



Long term trends of fish after liming of Swedish streams and lakes



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HIGHLIGHTS

- Fish was monitored at 1779 lakes and streams up to 42 years since first liming.
- Large-scale liming and monitoring revealed clear improvement at the national scale.
- Stream fish occurrence, species richness and abundance increased after liming.
- Few acid sites were left non-limed to permit consistent monitoring at reference sites.
- Species richness and abundance did not improve in lakes as in streams after liming.

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ABSTRACT

Thousands of Swedish acidified lakes and streams have been regularly limed for about 30 years. Standard sampling of fish assemblages in lakes and streams was an important part of monitoring the trends after liming, i.e. sampling with multi-mesh gillnets in lakes (EN 14757) and electrofishing in streams (EN 14011). Monitoring data are nationally managed, in the National Register of Survey test-fishing and the Swedish Electrofishing Register. We evaluated long-term data from 1029 electrofishing sites in limed streams and gillnet sampling in 750 limed lakes, along with reference data from 195 stream sites and 101 lakes with no upstream liming in their catchments. The median year of first liming was 1986 for both streams and lakes. The proportion of limed stream sites with no fish clearly decreased with time, mean species richness and proportion of sites with brown trout (*Salmo trutta*) recruits increased. There were no consistent trends in fish occurrence or species richness at non-limed sites, but occurrence of brown trout recruits also increased in acid as well as neutral reference streams. Abundance of brown trout, perch (*Perca fluviatilis*) and roach (*Rutilus rutilus*) increased significantly more at limed sites than at non-limed reference sites sampled before and after 1986. The mean species richness did not change consistently in limed lakes, but decreased in low alkalinity reference lakes, and fish abundance decreased significantly in limed as well as in non-limed lakes.

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1. Introduction

Thousands of Swedish lakes and streams have been regularly treated with limestone to mitigate acidification (Svenson et al., 1995). Liming became a large-scale and governmentally supported restoration program in the 1980's, and the number of local liming projects increased at the same time as large-scale measures were taken to decrease airborne emissions of sulfur and nitrogen compounds. The annual amount of limestone spread peaked at

more than 200,000 tons during 1998–2002. As an adjustment to decreasing acidification, the amount of lime decreased and recently stabilized at about 120,000 tons per year (Abrahamsson et al., 2013).

Deposition of sulfur and nitrogen has now decreased over Europe, although non-linearly related to reduced emission at regional scale (Fowler et al., 2007). Swedish soils are just slowly becoming less acid (Akselsson et al., 2013). Similarly, sulfate and acidity has decreased in streams and lakes (Fölster and Wilander, 2002; Futter et al., 2014), but many of the non-limed lakes are still acidified as compared to estimated reference pH. Slow and insufficient chemical recovery is probably one reason for slow or inconsistent biological recovery in Swedish lakes (e.g. Angeler and

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Johnson, 2012; Holmgren, 2014), but there are relatively few reports of evidence for partial biological recovery in non-limed streams (e.g. Monteith et al., 2005; Kowalik et al., 2007; Battarbee et al., 2014; Murphy et al., 2014).

Acid precipitation led to recruitment failure, declining abundance and eventually extinction of many fish populations in Scandinavia and North America (Schofield, 1976). Experiments revealed that fish species differ in sensitivity to low pH and acidity-associated aluminum (e.g. Skogheim and Rosseland, 1984; Poleo et al., 1997), and that reproductive and early life stages were most critically affected (e.g. Rask, 1984; Mc Cormick and Leino, 1999).

A main goal of the liming programs in Sweden was initially to restore waters identified as being of special value for fish and fishery (Bengtsson et al., 1980). Fish was also targeted in large-scale liming in Norway (Sandøy and Romundstad, 1995), and in other more recent plans to promote recovery of acidified waters (e.g. Josephson et al., 2014). Site-specific chemical targets for liming in Sweden currently focus on the most acid-sensitive species (Abrahamsson et al., 2013), in practice often Atlantic salmon (*Salmo salar*) in rivers, and roach (*Rutilus rutilus*) in lakes.

