Management in Practice

Fighting an invasive fish parasite in subarctic Norwegian rivers – The end of a long story?

Pål Adolfsen*, Helge Bardal and Svein Aune

Norwegian Veterinary Institute, Section for Environmental and Biosecurity Measures, Pb 4024 Angelltraa, 7457 Trondheim, Norway

Author e-mails: pal.adolfsen@vetinst.no (PA), helge.bardal@vetinst.no (HB), svein.aune@vetinst.no (SA)

*Corresponding author

Abstract

The introduced monogenean Gyrodactylus salaris (Malmberg, 1957) is categorized as one of the most severe threats against Atlantic salmon (Salmo salar Linnaeus, 1758) in Norway and has almost eradicated salmon populations in the Skibotn and Signaldalen Rivers in northern Norway. The parasite was unintentionally introduced to the Skibotn River in 1976 via release of infested Atlantic salmon smolt from Sweden. The parasite is restricted to freshwater and survives at most a few days without its host. Therefore, eradication of all hosts in the infected river systems has been the preferred strategy to eliminate the parasite. After two failed eradication attempts in 1988 and 1995, the parasite spread further to neighbouring rivers. This, along with several other failed rotenone treatments in Norway in the 1990s, resulted in severe criticism of the national eradication strategy for G. salaris. Still, the eradication program continued, and the failed eradication attempts were analysed for possible improvements. Arctic char (Salvelinus alpinus, Linnaeus, 1758) has proved to be a potential long-term host for the parasite and infested char were documented to have survived in small, groundwater-fed tributaries and ponds during the first two eradication attempts in the Skibotn River. Low limits on allowed rotenone concentrations set by the pollution control authorities might also have contributed to the failures. A third attempt at eradicating the parasite from River Skibotnelva was made in 2015 and 2016, using new knowledge about the parasite and its hosts, renewed strategies to map and deal with dilution from groundwater intrusion and an official acceptance of increased concentrations of rotenone. Treatments for two consecutive years was the main strategy improvement from previous eradication attempts. Water samples showed sufficient levels of rotenone concentrations at all sample points during the treatment periods. Significant efforts in collecting all possible surviving fish from the first-year treatment and screening them for G. salaris revealed no surviving parasites at the time of the second-year treatment. The national G. salaris eradication campaign includes a surveillance programme for eradication confirmation. The results so far are positive for the Skibotn Region, but the earliest an eradication confirmation can be issued earliest is 2021.

Key words: Gyrodactylus salaris, rotenone, Atlantic salmon, arctic char

Introduction

The monogenean parasite Gyrodactylus salaris is categorized as one of the most severe threats against Atlantic salmon in Norway (Anon. 2016). It has been detected in a total of 51 Atlantic salmon rivers in Norway since the
1970s, when it was introduced by live fish import from Swedish hatcheries. Genetic analyses of the different strains of the parasite have identified at least three different incidents that have resulted in infections in hatcheries and wild Atlantic salmon populations (Hansen et al. 2003). Eight rivers in two regions are still infected. In the remaining 43 rivers, the parasite is confirmed eradicated or the rivers are under post treatment surveillance.

In contrast to the Baltic strains of Atlantic salmon, the Norwegian (East-Atlantic) Salmon strains have no co-evolutionary history with this parasite and do not have the ability to control the infection. The infestation causes high mortality in the early freshwater life stages, and an average reduction of 86% (48–99%) of parr densities is documented in infested Atlantic salmon populations (Johnsen et al. 1999).

