S. coracina* $(r_s = -0.46)$ also had highest abundances in lakes with low fish abundance (Fig. 1 F), constituting large proportions (6–50 %) in Lillesjö, Klintsjön, Gyslattäsjön, Gölasjön, Hacksjön (0–26.1 individuals gill-net^{-1} day^{-1}). These lakes vary in their morphometry and water chemistry, and the only common factor is low fish abundance.

Table 3. The estimated optima (u), tolerances (t) and correlations (Spearman correlation coefficients, r_s) for fish density and biomass for the most frequent chironomid taxa (n = number of lakes from which the species were found).

Species	Density r_s	p	u	.t	Biomass r_s	p	u	.t	n
Ablabesmyia	-0.097	0.675	25	23	-0.004	0.988	959	959	18
Procladius	0.175	0.449	32	24	0.177	0.442	1,064	776	21
Cricotopus cylindraceus	0.170	0.460	43	30	0.068	0.769	1,305	659	10
Heterotanytarsus	-0.367	0.101	15	17	-0.258	0.260	658	714	14
Heterotrissocladius*	-0.094	0.686	26	18	0.044	0.848	1,253	894	12
Parakiefferella*	0.201	0.381	32	27	0.136	0.557	1,154	694	14
Psectrocladius*	-0.299	-0.187	26	18	-0.146	0.527	2,507	901	14
Zalutschia zalutschicola	-0.609	0.003	6	6	-0.597	0.004	214	279	7
Cladopelma lateralis	0.106	0.649	23	25	-0.022	0.924	695	703	12
Dicrotendipes nervosus	0.243	0.289	34	26	0.211	0.358	1,269	932	15
Harnischia	0.174	0.451	42	29	0.126	0.585	1,228	666	7
Lauterborniella	0.449	0.041	44	17	0.397	0.075	1,471	507	5
Microtendipes pedellus	0.073	0.752	32	28	0.101	0.664	1,115	865	15
Pagastiella	0.089	0.701	29	18	0.043	0.853	1014	587	8
Polypedilum I.	0.374	0.095	31	28	0.276	0.225	1,000	703	13
Sergentia coracina	-0.457	0.037	12	15	-0.427	0.054	400	546	10
Cladoytanytarsus mancus	0.391	0.080	37	26	0.259	0.257	1,170	664	15
Corynocera oliveri	0.619	0.003	59	16	0.491	0.024	1,179	111	6
Micropsectra	0.394	0.077	31	22	0.230	0.315	1,011	687	13
Stempellina	0.292	0.199	34	25	0.237	0.302	1,171	685	14
Chironomus anthracinus	-0.358	0.111	24	30	-0.402	0.071	674	735	15
Chironomus plumosus	0.333	0.140	59	18	0.457	0.037	1,835	253	4
Protanypus	0.497	0.022	46	20	0.489	0.024	1,603	459	5
Tanytarsus	0.322	0.155	31	27	0.229	0.317	1015	746	21

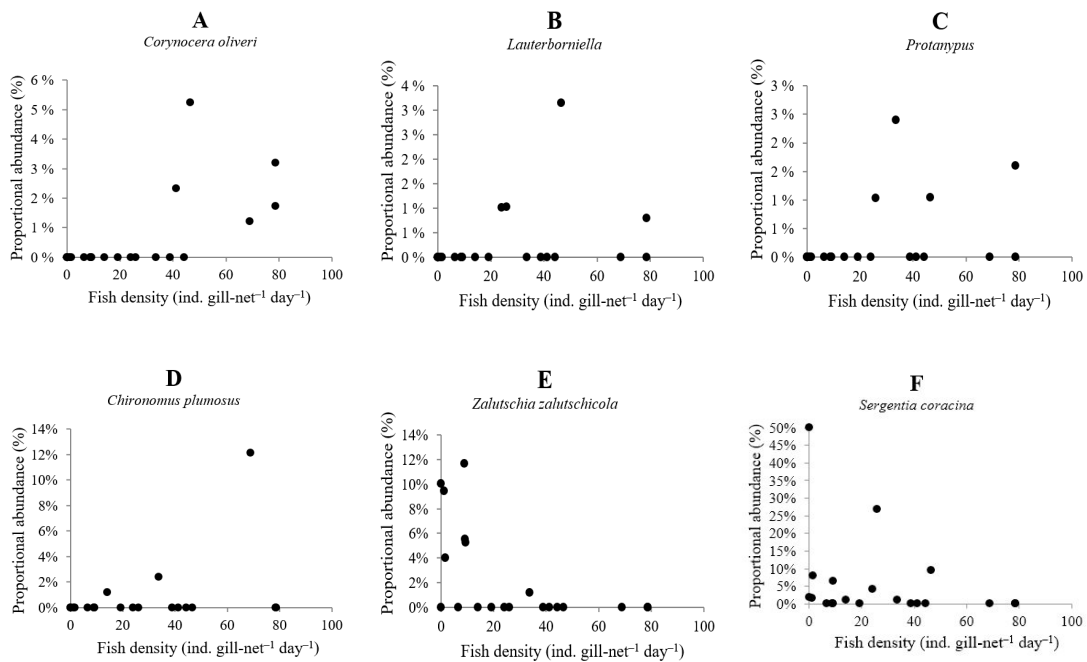*Genus optimum and tolerance is an average of two species.

