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Articles, letters and news relating to trout farming are always welcome and may be included in future issues.

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TROUT PRODUCTION

1998 SURVEY OF TROUT PRODUCTION IN SCOTLAND

The figures for rainbow trout, egg production and egg imports for Scotland were not available in time for inclusion in the July 1999 edition of Trout News. This data is now included here and is supplied from SERAD (Rural Affairs Department of the Scottish Executive, formerly SOAEFD) Annual Production Survey, 1998.

Rainbow trout were produced from 71 sites involving 56 companies with an overall production of 4913 tonnes in 1998 (4,653 tonnes in 1997) an increase of 260 tonnes on the previous year (a rise of almost 6%). Trends in production over the last 10 years are given in Table 1 below.

Table production

Table 2 gives trends in production for table fish over the past 6 years. Production in 1998 amounted to 4,069 tonnes representing an increase of 221 tonnes (5.7%) on the previous year.

Fish of less than 450 grams (<1 lb) continued to form the bulk of production (74%) and significant demand for large fish weighing >900g/2 lbs comprised 21.8% of production, primarily for smoking. The table trade accounted for 82.8% of total production.

Restocking production

Table 3 provides production data for the restocking trade for the last 6 years. Production for restocking increased by 39 tonnes (4.8%) to 844 tonnes representing 17.2% of the total production (17% in 1997).

Table 1. Total production for the period 1989-1998

Year	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998
Tonnes	3,512	3,183	3,334	3,953	4,023	4,263	4,683	4,630	4,653	4,913

Table 2. Production of table fish for the period 1993-1998

Year	<450 g < 1 lb	450-900 g 1-2 lb	>900g >2 lb	Total tonnes
1993	2,481	272	764	3,517
1994	2,376	288	1,038	3,702
1995	2,736	199	1,149	4,084
1996	2,701	181	1,002	3,884
1997	2,646	104	1,098	3,848
1998	3,009	173	887	4,069

Table 3. Production for the restocking trade in 1993-1998

Year	<450 g < 1 lb	450-900 g 1-2 lb	>900 g >2 lb	Total tonnes
1993	124	346	36	506
1994	125	337	99	561
1995	107	411	81	599
1996	188	484	74	746
1997	97	589	119	805
1998	69	538	237	844

Method of Production

Table 4 provides a breakdown of trout farms by system and scale of production. Compared with 1997 production from freshwater cages and freshwater ponds and raceways increased by 173 (10%) and 640 tonnes (38%) respectively. Production from freshwater tanks and hatcheries and seawater cages on the other hand decreased by 473 tonnes (72%) and 80 tonnes (13.7%) respectively. Freshwater production (4,409 tonnes) accounted for 90% of total Scottish production.

Production and manpower by region

The regional production and manpower information shown in Table 5 relate to Scottish Local Government Regions following their reorganization in 1996. These are shown in Figure 1.

Historically rainbow trout production has tended to be located in the south of Scotland due largely to the location of the pioneering producers and the relative easy access to markets. This pattern of development has continued and relatively few rainbow trout farms have been established in the north of Scotland.

Mean productivity ranged from 18.3 to 28.3 tonnes/person between production areas, productivity being greatest in the South and least in the Northern area.

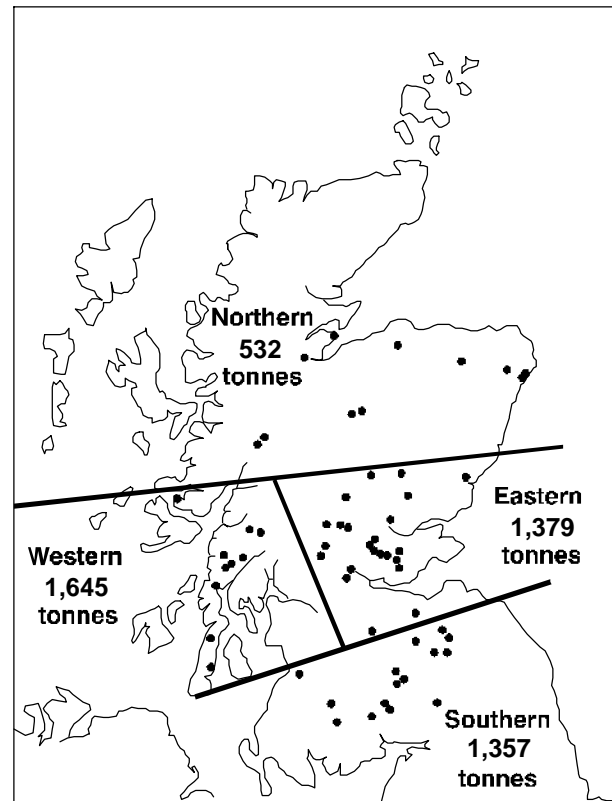


Figure 1. Map of Scotland showing the four trout production areas

Table 4. Analysis of rainbow trout farms by system and scale of production

Production method	Production grouping (tonnes) in 1998					Total tonnage	Total no. of sites*	% contribution
	<10	10-25	26-50	51-100	>100			
FW cages	1	4	0	2	5	1,902	11	38.7
FW ponds & raceways	4	7	4	5	9	2,323	20	47.3
FW tanks & hatcheries	7	2	0	2	0	184	19	3.7
SW cages	1	0	0	1	2	504	7	10.3
Total	13	13	4	10	16	4,913	57	100

* Excludes sites which specialise in the production of ova, fry/fingerling for on-growing

Table 5. Rainbow trout production and staffing by area in 1998

Area sites	No. of	Production			Mean tonnes/sites	Staffing			Productivity tonnes/person
		Table	Restocking	Total		F/T	P/T	Total	
North	10	411	121	532	53.2	17	12	29	18.3
East	21	1,069	310	1,379	65.6	39	11	50	27.6
West	20	1,503	142	1,645	82.2	45	14	59	27.9
South	20	1,086	271	1,357	67.8	36	12	48	28.3
All	71	4,069	844	4,913	69.2	137	49	186	26.4

Ova production

The number of eyed ova laid down for hatching from home-produced stock, from other sources within Great Britain and from foreign imports are given in Table 6 for the period 1993 to 1998. The proportion of ova laid down from GB broodstock increased to 2.6 million representing 10% of the total. The total number of eyed-ova laid down increased by almost 2.2 million (9.5%) on the 1997 figure.

Type of ova

Details of the number and type of ova laid down for hatching are given in Table 7. The preference for all female diploid stock was again evident, accounting for 92% of all ova laid down. Triploid ova laid down increased again in 1998 mainly for use by the restocking trade.

Table 6. Number and sources of ova laid down for hatching in 1993-1998

Year	Own stock	Other GB Stock	Total GB	Total foreign	Grand total	% GB
1993	1,830,000	405,000	2,235,000	17,509,000	19,744,000	11.3
1994	479,000	625,000	1,104,000	18,500,000	19,604,000	5.6
1995	165,000	360,000	525,000	20,310,000	20,835,000	2.5
1996	420,000	988,000	1,408,000	21,270,000	22,678,000	6.2
1997	1,232,000	837,000	2,069,000	21,434,000	23,053,000	9.0
1998	2,559,000	60,000	2,619,000	22,623,000	25,242,000	9.0

Table 7. Number and proportions (%) of ova types laid down for hatching in 1992-1998

Year	Total ova	All female diploid Nos. (%)	Triploid Nos. (%)	Mixed sex diploid Nos. (%)
1992	21,408,000	18,099,000 (85)	796,000 (4)	2,513,000 (12)
1993	19,744,000	17,261,000 (87)	1,396,000 (7)	1,087,000 (6)
1994	19,604,000	18,105,000 (92)	1,134,000 (6)	365,000 (2)
1995	20,835,000	19,546,000 (94)	1,170,000 (6)	119,000 (+)
1996	22,678,000	21,308,000 (94)	935,000 (4)	435,000 (2)
1997	23,503,000	21,118,000 (90)	1,386,000 (6)	1,000,000 (4)
1998	25,242,000	23,222,000 (92)	1,515,000 (6)	504,000 (2)

SUMMARY OF UK RAINBOW TROUT PRODUCTION IN 1998

Details of rainbow trout production both for the table trade and restocking are given in Table 1 below for England and Wales, Scotland and Northern Ireland.

Total production in 1998 amounted to 16,109 tonnes (16,037 tonnes in 1997) representing an increase of 72 tonnes on the previous year.