*Gyrodactylus salaris* is a viviparous, obligate parasite with a direct lifecycle. This means it lives its whole life on the host and gives birth to full-grown offspring that attach to the skin of the same host fish as the mother. Attaching with its specialized ophisthaptor, and feeding on the mucus and skin, the parasite damages the skin and kills the host if the total parasite burden gets too high. New hosts can be infested by direct contact with infested fish, by detached parasites drifting in the water column or sitting on the substrate. Detached individuals can survive for 132 hours at 3 °C. At higher temperatures survival time is shorter (Olstad et al. 2006). The parasite is restricted to freshwater, but can survive for shorter periods in brackish water, depending on salinity and temperatures (Soleng and Bakke 1997). *Gyrodactylus salaris* is capable of both asexual and sexual reproduction. This means that one single parasite can start an epidemic that spreads throughout the whole salmon population in a river or even a region of rivers, if interconnected with brackish waters. Saltwater in fjords and ocean acts as a dispersal barrier when the distance between rivers exceeds the parasites possible survival time at the actual salinity and temperature. Both anadromous and non-anadromous arctic char (*Salvelinus alpinus*) (Robertsen et al. 2007) and the non-native rainbow trout (*Oncorhynchus mykiss*) (Hansen et al. 2016; Paladini et al. 2021) have proved to be suitable permanent hosts for the parasite. Brown trout (*Salmo trutta*) has potential to be a temporary host, and may play a part in spreading the parasite both within and between rivers (Paladini et al. 2014).

The obligate lifecycle and a distribution restricted to the anadromous zone of freshwater systems makes eradication of *G. salaris* possible. Removal of all hosts by rotenone treatments, in some cases combined with migration barriers built to limit the anadromous zone, has been practiced since the early 1980s.

After successful eradications in small and medium-sized rivers during the 1980s, there were a series of failed eradication attempts in several more complex river systems from the late 1980s to the early 2000s. Among these setbacks were the failed attempts in the Skibotn River in 1988 and 1995.
Gyrodactylus salaris was detected again in 1999 and investigations concluded that infested fish probably survived as a result of insufficient rotenone concentrations because of groundwater influence (Brabrand and Koestler 2003). In 2000, G. salaris was also detected in the neighbouring River Signaldalselva and the tributary River Balsfjordelva. Prior to the treatments in 2015–2016, the parasite had never been detected in the River Kitdalselva, but it was considered potentially infected due to its location in the same estuary. This spreading of the parasite within the region more than doubled the length of infected river stretches. This was the backdrop when the Norwegian Veterinary Institute in 2013 on behalf of the Norwegian Environment Agency started planning for a new eradication project in the Skibotn Region.

The aim of this paper is to highlight the continuous evolution of the methods of mapping, planning and conducting G. salaris eradication operations. The result is a robust strategy that has proven to work at a large geographic scale in complex river systems, and which is here put to the test in treating what may be the most complex and challenging G. salaris infected region in Norway.

The Skibotn Region

The Skibotn Region (Figure 1) is located at 69°15′N; 19°54′E, in the municipalities of Storfjord, Lyngen and Balsfjord in Troms County, Norway. The climate is subarctic, with rivers populated with Atlantic salmon, arctic char, brown trout (Salmo trutta Linnaeus, 1758), three-spined stickleback (Gasterosteus aculeatus Linnaeus, 1758), burbot (Lota lota Linnaeus, 1758), European whitefish (Coregonus lavaretus Linnaeus, 1758), grayling (Thymallus thymallus Linnaeus, 1758), alpine bullhead (Cottus poecilopus Heckel, 1837) and pike (Esox lucius Linnaeus, 1758). The River Signaldalselva holds the majority of the aforementioned fish species due to a passage across the water divide with both a westward and an eastward outlet, connecting it to the Tornio watercourse.

The river valleys in the region have large glacial and fluvial deposits, resulting in large aquifers in contact with the rivers.

Materials and methods

In a rotenone treatment with the scale and complexity of the one in the Skibotn Region, a lot of factors could lead to failure. To eliminate or reduce the different risk factors, a set of different sub-strategies were implemented.