Figure 1. The proportion (%) of A) *Corynocera oliveri*, B) *Lauterborniella*, C) *Protanypus*, D) *Chironomus plumosus*, E) *Zalutschia zalutschicola* and F) *Sergentia coracina* in each lake plotted against the lake's corresponding fish density (CPUE).

3.2. Chironomidae weighted averaging model

For both the initial Chironomidae WA density and biomass models, Humlesjön's inferred abundance was less than 25 % of the observed fish abundance. Tolonen et al. (2012) considered Humlesjön to be an outlier in their regression models using *Chaoborus* mandible concentrations as the only predictor. Tolonen et al. (2012) used the observed fish abundances to be fitted against leave-one-out predicted abundances to evaluate Humlesjön, which they considered was an outlier after using Grubb's test for outlier detection. For these reasons, Humlesjön was considered to be an outlier and was not included in the final Chironomidae WA models.

The fish density inferred by the WA model explained 65 % of the variation in the observed fish density (Fig. 2 A), with a mean absolute error (MAE) of 11 individuals. The biomass WA model was slightly less precise, explaining 60 % of the observed variation (Fig. 2 B) with MAE of 363g per gill-net^{-1} day^{-1}. When the Chironomidae and *Chaoborus* models were combined, the density model slightly improved, ($R^2 = 0.70$, MAE = 11 individuals). The fish biomass model also improved, inferring the same amount as the density model ($R^2 = 0.70$, MAE = 367g).
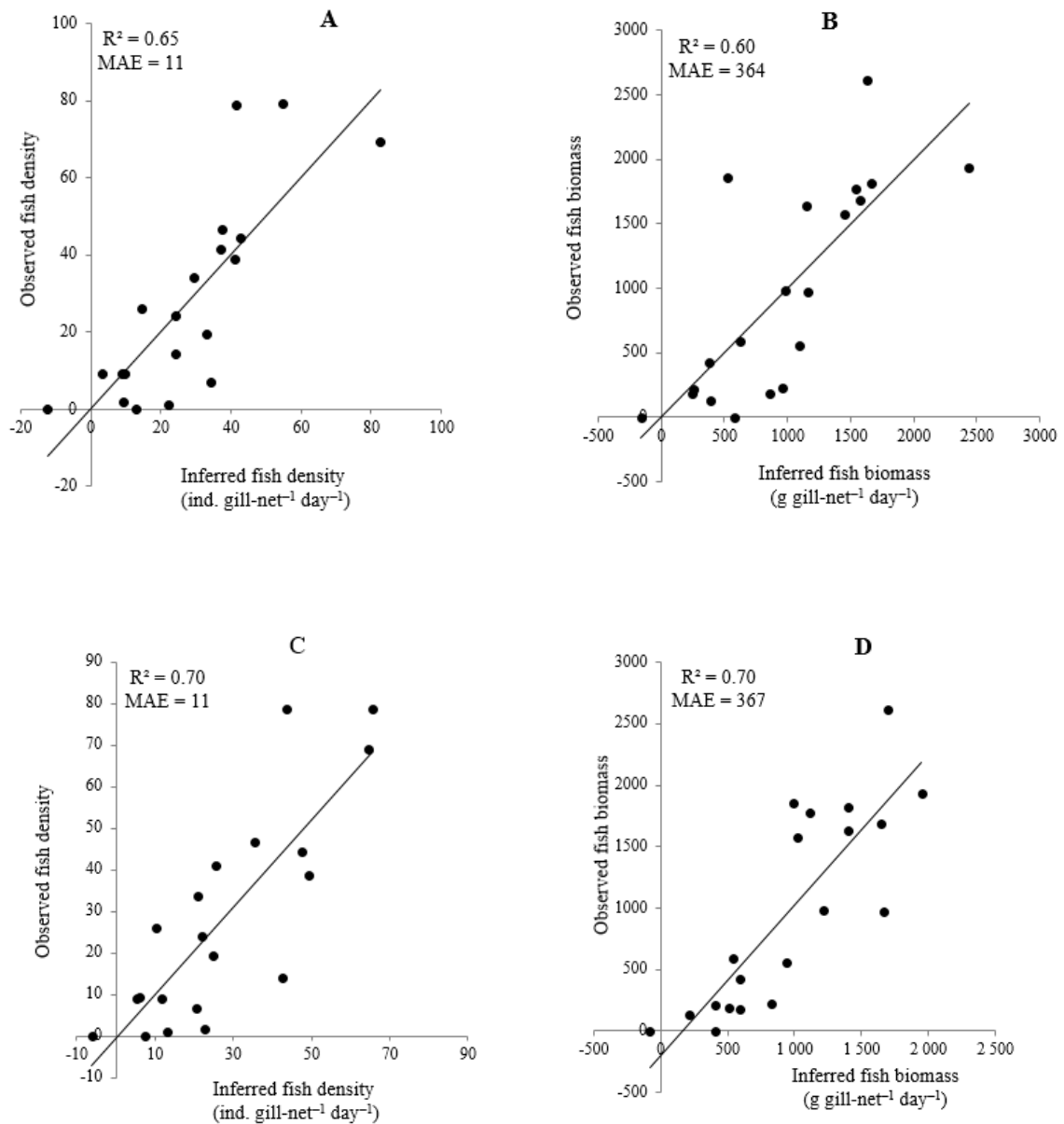
Figure 2. Relationship between the inferred and observed fish density (A, C) and biomass (B, D) using Chironomidae only (A, B), and the average of Chironomidae and *Chaoborus* models (C, D).