Table 1. UK Rainbow trout production for 1998

	Production in tonnes		
	Table	Restocking	Totals
England and Wales	6,980 (70.2%)	2,957 (29.8%)	9,937
Scotland	4,069 (82.8%)	884 (17.2%)	4,913
Northern Ireland	1,199 (95.2%)	60 (4.8%)	1,259
Totals	12,248 (76%)	3,861 (24%)	16,109

SCOTTISH RAINBOW TROUT EGG IMPORTS IN 1998

The number and source of imported rainbow trout ova for 1998 are given in Table 1 below. The total imported - over 22.6 million represents an increase of 1 million egg over the 1997 figure (21.6 m)

The quantity of out of season ova imported from South Africa has increased annually since 1994 and now represents over 51.2% of all imports into Scotland. In contrast, imports of northern hemisphere eggs have decreased by approximately one million, a difference of 8%.

Table 1. Number and source of trout ova imported into Scotland plus number of consignments (in brackets) by month in 1998

Month	Northern Ireland	Isle of Man	Denmark	South Africa	Total
January	400,000 (1)	820,000 (4)	600,000 (3)	-	1,820,000 (8)
February	-	500,000 (2)	300,000 (1)	-	800,000 (3)
March	-	-	2,600,000 (7)	-	2,600,000 (7)
April	600,000 (2)	-	1,700,000 (2)	-	2,300,000 (4)
May	350,000 (3)	-	500,000 (2)	-	850,000 (5)
June	-	200,000 (1)	-	600,000 (2)	800,000 (3)
July	-	-	-	10,985,000 (12)	10,985,000 (12)
August	440,000 (3)	-	-	-	440,000 (3)
September	190,000 (1)	-	-	-	190,000 (1)
October	25,000 (1)	50,000 (1)	-	-	75,000 (2)
November	-	803,000 (2)	-	-	803,000 (2)
December	60,000 (1)	901,000 (1)	-	-	961,000 (2)
Totals	2,065,000	3,274,000	5,700,000	11,585,000	22,624,000
Consignments	12	11	15	14	52

Total egg imports

The total number of eggs imported into the UK from foreign sources in 1998 amounted to a little over 61

million (66 million in 1997) representing a decrease of 5 million (8.2%) on the previous year.

ARTICLES

FARMED FISH QUALITY CONFERENCE BRISTOL, APRIL 1999

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*This article concludes Alasdair Scott's report of the Bristol Conference -
the first part appeared in Trout News No.28, July 1999.*

Slaughter techniques

Dr Ulf Erikson, a Norwegian consultant, spoke about the effects of pre-slaughter fasting, handling and transport. He described the metabolic changes that occur when fish are stressed, and concluded that current fasting strategies for salmonids pre-harvest do not significantly affect the quality of the harvested fish.

Investigations into Atlantic salmon transport revealed that loading and unloading were the major stressors and that there was an element of recovery from stress during transportation in well-boats. The quality of fillets appeared to be unaffected by transportation, except when the transport water was superoxygenated. Surprisingly, transport was better where carbon dioxide levels were allowed to increase, as this reduced the levels and toxicity of any ammonia excreted by the fish during transport. While high oxygen levels help maintain fish muscle pH and thereby increase shelf life, supersaturation of oxygen leads to soft muscle texture, mucous on the skin and an overall reduction in product quality.

Dr Erikson suggested that a system involving the use of CO₂ as a narcotic, together with chilling of transport water to 1°C during transport could maximise the quality of harvested salmon. This system reduces the activity of the fish by lowering their metabolic rate, thereby reducing physical damage during transport and acute stress response at unloading. This could reduce the strength and onset of rigor and increase the product shelf life, while maintaining the textural qualities of the fish. The welfare implications of using CO₂ and chilling were questioned, and it will be interesting to see how such technology develops in the face of increasing concerns over fish welfare.

Dr David Robb (University of Bristol) then spoke about humane slaughter and fish quality. He suggested there were two quality criteria to satisfy when harvesting fish. First, the ethical quality of the slaughter procedure and, second, the flesh quality of the harvested fish. Farmers should aim to produce high quality products by both criteria. Dr Robb then described some common slaughter procedures and the problems they pose.

Crowding, removal from water by hand or pump, killing in air, in ice slurry or in carbon dioxide all cause significant stress, and can lead to physical injury. The vigorous fish movements induced during harvesting and slaughter cause an increase in respiration rate, leading to lactate build-up, use of oxygen and ATP in muscle, and resulting in a reduction in muscle pH. The result is a more rapid and stronger rigor, increased drip loss and softening, and paleness of the flesh.

There is therefore a need to reduce pre-slaughter fish activity and develop instantaneous killing methods. The percussive stun and ike jime killing methods used in the salmon industry are appropriate methods, but no equivalents are yet available for smaller species.

Dr Robb suggested that the industry should adopt slaughter methods which took account of fish welfare, as these yield the best quality product.

Hans van der Vis (RIVO Holland) then spoke about slaughter of eels, again covering welfare and quality criteria. They demonstrated that the traditional method of killing eels by salting was both inadequate on welfare grounds and gave a poorer tasting product than experimental killing methods using anaesthesia and brain coring, or electricity. The use of anaesthetics pre-slaughter would not gain acceptance in the EU, but electrical killing methods warranted further research. They satisfy welfare and product quality criteria.

It did not go unnoticed that in this session there were contrary reports on the effects of pre-slaughter and slaughter stress on the quality of fish flesh. Those working on welfare aspects stated there were clear gains to be made in product quality by reducing stress at slaughter, the commercial consultant reported little if any evidence of flesh quality change while the research scientist reported contrary evidence on the subject in the literature. I was left with a clear impression that shelf life of the product could be improved if pre-harvest stresses are reduced, and that this may be a tangible gain for farmers faced with the inevitable task of improving fish welfare during harvest and slaughter. (see Tom Pottinger's article, this issue).

Primary processing

The fourth session of the meeting looked at post-harvest effects on quality. Anna Maria Bencze Røra from Aquaforsk spoke about primary processing, i.e. the evisceration and filleting of fish. This is done to improve hygiene and extend shelf life, to add value to the product, to improve the logistics of transport and distribution, and to separate the inedible part for use for other purposes. She highlighted how farmers' actions could influence the quality and yield of primary processed products. For example, reducing pre-slaughter stress delays the onset and reduces the strength of rigor, thereby enabling pre-rigor processing which can improve fillet quality and reduce the incidence of gaping. Reduction in the variability of fish condition factor at harvest significantly improves processing yields. Therefore there is scope for farmers to improve the yield and quality of primary processed products through breeding programmes, improved husbandry and improved slaughter handling methods.

Rhona Johnstone (Farne Salmon) then spoke about hygiene and decontamination of primary processing facilities. With respect to her own company she stressed how hazard analysis (HACCP) is used to identify major potential problems. These were tackled not only by targetting cleaning and testing at critical areas, but by establishing an appropriate work ethic within the company to deal with hygiene matters. Staff from management to cleaning staff are all made aware of the potential and actual problems by training and hands-on experience. Cleaning staff and process operators are all encouraged to take an active role in improving the quality of hygiene and decontamination work. The active involvement of all staff has helped ensure that appropriate hygiene standards are met within the processing operation.

Ingrid Urdeland from the Swedish Institute for Food and Biotechnology then spoke about lipid oxidation during the processing and storage of fatty fish. As discussed earlier, fish contain highly nutritionally beneficial polyunsaturated fatty acids. Their oxidation initially produces compounds of lower nutritional value, then compounds giving bad taste and odour problems, and finally products leading to poor muscle colour and tough texture. From various experimental studies, Dr Urdeland concluded that the following practices would reduce oxidation problems: avoid blood contamination at filleting; do not store fish on ice for more than 2 days; keep the skin on the fillets or deep-skin fillets to remove dark muscle; if freezing, keep skin on, pre-cook at low temperature (55°C), glaze and vacuum pack; if washing fillets, or rinsing minced fish, add antioxidants to washing water; pack minced fish in large packs, avoid oxygenation by glazing, vacuum packing and storing in darkness.

Brian Day of Campden and Chorleywood Food, then concluded the session by discussing modified atmosphere

packing (MAP) of chilled fish. MAP is a good method of reducing both oxidative and bacteriological degradation of fish products. It uses different gas mixes depending on the product packaged and the spoilage risks involved. Its advantages are increased shelf life, improved product hygiene, additive-free, quality image. Its disadvantages are the capital and unit packing cost and package volume. Overall, it is seen as an advantageous system where consumer preferences are for fresh chilled fish rather than a frozen product.

Quality assessment

Dr Ulf Erikson spoke about the measurement of rigor, which is an indicator of handling stress and product freshness, and provides useful information on slaughter and chilling routines. I will not detail methods here, but instead re-emphasise an interesting result, that where fish are rested at harvest, rigor formation is delayed by some 30 hours over fish typically exercised and stressed at harvest. This time reflects an effective difference in the potential shelf life of the product.