General results of liming projects have been reviewed (e.g. Henrikson and Brodin, 1995; Clair and Hindar, 2005), and reports on biological impacts of liming were recently synthesized both for rivers and streams (Mant et al., 2013) and lakes (Mant and Pullin, 2012). Positive effects on fish included increased diversity, with or without re-stocking of lost populations, and sometimes increased abundance or improved reproductive success. However, many reported liming projects were non-replicated case studies, without baseline and/or control sites, and often of short duration in terms of lime treatment and/or monitoring.

Bradley and Ormerod (2002) stressed the need for long-term monitoring of liming of acidified surface waters. The large-scale Swedish liming program offers a unique possibility to evaluate long term effects on fish at a national scale. Governmental funds support not only regular lime treatment, but also chemical and biological monitoring with standard methods (e.g. CEN, 2003; CEN, 2015), as long as the monitoring results are reported to national data hosts. Results from some Swedish fish studies of varying duration were included in the reviews mentioned above (e.g. Degerman and Appelberg, 1992; Degerman et al., 1992; Holmgren, 2001), but a national-scale analysis over more than 30 years of liming has not previously been presented.

In this study, we extended our perspective from just a few well studied sites, to evaluate more extensive monitoring data in large national fish databases. We expected that positive national-scale effects of liming on fish populations would, in most cases, dominate over any local site-scale changes. More specifically, we expected increasing fish occurrence and species richness, due to re-establishment of species that were previously lost during acidification. We further expected more regular recruitment of acid-sensitive species, and overall increased fish abundance. All predicted responses were tested using electrofishing data from streams. Responses on fish species richness and abundance in lakes were similarly explored using data from gillnet sampling.

2. Material and methods

2.1. Stream data

The main part of this study was based on stream fish data stored in the Swedish Electrofishing Register (SERS, Sers, 2014), representing autumn samples on annual basis or less frequently, with each site covering a wadable stream reach of at least 100 m² (CEN, 2003). Abundance was expressed as number of fish per 100 m², within each species caught, separately for age 0+ and >0+ for

brown trout (*Salmo trutta*) and Atlantic salmon (inferred from length frequency distributions), and for the sum of all occurring species.

Electrofishing sites were initially selected based on the following criteria; 1) samples from at least five occasions, 2) time span of at least seven years between first and last sampling date, 3) first sampling performed before year 2000, and 4) no known stocking of fish.

Catchments of selected electrofishing sites were delimited by using national digital elevation data. Limed sites and their first year of liming were identified, and information on amounts of limestone spread in the catchments methods of liming was retrieved by matching of electrofishing sites with the national database on liming (<http://kalkdatabasen.lansstyrelsen.se>). County administration boards provided existing data on pH and alkalinity from limed as well as non-limed reference streams. Electrofishing sites in non-limed streams were kept if available data made it possible to classify them in one of three reference groups; 1) acid (mean pH < 6.0 or minimum pH < 5.4), 2) low alkalinity (minimum pH > 5.4 and mean alkalinity < 0.5 meq L⁻¹), or 3) high alkalinity (mean pH > 6.0 and mean alkalinity > 0.5 meq L⁻¹).

2.2. Lake data

An additional part of the study was on lake fish data from the National Register of Survey test-fishing (NORS, Kinnerbäck, 2015), including sampling with benthic multi-mesh gillnets in late summer (generally after mid-July or in August). The prevailing type of multi-mesh gillnet changed gradually through time, during 1968–1990 from Lundgren type S (12 panels, bar mesh 10–75 mm, total area 54 m²) to Lundgren revised type S (14 panels, bar mesh 6.5–75 mm, total area 63 m², Hammar and Filipsson, 1985), and from 1991 the older types were increasingly replaced by the Nordic type (12 panels, bar mesh 5–55 mm, total area 45 m², Appelberg et al., 1995), used in a depth-stratified random design according to the current European standard (CEN, 2015).