3.3. Correlation of chironomid species traits with fish abundance

Four size and activity mode pairings showed significant correlations with fish abundance (Table 4). The smallest (< 0.3 mg) tube-dwelling clingers, including *Paralauterborniella* and *Cladoytanytarsus mancus*, correlated positively with fish density and appeared to be more abundant in lakes of low and medium fish density (Fig. 3 A). Small (0.3–3 mg) crawlers also showed a positive correlation with fish biomass, and made up about 25–75 % of the taxa in each lake (Fig. 3 B). This group encompassed a large number of genera including *Ablabesmyia*, *Tanytarsus*, *Micropsectra*, *Psectrocladius*, *Polypedilum* and *Heterotrissocladius*. The medium (3–10 mg) burrowers comprised of *Demicryptochironomus*, *Einfeldia*, *Stictochironomus* and *Protanypus* showed a positive correlation with both fish density and fish biomass (Fig. 3 C). The medium (3–10 mg) crawlers including *Macropelopia*, *Cryptochironomus* and *Parachironomus* showed a significant negative correlation with both fish density and fish biomass (Fig. 3 D).

Although not significant, the largest (> 10 mg) burrowers including *Chironomus*, *S. coracina*, *Glypotendipes* and *Stenochironomus* showed a negative correlation with fish abundance. Small burrowers such as *Z. Zalutschicola*, *Dicrotendipes*, *Cladopelma lateralis*, and the smallest crawlers including *Corynoneura*, *Stempellina* and *Parakiefferella* were all negatively, but not significantly, correlated with fish abundance.

Table 4. Spearman correlations (r_s) between taxa pairings and fish density and biomass.

Size (mg) + Mode	Density		Biomass		
	r_s	*p*	r_s	*p*	**n**
< 0.3 Crawler	-0.068	0.774	-0.076	0.750	20
< 0.3 Clinger	0.459*	0.042	0.298	0.202	20
0.3–3 Burrower	-0.235	0.319	-0.257	0.274	20
0.3–3 Crawler	0.429	0.059	0.545*	0.013	20
0.3–3 Clinger	0.234	0.320	0.037	0.877	20
3–10 Burrower	0.444*	0.050	0.548*	0.012	20
3–10 Crawler	-0.451*	0.046	-0.454*	0.044	20
3–10 Clinger	0.076	0.751	0.114	0.633	20
> 10 Burrower	-0.271	0.248	-0.334	0.150	20
> 10 Crawler	0.060	0.803	-0.020	0.934	20

