

**Management in Practice****Rotenone application and degradation following eradication of invasive roach (*Rutilus rutilus*) in three Norwegian lakes**Roar Sandodden<sup>1,\*</sup>, Svein Aune<sup>1</sup>, Helge Bardal<sup>1</sup>, Pål Adolfsen<sup>1</sup> and Terje Nøst<sup>2</sup><sup>1</sup>Norwegian Veterinary Institute, section for Environmental Restoration and Management, Pb. 5695 Sluppen, NO-7485 Trondheim, Norway<sup>2</sup>Trondheim Municipality, Environmental Unit, P.O. Box 2300 Torgarden N-7004 Trondheim, Norway

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**OPEN ACCESS****Abstract**

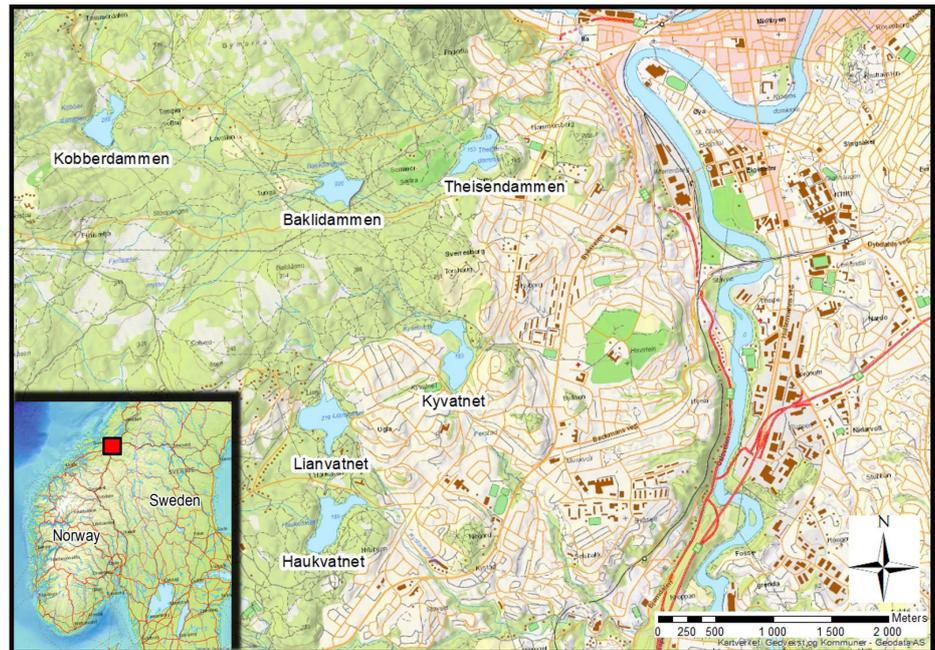
Roach (*Rutilus rutilus*) is indigenous to south-eastern Norway and alien to the rest of the country. In Trondheim municipality, in the middle part of Norway, roach was introduced into the Ila watercourse in 1881. Roach has a great potential to alter the ecosystem when introduced to new locations. The potential negative impact on potable water source quality and the prospect of permanently eradicating an alien species resulted in rotenone treatment of six lakes in Trondheim municipality. The rotenone concentration in the lakes was surveyed by water sampling until it could no longer be detected. A lethal concentration of rotenone at all test points was measured in all lakes during the survey period. Fourteen days after treatment, a near homogenous concentration was reached. The concentration reduction was similar in the lakes and relatively quicker during the first weeks after treatment. It was also consistent between depths except for the surface, where the concentration degraded more quickly. Rotenone degradation is a key factor when planning eradication efforts, and reports on this varies considerably between different locations. Despite application of rotenone in different depth strata, it took several days to reach homogenous concentration and several months and a fall turnover for the rotenone to break down and dilute below the detection limit in the lakes described.

**Key words:** invasive alien species, rotenone, CFT-Legumine, piscicide, rotenone dissipation

**Introduction**

Roach (*Rutilus rutilus*) is indigenous to south-eastern Norway and alien to the rest of the country (Huitfeldt-Kaas 1918). In Trondheim municipality, in the middle part of Norway, roach was introduced into the Ila watercourse in 1881 (Huitfeldt-Kaas 1918). The spread of roach to other lakes in Trondheim occurred in the 1970s and 1980s, and in the past 20 years, also to lakes outside Trondheim. No other populations of roach are known in middle parts of Norway, and it is likely that the population of roach in the Ila watercourse, and later the other lakes in Trondheim, have been the source of all introductions in the middle part of Norway.