Dosage strategy

The two target host species, Atlantic salmon and arctic char are known to be among the most rotenone sensitive fish species (Ling 2003; Stensli and Bardal 2014). Still, the minimum dose of rotenone was set to a relatively high
level (23 µg litre⁻¹ in 4 hours) to secure 100% mortality in conditions with significant local dilution from groundwater. All rotenone dosage referred to in this report was done with the liquid CFT Legumine 3.3%™ rotenone formula. Primary dosage stations were placed upstream from the migration barriers defining the treatment area in the rivers and larger brooks. Size of dosage station pumps was based on discharge at site (Watson-Marlow peristaltic pumps; 12v 100 series, 24v 300 series and 230v 700 series). The turbulence and flow of the river mixed and transported rotenone-treated water downstream through the treatment area. In parts of the river with

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**Figure 1.** The large map shows the rivers (in red) with *G. salaris* in the Skibotn Region. Orange marks rivers treated without findings of the parasite. All rivers and brooks potentially inhabiting salmonids south of the black line across the Lyngen-fjord were treated. Inserted map shows the location in Norway.
long transportation time, primary dosage stations were run in parallel to ensure that the whole section of river was dosed simultaneously with the riverbank and tributary treatment. Supplementing dosage stations were in some cases used to compensate for dilution, absorption and decomposition of rotenone. New supplementing dosage stations were set up when rotenone analyses documented minimum concentrations below the defined minimum level. In addition to this general dosage strategy, there were a variety of supplementary dosage techniques and strategies to face the special challenges in the region.

Treatment for two consecutive years

The aim was to perform the first treatment in a way that eradicated every host and parasite the first year. Still, the national strategy is to treat the entire infected region twice, with one year between treatments. This strategy was implemented as a standard for river treatments in the G. salaris eradication programme in Norway after 2003. Because of the history of the two previously failed treatments in the Skibotn River, a third year of treatment was considered as a possibility if the quality control and post treatment evaluation of the second year indicated a need for this.

Avoiding reinfection in treatment period

To reduce the risk of reinfection by fish migrating from untreated rivers to a treated river, the strategy was to treat all the rivers in the region within one week. In the three-river complex of Signaldalselva, Balsfjordelva and Kitdalselva, all in the same estuary, dosage stations was used as temporary chemical barriers until all of the rivers were treated. The chemical barriers were in the lower parts of the untreated neighbouring rivers so that potentially infected fish could not leave the untreated river without being exposed to lethal doses of rotenone, defined as minimum 33 µg litre⁻¹ for at least 30 minutes. Locations partially isolated from the rest of the treatment area were treated a short time (1–2 days) before the main treatment period, securing an overlap between lethal rotenone concentration in these locations and the connected parts of the main treatment area. In some cases, it was necessary to put up dosage stations to maintain rotenone concentrations in the pre-treated location until the desired overlap was achieved.

Nocturnal dosage

Atlantic salmon parr, as well as arctic char are known to display diel varying activity patterns, hiding in substrate cavities in periods (Amundsen et al. 2000; Adams et al. 2011; Holierhoek and Power 1995). To increase probability of exposing nocturnally active hosts to rotenone treated water, primary dosage stations were run so that the downstream area was exposed to lethal concentrations either the night and morning before the day of treatment,
or the following evening and night. This was done to ensure lethal rotenone concentrations also during dusk, night and dawn.

### Repeated treatments

Parts of the treatment area considered to be challenging were treated at least twice during each year’s treatment period. Locations temporary isolated from the rest of the watercourse were treated some days ahead of the main treatment period and were carefully examined for dead fish. This was done both as one of the repeated treatments, but also to even out the need of manpower per day. If no dead fish were found, that location’s treatment was defined as finished for the year. Locations where fish were found were treated again, once or twice, until no more dead fish were observed. Different crews normally conducted repeated treatments to minimize the risk of the same people repeating failures or misjudgements.

If rotenone analyses detected insufficient rotenone concentrations, this also triggered a local repeated treatment.