Dr Robb then discussed the measurement of colour in fish flesh. He showed how the methods chosen varied in accuracy and cost and stressed that the methods used should be tailored to the particular need. He also emphasised the need for screening and training of assessors whatever technique was used. The use of colour reference cards was seen as the most generally useful method for fish farmers and their customers.

Mr Kurt Fjellanger of Nutreco spoke about proximate analysis of fish. He stressed that quality assessment was dependent upon a number of variables, particularly market requirements, and the location within the fish from which tissue for analysis was taken. Even where the requirements were defined and known target tissue is used, the methodologies available to carry out proximate analysis could lead to different results. He therefore concluded that there was a need to establish standards across the industry and document how, for example, fat level and fat type affected product quality.

Rob Sinnott of Trow discussed carcass quality monitoring on farms and at primary processing. He suggested that regular monitoring of useful carcass quality parameters should take place. On farms it would enable the farmer to build up a database of year-to-year variations, seasonal trends, relationships between factors, etc. This can be used in broodstock selection programmes, in predicting ultimate carcass quality at harvest, and enable carcass quality to be manipulated prior to harvest, e.g. pigmentation and fat levels. This would ultimately minimise production costs and enable proper communication with customers about carcass quality variation.

For primary processors, monitoring would enable quality specification to be assessed against that requested or required. The database of information

gathered would allow prediction of the product behaviour during processing, account for seasonal effects in fish quality, and enable product specification standards to reflect what could be reasonably achieved on the farm. Between the farms and primary processors it should be easy to identify how best to produce fish to the specifications required by various customers. The level of monitoring required on farms needs to be a compromise between cost and the statistical validity of the data collected. It was suggested that 2 samples of 10-30 fish from each stock on table trout farms and 3-4 similar samples on large trout/salmon farms would be realistic. Parameters to monitor would be growth rate, appearance, condition factor, carcass fat, flesh colour, flesh oiliness, and gonadosomatic index.

The next two talks by Dr Lisbeth Johansson (Uppsala University) and Geoff Nute (University of Bristol) covered sensory analysis and preference testing, respectively. While not wishing to cover these talks in any detail here, both were impressive in illustrating the complexity of the subjects discussed. Having seen the care taken to ensure that sensory analysis panels carry out their work objectively and to uniform standards, I will be more likely to accept their evidence in future. The news that stated preferences could reflect the quality of the test used rather than true preferences of those tested, came as little surprise to those of us used to analysis of statistics, but should have been noted by anyone in the industry seeking consumer preference information. Bringing the correct question and correct statistical test together is an essential part of any survey of customer preference.

Strategies for improving quality

The final session was opened by Peter Howgate, who spoke about aquaculture product safety. He stressed that food quality and food safety are of major concern to the public. The public rely on major retailers to make proper evaluations of product safety. This drives the supermarkets to produce and implement quality assurance schemes. The industry needs to take these matters seriously, as it cannot withstand the sort of food safety scares seen in other areas. The good news from this talk was how little risk there was in food safety terms from temperate finfish aquaculture. In contrast, the farming of tropical fish or shellfish farming pose much greater food safety risks. As food safety concerns in farmed fish are often less than those of equivalent wild fish, there exists the scope to further promote the safety of farmed fish.

Allen Greenhalgh from Food Certification Scotland Ltd then spoke about the benefits to be derived by the industry from participation in quality assurance schemes for its products. He illustrated the development of the Scottish Quality Salmon scheme, from the difficulties of its inception to the practical problems in its implementation by farmers involved and by scheme

inspectors responsible for ensuring that standards are maintained. He concluded that the maintenance of a quality image for farmed salmon alongside that of the wild fish has been a significant benefit to the whole industry, and especially so to those in the SQS scheme who were able to sell their product at a premium. He advocated the development of QA schemes wherever possible.

Conclusion

Dr Ron Roberts (University of Idaho) gave an overview and opened with a comment that it had taken 30 years for the industry to raise quality to the top of its agenda, but having done so it was clearly timely given the range of problems identified during the meeting. He stressed that, although salmonids were inherently safe as a foodstuff, there was a real need to promote food safety through quality assurance schemes, and avoid the sort of tabloid scares that have affected other agriculture sectors. The industry needs to show that its products are better than pork, poultry, etc.

In looking at individual contributions, he remarked on the enormous potential of the study of muscle structure, but stressed even more the value of applied research to answer the industry's major existing problems. He commented on the study of downgrading which showed spinal development problems at high levels, and later papers showing the knock-on effect of these for processing plants. These problems are prime targets for research.

There were many inter-related problems brought to light which the industry needs to address, and almost all require near-market applied research. Dr Roberts suggested that given the lack of funds in fish farming, it is clear that the profit-making feed companies and supermarket chains need to become involved in payment for some of this applied research, particularly as they stand to make significant benefits from any advances made.

Apart from the terrible jokes, I approved of much of what Dr Roberts said. I would have added a note that the welfare of fish was clearly an increasing concern, but with the news from this conference that there may be benefits to the industry from improvements in welfare at harvest and slaughter, there should be an internal incentive as well as external pressure to investigate improved slaughter methods.

Overall, a very interesting and potentially useful meeting for most participants. It was a shame there were so few trout industry representatives present, as I believe that industry has much to learn about the production of quality products.

The papers given at this conference are expected to be published by Blackwell Science Ltd in book form which should provide a valuable reference for trout and salmon farmers interested in aspects of fish flesh quality.

BRITISH TROUT FARMING CONFERENCE, SPARSHOLT 1-3 SEPTEMBER, 1999

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Introduction

This thirteenth conference to be held at Sparsholt following the move from its original venue at Two Lakes Fishery, Romsey in 1987 was attended by around 200 delegates (the same as last year), 90 of which (45%) were fish farmers. The conference was wide ranging as usual, but the main theme addressed the problems facing the trout industry by seeking possible solutions in other sectors that are similarly affected namely the salmon and the Danish trout industry as well as the beleaguered pig industry. Other species considered included Arctic charr and sturgeon production in Canada and France respectively and aspects of the biology and exploitation of eels under the title Eel: The Eurofish, the 1999 Buckland lecture given by professor Morriarty. The conference hosted the usual trade exhibition with a capacity 20 companies displaying a range of products and included in the programme for the first time was a 'suppliers forum' in which exhibitors were invited to give a short 10 minute talk on products new to the industry.

European trout markets

The first speaker, Inge Vibeke Christensen, a trout farmer whose father was responsible for establishing the Christensen strain of rainbow trout in the 1950s described the European trout market with particular reference to the Danish industry. Trout production in Denmark fell from 34,000 to 32,000 tonnes in 1998. The majority (86%) is exported mainly to Germany as live, fresh/frozen or smoked at a value of between 700 and 800 million d.kr. The industry employs 3000 people and production is mainly carried out in earth ponds (73%) with the remainder from concrete ponds or a combination of the two. Trout prices have decreased since 1992 but stabilised in 1997.

She identified the main problems currently facing the industry which were low prices and difficulties with environmentalists and politicians who perceived trout farming as environmentally damaging thereby creating a poor image for the industry as a whole. Stringent environmental legislation introduced in 1989 halted expansion of the industry by placing limitations on trout feeds (ingredients, quotas and conversion rates), discharge levels and effluent quality. As a result the number of operational farms in Denmark has declined from 500 in 1989 to an estimated 420 farms in 1998 a fall of 15% in 9 years. The impact has been less severe on production as farmers have been forced to become more efficient.

On the question of image she considered trout had a good reputation in Europe, a recent survey indicating 18% would buy more trout over the next 5 years. Convincing consumers that trout is a nutritious and healthy food was not sufficient however, and what was really required was a more pro-active industry. There should be more information on prices in Europe, quality assurance schemes should be introduced (Denmark will be producing one this year) and more unified promotion campaigns are required which target not only the general public but students (the future market), government institutions (legislators) and the press. FEAP involvement was considered vital in providing a proactive information source for promoting the European trout industry and its value had already been demonstrated in facilitating a COP for large trout.

Lessons from the salmon industry?

The next speaker, George Hide, a lecturer at Sparsholt college and former salmon farmer, then gave a talk entitled 'The salmon industry: are there lessons for trout?'. He began by describing the Scottish salmon industry which in contrast to trout had shown dramatic growth over the last decade. In 1990 the industry stood at 32,350 tonnes increasing 3.5 fold to over 115,000 tonnes by 1998. Over the same period productivity in terms of tonnes/man increased from 22 to 77, yield per smolt from 1.55 to 3.2 kg, food conversion efficiency from 2.5 to 1.3 and mortality in the sea water phase reduced from 40 to 10%. These increases were the result of improved growth of smolts which when introduced to sea water at a weight of 50 gms required 18 months to reach 2 kg in 1990 compared to 8 kg over a similar period now. The industry is now able to produce 3-4 kg salmon on a consistent basis which was not possible a few years ago.