Roach has a great potential to alter the ecosystem when introduced to new locations through high propagation potential, wide temperature tolerance,



**Figure 1.** Lake Lianvatnet, Haukvatnet, Kyvatnet, Kobberdammen, Baklidammen and Theisendammen in Trondheim municipality.

and a broad nutrition spectre (Kottelat and Freyhof 2007; Winfield et al. 2011; van der Kooij and Redford 2012). This has negative consequences for biodiversity and water quality (Forsgren et al. 2018). Dense roach populations will result in a significantly reduced zooplankton community and contribute to increased circulation of organic matter (Vøllestad 1985; Nøst and Langeland 1994). This results in larger phytoplankton biomass, with reduced light penetration in the water and increased oxygen consumption. Roach is categorized as a high impact species in the Norwegian alien species list ([www.biodiversity.no](http://www.biodiversity.no)) Trondheim municipality assessed the risk of further spread to potable water sources as highly likely. In 2004, an attempt to eradicate the roach from the lakes in the Ila watercourse by draining failed. This attempt involved partial draining with liming of the residual water. As an alternative, the municipality decided to try a rotenone treatment to remove roach. A similar rotenone treatment was carried out in lake Vikarauntjønna in Trondheim municipality in 2014. This was also due to risk of spreading to the Trondheim's potable water source. The treatment was a success (Bardal 2015).

Rotenone is currently the only effective piscicide approved by Norwegian and European environmental authorities to eradicate fish species in freshwater. It is the only piscicide allowed for essential use according to the Biocidal Products Directive ([www.europa.eu](http://www.europa.eu)). Due to roach's potential negative impact on potable water quality and the prospect of permanently eradicating a foreign species from the region, rotenone treatments were applied to the six roach lakes in Trondheim municipality. The six lakes were Lianvatnet, Haukvatnet, Kyvatnet, Kobberdammen, Baklidammen and Theisendammen (Figure 1).

Rotenone is a naturally occurring substance derived from the roots of tropical plants in the *Leguminosae* family (USEPA 2007) and has been used for centuries to capture fish for food in Southeast Asia and South America where these plants naturally occur (McClay 2000; Ling 2003). Rotenone is non-persistent in the aquatic environment and is degraded by photolysis and hydrolysis (Finlayson et al. 2014). Rotenone is poisonous to organisms to varying degrees, but gill-breathing species are particularly vulnerable. Rotenone is rapidly absorbed across the gill epithelium and blocks oxygen use by cells (Koopman et al. 2005). Rotenone is highly toxic to fish and certain invertebrates (Fukami et al. 1969). Vinson et al. (2010) reviewed the effects of rotenone treatments on invertebrate communities and showed that sensitivity to rotenone varies considerably between taxonomic groups. Studies show that non-tolerant and rotenone sensitive aquatic invertebrates typically experience a short-term reduction in abundance (Arnekleiv et al. 2001). The reestablishment of most taxa is rapid and often complete after a year (Eriksen et al. 2009; Arnekleiv et al. 2015). Long-term studies following rotenone treatments in standing water show that zooplankton recovery time varies between three to twelve months and species composition recovery usually takes place within a year (Arnekleiv et al. 1997, 2015; Eriksen et al. 2009; Kjærstad et al. 2018; Marr et al. 2019).

Rotenone persistence in temporal natural waters varies from a few days to several months depending on the season. In a typical subarctic Norwegian forest lake it usually takes a year and two turnovers for the rotenone to disappear (Sandodden et al. 2019). The half-life of rotenone is longest in winter, but may decrease to as little as a few hours in summer. Fishery managers must carefully plan applications to consider these factors.

One of the conditions for allowing rotenone treatment was that Trondheim municipality should facilitate the re-establishment of biological diversity as soon as possible. A release plan for trout was therefore prepared, which is considered to be the local fish species in the area. Three age classes are set out (young of the year, age 1 and 2 year), where the number of fish is adapted to the size and production potential of the individual lake.