### Organisation and planning

All mapping and planning of the treatment was done by a working group consisting of six persons from the Norwegian Veterinary Institute. The Environmental department at the County Governor in Troms County was locally in charge of the national eradication program for *G. salaris*, administrated and financed by the Norwegian Environmental Agency.

The region was divided into smaller units, each unit of manageable size for one-day treatment. In total, the region was divided into seven consecutive days of treatment (Table 1). Each day of treatment was mapped and planned by two people. The same two people became operation leaders of that unit from a headquarters with VHF-radio contact with all the treatment teams in the field. In this way all treatment teams could address any question about the maps or treatment instructions to the operation leaders that had done the mapping and planning of that unit. The three groups of two operation leaders were rotated so that they led the treatment every third day, which left them time the day before to update plans and instructions, and the day after to check out the reports from the treatment teams.

One person assisted the treatment leaders, being responsible for the discharge measurements, dosage calculations and other practical and logistical tasks.

### Table 1. Treatment schedule of the region by treatment day and date.

| Treatment area                                      | Treatment day | Date 2016 |
|----------------------------------------------------|---------------|-----------|
| River Skibotnelva, Day 1                           | 1             | Aug 24    |
| River Skibotnelva, Day 2                           | 2             | Aug 25    |
| Rivers in the fjord and pre-treatments in River Signaldalselva | 3             | Aug 26    |
| River Signaldalselva, Day 1                        | 4             | Aug 27    |
| River Signaldalselva, Day 2                        | 5             | Aug 28    |
| River Kitsalselva                                  | 6             | Aug 29    |
| River Balsfjordelva                                | 7             | Aug 30    |
Quality control

The quality control in the project consisted of different measures before, during and after the treatment period. Maps and plans went through a quality check involving people with knowledge of the local river geography and members of the planning group. Inspection of locations of special importance, like for instance the upper migrating barriers in the watercourse defining the treatment area was done at different water levels, and by different people to ensure solid conclusions were made. Juvenile salmonid fish were sampled and preserved in 96% ethanol. During the first year treatment, the focus was to sample and examine fish from the periphery of the treatment area to get data on the prevalence of infected hosts outside the main river. This was considered important data to adjust the treatment plans for the following year’s treatment.

During the second year of treatment, the focus shifted to look for juvenile salmonids older than one year, which indicated survival due to insufficient treatment during the previous year. To be able to sample a maximum number of potentially surviving fish, two teams of free divers searched the deeper parts of the rivers at the end of the dosage period. All juvenile salmonids sampled were examined in a stereo loupe for *G. salaris*.

A lab for rotenone analysis was established at the headquarters of the operation, about 90 minutes by car from the furthest parts of the treatment area. Water samples were collected and analyzed (Sandvik et al. 2018) typically every second hour throughout a 10 hour period. It was therefore possible to adjust dosage during treatment and extend the treatment duration if necessary or put up a supplemental dosage station.

Treatment teams had to fill in a checkout schedule after completing their daily tasks. Schedules were cross-checked against the plans by the operation leaders at the end of each day to ensure that every task in the treatment plan was completed. After the first year treatment, an evaluation was conducted based on observations, fish sampling, parasite examinations, rotenone analyses and feedback from the treatment teams. This was the basis for optimization of the treatment plans for the second year treatment. An evaluation based on the findings in the second year of treatment, along with results from electrofishing and partial treatments in the spring of 2017 was the basis of the decision on whether there should be a third year of full scale treatments.