Lakes were first selected based on the following criteria; 1) multi-mesh benthic gillnet samples representing the whole lake and all of its available depths, 2) samples from at least two years, 3) time span of at least five years between first and last sampling date, and 4) first sampling performed before year 2000.

The dataset was further reduced to include only lakes with recorded status as limed or non-limed, and limed lakes were kept if the first year of liming was available in the fish database NORS. Available data on pH and alkalinity were retrieved from a national database managed by the Department of Aquatic Sciences and Assessment (<http://webstar.vatten.slu.se/db.html>). As for streams, non-limed lakes were kept if available data made it possible to classify them in one of three reference groups; 1) acid (mean pH < 6.0 or minimum pH < 5.4), 2) low alkalinity (minimum pH > 5.4 and mean alkalinity < 0.5 meq L⁻¹), or 3) high alkalinity (mean pH > 6.0 and mean alkalinity > 0.5 meq L⁻¹).

2.3. Data analysis

The following fish metrics were retrieved for each stream sample; 1) fish occurrence (yes = 1 or no = 0), 2) species richness (number of caught fish species), 3) occurrence of brown trout recruits (age 0+ estimated from length distribution, yes = 1 or no = 0), and 4) abundance (estimated number of fish per 100 m²). Abundance was estimated separately for each species, separately for age 0+ and >0+ within brown trout and Atlantic salmon, and for the sum of all observed species.

Only two fish metrics, species richness and total fish abundance, were retrieved for each lake sample; i.e. 1) species richness as for

streams, and 2) abundance (number of fish per unit effort, NPUE). One unit of effort was initially defined as one gillnet and one night of fishing, including dusk and dawn. Finally NPUE was transformed to a standard gillnet area of 45 m² (NPUE₄₅), to increase comparability of data from different time periods.

The main approach was to compare fish response metrics between groups of sampling events, based on time (years) elapsed since first liming. As most sites were not sampled on an annual basis, samples at limed sites were grouped as occurring before liming, in the year of first liming (year 0), or in any of five time periods after first liming (1–4, 5–8, 9–12, 13–16 or > 16 years, Table 1). No limed group had fewer than 100 observations, and groups with no overlap in the 95% confidence intervals (c.i.) of estimated mean values were considered to be significantly different. Samples from non-limed sites were grouped in similar time periods, but consistently setting the median year of first liming (1986) as year 0. There were far fewer observations at non-limed compared to limed sites, and very few non-limed sites sampled before 1990. Informative mean values were therefore not expected until observations at acid and low alkalinity reference sites more generally exceeded 30 samples within time periods 9–12, 13–16 or >16 years after 1986.

We ran linear regression analyses to further test for temporal trends indicating recovery. For each fish metric, annual percentages or mean values were weighted by the number of sites sampled. The fish metric was used as dependent and time lapse (year) since first liming as independent variable (or since 1986 for non-limed sites). We used the adjusted r-square statistic as it takes into account the number of observations and parameters in the model, P-values < 0.05 were interpreted as significant trends.

Change in stream fish abundance was also analyzed at the site level, by comparing mean values before and after liming with t-tests. Abundance at reference sites were similarly compared before and after year 1986, which was the median year of first liming. The t-values were transformed to correlation coefficients (Rosenthal, 1994), which were used in meta-analyses (Rosenberg et al., 2010).

To facilitate interpretation of observed stream fish responses at limed sites, minimum pH was identified for site and year with at least four water samples. The proportion of sites at which minimum pH fell below two threshold levels (6.0 and 5.6) was calculated at the year of first liming and for each year after first liming. Linear regression was used to test for temporal trends, with

proportion of limed sites with low minimum pH as dependent and year after liming as independent variables. The critical limits at pH 6.0 and 5.6 correspond to two different pH targets used in Swedish guidance for liming (Naturvårdsverket, 2010).