A rotenone monitoring program was performed, which included all the treated lakes, but only three were monitored until the rotenone degraded below the detection limit. The water chemistry was most similar in the lakes that were monitored until full rotenone degradation was achieved. In the remaining portion of this paper, only the results from those lakes that were monitored until complete rotenone degradation occurred are presented and discussed.

Rotenone degradation is a key factor when planning eradication efforts, and reports on this varies considerably between different locations. Water chemistry and the hydrology characteristics of the location are essential factors. Despite application of rotenone in different depth strata, it took

**Table 1.** Water quality parameters of the surface water of the lakes august 2017.

Lake	Total Phosphorus ( $\mu\text{g P/l}$ )	Total Nitrogen ( $\mu\text{g N/l}$ )	Calcium ( $\text{mg Ca/l}$ )	Colour ( $\text{mg Pt/l}$ )	Alkalinity pH	Turbidity (FTU)	Transparency (m)
Lianvatnet	6,5	250	22,7	21	8,0	0,6	4,2
Haukvatnet	4,6	220	23,8	20	8,0	0,6	3,5
Kyvatnet	8,5	330	28,6	26	7,9	1,2	3,0

several days to reach homogenous concentration and several months and a fall turnover for the rotenone to break down and dilute below the detection limit in this lakes described.

## Materials and methods

### *The Lakes*

The treatment area consisted of six lakes with a surface area of 8.2–11.1 hectares. Maximum depth ranged from 9 to 16 meters. Lianvatnet drains into Haukvatnet. Haukvatnet and Kyvatnet drain into River Nidelva and the Trondheim Fjord. Lake Lianvatnet has a surface area of 11.1 ha and a catchment of 1.05 km<sup>2</sup>. The volume of the lake is calculated to be 416,140 m<sup>3</sup>. The maximum depth is 15 meters, but most of the lake is shallow. Large areas have depths less than 2–3 m. Lake Haukvatnet has a surface area of 10.2 ha and catchment of 2.67 km<sup>2</sup>. The volume is 617,720 m<sup>3</sup>. Maximum depth is 16 m. The Lake Kyvatnet has a surface area of 9.7 ha and catchment of 1.12 km<sup>2</sup>. The volume is 462,830 m<sup>3</sup>. The lake has two pools, a big and deep pool in the south, and a smaller pool in the north. The northern pool is shallow (maximum depth 2 m) and the surface is largely covered in macrophytes. The southern pool has a maximum depth of 15 meters.

### *Water chemistry*

Chemical analyses were carried out on samples of surface water in August 2015 (Nøst 2018; Table 1). A thermocline was established between 4 to 5 meters depth. Lianvatnet, Kyvatnet, and Haukvatnet have quite high content of calcium (> 20 mg Ca/l) compared to other lakes in this part of Norway. The colour of the water of the three lakes was between 20 and 30 mg Pt/l, which indicates oligohumus conditions. The content of total phosphorus and total nitrogen indicates nutrient-poor conditions in Haukvatnet and Lianvatnet, 4.6  $\mu\text{g P/l}$  and 220  $\mu\text{g N/l}$  and 6.5  $\mu\text{g P/l}$  and 250  $\mu\text{g N/l}$  respectively, while Kyvatnet is slightly more nutritious with 8.5  $\mu\text{g P/l}$  and 330  $\mu\text{g N/l}$ . The transparency in the lakes is typical of lower-lying forest lakes and is in the range 3–4.2 m. The pH is slightly basic, between 7.9 and 8.0. Turbidity in the water is relatively low in Lianvatnet and Haukvatnet 0.6 FTU, while in the Kyvatnet it is 1.2 FTU. Measurements of oxygen content showed that the waters had virtually oxygen-free (< 5% saturation) environments close to and at the bottom.