Mapping groundwater

Several groundwater-fed springs, ponds and sections of the main rivers were previously documented in the investigations after the failed treatments in 1988 and 1995 (Brabrand and Koestler 2003). There was still a need for a total large-scale mapping of all waterbodies with significant groundwater influx with the potential to dilute rotenone concentrations to
Figure 2. Mapping of groundwater influx in the River Signaldalselva. The mapping was done by parallel logging of GPS position and temperatures along the riverbanks at late summer, the time of year with the highest temperature contrasts between surface water and upwelling groundwater. Photograph by Norwegian Veterinary Institute.

sub-lethal levels. This was done by water temperature mapping along the riverbanks of the main rivers and larger tributaries throughout the whole region, logging temperature contrasts indicating significant groundwater intrusion. The temperature mapping was done in late summer when the temperature contrast between groundwater and surface water was expected to be the highest. Special equipment was developed for this task, consisting of a general purpose data logger unit (Squirrel 2010, Grants Instruments) coupled to a high sensitivity, fast responding thermocouple temperature sensor mounted in the tip of a rod (Figure 2). Temperature anomalies were marked on paper maps and located by GPS. A GPS track-log was recorded in parallel with the temperature logging to facilitate the merging of temperature and location data for later analysis. The same riverbanks were also surveyed in the winter, and anomalies in ice and snow cover indicating significant influx of warmer groundwater were mapped.

Groundwater flow was also registered and located by GPS during the general mapping of all waterbodies accessible for fish in the infected area. Groundwater was often visually detectable as springs, brooks and ponds along the rivers and in side-channels with increasing flow.

Reversing groundwater flow through flooding

The Skibotn River is regulated for hydropower production. The main reservoir is located upstream from the infected part of the river, giving a possibility to manipulate the river discharge. For the upper half of the river,
Figure 3. Spraying the riverbank of the River Signaldalselva with water of high rotenone concentration. The iconic mountain Otertind in the background. Photograph by Dag H. Karlsen.

water was released directly from the reservoir, and for the lower half, the water was discharged through the hydroelectric power plant by running the turbines on maximum effect. The strategy was to create a sudden increase in discharge and water level in the river that rises above the surrounding water table, thus reversing the groundwater flow for a period. Juvenile fish hiding in substrate cavities in the hyporheic zone would then be exposed to rotenone treated water. Just ahead of and during the period with high discharge, the rotenone dosage was increased to secure relatively high concentration in the water that intruded the river substrate. Besides the reversing of groundwater intrusion, the aim was to create a flow of rotenone treated water through groundwater-fed side channels, alcoves and ponds along the riverbanks, as well as to make enough turbulence to break through the stagnated layers of cold groundwater in some deep pools.

**Spraying riverbanks**

In unregulated tributaries and in the other rivers of the region, there was no way to create artificial floods like in the Skibotn River. Based on the supposition that most of the groundwater influx is along the banks, the riverbanks were sprayed with large quantities of water with a concentration of approximately 500 ppm of 3,3% rotenone formula (CFT Legumine™). Boat mounted fire-fighting pumps (Sanmit 112HT) were used for the riverbank spraying (Figure 3), while backpack mounted pumps (Figure 4) where uses in locations inaccessible by boat mounted equipment. The aim was to create a temporary reservoir of rotenone in the riverbank, seeping into the outflowing groundwater.
Figure 4. Spraying a groundwater-fed side channel of the Skibotn River with portable backpack mounted pump. Surviving *G. salaris* infested arctic char was found in this location after the previous treatments in 1988 and 1995. In 2015 and 2016 this and similar locations was treated several times by different teams using both Vectocarb, CatSan hygiene litter saturated with CFT-Legumine and conventional spraying with water of high rotenone concentration. Photograph by Dag H. Karlsen.

**Heavy rotenone; Vectocarb™ and CatSan hygiene litter**

Two newly developed techniques for additional dosage in parts of rivers, brooks and ponds with groundwater influx were used in this project (Haukebø et al. 2018). The first was Vectocarb™, a calcium carbonate powder with a high capacity for absorbing the liquid rotenone formula CFT Legumine™. It sinks to the bottom where the rotenone can bleed into the upwelling groundwater. The Vectocarb™ powder was saturated with the CFT Legumine™ and further diluted with water to form a slurry liquid that could flow through the ejector system on the boat-mounted pumps (Sanmit 112HT) used to distribute the slurry over the areas to be treated. Originally Honda WX 10 pumps were assigned for portable use in tributaries, but they malfunctioned because of gravel and pebbles coming in the suction hose.