3. Results

3.1. Streams

Selection criteria were fulfilled for 1023 limed and 195 reference electrofishing sites with on average 14.2 and 17.0 sampling dates per site (Table 1). 300 limed sites were sampled before liming (1–15 times, mean 2.4), 256 sites were sampled in the first year of liming, and all sites were sampled in one or more 4-year groups after liming. Only 10 and 9 non-limed streams were sampled before and in the median year of first liming (1986), with 3–4 streams within each of the acid, low alkalinity and high alkalinity groups.

No fish were recorded at 11.5% of limed sites prior to first liming, compared to at least one fish in each sample at the few non-limed sites before 1986. The percentage of no fish occurrence was significantly reduced already 1–4 years after liming (Fig. 1a). Samples with no fish also appeared in each group of non-limed sites, when more sites were sampled in later time periods. The proportion of sites with no fish continued to decrease with time after first liming. After 9–12 years it was significantly lower than at acid reference sites, but not significantly different from low alkalinity reference sites. Linear regression revealed a significant decrease of % no fish occurrence in limed streams, as well as in high alkalinity reference sites, but no significant trends in the two other groups (Table 2).

A reversed pattern occurred for observed species richness, which increased with time after liming (Table 2), from an average of less than two species before liming, to more than 2.6 species after > 16 years (Fig. 1b). There were no significant trends in species richness in non-limed groups. A significant difference between groups appeared from 9 to 12 years, and since then species richness remained consistently lower at acid reference sites compared to the other three groups.

The occurrence of brown trout recruits increased as expected after liming, and % recruit occurrence also increased since 1986 in each of the non-limed groups (Table 2, Fig. 1c). Again high within-group variation and low sample size in non-limed groups

Table 1

Number of study sites and fish sampling dates (N) in streams and lakes, and average number of sampling dates per site. Numbers are given for each group of sampling event as well as in total for non-limed and limed sites, respectively. Number of sampling dates in lakes are generally fewer for comparable estimates of NPUE (N^b) than for observed species richness (N^a), as explained by the foot notes.

Group	Streams			Lakes				
	Sites	N	N site ⁻¹	Sites	N ^a	N ^a site ⁻¹	N ^b	N ^b site ⁻¹
Ref. high alk.	35	539	15.4	13	101	7.8	77	5.9
Ref. low alk.	103	1355	13.2	52	459	8.8	452	8.7
Ref. acid	57	881	15.5	36	301	8.4	292	8.1
Ref.(Total)	195	2775	14.2	101	862	8.5	821	8.1
Before	300	725	2.4	294	482	1.6	438	1.5
Year 0	256	261	1.0	116	118	1.0	100	0.9
Year 1–4	553	1417	2.6	252	293	1.2	258	1.0
Year 5–8	685	1762	2.6	357	423	1.2	394	1.1
Year 9–12	779	1998	2.6	361	420	1.2	406	1.1
Year 13–16	867	2323	2.7	354	412	1.2	402	1.1
Year > 16	900	6231	6.9	505	1066	2.1	1056	2.1
Limed(Total)	1029	14717	17.0	750	3214	4.3	3054	4.1

^a Species richness.

^b NPUE, transformed to a standard gillnet area of 45 m².