The proportions of taxa sizes differed within lakes of varying fish abundance. Of the two fishless lakes, Lillesjö is dominated by large taxa (Fig. 4). On the other end of the spectrum, lakes with high fish abundance had low abundances of large taxa, with higher abundances of small taxa. The lakes in the middle range appeared to have a substantial amount of the smallest taxa, as well as medium-sized taxa. In all lakes, the small (0.3–3 mg) taxa are most abundant, but this is because a majority of the taxa (43 species) are included in the small category, compared to the other size categories ranging from 7–17 species (Appendix 1). The category Ungrouped, represented 0–8 % of the total taxa in each study lake, included head capsules that could not be identified to a genus and were categorized into their appropriate subfamily or tribe (Chironominae, Orthocladiinae, Tanypodinae and Tanytarsini).
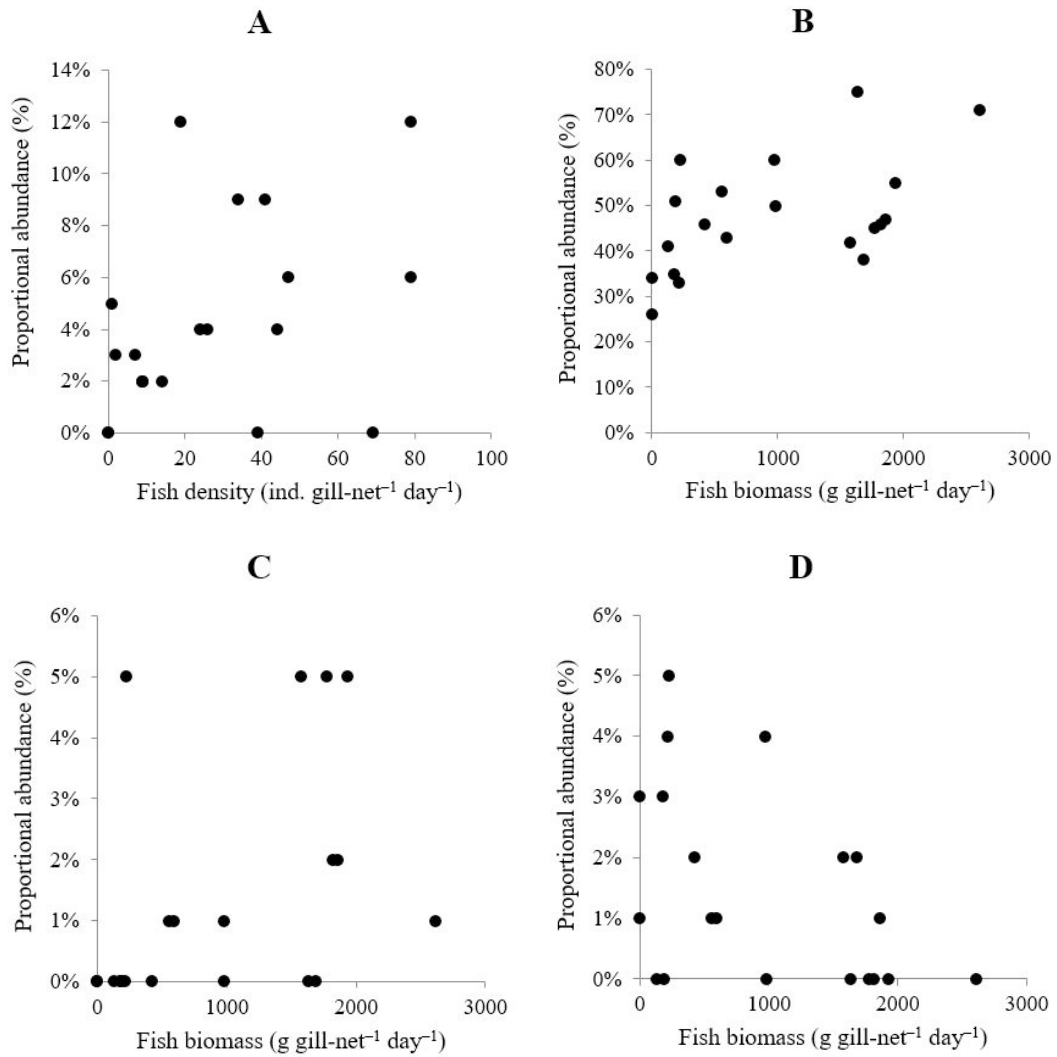
Figure 3. The proportion (%) of A) < 0.3 mg clingers, B) 0.3−3 mg crawlers, C) 3−10 mg burrowers and D) 3−10 mg crawlers in each lake plotted against the lake's fish density or biomass (CPUE).
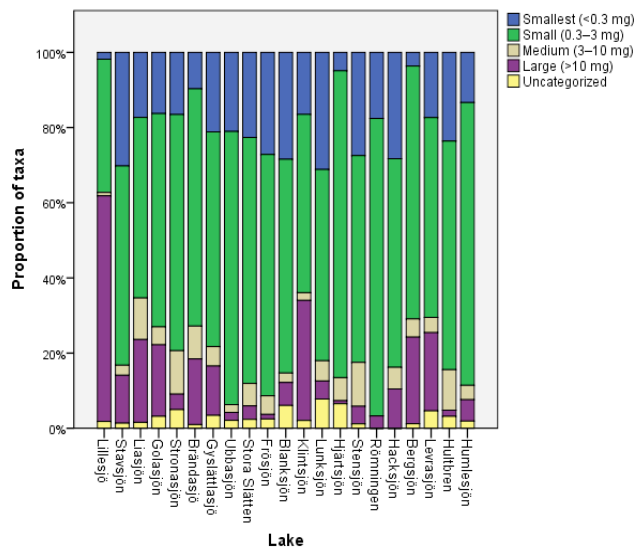

Figure 4. The proportions of chironomid larval size categories in each lake arranged in ascending order of fish abundance.

3.4. Redundancy analyses and variation partitioning

Roach and ruffe density were selected as the only significant (p < 0.05) variables among the fish abundance parameters (other candidate variables were total fish, perch, bream, rudd and tench densities) in the first phase RDAs with forward selection. Relative depth was the only significant (p < 0.05) variable among the lake morphometry parameters. Among the water quality variables, the species composition of chironomids was significantly (p < 0.05) associated with Secchi depth and pH.

