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NATURAL INFECTION OF FARMED ATLANTIC SALMON, *Salmo salar* L., PARR BY GLOCHIDIA OF THE FRESHWATER PEARL MUSSEL, *Margaritifera margaritifera* L.

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During December 1986 we examined a high prevalence of white raised spots on the gill lamellae of a population of farmed Atlantic salmon, *Salmo salar* L., parr reared in tanks. A moderately heavy invasion of the glochidial stage of *Margaritifera margaritifera* L., was identified. This is the first reported occurence of this condition from Scottish fish farms.

The freshwater pearl mussel, *M. margaritifera* lives in fast flowing cool waters which are relatively low in calcium, thus restricting its range in Britain to rivers and streams to the west and north of the country (Young & Williams, 1983). A hookless, subovate glochidial stage is released annually in large numbers and is parasitic on the gills of the Salmonidae and occasionally other fish and this maintains the upstream adult populations (Bjork, 1962). The reproductive biology of the freshwater pearl mussel is reviewed by Young & Williams (1984).

The non-swimming glochidia undoubtedly reached the gill lamellae of the parr passively in the ventilating current. The consequence of prolonged attachement of the glochidia on the parr was unknown, but it was believed the glochidia would remain on the gills and continue their development until the temperatures rose during the following year. This study was designed to investigate whether there was a significant risk of growth being compromised or significant mortalities resulting from secondary infections and fluid loss following sloughing of the glochidia from the gills during their natural migration from the fish.

In addition a variety of methods of removing the glochodia from the gills were examined. Due to the size of the farm and the large number of fish involved any treatment methods would (a) need to have practical applications on the fish farm (b) have to achieve a high degree of success to justify the effort and (c) use chemicals and methods recognized for use on fish farms. The histopathology of this host-glochidia interaction is poorly described for Atlantic salmon and therefore was also included in this study.

Trials were either carried out within the affected fish farm or on a population of fish moved from the farm to an experimental site. The following treatments were carried out on at least 20 parr in each experiment: (a) Bathing in salt water at concentrations of 5.4, 10.1 and 24.2 ‰ for 1, 3 and 9 hours and in 33.3 ‰ for 24 hours, (b) bathing in 0.5 mg l^{-1} $CuSO_4$ for 1h, (c) bathing in 5 mg l^{-1} Nuvan for 1h followed by 1 mg l^{-1} Roccal for 1h. Fish were also held in water raised from 3-5°C ambient at the time of sampling to 10°C for three weeks in an attempt to stimulate natural detachment of the glochidia and to gain early evidence of any associated problems. Entire gill arches (hemibranch) were carefully dissected from treated and control groups at 1, 2 and 3 weeks post treatment and fixed in buffered

20% formalin. The total number of glochidia on the ventral and dorsal surface was recorded using low power magnification. Both right and left gill arches were examined. No variation in the number of glochidia was found between the left or right side so only the right side was processed during subsequent sampling. Adjacent gill arches from these fish were sectioned and stained with haemotoxylin and eosin (H&E) and the number of glochidia present in a mid-tissue section recorded. The development of the glochidia and the host response was examined histologically. Monthly sampling of the farmed stock was carried out between December and May. Growth of the farmed fish was continually monitored.

Bathing in sea water, Nuvan and Roccal, $CuSO_4$ or raising the water temperature did not kill or dislodge a significant number of glochidia from the parr when compared with control groups as determined by counting cysts on whole gill arches and examining histological sections. No mortalities occured following these treatments. During regular monitoring of the farmed stock there were no significant mortalities attributed to the glochidia, no secondary infections and growth was not compromised in comparison to uninfected parr reared in tanks on the experimental site.

The mean number of glochidia on the gill arch of the parr at the start of this outbreak through smolting in the following May/June, is reported in Table 1.

The following year class (1987 hatch) was similarly sampled beginning in September.

In December, at least 90% of the 1986 hatch were carrying a moderate burden of glochidia which had encysted on the gills, predominantly on the middle section of each hemibranch (Fig. 1), but occasionally they were attached to the gill rakers. The glochidia appeared smooth, opaque-white, and slightly ovoid. A maximum number of 736 glochidia per fish (calculated by multiplying the number of glochidia / gill arch by the number of gill arches) was recorded in this study.

Meyers & Millemann (1977) reported that the LD_{50} for Atlantic salmon fry is 2300 attached glochidia, hence it was concluded that the lack of significant mortalities found among the farmed fish was due to the lower level of infection and the larger size of the farmed fish. By January the number of glochidia on the gill lamellae had declined by over 50%, although in subsequent sampling through to May there was no further significant decline. In June when the 1986 S1 stock had completed smolting the mean number of glochidia per gill arch had declined by 84%. There was no significant difference in the number of glochidia on the ventral or dorsal gill arch of the potential S1 or S2 parr (Table 1). There were no glochidia on the S1 post-smolts in September (Table 1). This data was confirmed by the counts carried out on the stained gill arch sections. The S2 population were killed in the summer so further data on this population is not available. Although the adult mussels were translocated prior to July from the water intake of the farm and from 500m of the river upstream the 1987 hatch became infected by glochidia in September. It was believed the 1986 hatch had been infected at a similar time. A comparison of the two years data suggests that a significant number of glochidia are lost during the first few months post-attachement (Table 1). The loss of the glochidia during this time was considered to result from incomplete attachement and may have been aided by respiratory movements.

The histopathology of the glochidia infection in Atlantic salmon parr appears to be similar to the observations made by Karna & Millemann (1978) who reported on experimentally infected alevin chinook salmon, *Oncorynchus tshawytscha*

Walbaum. In the Atlantic salmon examined here, each glochidia was covered by a host epithelial layer which rapidly developed into an extensive hyperplasia, gradually proliferating to involve adjacent lamellae, particularly when the glochidia continued their development to juvenile mussels during the spring (Fig. 2).