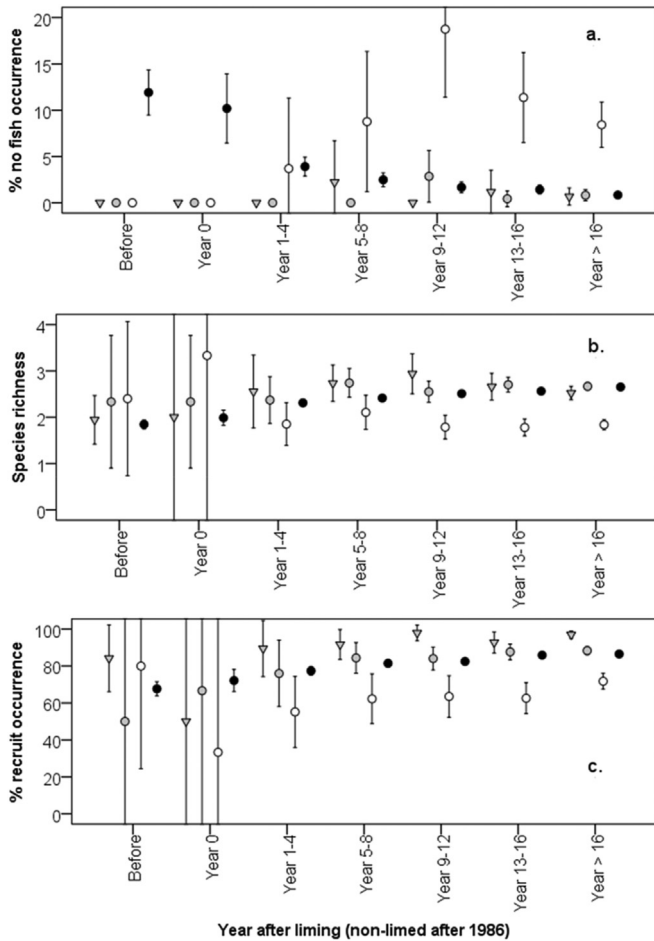


Fig. 1. Mean \pm 95% c.i. of a) % of no fish occurrence, b) observed fish species richness, and c) % occurrence of brown trout recruits (age 0+), in limed streams (black circles) and three groups of reference stream sites (acid: white circles, low alkalinity: grey circles, high alkalinity: grey triangles). Observations in each group are grouped in relation to year of first liming (year 0).

prevented detection of average differences between limed and non-limed groups before and in the first years after liming. In more recent years, however, the acid reference streams still had significantly lower recruit occurrence than the other groups.

Table 2

Average trends after liming, in % no fish occurrence, species richness and % occurrence of brown trout recruits, within limed stream sites and for three groups of non-limed reference sites. The estimated intercept in each linear regression represent year 0 as in Fig. 1, and the regression coefficient estimates the average annual change.

Group of streams	Regression	Numbers of years	Adjusted r-square	P-value
% no fish occurrence				
Limed	3.274–0.110*year	42	0.633	<0.001
Ref. acid	14.56–0.255*year	27	0.121	0.099
Ref. low alk.	16.11–0.041*year	27	0.002	0.313
Ref. high alk.	3.85–0.0145*year	27	0.070	0.042
Species richness				
Limed	2.338 + 0.0139*year	42	0.681	<0.001
Ref. acid	1.845–0.00031*year	27	–0.040	0.956
Ref. low alk.	2.805–0.0083*year	27	0.038	0.983
Ref. high alk.	2.887–0.0161*year	27	0.096	0.064
% recruit occurrence				
Limed	70.34 + 0.368*year	42	0.605	<0.001
Ref. acid	41.74 + 0.809*year	27	0.209	0.010
Ref. low alk.	75.96 + 0.391*year	27	0.150	0.026
Ref. high alk.	80.70 + 0.606*year	27	0.309	0.002

At the site-level, the abundance increased significantly after liming for all studied species and age groups (Fig. 2), except for decreased abundance of grayling (*Thymallus thymallus*). Some changes also occurred at non-limed reference sites, when comparing samples before and after the median first year of liming. Abundance increased for crayfish (*Astacus astacus* and *Pacifastacus leniusculus*), pike (*Esox lucius*), burbot (*Lota lota*), European eel (*Anguilla anguilla*), European minnow (*Phoxinus phoxinus*), lampreys (*Lampetra* spp.), bullheads (*Cottus* spp.) and age 0 + salmon. In contrast, abundance of roach and older (>0+) brown trout decreased at the non-limed sites. More importantly, abundance of roach, perch (*Perca fluviatilis*) and both age groups of brown trout increased significantly more at limed than at non-limed sites, and this was also found for the total abundance of fish.