These dramatic gains, he continued, were the result of improvements in three key areas of production - nutrition, health control and husbandry techniques. The main changes in nutrition have been a reduction in carbohydrate from 15 to 7% and an increase in oil levels, now typically 30-33% as against 20% in the early days. This produces a more energy dense diet which coupled with the use of high quality protein from fish meals enables a more efficient conversion of protein to fish flesh combined with less pollution. Also the way fish are fed in terms of when and how has changed from the traditional hand feeding to the use of automatic feeders. Salmon show peak appetite at dawn and dusk and feeders can now be programmed to deliver the maximum number of meals at these times. The use of underwater cameras to detect uneaten pellets has also

helped to reduce wastage. The latest interactive feeders employ sensors at the net bottom which switch off the feeder when uneaten pellets are detected. The use of these systems has significantly improved both growth rate and food conversion efficiency.

Fish health has greatly improved over the past decade by controlling furunculosis and sea lice which together caused mortality rates of 40%. Control has been achieved by the use of prophylactic strategies such as site fallowing to break the disease cycle, vaccination against furunculosis and a national treatment strategy for sea lice. This involves grouping farming operations into zones of which 17 have now been set up in Scotland. In each zone the breeding lice population is kept as low as possible and relies on the full co-operation between neighbouring farms when treatment is required.

The third major area of improvement has been in husbandry methods during the freshwater phase involving the use of heated units, photoperiod manipulation and improved grading. These have enabled larger smolts to be produced over a nine month period from late summer to May the following year. Photoperiod control has also boosted the seawater growth phase in which continuous light during the first sea winter reduces grilising and increased growth by 20%. Breeding programmes have not been undertaken in the past but two of the largest salmon companies have started family selection programmes based on growth rate, maturation age, pigmentation and fat content.

The biggest set-back to the Scottish salmon industry has been the outbreak of Infectious Salmon Anaemia (ISA) in 1998 when 25 sites were either suspected or had the disease. It has been estimated that £25 million has been lost in compulsory slaughter from this EU list I disease since the outbreak. The legislation is designed to eradicate the disease but because there is no compensation, he said this could represent the biggest threat to the industry than the disease itself.

As to the lessons for the trout industry the speaker said that the huge gains in performance and health management over the past 10 years equally applied to both industries. The increased co-operation of neighbouring salmon farmers in eradicating disease had been a great help and this could be usefully applied to trout farming. The selective breeding programme and ISA experience were also key areas to highlight, if for example, VHS or IHN were to hit the trout industry. But at the end of the day the key issue to address for both industries was clearly the low market price of fish which in some cases was close or below the production costs.

Fish and shellfish cultivation review

Richard Slaski from the British Halibut Association (BHA) then gave his review of fish and shellfish cultivation which was used in the recent MAFF review of

its R and D requirements for aquaculture. He said the review gave some insights into what technical issues were holding back GB aquaculture at present and identified the gaps in defensive strategic research requirements. Covering both fresh-water and salt-water species, the report was weighted more towards marine aquaculture although there were common issues that applied to both sectors which included the environment, welfare and economic pressures, safety and public perception.

He then considered the fledgling halibut industry in some detail which he described as the last new species remaining for cultivation in the UK. The BHA, which functioned as a research club, has been in operation now for some 12 years and is currently looking at getting the hatchery production of halibut up and running. Projections for the year 2009 was for 300,000 juveniles and a production of some 10,000 tonnes of fish. The market for halibut was seen as a future growth area serving the food service sector with an aspirational and differentiated product that was both healthy and good tasting. It was intended the market would be served with uniform 220 g portions of fish either as fillets or steaks, without bones, frozen for convenience and available 52 weeks of the year. To achieve this output large production units would be required which have the necessary economy of scale and consistency of supply. Single year classes with site fallowing in between to reduce the incidence of disease would be the normal production strategy. He saw the role of a trade association as fostering an operating climate, always looking over the next hill and supplying marketing information and strategies. Crucially the membership must be pro-active.

European and global trends were then briefly reviewed for the major species. Salmon production had shown phenomenal growth in recent years but this had been accompanied by a steady decline in price and there was now a need to reduce costs. The price for turbot, although initially very high was now plummeting and the same applied to bass and bream. Tilapia production was variable but the value was steadily rising while eel prices had reached a plateau and were now falling. Of all the cultivated species rainbow trout had the most stable value pattern of all. He considered the trout industry was very well represented by the BTA. It was recognised the industry faced a considerable business challenge with more technical changes ahead. He said the trout industry must define the help needed, make forward plans and seek help from the research sector. The Promar report provided a future vision for the industry and there were already important research projects under way or being considered such as algal taint, stocking densities and selective breeding.

Arctic charr production

Tim Fleming of Icy Waters Ltd, a Canadian based company located in Whitehorse, Yukon then gave an account of the company's involvement in Arctic charr production. He described charr as the 'top end' of

salmonid fish in terms of flavour and was a highly rated species in Canada. A cousin of the brook trout and lake trout he described four domesticated strains of charr from Canada and North America from which the company was developing its commercial lines. These were Fraser river, Nauyuk, Yukon Gold and Tree River the last being the largest Arctic charr strain in North America.

In terms of cultivation, charr have many valuable attributes. They are able to be fully domesticated and grow well in tanks at an optimum temperature of 12°C but require a reduced temperature of 6°C for broodstock holding and egg incubation. Above 15°C they exhibit a poor feeding response and at 21°C all feeding stops. They have a low food conversion rate of between 1.1 and 1.2 and tolerate high stocking densities which lend themselves well to cultivation in re-circulation units. Growth is poor at stocking densities of less than 60 kg/cubic metre and for normal commercial production densities of 100-150 kg/cubic metre are recommended although stocking rates of 321 kg/cubic metre have been achieved. Because of these high stocking rates special attention must be paid to adequate oxygen supplies. Charr generally have good resistance to disease but are more susceptible to furunculosis. Fast growth rates have been achieved in some strains with each year class showing between 10-15% better growth.

Icy Waters marketing strategy for farmed charr is to have a product or propriety species not just a commodity. This is facilitated by the fact that there is little or no competition from captive fisheries (only 25 tonnes are caught annually from Lake Geneva for example) and permission is required from the Inuit population of Canada to remove fish from the wild. In order to retain control of production the company sells only triploid (sterile) ova for on-growing.

Disease certification has been obtained for the Yukon Gold and Tree river strains for export but for production purposes a hybrid cross has been developed between these two strains for commercial release in the year 2000. This hybrid is claimed to have a 25% better growth rate than either of the two parent strains. The hatcheries at Cloan and Caledonian already hold Yukon Gold charr and Stirling University is currently negotiating a contract to produce them at its Howiton hatchery where water at 6°C is also available.

More lessons for trout?

The next speaker, James Miles-Hobbs of Andersons, a firm of farm business consultants outlined the current problems affecting the pig industry. He said the output of pigs had always been cyclical in nature with peaks and troughs in production as the price of pig meat varied. The latest crises had been particularly severe however but production had started rising again since September, 1998. In contrast trout demand levelled off in the late 1980s and had remained static since.

The greatest problem affecting the pig industry, he continued, was competition from abroad where production costs were lower. To produce a 32 kg weaner pig, for example, costs 100 pence per kg in the UK as against 49 pence in the USA. To combat this differential it was essential to respond to the needs of the UK consumer in the various retail outlets (supermarkets, fast food and catering). The key would probably be in the processing and food manufacturing industries in developing new product ranges utilising other meat not wanted by the retailers who only require selected parts. A guaranteed supply would also be essential. This would require greater integration of farmers with the processors in terms of tighter grading and larger volumes of more even pigs and increased co-ordination of supply. A more contracted supply would take out the highs and lows of production and help to ensure a reasonable return for efficient producers. Guarantees on consumer concern for the environment and animal welfare must be met as well as the development of value added products, niche markets and organic production to help ensure long term stability in the pig industry.

In comparison with trout the pig industry was much bigger, worth £880 million as against £26 million, therefore a larger marketing spend was available with more dedicated value added processors and better product innovation. In addition there was still further genetic potential to improve stock performance at lower cost providing 50% more growth rate for the top 10% of producers. Against this was the fact that pig meat was an international commodity and therefore prone to world market fluctuations outside the control of the industry.