In smaller brooks and ponds inaccessible by rafts and heavy pumps, CatSan™ hygiene litter saturated with CFT Legumine™ was used in the same way as the Vectocarb™ slurry, except it was spread manually from buckets.

**Rotenone discs**

In springs and brooks that needed continuous dosage over some time, the usual technique was to place drip stations consisting of a 20 litre plastic can with a thin siphon tube that doses diluted CFT Legumine™ over a 4-hour period. In the Skibotn Region, there were a large number of this type of waterbodies, often located far away from roads. Several 20 litre plastic cans
make a very bulky load for a person walking through difficult terrain, and operators have to go back to retrieve the cans after use. This led to the development of the rotenone disc (Figure 5) The disc is a handy, pocket sized “dosage station” consisting of CFT Legumine™ and melted hand soap mixed in a 60:40 ratio and left to cure in a petri dish.

Prior testing showed that a 100–120 gram disc placed in a brook with a 3 l s⁻¹ discharge would dissolve gradually and release the CFT Legumine in a relatively steady rate over a period of 1–2 hours (Helge Bardal, unpubl. data). The number of discs necessary at each location was determined by an on-site evaluation of the discharge.

**Results and discussion**

The total amount of CFT Legumine used was 11 212 litres. The total cost of the treatments are 34,2 million NOK (approximately 3,7 million Euro). Table 2 gives an overview of kilometres rivers treated, annual and treatment discharges, man-hours, and CFT Legumine used in all rivers in the region.
Table 2. Overview of total kilometres rivers treated, annual average and treatment discharges, man-hours, and CFT Legumine used in the 2015–2016 rotenone treatments in the Skibotn Region.

| River name       | Treated river stretch (km) | Average annual discharge (m³/s) | Discharge during treatment 2015 (m³/s) | Discharge during treatment 2016 (m³/s) | Man-hours treatment 2015 | Man-hours treatment 2016 | CFT Legumine used (liters) 2015 | CFT Legumine used (liters) 2016 |
|------------------|-----------------------------|---------------------------------|--------------------------------------|--------------------------------------|-------------------------|-------------------------|---------------------------------|---------------------------------|
| Skibotnelva      | 24                          | 19,8                            | 23,8                                 | 22,8                                 | 1632                    | 2196                    | 2340                            | 2513                            |
| Signaldalselva   | 35,3                        | 16,4                            | 20,5                                 | 5,4                                  | 1992                    | 2364                    | 2101                            | 1409                            |
| Kridalselva      | 16                          | 4,4                             | 4,3                                  | 2,1                                  | 756                     | 816                     | 346                             | 149                             |
| Balsjordelva     | 10                          | 2,5                             | 5,4                                  | 3,9                                  | 756                     | 792                     | 850                             | 471                             |
| Other rivers     | 9,4                         | 0,4–4,5                         | 0,6–9,4                              | 0,3–9,0                              | 420                     | 384                     | 584                             | 449                             |

The Skibotn River is regulated, so the discharge was planned by the operation leaders and is nearly the same for both years. In River Signaldalselva there is no regulation, and the discharge was almost four-fold in 2015, compared to 2016, due to rainy conditions. Increased discharge generally does not require more workforce, only an increase in CFT Legumine dosage.

The total effort per river is calculated in man-hours, and is based on an average of 12 hours per person per day. On average 50 persons (42–54) per day worked in treatment teams, directly involved in treatment operations. 21 persons had other duties per day, including operating leaders, staff for operating leaders, equipment storage and maintenance service, transport, welfare team (soda, snacks and waffle service to treatment teams), discharge measurements, VHF communication operation, safety representative, laboratories for chemical analysis, and laboratories for dead fish registration. Crew for dead fish collecting is not included; it varied depending on rivers size and aim of collection. The increase in workforce in 2016, based on the experiences from 2015, did slightly increase the number of treatment teams per day, but the main increase in workforce was pre-treatments. The pre-treatments aimed to even out and lower the number of treatment teams each day. Board and lodging was at a local hotel.

