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VACCINATION OF ARCTIC CHAR (<u>SALVELINUS</u> <u>ALPINUS</u>) AGAINST VIBRIOSIS

by

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ABSTRACT

Arctic char of six different strains have been vaccinated against vibriosis and a relative protection (RPP) of 98% was demonstrated by waterborne challenge.

This species is highly susceptible to vibriosis and the gross pathology is similar to vibriosis in rainbow trout. Sex maturation and genetics may have some impact on disease resistance

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INTRODUCTION

Currently, commercial farming of salmonids in Norway is almost entirely confined to Atlantic salmon (<u>Salmo salar</u>) and rainbow trout (<u>Salmo gairdneri</u>). However, other salmonid species such as Arctic char (<u>Salvelinus alpinus</u>) are available. Arctic char is exposed to seawater when reared in brackish water or when reared in freshwater treated with seawater due to low pH. When exposed to seawater there is a risk of exposure to <u>Vibrio</u> <u>sp</u>. The Arctic char is highly susceptible to vibriosis (Østhus 1976; Grotnes 1987) and this study was undertaken to evaluate the effect of vaccination against vibriosis.

MATERIAL AND METHODS

Fish

Six different strains (two northern anadromous, four southern landlocked) of fin clipped Arctic char were used in the experiment. At the time of vaccination, the fish were kept in 1 m³ tanks in freshwater at 12-14 °C. Two weeks before challenge the fish were transferred to 400 l aquaria and the salinity was adjusted to 6 ppt with UV-treated seawater. They were fed Ewos (TM) dry pellets and were actively feeding before challenge. The temperature was adjusted to 9 °C. Relative gonad length (RGL) was used as an unbiased estimate for maturation.

$RGL = \frac{Length of swollen gonad}{Total abdomen length}$

<u>Vaccine</u>

A commercial available vaccine (Vibriovaks^R Leo vet.) containing trypsin treated Norwegian serotypes of <u>Vibrio anquillarum</u> was used for vaccination. Vaccination was performed by intraperitoneal (i.p.) injection of 0.1 ml undiluted vaccine.

The unvaccinated control fish were marked by injecting subcutaneous Alician blue creating a blue spot on the abdomen posterior to the pectoral fins (Johnstone 1981).

After vaccination the fish were kept for ten weeks at 12 - 14 °C before challenge.

Challenge

A waterborne challenge was performed by exposing the fish to 1.6 x 10^G bacteria ml⁻¹ for 80 minutes, and the temperature was raised from 9 °C to 12 °C. After challenge, the fish were kept at 12 °C and 6 ppt salinity. The bacteria used for challenge was a Norwegian serotype of <u>Vibrio anquillarum</u> (NCMB 2129) isolated from rainbow trout (Egidius and Andersen 1977). Biochemically on API 20 and API 50 this bacteria is identical to <u>Vibrio anquillarum</u> isolated during two outbreaks of vibriosis in farmed Arctic char in 1987 and 1988.

A total of three aquaria with a mixture of all six strains were challenged. Two with vaccinated fish, containing 80 fish each and one with a mixture of 30 vaccinated and 51 unvaccinated fish.

Dead and moribund fish were collected. Cultivation of bacteria from the kidneys was attempted on tryptone soy agar (TSA) with 15 ppt NaCl incubated at 20 °C. Four weeks after challenge 20 survivors from each group were sacrificed and examined for presence of bacteria in the kidneys.

Protection was calculated as relative percentage protection (RPP).

 $RPP = (1 - \frac{Mortality in vaccinated}{Mortality in controls \%}) \times 100$ Length, weight and maturation were registered for all fish.

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RESULTS

Four days after challenge unvaccinated fish began dying. The most common external symptoms were haemorrhages on the fin base, a red and swollen anal region and haemorrhages in the head region. Petechia on the abdomen and on the operculum was also seen. Internally the spleen was enlarged and swollen and occasionally entirely liquified. Ascites, haemorrhages on the swim bladder and peritoneum, inflamed intestine and extensive haemorrhages on the liver were common findings (Table 1).