Industry review

The next speaker, John Giles of Promar International, began his review of the trout industry by outlining the background to the report which the BTA had commissioned back in November, 1998 for completion by the following March. The study was commissioned because there was a clear need for change within the industry - it was not processor driven, he said. He explained that Promar were not trout specialists but had wide experience in most other agricultural food producing industries and could, therefore, provide a fresh approach to the problem.

There were many production related issues which were causing concern and it seemed inevitable there would have to be a rationalisation in which the industry would contract over the next 3-5 years to about 150 trout farms in the UK, a reduction of around 25-30%. There were opportunities, he said, to improve the supply chain management and review the costs of production suggesting a 10% improvement in food conversion rates was possible. There was also scope for creating joint initiatives and closer R and D partnerships, particularly in the food manufacturing sectors.

He then reviewed the strengths and weaknesses of the industry which had been revealed by the survey. On the plus side was its disease free status (cf. Salmon), although there were some areas for concern, and trout had a good image as a healthy, convenient and versatile food. Being a domestic product it had a regional image with some good companies and individuals. There was also a considerable interest from the retail and food service sectors who were actively looking for new products and initiatives. On the debit side there was very limited marketing going on and the state of the industry had produced much short term thinking. There was limited added value and the quality mark schemes and COPs, of which there were several, were in danger of fragmentation in a small industry. He thought the all-female and sterile methods of trout production could be viewed by the public as 'frankenstein' foods creating a potential PR bombshell. The leadership was viewed with varying degrees of uncertainty and the whole industry had become very inward looking in a way quite unlike any other Promar had investigated. Consumer profiles had shown trout was viewed as dull because of a lack of product development and in general the industry had stood still in the face of competition from the salmon, poultry, lamb and pig industries.

Marketing opportunities were then explored starting with the retail sector which in general was satisfied with existing suppliers. Niche markets existed for different sorts of trout (the web was useful here) and there was much scope for adding value to commodity products (recipe dishes, pate, fillets and steaks etc) but these had not been fully exploited. The food service sector (hotels and restaurants) was another growth area which was expanding by 30% a year and there were potential export markets to the US, Japan and the EU. In all these opportunities, he said, the industry must bend over backwards to supply what is required and be prepared to invest heavily. Speculative production had only a limited future. In conclusion, he said a 10% increase in trout production over the next 3 years was achievable, there were alternatives to the major retailers and the industry really must change if it was to survive, grow and compete.

BTA perspective

The final talk of the first day was given by Mark Davies, chairman of the BTA. He began by stating that what he was about to say the audience had heard many times before. It was not just the trout industry in trouble but also sheep, pigs and chickens all of which were currently losing money. The trout industry had been in this position about 15 years ago when several uneconomic sites went under and the same thing was likely to happen now. Although sales had shown a modest increase, indicating production was not being stockpiled, the present scenario suggested 25% of trout farms would go.

The realities, he said, were known to most people but were worth repeating. The luxuries of the past, when

huge profits were possible, are now gone. There was over capacity of production and processing and table farmers were becoming more remote to the needs of the public. With 70% of trout now sold to multiple retailers via the processors the margins and specifications were becoming ever tighter. In spite of this there were still poor quality tasting trout being sold on the market.

Despite the current problems he considered the trout market was still capable of growing but not overnight. In the presence of fierce competition from other food producers, it was necessary to think laterally on cost saving and efficiency measures. The industry as a whole must be technologically driven without compromising quality.

He outlined the important issues that needed addressing. There was more data required on environmental interactions, producers must understand the market better and respond to its requirements. Co-operation was needed in marketing and promotion. Table producers must interact more with the fry and fingerling producers because without this base supporting the on-growers the industry would be in serious problems. He summarised the three main areas to concentrate on; quality (getting it right every time because quality sells), marketing (giving the markets what they want and by supporting the product eg promotional material, investment in processing and packaging etc) and production (saving costs and examining how, when and why fish are grown).

Since publication of the Promar report plans were being laid by the BTA and MAFF to help the industry as much as possible but he said it was up to everyone to look critically at their own situation and try and improve their businesses. The message both from the BTA and Promar was clearly 'survival of the fittest'.

Open forum

The last session of the afternoon was reserved for an open forum, which, under the general title of 'the trout industry into the next millennium' the audience was invited to raise issues of general concern for consideration by a panel made up of the day's speakers.

The first topic raised concerned fingerling supplies. Fingerlings currently cost around £45 per thousand to produce and unless the returns on this did not improve it was thought many businesses in this sector could go. The biggest threat to the fry producers, it was suggested, was due to high mortality rates caused by RTFS. A serious problem caused by the recession in the industry had been with cancellation of fingerling supplies because of the failure of farmers to shift stocks. Mark Davies suggested that this sector should be looked at as part of the production chain in which the supply of fry and fingerlings should arrive automatically as and when required.

Tim Small of Lechlade Trout Fisheries then queried why the UK trout processors were not concentrating more on single portion meals since there was now a huge demand for these products. In Denmark 6,000 tonnes of smoked fillets are produced for the German market which equated to 12,000 tonnes of trout in the round - about 12% of total production. Moreover trout produced for the smoking sector fetched an additional premium equivalent to 20-30 pence for the raw material.

There then followed a discussion on trout promotion and the role of trade associations. The Danish trout industry spends less on promotion than the UK and none of its processors are members of a marketing association. However advice on processing was considered necessary but funds are not available to carry out the required research. Mark Davies responded by saying the role of the BTA was to act as an enabler in providing market information, any product innovation should

come from within the industry. The BTA was a small organisation with a relatively small budget. Its main role was representational in Europe, on legislation and monitoring. Product development was most successful when carried out in conjunction with the customer and must be driven by the membership. If there was a will then the trade association could step in and help. The session ended with some discussion over the question 'is there a north/south divide of the trout industry'. The Scottish industry which started later in the early eighties was considered more cohesive with a large marketing association under the auspices of Scottish Quality Trout. The equivalent English marketing group was considered less cohesive and should take an example from the Scottish industry.

The papers given on the final day of the conference which included eels, sturgeon and trout health issues will be reviewed in the July edition of Trout News.

ASPECTS OF CARCASS QUALITY AND ITS MONITORING IN RAINBOW TROUT

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Introduction

Most of the rainbow trout farmed in the U.K. is destined to reach the consumer by way of the major high street multiples. Although trout may be sold whole or with gut and gills removed most British grown trout is now sold in the form of fillets. The visual quality of the flesh has therefore become more marketable than the appearance of the whole fish. Trout make a quick nutritious and easy meal and are ideally suited to the microwave oven. The shopper expects the product to represent good value and to be pleasing to the eye, safe, well packaged and of a convenient size. Once cooked the product must taste good and look good on the plate. If all these criteria are met then it is very likely that the product will be eaten on a regular basis. The trout farmer and fish processor can control some of these criteria and influence others.

There are two primary reasons for monitoring flesh quality. The first is to ensure that fish are not sold until they exceed the minimum quality specifications required by the market. The second is to provide information that can be used proactively to implement a strategy of remedial action for positively influencing the quality of the fish so that they do meet the market specifications. There is, however, no magic wand with regard to modifying carcass quality in farmed fish and it is therefore important that all links in the supply chain are aware of what is actually possible. This short article will review the potential for using carcass quality monitoring as a tool for the production of high quality rainbow trout.

The shape of a trout is determined by hereditary factors, but also by its feed intake. There are several research papers confirming the influence of hereditary factors on, for example, flesh colour and carcass fat (Gjerde, 1987 and Gjerde and Schaeffer, 1989). The carcass quality of trout is also affected by physiological factors, such as maturation, and trout will alter in shape and appearance and lose flesh colour, as well as being unpleasant to eat, as they get close to maturation.

Carcass Quality Monitoring

Monitoring of carcass quality characteristics provides information that the farmer can use to manipulate fish quality by feed management. A carcass quality-monitoring programme should be designed to make regular measurements of useful fish quality characteristics. Such a programme should normally include recording the weight, length and yield of fish sampled. It would also make an assessment of the degree of maturation and would measure the colour of the flesh, fat content, and assess the texture of the flesh.