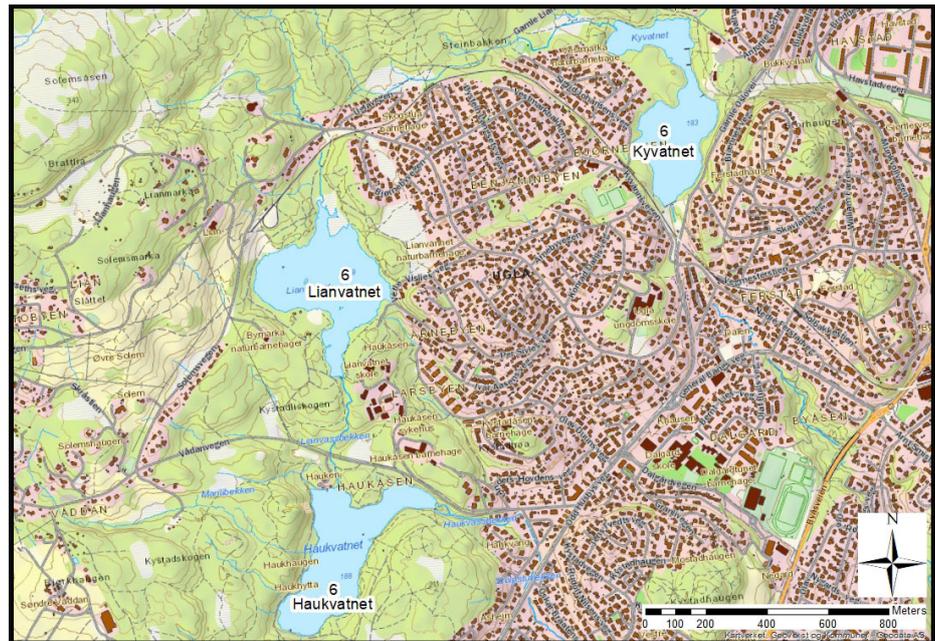
**Table 2.** CFT-Legumine dosage below the thermocline, at the surface, lake shorelines and in tributaries in Lake Lianvatnet, Haukvatnet and Kyvatnet.

Lake	Dosage	CFT-Legumine® (L)
Lianvatnet	Below thermocline (6 m)	80
	Surface	400
	Shorelines	80
	Tributaries	3,5
	<b>SUM</b>	<b>563,5</b>
Haukvatnet	Below thermocline (6 m)	240
	Surface	600
	Shorelines	40
	Tributaries	3
	<b>SUM</b>	<b>883</b>
Kyvatnet	Below thermocline (6 m)	140
	Surface	482
	Shorelines	30
	Tributaries	6
	<b>SUM</b>	<b>658</b>

### *Rotenone concentration and application*

Lake Lianvatnet and Haukvatnet were treated on 19.09.17 and Lake Kyvatnet on 20.09.17. 2104.5 l the rotenone-containing chemical, CFT-Legumine® 3.3% was used. Ling (2003) reviewed the rotenone tolerance for several species. Roach LC5024h was found to be 24.5 µg/l rotenone. In this treatment exposure time would exceed 24 hours. To account for degradation and dilution before reaching homogenous concentrations and to achieve a quick kill, the treatment concentration was set to 49.5 µg/l rotenone for the total water volume to be treated. This concentration has also been used in successful rotenone treatment to remove roach in Norway (Bardal 2015). Lakes were dosed according to the water-volume at different depths and on the surface including lake shores and peripheral water bodies (Table 2).

All lake inlets were included in the treatment area. The CFT-Legumine was distributed in the lakes using boats equipped with gas powered fire-fighting pumps. The pumps used water from the lake and were mixed with CFT-Legumine in the boat before distribution. The dosing was carried out by attaching the flushing hose just below the surface of the bow of the boat, so that the engine behind contributed to the mixing of rotenone and water. The surface was defined as consisting of the water above the thermocline established at approximately 6 meters. The shorelines were flushed with diluted (100 ppm) CFT-Legumine by the use of the boat mounted pumps. Another boat was used for dosage below the surface layer. This layer was defined as consisting of water below the thermocline. Rotenone was distributed by using a weighted long hose with several nozzles. Lead weights was attached at the end of the dosing hose to make it sink. Four nozzles distributed the rotenone at the bottom three meters of the dosing hose. An echo sounder was used during this dosage for a continuous control of the depth the dosing hose. This to ensure that a correct amount of rotenone was distributed at the correct depths in the lake.



**Figure 2.** Water-sampling station 6 in Lake Lianvatnet, Haukvatnet and Kyvatnet. Station 6 in each lake had the most frequent sampling regime and was chosen as a reference location for the vertical distribution of rotenone in the lakes.