The frequency of low minimum pH at limed sites decreased with increasing time after first liming (Fig. 3), indicating successive adjustment of lime treatment in relation to chemical targets of liming.

3.2. Lakes

Data selection revealed 750 limed and 101 reference lakes with on average 4.3 and 8.5 sampling dates per lake (Table 1). 294 limed lakes were sampled 1–13 times before liming (mean 1.6). 116 lakes were sampled in the first year of liming, and as for streams, all limed lakes were sampled in one or more 4-year groups after liming. Only 30 non-limed lakes were sampled before the median year of liming and 8 of them in the median year 1986.

In contrast to streams, mean fish species richness in limed lakes was significantly lower before first liming than in low alkalinity reference lakes sampled before 1986 (Fig. 4a). In all time periods after liming, the limed lakes had consistently higher species richness than acid reference lakes, but significantly lower than in low and high alkalinity lakes. The mean species richness did not change monotonically with time since first liming, but a decreasing trend appeared in the low alkalinity reference lakes (Table 3). In contrast to similar species richness in low and high alkalinity streams, mean species richness was always considerably higher in high alkalinity lakes compared to other lake groups.

Mean fish abundance (NPUE₅₄₅) seemed to increase and peak 5–8 years after first liming (Fig. 4b), when it was higher than in acid lakes but not different from low alkalinity lakes. Mean NPUE₅₄₅ was generally about 2–4 times higher in the group of 13 high alkalinity lakes than in all other groups. In longer time series the mean NPUE₅₄₅ decreased significantly in limed as well as in all groups of

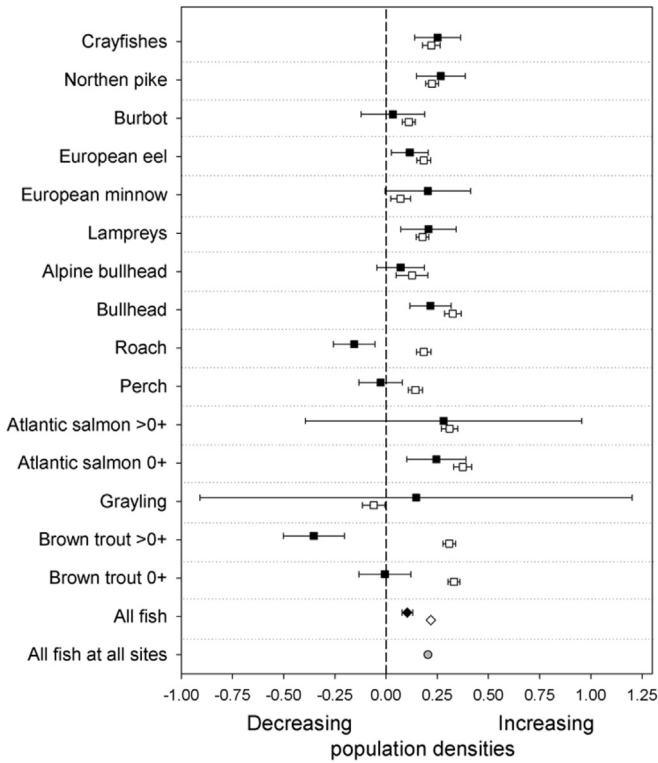


Fig. 2. Effect sizes (mean ± 95% c.i.) of changes in abundance after liming of streams, for the most frequently observed species or within age groups for salmon and brown trout, and for total abundance for all fish species caught. Effect sizes are shown separately for limed sites (white symbols) and non-limed reference sites (black symbols), and additionally the effect for all fish is shown for all sites together (grey symbol). Non-significant changes appeared whenever 95% c.i. overlapped the vertical reference line.

reference lakes (Table 3), reflecting higher weight of more lakes sampled in the latter time periods.