Rainbow trout grown in the U.K. are mainly harvested between 300 and 500 grams. The fish are therefore typically on the farm for a period of 9 to 12 months and the frequency of sampling must take account of this. It is important, where practical, to look at the trout two months prior to harvest as this will allow some time to influence fish quality should it be considered necessary. The numbers of fish sampled for a particular stock at a

particular point in time should be large enough to let the farmer know the frequency of a number of carcass characteristics occurring across the whole population of fish. Statistical tables give the sample number required to expose a particular carcass trait at a particular confidence limit. In a farm population of one stock numbering 100,000 trout this could mean looking at a sample of 317 fish (square root of $n + 1$). This would be unacceptable to most farmers and it would take a considerable time to carry out the operation. Taking 5 fish a time is too few to represent the variability within the population, so is there a compromise? My own experience would suggest that 30 fish a time would typically provide a reasonable amount of information. Even for this number of fish it could take more than two hours for an experienced monitor to take readings and record the results. On a practical level it helps enormously to have two people carry out the operation, one to handle the fish and the other to record the results. The immediate entry of the data on to a waterproofed portable computer is recommended.

Flesh quality characteristics

Details of the trout quality characteristics that should be included in a quality-monitoring programme are given below. The trout processing plant is an ideal situation to monitor and record the carcass quality characteristics of the fish passing through it.

External Appearance

Since the carcass-monitoring programme requires the killing of the sampled fish it is recommended to make full use of them. The trout should be closely examined paying particular attention to the gills, skin and anal region. The presence and location of skin damage or

lesions should be recorded along with a note on the state of the fins. The gills structure should be closely looked at and any obvious external parasites removed for more detailed examination. Any bloody discharge from the hindgut should be considered as an indicator of the presence of a bacterial pathogen within the stock. If disease is suspected then the farm veterinarian should be contacted for advice.

Flesh Colour

One of the problems facing retailers is obtaining supplies of consistently pigmented fillets. An example of this is given in Figure 1 below which shows the percentage of fish at a processing plant failing to meet the minimum requirements for flesh colour over a one-year period. This type of information can be of real help when investigating the causes of any fluctuation in flesh quality. It could be argued that the consistency of flesh colour is more important than the actual colour of the fillets. It has been shown that there is a tendency for the shopper to pick the reddest-coloured fillets first (Koteng, 1992). The colour of the flesh was found to be associated positively with flesh quality.

The regular measurement, of flesh colour serves to let the farmer know how the flesh colouration is developing and whether the amount of dietary pigment fed should be increased before the fish are harvested. In order to achieve an acceptable flesh colour for trout fillets it is usual to feed a diet containing 40 to 50 mg of pigment per kg of feed from a weight of 80-100 g until harvest.

The two pigments currently licensed for use in farmed trout are astaxanthin and canthaxanthin and these can be fed either separately or as a mixture. The maximum amount of pigment that is legally permitted in feed for salmonids is restricted to 80 mg/kg for canthaxanthin

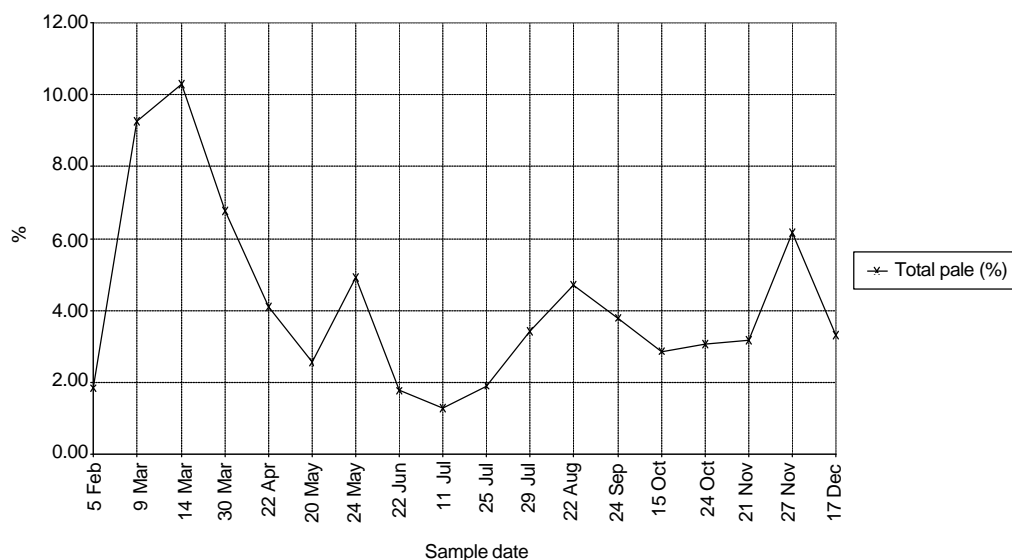


Figure 1. Seasonal trend in fillet flesh colour at a specific farm site

and 100 mg/kg of feed for astaxanthin or a mixture of the two pigments. This gives a good degree of latitude for increasing dietary pigment levels if the results from the carcass quality monitoring indicate it is necessary. Research by Trouw has shown that it is typically not cost-effective for pigment levels to exceed 65 mg/kg of feed in trout diets as the efficiency of pigment deposition decreases with increasing dietary pigment levels. Although both pigments are legal it is always advisable to consult potential fish processors or buyers for advice on which pigmentation regime to use.

For rainbow trout farmers producing fish for the cold smoking market (>2 kg) a further pigmentation strategy is recommended. If the sampling programme shows the fish population has reached an acceptable level of pigmentation then the flesh colour can be maintained by reducing dietary pigment levels to 30 mg/kg of feed. This can result in significant savings on pigment costs for the farmer but it is very important to monitor the colour of the population to ensure that flesh colour is being maintained at or above the target level.

Flesh Colour can be measured visually using the Roche Fan or objectively using an instrument such as the Minolta Chroma Meter. For the flesh colour standard to target it is always advisable to consult the intended fish processor. The typical visual colour scores expected for trout products ranges from 25-30 with the lower end of the scale associated with fresh products and the higher end for value added products, such as cold smoked trout fillets. Flesh colour is normally measured from a fillet or outlet and should always be measured at the same point. The method of measurement and the position where the measurements are taken should be agreed with the fish purchaser. The visual flesh colour is typically best in the muscle near the tail and lowest in the belly area. The mid-dorsal muscle area would typically have an intermediate colour.

• **Muscle Fat**

Excess muscle fat is rarely a problem for trout farmers producing table-sized trout. Low muscle fat levels are found in fish that have been held for long periods at constant weight or in fish that have been grown very slowly. Rainbow trout exhibiting 'soft' muscle texture are often deemed excessively fat; in fact the very opposite is usually true. Muscle fat is inversely proportional to muscle moisture and fish with low fat levels may contain high moisture levels and may be 'spongy' for this reason.

Maturing trout will lay down fat during the autumn of the year before maturation but this will be rapidly diminished with the onset of maturation.

Unfortunately for the processors of large trout and salmon the condition factor of rainbow trout has only a loose

relationship with muscle fat. A stronger relationship between the two would make the online grading of fish, for fat content, at the factory very much easier.

Measurement of muscle fat can be made on live anaesthetised trout using a Torry Fat Meter, which uses a microwave sensor to measure the amount of moisture in the fillet. As moisture and fat are inversely proportional in the muscle this moisture measurement can be automatically converted to a measurement of fat. Another relatively simple method is to take a weighed cut of muscle and dry this to constant weight. The new weight will give the moisture level and, as above, this can be converted to give a measurement of fat. Alternatively samples of flesh can be sent to a laboratory for proximate analysis. Whatever method is used it is most important to use a standard sampling protocol. The feed supplier can advise on the most appropriate method to use in a particular situation. The data collected can be used in various ways and Figure 2 shows the relationship between fish weight, muscle fat and season portrayed in a contour format. This shows that the fat level for a 500 g trout is lowest in the summer and highest in the autumn and winter.

• **Flesh Texture**

In common with other livestock, fast growing young rainbow trout have fewer cross linkages in the connective tissue holding muscle fibres and muscle blocks together than slower growing, and therefore older, fish of the same size. Careful handling of these fish is very important in reducing the incidence of muscle gaping. Fast grown fish may have more glycogen stored in the muscle and this can be converted into lactic acid following death. The minimum industry recommended starvation period should be strictly observed in order to reduce potentially damaging enzymatic activity in the muscle following harvest. Although there has been much conjecture regarding the effects of slaughter method on flesh quality it has been reported that flesh texture is not correlated to the slaughter method (Azam et al., 1989). The major factor negatively affecting fillet texture has been reported to be the storage time following the passing of rigor mortis (Faergemand et al., 1995). Rapid chilling of fish to less than four degrees centigrade post harvest and the maintenance of chilling throughout the transport and processing of them is crucial for optimising fish quality. Good harvest techniques, such as minimising stress pre-harvest, coupled with rapid chilling will help to delay the onset of rigor.