Table 1. Gross pathology of fish dying of vibriosis

Gross pathology	No of fish affected/no of fish dying of vibriosis
Haemorrhages on the fin base	13/16
Red and swollen anal region	7/16
Petechia on the head region	8/16
Petechia on the abdomen	2/16
Petechia on operculum	2/16
Enlarged/swollen spleen	13/16
Haemorrhages on swim bladder	8/16
and peritoneum	
Haemorrhages on the liver	8/16
Ascites	8/16
Inflamed intestine	8/16

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In the mixed group, 18 of the non vaccinated fish and none of the vaccinated died (Fig.1). In the two other groups, two fish died. This gives a RPP of 98 %.



Fig 1. Cumulative mortality in non vaccinated Arctic char challenged with <u>Vibrio anguillarum</u>

<u>Vibrio anquillarum</u> was cultivated from the kidneys of all dead and moribund fish except for one unvaccinated and the two vaccinated. These fish showed no symptoms of vibriosis. No bacteria were cultivated from the survivors and they showed no sign of vibriosis.

There was no size-selective (weight) mortality observed in the control group (Students t-test, p=0.14). The mature or maturing fish pooled were not significantly more susceptible to vibriosis (Mann-Whitney, p> 0.05). The mortality was 26.7% among non-mature control fish, compared to 39.4% of mature/-maturing control individuals. The mortality among mature/-maturing males in control was 23.1%, among mature/maturing females was 45.5%.

The mortality in the control group was not significantly strain specific. However, when the two northern anadromous strains are pooled, 42.9% of the unvaccinated fish died compared to 25.0% of the four southern landlocked strains.

DISCUSSION

Arctic char can be successfully vaccinated against vibriosis by i.p. injection.

When vaccinating rainbow trout (<u>Salmo gairdneri</u>) against vibriosis, injection is superior to dip and bath when vaccination methods are compared (Horn <u>et al</u>. 1982, Håstein and Refstie 1986). This might also be true for the Arctic char. In this experiment i.p. injection gave a RPP of 98 %. A single dip vaccination seems to be unsatisfactory (Barnung and Holm 1988).

The bacteria used for challenge was a Norwegian serotype of <u>Vibrio anquillarum</u> (NCMB 2129). This serotype has the same biochemical characteristics (API 20, API 50) as <u>Vibrio anquillarum</u> isolated during natural outbreaks of vibriosis in Arctic char. A protection satisfactory for commercial aquaculture is therefore to be expected.

Arctic char is highly susceptible to vibriosis and the disease can be transmitted through the water. The gross pathology is similar to vibriosis in rainbow trout where a swollen spleen is most characteristic (Egidius 1987) .

Of the non vaccinated fish, the mature/maturing females were more susceptible to vibriosis than the non matures and the mature/maturing males. When non vaccinated fish are exposed to seawater and thus running a risk of catching vibriosis, the percentage of mature/maturing females in the population may have some impact on the survival rate. The same tendency might be expected for the males, although not shown here.

The six different strains did not differ significantly in susceptibility to vibriosis. However, when pooled, the northern anadromous strains seemed to be more susceptible than the southern landlocked strains. This should be considered when choosing strains to be reared in brackish water. Vibriosis appears to be a limiting factor when Arctic char is reared in brackish water (Østhus 1976). Vaccination is therefore highly recommended. If vaccination is not possible, southern landlocked strain with late maturation might be the best choice.

Nordeng (1983) demonstrated that the potential for anadromy exists in populations of Arctic char in the southern non anadromous area in Norway. He states that one reason for resident behaviour is the ureter fluke <u>Phyllodistomun conos-</u> <u>tomum</u> olsson. However, if infections really affects the migration behavior in char, vibriosis may be a disease of importance. At least in culture, vibriosis is the most serious disease. REFERENCES

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