4. Discussion

This study was the first on a national scale to confirm the

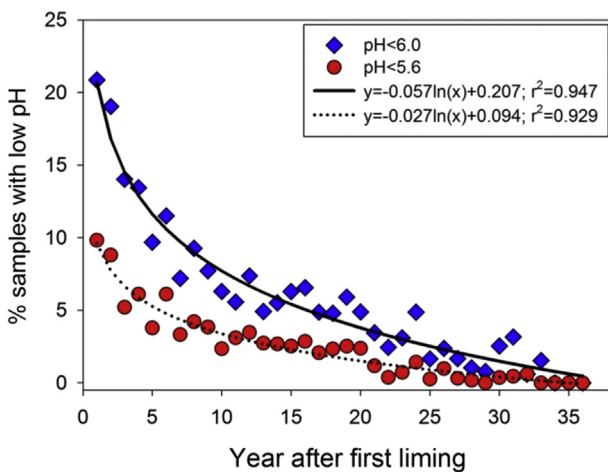


Fig. 3. Proportion of stream sampling dates with annual minimum pH < 6.0 and pH < 5.6, respectively, in relation to year after first liming. Regression lines were fitted after ln(X)-transformation. N = 37 year groups.

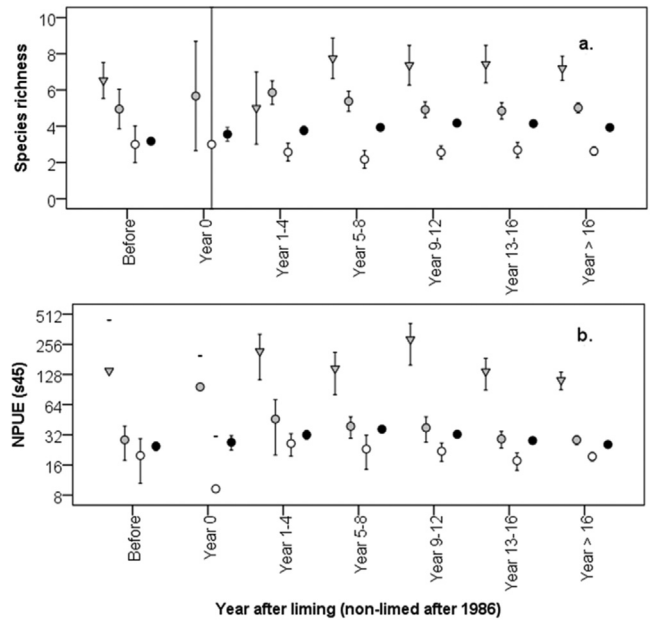


Fig. 4. Mean ± 95% c.i. of a) observed fish species richness, and b) numbers of fish per 45 m² gillnet [NPUE(s45)], in limed lakes (black circles) and three groups of reference lakes (acid: white circles, low alkalinity: grey circles, high alkalinity: grey triangles). Observations in each group are grouped in relation to year of first liming (year 0). The y-axis in b) is shown in log₂ scale to enhance readability of differences between lake groups.

generally expected effects of liming on fish in Swedish streams, but less general and consistent patterns was observed for fish in lakes. For streams, the trends were increasing for fish occurrence, species richness, occurrence of brown trout recruits and fish abundance. The large scale monitoring program thus included a sufficient number of limed sites to detect the significant improvement with time since first liming, despite high variance at local scale. This study, however, also revealed insufficient monitoring of non-limed reference sites, especially before 1990. This is possibly explained by subsequent lime treatment of almost all acid sites discovered in the early monitoring programs. The stream study was facilitated by standard sampling of hard-bottom and wadable stream reaches in all sites and years before and after liming (CEN, 2003), i.e. reaches most suitable for brown trout recruits. Electrofishing by wading is an established and suitable method for sampling of age 0 + fish in streams, especially for the most frequently occurring brown trout. We therefore had good data to see recruitment response to decreasing frequency of low pH in limed streams.