Continuous drip stations were used in streams. A drip station consists of a 20-l plastic-jug with a dosing hose used as a siphon. Using 8 m thin hose and a drop of 60 cm from the bottom of the jug to the end of the hose, they supplied a linear and constant dosage of 20 l diluted (according to discharge) CFT-Legumine® for four hours. In the tributaries, consisting of small streams, seeps, and small pockets of standing water, common watering cans were used to dispense diluted (1000 ppm) CFT-Legumine®

### *Water sampling*

Water samples were withdrawn from a boat using a Kemmerer water sampler at set locations and depth to document lethal concentrations of rotenone in the treated lakes the days following treatment. Samples of 60 ml of water were collected in dark glass bottles, refrigerated at 4 °C and brought to the lab within two days of collection. The sampling was performed 2, 5, 8, 14, 21, 30, 45, 70, 95, and 120 days after completing the treatment in lakes Lianvatnet, Kyvatnet and Haukvatnet respectively. Rotenone concentration was monitored until it was no longer detectable.

Fourteen water samples were collected during the first sampling day in each lake. After two full samplings, the number of samples at each sampling was reduced by sampling only Stations 6 and 9 in each lake. Station 6 had the most frequent sampling regime and was chosen as a reference location for the vertical distribution of rotenone in the lakes (Figure 2).

### *Chemical analysis*

Rotenone concentration analysis was performed by the Norwegian Veterinary Institute using liquid chromatography with UV detection according to the

method described in Sandvik et al. (2018). The UFLC-UV system from Shimadzu (Kyoto, Japan) was equipped with two LC-20AD pumps, a SIL 20A automatic sample injector, and a SPD 20A DAD detector. Rotenone ( $\geq 95\%$ ) and deguelin ( $\geq 98\%$ ) standards were obtained from Sigma Chemical (St. Louis, MO, USA).

Rotenone was quantified using an external four-point calibration curve in the concentration range 1–250  $\mu\text{g/L}$ . The limit of detection (LOD) and the limit of quantification (LOQ) were defined as the minimum concentration generating a signal-to-noise ratio (S/N) equal to 3 and 10, respectively. The limit of quantification was 1  $\mu\text{g/l}$  in water samples diluted with acetonitrile (water-acetonitrile, 50:50). A pre-concentration step using Spin-X® centrifuge filters allowed quantification levels of 200 ng/l.

## Results

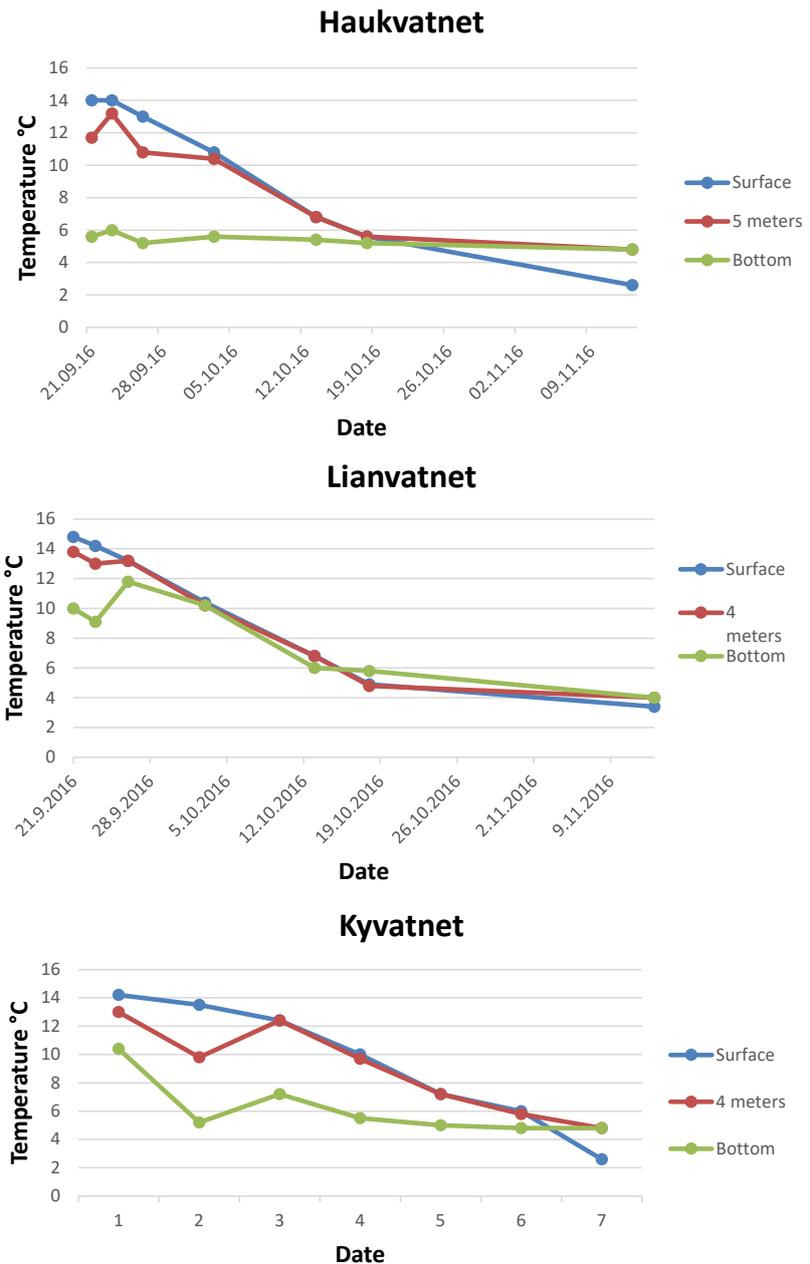
Temperature difference between the surface and the thermocline (5 m) on the day of treatment measured 4.5, 3.4 and 2.5 °C surface in Lake Haukvatnet, Kyvatnet and Lianvatnet, respectively (Bardal et al. 2018). Fall turnover in the lakes happened during late October (Figure 3).