In contrast, Fustish & Millemann (1978) found in experimentally infected coho salmon *O. kisutch* Walbaum that sloughing of the glochidia was nearly complete by 14 days post infection. Meyers, Millemann & Fustish (1980) suggested that apparent resistance to the glochidia in coho salmon may be due, in part, to humoral factors. In the present work, where encystment had occured at the tips of the Atlantic salmon gill lamellae they were often clubbed and bent, the cellular proliferation fusing with other adjacent lamellae. Despite a well developed hyperplasia the glochidia only appeared to have sloughed from the filament tips, and this was recorded histologically as empty cysts with ruptured walls associated with the presence of aneurysms. Haemorrhaging from the gills and lifting of the epithelium on secondary lamellae was noted in the present study, particularly in those fish where a large number of glochidia had attached.

It is concluded that although the appearance of the glochidia on the gills is spectacular and a localized pathology occurs, the survival and growth of the parr in fresh water and of the smolts on their subsequent transfer to sea water was not measurably impaired at the intensity of glochidia infection recorded.

Summary

A high prevalence and moderately heavy invasion by mussel glochidia of the fresh water pearl mussel, *Margaritifera margaritifera* on the gills of farmed Atlantic salmon, *Salmo salar* parr is reported. Attempts to remove the glochidia by raising the salinity, bathing in Nuvan followed by Roccal; $CuSO_4$ bath treatment; or raising temperature, had no success. There was no significant mortality, reduction in growth, or serious pathology of the gills of infected fish in fresh water or after smolt transfer to sea water.

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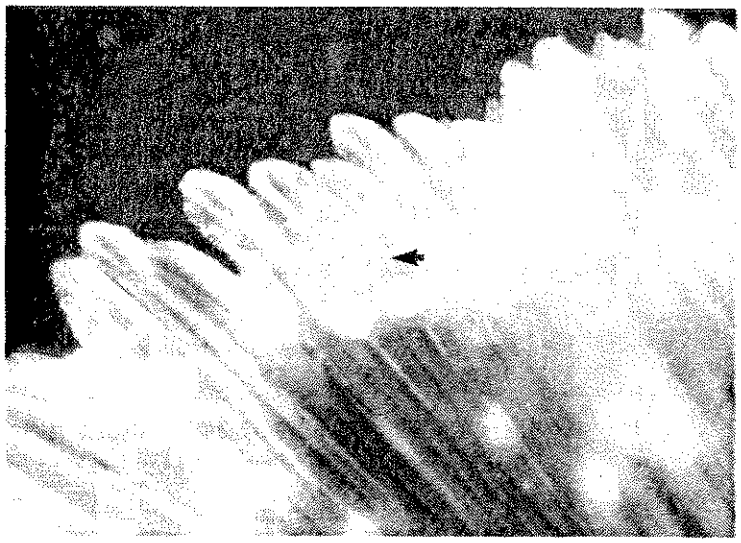
Table 1: Mean number of glochidia of the fresh water pearl mussel *Margaritifera margaritifera*, counted on a single gill arch of farmed Atlantic salmon, *Salmo salar* parr

Month	Mean number of glochidia Potential S1's		Number of fish examined	Mean number of glochidia Potential S2's		Number of fish examined
	Ventral gill arch	Dorsal gill arch		Ventral gill arch	Dorsal gill arch	
December	50	45	4	NS	NS	-
January	22	20	7	35	20	4
February	21	21	8	29	26	9
March	27	28	6	20	18	6
April	30	31	9	15	9	4
May	21	15	6	18	12	6
June	8	8	10	9	14	4
September *	0	0	5	NS	NS	-
September **A	77	60	4			
September **B	50	41	4			

* Post smolts NS = not sampled
** 1987 hatch A = 6 mm grade B = 7 mm grade



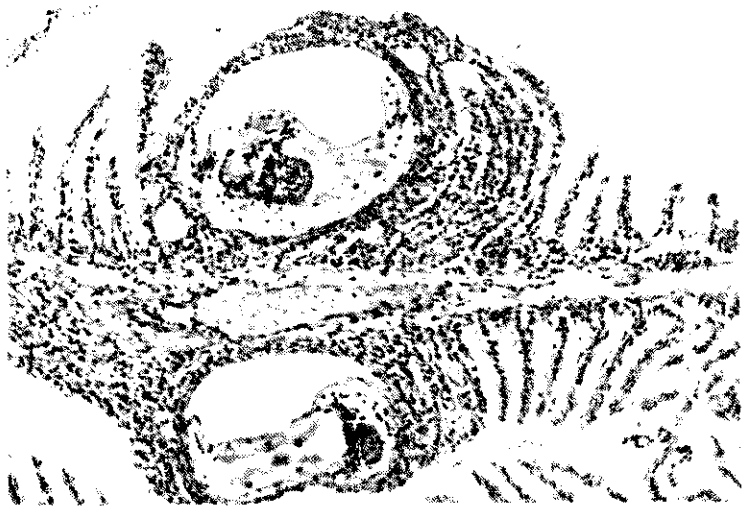


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Fig. 1. Dissected gill arch from a farmed Atlantic salmon, *Salmo salar* with the attached mussel glochidia of *Margaritifera margaritifera*. A hyperplastic reaction (arrowed) can be seen covering the glochidia.

Fig. 2. *Margaritifera margaritifera* glochidia attached to the secondary lamellae of a farmed Atlantic salmon, *Salmo salar* parr sampled in January. (H&E, x145).