The result of the eradication project in the Skibotn Region is not yet known. In most cases of failed *G. salaris* eradications, the parasite has been detected 2–3 years after the treatment. In the Skibotn Region in the 1988 and 1995 treatments, the infection reappeared respectively after 4 and 3 years. The post-treatment surveillance program cannot conclude on eradication confirmation until 5–6 years after the last treatment, meaning 2021–2022. Still, it can be useful to describe the results and evaluate every strategy choice within the project before the ultimate answer is ready.

It was impossible to quantify the risk of reinfection from neighbouring untreated rivers during the treatment period. The probability of fish migrating from infected untreated rivers and re-infecting newly treated nearby rivers during a short treatment period was small, but still worth eliminating by simultaneous treatment periods. Studies of sea trout and anadromous arctic char in the region have proven that adult individuals of these species can migrate between the rivers in a time that allows *G. salaris* to survive and spread between the rivers in periods with low salinity (Jensen 2013).
Mapping groundwater influx to the treatment area by thermal contrasts between groundwater and surface water confirmed the observations of earlier studies in the river (Brabrand and Koestler 2003). It also revealed previously unknown parts of the rivers with massive influxes of groundwater, both in the River Skibotnelva and in the other infected rivers. Locating and planning special treatment strategies for these parts of the watercourses was probably one of the most important measures to be able to succeed with the eradication program.

Quality control revealed several incidents or failures that separately could have altered the result of the eradication project. Rotenone analyses in the Skibotn River showed both low concentrations downstream from a primary dosage station because of wrong instructions to the dosage station operators, and sections of the river with far more rotenone dilution/loss than expected. Concentrations were adjusted to correct levels within the same day. In the previous failed treatments these corrections were not possible, and with the relatively low rotenone concentration allowed by the pollution authorities in 1995, failure was a very probable result.

A planned extra treatment in a spring fed brook in the first year of treatment revealed a high number of surviving fish due to insufficient treatment. The strategy of changing crew for repeated treatments might have been important in this case. Cross-checking the reports of the treatment teams in one case revealed an untreated side-channel in the Skibotn River.

By treating every possible location partly or temporarily isolated from the main rivers ahead of the main treatment period in 2016, it was possible to even out the need for manpower between days, keeping the total numbers of crew down. The pre-treatment of this type locations gave useful information about where fish possibly had survived during the first year of treatment.

In the River Kitdalselva, *G. salaris* was found for the first time on juveniles of Atlantic salmon and arctic char killed in the treatment. These findings confirmed the assumption that this river was infected, despite no previous detections.

During the second year treatment, free diving teams sampled a relatively large number of small arctic char and brown trout suspected to be survivors of the first year treatment. Age determination showed that these individuals were relatively old compared to the juveniles in the anadromous population, and they were most likely fish who had migrated from resident populations upstream of the treatment area. No *G. salaris* was found during the examination of the sampled material, nor were any juvenile Atlantic salmon detected in the river the second year. This led to the conclusion that the parasite was probably eradicated already after the first year of treatment. The quality control based on the sampling of the diving teams was thereby the main basis for the decision not to plan a third year of treatment.
Visual observations during the artificial floods of rotenone-treated water confirmed that the discharge was high enough to fill the entire riverbed, including groundwater-influenced side channels previously without throughput from the main river. The alternative to flooding the riverbed with rotenone treated water would have been the highly resource demanding and time-consuming spraying of the riverbanks and side channels with rotenone treated water. Being released from this task, there were more resources to put into repeated treatment of challenging areas in tributaries and wetlands along the river.