The RDA ordination combining significant variables of each three variable group explained significant (p < 0.001) amount of variation in the chironomid species composition of study lakes (Table 5). In the RDA ordination of chironomid assemblages, relative depth and roach density correlated strongly with the first axis (Fig. 5). Secchi depth and ruffe density associated most strongly with the third axis. Stavsjön and Lillesjö, the two fishless lakes are separated from the other lakes along the first ordination axis. The lakes with varying fish density form clear clusters along the first ordination axis, appearing to differ from one another in their amount of ruffe and roach, pH, and Secchi depth. Lakes of medium (10–40 ind. gill-net^{-1} day^{-1}) fish density seem to form a group along the second axis correlated with ruffe density and Secchi depth. Although the lake groupings of high, medium and low fish abundance overlap with each other, the fishless lakes appear to have their own distinct grouping, indicating their different chironomid assemblages.

Table 5. Statistics for the RDA Ordination axes and the variation partitioning.

Axes	1	2	3	4	5	P-value	df
Eigenvalues	0.069	0.0387	0.030	0.018	0.016		
Proportion Explained	0.404	0.226	0.175	0.103	0.093		
Cumulative Proportion	0.404	0.630	0.805	0.907	1.000		
Biplot scores (axis correlations) for constraining variables:							
Secchi depth	0.323	0.506	0.769	0.221	0.002	0.011*	1
Relative depth	0.867	0.023	-0.007	-0.460	0.196	0.024*	1
pH	-0.554	0.455	0.445	-0.531	0.073	0.037*	1
Ruffe density	-0.451	-0.407	0.713	-0.225	-0.269	0.006**	1
Roach density	-0.774	0.140	0.260	0.004	0.560	0.392	1
Residual							15

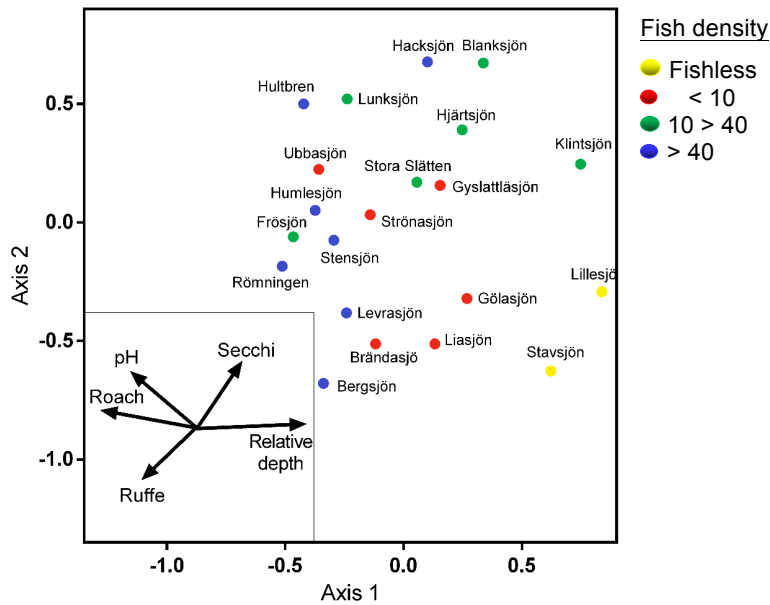Significance codes: 0 '***' 0.001 '**' 0.01 '*'

Figure 5. RDA ordination of the chironomid assemblages in study lakes, with differently colored symbols indicating different levels of fish density (ind. gill-net^{-1}day^{-1}). The environmental variable arrows are showing the correlations between the ordination axes and environmental parameters.

The variation partitioning based on RDA suggested that water chemistry, morphometry and fish density all have an independent effect on the chironomid taxonomic composition (Fig. 6). Water chemistry factor had an independent influence of 7 %, morphometry 3 %, and fish density 4 % on chironomid assemblages. The shared effects between all variable groups and between morphometry and fish density were 3 % and 2 %, respectively. Unexplained residual variation was the remainder 82 %.
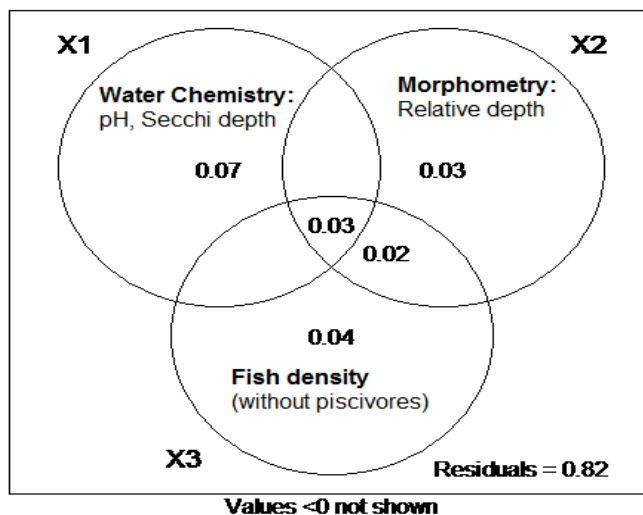

Figure 6. Venn diagram of RDA variation partitioning showing the individual (unique) and shared fractions explained (the adjusted r squared values) by the three factors: water chemistry, lake morphometry, and fish density, of the variation in the chironomid assemblage composition.