Concentrations above LC5024h was reached in all depths during the sampling period. On the other hand, concentrations on the surface were above the set treatment concentration, while concentrations at 4m, 5m, and at the bottom was below. Rotenone dissipated quickly from the treated lakes and most of the rotenone was gone within 70 to 95 days post treatment. Rotenone concentration 2, 5, 8, 14, 21, 30, 45, 70, 95, and 120 days after finishing the treatment in lakes Lianvatnet, Kyvatnet and Haukvatnet are presented in Figure 4.

A relatively higher proportion of the rotenone was distributed on the surface. The surface rotenone concentration was relatively high during the first two samplings two and four days following the treatment. The amount distributed was calculated to achieve lethal concentrations both above and below the thermocline. Four days after treatment full mixing had not yet occurred in this layer of water. In the third and fourth sampling, eight and fourteen days after treatment, the mixing was better and a near homogenous concentration had been reached. Both wind mixing and water currents in the lakes are assumed to have contributed to this. There was no ice formation on the lakes in this period. The concentration reduction was similar in the lakes and relatively quicker during the first weeks after treatment. These findings seem quite consistent between depths, except for the surface where the concentration degrades more quickly.

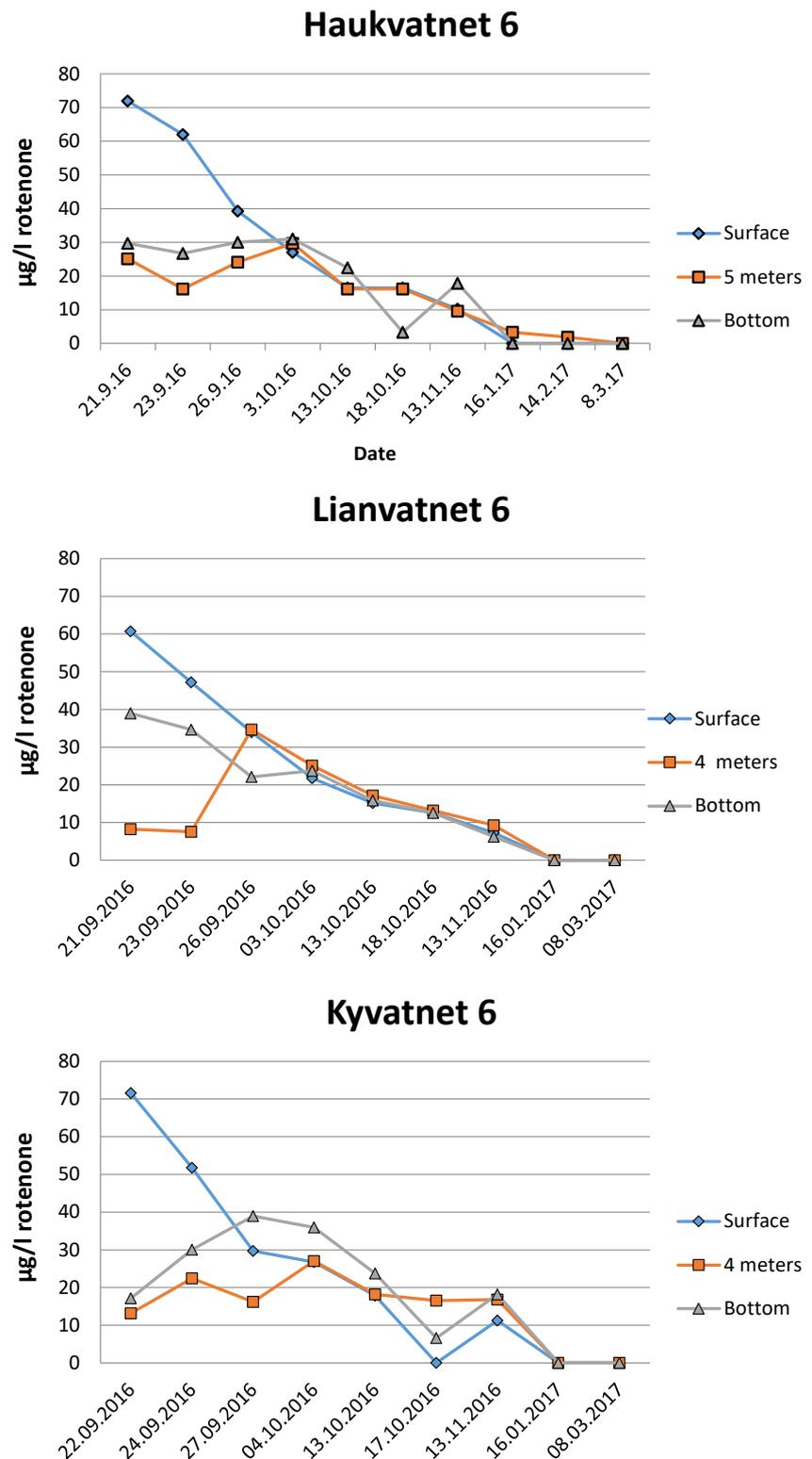
## Discussion

The persistence of rotenone in water is dependent upon several factors. Specific data are not available to address all degradation parameters under



**Figure 3.** Water temperature (C°) from late September until early November in Lake Haukvatnet, Lianvatnet and Kyvatnet. The highest depth represents the bottom in all three lakes.