Factors affecting carcass quality and yield

• **Yield**

Few trout are now sold in the round. Rainbow trout are sold as either gutted, or as gilled and gutted. The

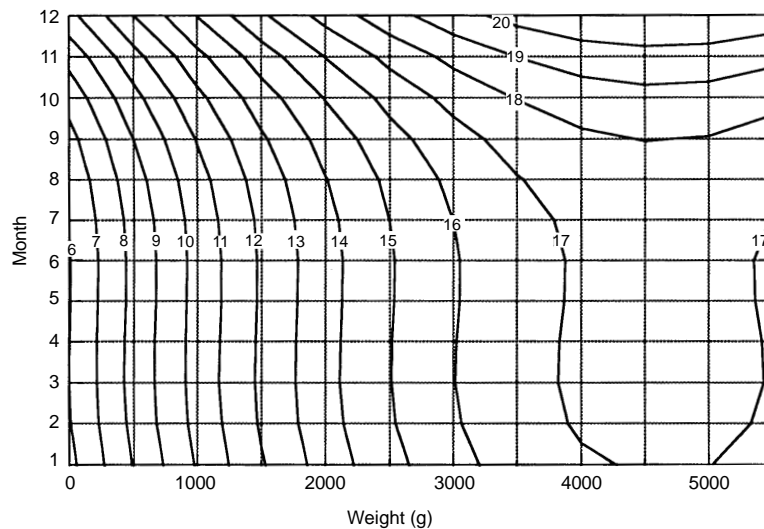


Figure 2. Relationship between weight, time of year and fat (% body weight) for rainbow trout

resulting yield, that is what is actually sold, will vary between populations of fish and with the time of year. The yield for immature trout can be less in the autumn than in the summer. This coincides with the tendency for fish to lay down visceral fat in the winter as an energy store to be drawn from during hard times in winter and at maturation.

Prolonged starvation of more than one week will improve the yield but at a cost to the farmer in terms of lost growth. The maximum forty eight-hour period of starvation recommended in the Farm Animal Welfare Council (FAWC) report will have a minimal effect in reducing either muscle fat or in improving yield.

• **Sexual maturity**

In the past most trout were grown from mixed sex populations and precociously maturing males were a real problem. At maturation these fish would lose pigment and muscle fat and would change external shape as well as darkening in skin colour. Following maturity they would be susceptible to disease and fungal infections and many fish would die before the maturation process had run its course.

In order to monitor the degree of sexual maturity the Gonadal Somatic Index (GSI) may be taken. This is a measure of the weight of the gonads, that is the ovaries or testes of the fish, expressed as a percentage of the total round starved body weight of the fish. Monitoring GSI in all female stocks can give interesting results. Some years ago, for example, I was investigating a problem with poor flesh colour in rainbow trout at a fish-processing factory only to discover that a small but significant proportion of the “all female” stock were in fact maturing males.

From a practical point of view it is not necessary to weigh the gonads of table sized trout but it is important to look at the gonadal development to make sure that no maturation problem is imminent. It is also important to remember that all female stocks may still mature before the projected harvest period if kept for long periods.

Nutrition

The farmer can influence carcass quality by adopting feed management practices to modify the final composition of the flesh. The protein composition of the diet has little effect on the composition of the muscle but the fatty acid profile of dietary oils will be reflected in the fatty acid profile of the fish muscle (Thomassen and Røsjø, 1989). Trouw have, however, carried out in depth research in this area and has shown that the fatty acid profile of rainbow trout can be modified by partial replacement of marine fish oils with certain vegetable oils without negative effects on the major sensory characteristics of fillets. The amount of energy consumed by trout will influence its growth rate, carcass fat and flesh texture. Fish fed as much as they will eat will have deeper bodies and a higher condition factor than slower grown fish. The condition factor of fish (C.F.= weight/ length cubed) is an expression that takes into account the weight and length of the fish. The effects of feed ration levels and age on sensory characteristics, fillet lipid content and fatty acid composition of rainbow trout composition have been studied by Johansson et al. (1995). They found that the ration level had no effect on the intensity of fresh smell or any sensory characteristic of taste or consistency of fish at ration levels over 75% of the maximum expected appetite. At ration levels below 50% of appetite there were significant negative effects on fresh taste and

firmness. They also found that the intensity of fresh smell increased with increasing age, independent of ration level but that it had no effect on firmness or juiciness at rations above 50% of appetite.

A useful indication of nutritional balance in fish is to measure the Hepato Somatic Index (HSI) which is a measure of the weight of the liver expressed as a percentage of the total round starved weight of the fish. In trout the HSI typically ranges from 1-2 per cent of the starved body weight, so that a deviation from the expected value may be a pointer toward a nutritional imbalance within the fish or in its diet. For this reason it may be useful for the farmer to keep a record of the HSI normally found in his stocks.

• **Season**

I have already made reference to the influence of season on carcass fat and yield (Figure 2). This is also true for flesh colour; Figure 3 shows that the astaxanthin content of trout muscle can vary throughout the year and may be lowest in the summer months. Some farms appear to achieve better flesh colour, for example, than others even when the same stocks and diets are used; the reasons for this are not always easily understood. A regular monitoring programme carried out at different times of the year will allow the farmer to build up a comprehensive picture of carcass quality trends in the stocks he uses and on his own site.

Another problem very much linked to the seasons is muddy taint in trout. This is generally caused by a metabolite from blue green algae called geosmin, which is released into the water and picked up mainly through

the gills and skin of the fish. It mainly occurs during the spring and early summer months (May to late June) particularly when the weather has been hot and dry. This leads to low water levels which concentrates nutrients in the water stimulating algal growth and hence geosmin production.

Trout should not be marketed if tainting is suspected and where the problem is known to occur fish should be sampled, on a weekly basis, during vulnerable periods for muddy taint. Two or three fish should be cooked in a simple way such as steamed or microwaved and tasted for any off flavours. There is little that can be done if contamination occurs but to wait until the problem goes away. However one recognised approach is to purge the taint from the fish by transferring trout to clean tap or bore-hole water, if available, for at least 5 days.

A straight forward protocol for on-farm assessment of taint in trout has been devised by Steve Kestin and Paul Warriss at the Department of Clinical Veterinary Medicine, University of Bristol, Langford, Bristol, BS18 7DY.

• **Disease**

Another good reason for monitoring fish regularly is to highlight the presence of disease. In the South of England Proliferative Kidney Disease (PKD) has been shown to have an impact on flesh colour (personal observation). From the information gained from sampling it may be possible to avoid selling fish when the likelihood of mark-downs, or rejections, based on poor flesh quality at the processing factory, is higher.

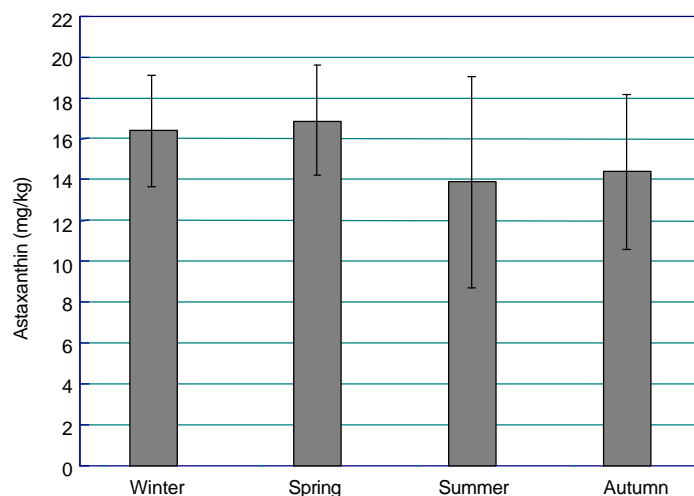


Figure 3. Seasonal variation in astaxanthin content in 2.5-3.5 kg rainbow trout 1996/97

Conclusions

I thoroughly recommend the principle of a carcass quality-monitoring to trout farmers as it allows potential problems to be recognised and dealt with early. Feed suppliers generally have technical advisers who can help to train farm staff to carry out this task. For further information on carcass quality monitoring programmes please contact me at the address given at the front of this article or by telephone on 01606 561090.