4. DISCUSSION

The Chironomidae WA model inferred reasonably well the fish abundances of the 20 study lakes. The Chironomidae models performed well on their own, and their performance was apparently better than the model based on *Chaoborus* and morphometry predictors (c.f. Tolonen et al. 2012: fig. 5). Chironomidae models improved slightly when combined with *Chaoborus* models. These combined Diptera models explained both 70 % of the fish abundance, and accounted for 5 % and 10 % more of the fish density and biomass, respectively, than the Chironomidae model alone. Hence, the accuracy of *Chaoborus* presence/absence models developed earlier (Tolonen et al. 2012) could be improved by also examining chironomid composition. Previous studies have indicated shortcomings in the presence/absence model due to a finding a limited number of *Chaoborus* mandibles (Palm et al. 2011). *Chaoborus* are usually associated with eutrophic or dystrophic conditions, and are less abundant in oligotrophic lakes (Thienemann 1925). Incorporating Chironomidae into *Chaoborus* models could possibly allow the model to be used in a broader variety of lake types, with a better sense of validation in scenarios when there is a lack of *Chaoborus* mandibles.

One limitation of the present data and the models developed is the shortage of study lakes with high fish abundance. The lake with the highest fish abundance within the data set was Humlesjön, which unfortunately was considered an outlier and removed from the Chironomidae WA models. Tolonen et al. (2012) hypothesized that the high near-bottom-turbidity caused by sediment resuspension of benthivorous fish provided refuge for the phantom midges to avoid predation, and this could also have been true for chironomids. In both the Chironomidae model and the *Chaoborus* model, the inferred estimate was severely lower than the observed fish abundance in the lake. After removing Humelsjön, the lake with the second highest fish abundance of 78 ind. gill-net^{-1} day^{-1} became the top range within the model. Therefore, it is possible that reliability of this model is limited to only small to medium–sized (3–383 ha) temperate lakes with a fish density less than 100 ind. gill-net^{-1} day^{-1}.

Of the six taxa that significantly correlated with fish abundance, all but *Z. zalutschicola* and *C. plumosus* tend to prefer the littoral zone of lakes (Brooks et al. 2007). *Z. zalutschicola* has been recognized as an acid indicator (Brodin 1986, Uutala 1986, Brooks 2005), and its high abundance in these lakes may be linked to the acidified environment, which consequently is also the main factor responsible for the expiring of fish. *Phaenopsectra*, which has also been recognized as an acid indicator (Brodin 1986, Brooks 2005), had the highest abundances in Stavsjön (5 %), and Brandasjö (5 %), also sharing high abundances of *Z. zalutschicola*. *C. plumosus* was likely able to coexist with fish due to its activity mode of burrowing, providing protection from fish predation, despite its large size.

Correlations between fish abundance and the species traits (body size and activity mode pairings) were only significant for the smaller-sized chironomid taxa pairings. This may possibly indicate that the abundances of small- and medium-sized free-living chironomids are more influenced by fish abundance than the larger-sized chironomids. Although some researchers, as cited by Diehl (1992), have reported significant effects on the biomass, abundance, and diversity of benthic macroinvertebrates by benthivorous fish (Crowder & Cooper 1982, Morin 1984, Mittelbach 1988, Gilliam et al. 1989, Power 1990), other researchers of mainly lotic studies have found little effect of fish on benthic communities (Thorp & Bergey 1981, Allan 1982, Reice 1983, Flecker & Allan 1984). In

the case when benthic invertebrates are depleted, Crowder & Cooper (1982) thought it was due to their behavior and movement, rather than their size (also supported by Diehl 1992).

In the present study, medium sized (3–10 mg) burrowers appeared more protected from fish predation, and they correlated positively with fish abundance. This group also included two of the six species that correlated significantly with fish abundance, *Corynocera oliveri* and *Protanypus. Protanypus, Stictochironomus, Einfeldia dissidens* and *Prodiamesa* are all characteristic inhabitants of the littoral zones of lakes (Brooks 2007). Exhibiting an opposite trend to the medium-sized burrowers, the medium-sized (3–10 mg) crawlers negatively correlated with fish abundance, likely due to their activity causing them to be more vulnerable to fish predation. The largest burrowers (> 10 mg) showed a weak negative correlation with fish abundance. However, *C. plumosus*, the large profundal burrower, correlated significantly with fish abundance, and was absent from fishless lakes. The smallest burrowers (0.3–3 mg) also showed a weak negative correlation with fish abundance. The lack of a significant correlation for both the small- and large-sized burrowers may indicate that there are other factors influencing their abundance, and fish abundance may not be a major factor.