actual use conditions, but rate of rotenone degradation in water is mostly determined by temperature (USEPA 2007), and also by turbidity and by the levels of light, transparency, alkalinity, and oxygen (Dawson et al. 1991; Robertson and Smith-Vaniz 2008). Dilution also contributes to decreasing concentration beyond detectable levels of rotenone following treatments. Bioavailability of rotenone is reduced because of strong adsorption to sediments, plants, and particulate matter in treated waters. (Turner et al. 2007). Lethal levels are a combination of concentration and exposure time and thus are not easy to define. A conservative approach using a high concentration of rotenone distributed at all depths of the lakes was applied during this treatment, aiming for a quick fish kill to enhance the likelihood



**Figure 4.** Rotenone dissipation at location 6 in Haukvatnet 2, 5, 8, 14, 21, 30, 45, 70, 95, and 120 days following treatment.

of total eradication. Before homogenous and lethal concentrations of rotenone are reached, stagnant fish in pockets of water can survive. If they stay there long enough, sub-lethal concentrations caused by degradation and dilution in other pockets can appear.

Rotenone concentration decreased rapidly in the first few days following treatment. This is probably due to the effect of organic matter that binds and breaks down rotenone. Then, rotenone dissipation occurred more slowly, controlled by temperature and light conditions. The dissipation was slower but consistent with the results of Finlayson et al. (2014), investigating the fate and behavior of rotenone in Diamond lake, Oregon USA. During late fall and onset of winter, the temperature in the lake decreased until fall turnover effectively resulted in homogenous concentration. Furthermore, the rotenone concentration decreased evenly, and the turnover had no apparent effect on the speed of degradation. Turnover primarily had a beneficial effect on faster mixing, and may have a positive effect on the rate of degradation of rotenone where there were differences between surface and bottom layers, due to either temperature stratification or lack of light penetration or oxygen in deeper areas of the lakes.