The results show some immediate improvement after liming of streams, and continuous recovery with time since first liming. The cause of the lag in improvement may be that it takes time for fish to recolonize habitats and may also be due to an initial low number of spawners when the species already occurred at a site. Also, the liming methods and doses have been improved successively (e.g. Alenäs et al., 1995). Initially episodes of low pH may have disturbed the fauna (cf. Fig. 3). Bradley and Ormerod (2002) found that the recovery of invertebrates in Welsh streams was hampered by such acid episodes. The entire Swedish liming program has been repeatedly revised with successively better practices for choice of methods and doses, e.g. after an initial focus on lake liming, supplementary liming with dosers and on wetlands were more frequently used. Improved liming practice is probably contributing to the observed decreasing frequency of critically low pH-values with time since first liming.

Table 3
Average trends after liming, in species richness and fish abundance (NPUE₅₄₅), within limed lakes and three groups of non-limed reference lakes. The estimated intercept in each linear regression represent year 0 as in Fig. 4, and the regression coefficient estimates the average annual change.

Group of lakes	Regression	Numbers of years	Adjusted r-square	P-value
<i>Species richness</i>				
Limed	4.136 + 0.0012*year	40	-0.024	0.790
Ref. acid	2.595 + 0.0123*year	29	0.012	0.253
Ref. low alk.	5.282-0.0247*year	25	0.115	0.037
Ref. high alk.	6.873-0.0054*year	22	-0.039	0.866
<i>NPUE₅₄₅</i>				
Limed	2.271-0.0038*year	40	0.230	0.001
Ref. acid	2.206-0.0085*year	29	0.210	0.009
Ref. low alk.	2.388-0.0097*year	25	0.256	0.004
Ref. high alk.	3.871-0.0335*year	22	0.319	0.004

Whole-lake gillnet sampling includes all benthic lake habitats (CEN, 2015), and therefore reinforces any differences in natural preconditions between the lakes within and between study groups. A long term study like this, also suffers from sampling bias due to older gillnet types used before and in the first years after first liming, only partly solved for by correcting CPUE to a standard gillnet area. Step-wise changes included a decrease in minimum mesh size and an increased proportion of smaller mesh-sizes. Such changes imply that small, young and potentially abundant fish have been more efficiently caught in the most recent decades, i.e. when sampling most limed lakes after ten or more years after first liming.

Differences in species richness and abundance between our study lake groups may be influenced by factors like lake size and productivity, thus masking the expected effect of liming. The highest richness is usually found in larger and deeper lakes in warmer areas, and more eutrophic lakes have higher fish density (e.g. Brucet et al., 2013). The decreasing NPUE₅₄₅ in all studied groups of lakes possibly reflect a generally observed oligotrophication, with stronger decrease of total phosphorous in limed than in non-limed lakes in Sweden (Hu and Huser, 2014). This effect could, however, not be tested in specifically in our study, due to lack of data on total phosphorus for many of the studied lakes and years.

Roach is one of the most acid-sensitive fish species in Swedish lakes, and many roach populations were previously lost due acidification (Bergquist, 1991). The probability of catching roach less than 10 cm in lakes decreases at low pH and at high concentrations of inorganic aluminum (Holmgren and Buffam, 2005). Age 0 + roach, and other small-sized species, are not efficiently sampled by gillnets, and the youngest age classes in the catch may overlap in size. Age 1 + roach may then be larger than 10 cm at low population densities, and in denser populations roach less than 10 cm may include several age classes. Therefore ageing, by counting annual patterns in otoliths, scales or bones, is needed to evaluate recruitment success in limed and recovering lakes (Holmgren, 2013).

The long term monitoring of fish in streams showed significant recovery after liming. The fish fauna successively approached mean values as in non-limed reference streams, while fish recovery was less consistent in non-limed acid streams. No general recovery trends appeared in the limed lakes, although species richness and fish abundance were higher in limed than in non-limed in the most recent decades. The difference may be due to the fact that sampling fish with gill-nets normally leads to low or no catch of the youngest age classes, whereas electrofishing is aimed at recruitment which is improved by liming. Also the effects of acidification will often be more pronounced in streams where acid surges during snow-melt may lead to low pH and elevated levels of aluminum.

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