The smallest (< 0.3 mg) clingers correlated positively with fish abundance. The small proportion of large taxa in lakes with high fish abundance is likely a result of fish predation, benefitting the smallest taxa, as hypothesized by Uutala (1986) and Langdon (2006). These small sized taxa are not favored prey by fish. Small-sized taxa may also be indirectly "protected" by fish, which feed on large-sized predatory chironomids and other invertebrate predators, such as Odonata and Sialidae larvae, which feed on smaller invertebrates including chironomids. Crowder & Cooper (1982) also observed fish to alter the invertebrate community composition by removing large invertebrate predators (Odonata) and herbivores (Hyalella) to benefit smaller forms. This also supports Gilinsky's (1984) finding of the predatory, large chironomid, *Clinotanypus*, showing a negative response to fish predation, contributing to an increase of non-predatory macroinvertebrates. Gilinsky (1984) recognized the decline of *Clinotanypus* could be due to competition with other dominant predaceous chironomids in this system, including *Procladius*, but found no significant correlations between any of the three chironomids in her study, suggesting that predation between chironomids is not important.

The smallest crawlers (< 0.3 mg) comprised of *Corynoneura, Stempellina, Parakiefferella* and 9 other taxa showed a negative correlation with fish abundance, although not significant. *Corynoneura*, a free-living chironomid was inferred by Uutala (1986) and Langdon et al. (2006) to be more susceptible to fish predation and larger chironomid predation. In this study, *Corynoneura* was found in high abundances in Ubbasjön of low fish abundance and fishless Stavsjön, but also in Levrasjön of high fish abundance. Although the smallest crawler category correlated negatively with fish abundance, a few of the smallest taxa in this size category appeared more abundant in lakes of fish abundance. *Stempellina*, weighing 0.15 mg, was most frequently found in lakes of medium fish abundance (between 19.2 and 78.8 ind. gill-net^{-1} day^{-1}), absent from fishless lakes, and found in small proportions from only one lake of low fish abundance. *Parakiefferella*, weighing 0.06 mg, showed similar patterns.

The small 0.3–3 mg crawlers comprised of 28 taxa including *Tanytarsus, Micropsectra, Psectrocladius, Polypedilum* and *Procladius*, showed a significant positive correlation with fish abundance. Despite the correlation, it is difficult to assess to what degree the taxa in this group are truly influenced by fish abundance because they constitute a major proportion in every lake. One genera of this group, *Procladius* was hypothesized to

be more abundant in fishless lakes than lakes with fish because they are susceptible to fish predation due to their relatively large size and high activity (Serra-Tosio 1978, Uutala 1986, Heiri & Lotter 2003, Langdon et. al 2006). However, *Procladius* showed no significant trend with fish abundance. *Procladius* had the highest proportions, ranging from 13–27 %, in lakes of high and medium fish abundance, while the five lowest proportions were from lakes with varying of fish abundance. Hence, if *Procladius* is sensitive to fish predation, it was not evident in these study lakes.

The size categories that correlated positively with fish abundance were of relatively small size. It is possible that the smaller taxa of the littoral zone may be more dependent on fish for survival. Diehl (1992) cited that in the presence of fish, large predatory invertebrates are the first excluded or greatly reduced in the pelagic zone (Stenson 1978, Erikson et al. 1980, Evans 1989), in contrast to large benthic predatory invertebrates, which can coexist with fish if they exhibit the appropriate behaviors (Morin 1984, Blois-Huelin et al. 1990). Diehl (1992) indicated one could hypothesize the degree of omnivory and competition between invertebrate-feeding fish and predatory invertebrates is higher in the littoral benthic than in pelagic communities. This hypothesis supports my findings of smaller chironomid size groupings showing stronger correlations with fish abundance than the other larger size groupings, and is also align with my observation that many taxa of these groupings are characteristic of the littoral zone. If this hypothesis were true, chironomid taxa of the littoral zone may be most appropriate and precise for modeling fish abundances.

The RDA ordination shows that the chironomid assemblages in lakes with abundant fish populations are distinct from the assemblages in fishless lakes. Although water chemistry variables showed the strongest unique effects on the chironomid species composition, the apparent independent influence of fish was also significant, and apparently stronger than that of lake morphometry. Associations between chironomid taxa and certain water chemistry and lake morphometry properties have been studied in depth, including reconstruction methods such as chironomid transfer functions to infer total phosphorus, total nitrogen, and chlorophyll-*a* (Brodersen & Quinlan 2006). Other taxa have been associated with lake morphometry characteristics, specifically lake depth by Brundin (1958), Sæther (1975), Walker & Mathewes (1989), Walker & MacDonald (1995), and Brodersen & Quinlan (2006).

5. CONCLUSIONS

Chironomids have been used extensively for reconstruction of environmental conditions including e.g. water chemistry, but not previously for reconstructing fish populations. Incorporating Chironomidae remains could potentially serve as a revolutionary new way to assess if a fish population in a given lake has changed over a specific time period. This tool would be crucial for setting reasonable restoration targets for fish in lakes that have experienced negative changes due to a human influence.