The technique applied when distributing the rotenone is obviously of great importance when aiming for a homogenous distribution and homogenous concentrations before the rotenone degrades and dilutes below lethal levels. Applying rotenone at different depths and below an established thermocline is especially important. Surface application alone will increase the likelihood of failure because of non-homogenous concentrations and more detrimental effect on non-target organisms mostly found above the thermocline.

These results show a slower mixing of water in the surface area (0–6 m) than expected. This shows that this layer should have been divided in two, with distribution both at the surface and at 3 m depth. The mixing above the thermocline at 6 m was slower than we have experienced before. A possible explanation is a relative high temperature difference from the surface to the thermocline in the lakes during the treatment and the fact that a relatively higher proportion of rotenone than intended was distributed on the surface.

Kjærstad et al. (2019) monitored the invertebrate community following treatment. This study showed a decrease in the number of individuals of invertebrates. In 2018, the number of individuals had increased and was higher than the densities in both 2015 and 2017 in the treated lakes. However, the increase in total numbers was largely due to an increase in Chironomidae larvae and in some cases also Ostracods. Several of the species that were missing in 2017 were again registered in 2018, but some species and groups were still missing. Rotenone-treated streams appear in 2019 to have re-established a diverse benthic fauna that is approximately equal to the expectation of natural state and/or measured pre-condition. (Bergan 2020). Schnee (2019) investigated the long term effects of rotenone on the assemblages of aquatic benthic macroinvertebrates in outlet streams of 13 alpine lakes in Montana, USA. He concluded that aquatic systems will recover in reasonable time and provide sufficient and stable sources of

food to sustain the restocked fish populations 1 year after rotenone treatment. Furthermore, aquatic systems will recover and overall ecosystem health will recover to pre-treatment conditions within 1 to 2 years.

Kjærstad et al. (2020) presents results from follow-up surveys from the treated lakes in 2019.

The zooplankton is well re-established after the rotenone treatment. In most lakes, there has been a shift from strong dominance of crayfish (Copepoda) before treatment to a correspondingly strong dominance of water fleas (Cladocera) after. In the littoral zone, the vast majority of species from before the rotenone treatment have been rediscovered and a significant number of new species have emerged. All lakes had more species of small crayfish in the littoral samples in 2019 than before the treatment. The total amount of benthic animals was reduced in 2019 in most lakes, which is set in connection with grazing from fish that were released in several of the localities in the autumn of 2018. In most of the lakes the light penetration had increased following the rotenone treatment.

No roach has been found after the treatments were performed. No systematic monitoring has been performed, but in Lake Lianvatnet and Haukvatnet, eDNA sampling during 2017 found no roach DNA (Majaneva Nina *unpublished*). Trondheim municipality have had no reports of roach after the treatments (Nøst *pers. comm.*). Trout populations have been re-established in all lakes and fishing is already popular. Spawning success for trout has been documented in 2019 and 2020 (Nøst *pers. comm.*). There is also a lot of activity in and around the three lakes. They are used for swimming during the summer and there are trails and exercise areas at and around all three lakes. The treatments created a lot of attention and many of the users and residents around the lakes have had a close eye on the lakes to see if roach reappear. The roach was very visible in all lakes when it was present before the treatments. On the basis of this we feel that it is highly likely that the roach have been eradicated from the three lakes, despite that the post-treatment monitoring effort has been low.

In summary, based on studies of the effect of rotenone treatments carried out in Norway over the past 25 years, it can be concluded that they have had varying effects on aquatic fauna beyond fish, but that all effects have been relatively short-lived. The degree and duration of the negative effects are probably affected both by the time of treatment, the extent of untreated refuges upstream/outside the treated area and the physical/chemical conditions around the treatment (temperature, water flow, and residence time and dosage concentration).

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## Author's contribution

R.S. – research conceptualization, sample design and methodology, investigation and data collection, data analysis and interpretation, ethics approval, funding provision and writing and editing; S.A. – research conceptualization, sample design and methodology, investigation and data collection, and writing and editing; H.B. – research conceptualization, investigation and data collection, and writing and editing; T.N. – investigation and data collection and editing.

## Ethics and permits

The Norwegian Environmental Agency gave according to Norwegian law a permit to distribute the rotenone containing chemical CFT-Legumine in the lakes described in the paper.

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