Temperature logging in a groundwater-fed riverbank documented the effect of the intrusion of surface water at a substrate depth of 10 cm (Figure 6). We assume that the rapid temperature rise indicates an instant intrusion of surface water through the substrate mainly consisting of coarse gravel and pebbles. The higher temperature equaled the surface water and lasted as long as the artificial flood, indicating that rotenone treated surface water kept on intruding the substrate as long as the water level in the riverbed was higher than the surrounding groundwater table. We also assume that substrate consisting of finer organic matter that could have absorbed significant amounts of rotenone do not have the cavities necessary to act as refuges for juvenile salmonids. This means that infected juvenile salmonids hiding in cavities in groundwater-fed substrate were very likely to be exposed to lethal doses of rotenone.
Figure 7. Increase in rotenone concentration in water samples along the riverbank as result of spraying the bank with water of high rotenone concentration.

Spraying the riverbanks with water of high rotenone concentration has been a standard procedure in G. salaris eradication projects since 2003 and was implemented parallel to the two years of treatment strategy. In River Signaldalselva the effect was documented at two water sampling locations where samples were taken a short time after the riverbank spraying teams had passed. The rotenone concentrations along the riverbank peaked at 176 and 214 µg/litre in these samples, 5–7 times the general concentrations (Figure 7) The one-hour sampling frequency do not document the duration of the highest elevated concentrations, but they remain higher than the threshold level also for the next two succeeding samples. This shows that the riverbank spraying positively affected the rotenone concentration along the riverbank for at least 3 hours. A dose of 25 µg/l in 30 minutes has proven to be sufficient for 100% mortality of Atlantic salmon juveniles (Mo 2000).

The two different “heavy” inert media for carrying rotenone, Vectocarb™ and CatSan™ hygiene litter were used in small-scale tests (Haukebo et al. 2018) in the first year of treatment and in full-scale for the first time in the second year of treatment in the Skibotn Region. The manual application of the CatSan™ hygiene litter proved to be an easy and robust strategy for smaller groundwater upwelling areas and wetlands with only a few centimetres of water, where there was not enough water for the pumps to work properly. Application of the Vectocarb worked fine when using the same type of fire pumps as for the riverbank spraying. However, because of the very fine grain of the slurry particles, the plume of slurry was transported some distance downstream before it settled in the river substrate. To reduce the
downstream drift of the media, the slurry was distributed close to the bottom upstream from the treatment area, rather than distributing it over the water surface.

The use of the rotenone disc was a huge simplification compared to the traditional drip-stations, making the crew able to dose a large number of small, fast running springs and brooks spread over large distances in an efficient way. Rotenone discs and CatSan™ hygiene litter serve distinct purposes. Rotenone discs can only be used in running water, and it was not known if CatSan™ hygiene litter could hold lethal concentration in running water over time. CatSan™ hygiene litter was ideal to spread on larger surfaces with diffuse sources of upwelling water.

**Conclusion**

The constant improvement in strategies and techniques of rotenone treatments since the 1980s and 1990s has led to a robust method with the potential to eradicate the *G. salaris* hosts from large and complex regions of rivers. The main improvement of dosage strategy has probably been treatments for two consecutive years. For the Skibotn River, flooding the riverbed with high discharge from the hydropower reservoir eliminated dilution effects from groundwater along banks and in deep pools. The procedures for planning and quality control developed during the last decade of eradication projects led by the Norwegian Veterinary Institute has made eradication campaigns of this scale possible.

Assuming that the third eradication attempt in the Skibotn Region was a success, the introduced *G. salaris* is now restricted to two regions in south-eastern and north-western Norway, with a total of eight salmon rivers left to be treated. New strategies using chlorine as the main chemical are under development, potentially giving a method non-lethal to fish. The principles and techniques of the large scale rotenone treatment against hosts of *G. salaris* are also transferable to other freshwater fish eradication projects.

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