The WA model constructed in this study proves that chironomid subfossils may have the potential to serve as reliable predictors of fish abundance. Chironomidae species responses and species traits were further examined to better decipher why certain species reacted positively or negatively to fish. This study suggests the relationships of the studied species traits (body size and activity mode) with fish abundance may stem not only from direct predation effects by fish, but also from indirect effects through large invertebrate predators. Small-sized clingers and crawlers showed a significant positive correlation with fish abundance, likely because fish "protect" them by feeding on the large-sized

invertebrates, potential predators of small-sized chironomids (Crowder & Cooper 1982, Uutala 1986, Diehl 1992, Gilinsky 1992). However, the patterns of body size and activity mode observed in this study provided rather weak support for the hypothesized fish predation effects on chironomid species traits. In addition, the RDA test showed at least a fraction of the chironomid taxa community variation can be linked with fish, independent of environmental factors.

Overall, it is apparent that chironomid taxon composition can be used to infer fish abundance in lakes. Past studies have repeatedly confirmed that chironomid composition differs between fishless lakes and lakes with fish (Gilinsky 1984, Uutala 1986, Johnson et al. 1990, Langdon et al. 2006). This study shows that use of Chironomidae remains can be expanded to reconstructing fish abundances in lakes. Although this model has not been tested using independent spatial and/or temporal data, the performance of this model shows that there is a potential to be used for paleolimnological applications. Lastly, further studies examining Chironomidae predator-prey dynamics could help to explain the indirect and direct effects responsible for the differing taxa compositions between fishless and fish abundant lakes. By studying predator-prey dynamics and other biotic factors affecting the abundance and species composition of chironomids, the WA modeling approach could be made more precise in its ability to assess fish abundance.

ACKNOWLEDGEMENTS

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APPENDIX 1.

List of all species and genera used in the Spearman correlation size and activity mode pairings. Genera written according to Wiederholm (1983), species level written according to (Brooks et. al 2007).

< 0.3 Crawler	0.3−3 Crawler	3−10 Burrower
Corynoneura coronata-type (0.02)	*Rheocricotopus effuses* (0.39)	*Demicryptochironomus* (3.06)
C. lacustris	*Eukiefferiella claripennis* (0.51)	*Einfeldia dissidens*-type (3.56)
C. lobata	*Apsectrotanypus* (0.66)	*Stictochironomus* (4.11)
Stempellinella – Zavrelia (0.04)	*Micropsectra*	*Protanypus* (4.73)
Thienemanniella clavicornis-type	*Paratanytarsus – Tanytarsus*	*Prodiamesa* (6.15)
Heterotanytarsus (0.06)	*Pentaneurella katterjokki*	
Parakiefferiella bathophila	*Telmatopelopia*	**3−10 Crawler**
P. smolandica	*Procladius* (1.03)	*Parachironomus frequens* group (3.06)
Synorthocladius	*Hydrobaenus*	*P. vitiosus*
Lauterborniella	*Harnischia*	*P. varus*
Djalmabatista (0.15)	*Tanytarsus*	*Macropelopia* (4.73)
Limnophyes – Paralimnophyes	*T. pallidicornis*-type	*Cryptochironomus* (6.96)
Stempellina	*T. lactescens*-type	*Monodiamesa*
Monopelopia (0.29)	*T. chinyensis*-type 1.	
	T. mendax-type	**3−10 Clinger**
< 0.3 Clinger	*T. lugens*-type	*Microtendipes pedellus*-type (6.96)
Pagastiella (0.06)	*Heterotrissocladius grimshawi* (1.27)	*Endochironomus* (8.80)
Paralauterborniella (0.15)	*H. marcidus*	
Cladotanytarsus mancus-type 1.	*Polypedilum* I.	**> 10 Burrower**
	Polypedilum II.	*Glyptotendipes* (10.96)
0.3−3 Burrower	*Polypedilum* V.	*Sergentia coracina*-type.
Paratrissocladius (0.39)	*Chaetocladius*-type B (1.54)	*Stenochironomus* (10.96)
Paratendipes nudisquama-type	*Natarsia*	*Chironomus* 1st Instar (37.76)
Cladopelma lateralis-type (0.51)	*Paracladopelma*	*C. anthracinus*-type
Zalutschia zalutschicola	*Ablabesmyia* (2.21)	*C. plumosus*-type (50.15)
Chernovskiia Orbicus (0.66)	*Psectrocladius sordidellus*-type	
Microchironomus	*P. (Monopsectrocladius) septentrionalis*-type	**> 10 Crawler**
Dicrotendipes nervosus-type (2.21)	*P. (Mesopsectrocladius) barbatipes*-type	*Anatopynia* (16.38)
D. notatus-type		
Pseudochironomus		**Uncategorized**
Tribelos (2.61)	**0.3−3 Clinger**	*Omisus caledonicus* (2.21)
	Corynocera oliveri-type (0.51)	*Microcricotopus*
	Cricotopus (0.83)	*Pentaneura*
	C. cylindraceus-type	Chironominae subfamily
	C. (Isocladius) intersectus-type	Orthocladiinae subfamily
	Phaenopsectra (1.27)	Tanypodinae subfamily
		Tanytarsini tribe

Potential maximum dry body weight in parentheses according to Tolonen et al. (2016).