References

- AZAM, K., MACKIE, I. M. AND SMITH, J., 1989. The effect of slaughter method on the quality of rainbow trout (*Salmo gairdneri*) during storage on ice. *International Journal of Food and Technology*, 24, 69-79.
- FAERGEMAND, J., RØNSHOLDT, B., ALSTED, N. AND BØRRESEN, T., 1995. Fillet texture of rainbow trout as affected by feeding strategy, slaughtering procedure and storage post mortem. *Water Science and Technology*, 31 (10), 225-231.
- GJERDE, B. AND GJEDRUM, T., 1984. Estimates of the phenotypic and genetic parameters or carcass traits in Atlantic salmon and rainbow trout. *Aquaculture*, 36, 97-110.
- GJERDE, B., 1987. Predicting carcass composition of rainbow trout by computerized tomography. *Journal of Animal Breeding and Genetics*, Vol. 104, 121-136.
- GJERDE, B. AND SCHAEFFER, L.R., 1989. Body traits in rainbow trout II. Estimates of heretabilities and of phenotypic and genetic correlations. *Aquaculture*, 80, 25-44.
- JOHANSSON, L., KIESSLING, A., ÅSGÅRD, T. AND BERGLUND, L., 1995. Effects of ration level in rainbow trout, *Oncorhynchus mykiss* (Walbaum), on sensory characteristics, lipid content and fatty acid composition. *Aquaculture Nutrition*, 1, 59-66.
- KOTENG, D.F., 1992. Markedsunders Økelse Norske Laks. FNL, Bergen, Norway.
- LAVETY, J., AFOLABI, O.A. AND LOVE, R.M., 1988. The connective tissues of fish. IX. Gaping in farmed species. *International Journal of food Science and Technology*. 23, 23-30.
- THOMASSEN, M. AND RØSJØ, C., 1989. Different fats in feed for salmon: Influence on sensory parameters, growth rate and fatty acids in muscle and heart. *Aquaculture*, 79, 129-135.

CAN HUSBANDRY STRESS AFFECT FLESH QUALITY IN FISH?

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Introduction

Many husbandry procedures unavoidably involve a degree of stress and in most cases are accompanied by physical exertion on the part of the fish (and farmer). Even minor disturbances can cause enormous changes in the physiological status of fish. Because stressed fish do not grow well and may exhibit susceptibility to disease there is widespread awareness of the potential problems arising from husbandry stress, particularly repeated or chronic stressors. But can stress (and exercise) also cause undesirable changes in the characteristics of the flesh of fish?

Flesh quality is defined by a number of factors, among which are fat content, the distribution of fat within the body, flesh colour, and flesh texture. These characteristics are determined in part by the genetic makeup of the fish, and in part by the frequency, timing and quality of feeding. Evidence from other intensive animal rearing industries (cattle, pigs, poultry) indicates that stress, and exercise, can also influence flesh quality.

The relationship between stress and flesh quality in fish has attracted less research effort than other areas of aquaculture, and what results there are tend to be somewhat contradictory. The aim of this article is to provide a brief overview of what is currently known and attempt to reach some generalised conclusions.

Stress and exercise - physiological effects

To understand how stress and exercise can affect flesh quality in fish it's necessary to consider some of the changes in the physiology and metabolism of fish which stress and exercise can cause.

The reasons why fish respond to a stressful stimulus and the sort of response they mount have been discussed at length elsewhere³. In terms of metabolism the net effect of activating the stress response can be described as a switch from an anabolic state (in which tissues are actively synthesised) to a catabolic state (in which the

breakdown of tissues may occur). One important role of the hormones which are released as part of the stress response is to mobilise stored energy (fats and sugars) to provide the fish with the metabolic fuel necessary to help it overcome whatever challenge it is facing. Therefore, a major effect of stress can be characterised as the redistribution of stored energy resources. As you would expect, the depletion of stored reserves is greater in fish which are exposed to prolonged or repetitive stressors than fish which are exposed to brief stressors than fish which are exposed to brief stressors.

Not all routine husbandry procedures are necessarily perceived as stressful by the fish and don't, therefore, evoke a neuroendocrine stress response. However, if the procedure causes the fish to exercise (forced swimming) then the mobilisation of energy resources and accumulation of waste products will occur, just as is the case for stressed fish. Many biochemical and metabolic events are common to both exercised fish and to fish exposed to stressors.

Biochemical changes in muscle

It is the metabolic changes which take place in muscle tissue during exercise which appear to be directly responsible for subsequent alterations in flesh quality. Although complex, these changes can be summarised quite simply. When fish are exercised, and muscles are caused to contract, this work is powered by **adenosine triphosphate (ATP)** which is broken down to liberate energy contained within the molecule. This reaction generates **adenosine diphosphate (ADP)** together with free phosphate. The supply of ATP is regenerated from ADP by **phosphagens**, mainly **phosphocreatine** in muscle. However, the phosphagen pool is limited. Therefore, in 'red' muscle which has a predominantly aerobic metabolism ATP is also generated from the complete oxidation of carbohydrates, fats and amino acids in the **tricarboxylic acid cycle** while in 'white' muscle (the bulk of muscle mass in fish) which is adapted to anaerobic metabolism ATP is generated by the catabolism of **glucose to lactic acid** via the **glycolytic pathway**. **Glycogen** stored within the muscle is broken down to its constituent glucose to fuel the anaerobic pathway in muscle. The **lactic acid** which accumulates within the muscle is utilised during recovery to replenish the depleted muscle glycogen.

Thus the net effect of exercise within the bulk of fish skeletal muscle is a depletion of phosphagen and glycogen reserves and an accumulation of lactic acid with a resulting decline in muscle pH.

The effects of stress and exercise on flesh quality

How exactly do the biochemical changes described above affect flesh quality? There has been a considerable amount of research into changes in meat

quality arising from stress and exercise in cattle and pigs, and to a lesser extent in poultry, but relatively little research on this subject in fish. There is a general consensus that stress and exertion are linked to reduced flesh quality in animals and that this is primarily associated with disturbances prior to slaughter. Two factors appear to be important in determining flesh quality. If muscle glycogen levels are depleted excessively prior to death then the level of muscle metabolism post mortem is reduced resulting in the production of less lactic acid and a relatively high muscle pH. This can affect the appearance and water holding capacity of the muscle. If post mortem muscle metabolism is too high, or ATP levels are depleted excessively before death, then a reduction of muscle pH occurs, affecting the structure of the fillet, and the rate at which rigor mortis occurs is increased.

Therefore, the severity, duration, and timing of husbandry-related stressors will determine how flesh quality in fish will be affected. The likelihood of aquacultured fish being exposed to a severe chronic stressor is limited. Because of the impact of chronic stress on growth, feeding behaviour and the immune system, stress-related problems become apparent well in advance of slaughter and will be addressed. Nonetheless, it is worth considering what the direct effects of long-term, or intermittent acute stress, on flesh quality might be. Because of the catabolic effects caused by exposure to a long-term stressor the metabolic effects will be similar to those of starvation, with the mobilisation of stored energy reserves and possible muscle wasting. Published data suggest that the effects of prolonged food withdrawal, in the absence of additional stressors, varies in fish according to species.

Effects of long-term stress or starvation on muscle quality

When Atlantic salmon were starved for up to 86 days (at 4°C) prior to slaughter the glycogen content of white muscle at slaughter was reduced together with fillet fat and protein content. During post-slaughter storage on ice, muscle lactate and glycogen levels also decreased, accompanied by an increase in muscle pH. Despite these biochemical changes the effects of starvation on the texture, freshness and colour of the fillets were marginal and effects on fat content were masked by large variation in fillet fat content between and within individuals^{4,5}.

In contrast to the Atlantic salmon, in rainbow trout which were starved for up to 2 months prior to slaughter a reduction in the total lipid content of the fillet was accompanied by a detectable difference in eating quality with a putrescent odour and muddy taste being reported⁹.

However, shorter periods of starvation (<18 days) did not alter the fat content of rainbow trout fillets^{6,13}. In rainbow

trout subjected to a 10.5 hour period of transport in O_2 -saturated water following an 8-day period of starvation there was no effect of transport or starvation on white muscle lactic acid content, glucose content, or glycogen content. Nor was there any effect of transport on the texture of the fillet (hardness, elasticity, breakpoint)¹³. Starvation does not affect the concentration of phosphocreatine and ATP in rainbow trout muscle¹⁰ which is of some significance when considering the effects of pre-slaughter exertion/stress (see below).

Finally, at stocking densities high enough to cause a reduction in growth rate (16 kg m^{-3} rising to a maximum of 43 kg m^{-3}) relative to fish maintained at lower density (8 kg m^{-3} rising to a maximum of 25 kg m^{-3}) no significant effects were observed on muscle in terms of water content, protein, total lipids and fatty acids, and amino acid composition¹⁷.

Overall, these data suggest that moderate periods of food withdrawal prior to slaughter have no significant effects on fillet quality.

Effects of short-term stress and exercise on flesh quality

Severe exercise, or acute stress, causes considerable redistribution and depletion of metabolites in muscle. However, recovery from brief but severe exercise is rapid in most fish, requiring a maximum of about 8 hours. So the exposure of fish to acute stressors is unlikely to have significant effects on flesh quality unless it occurs very close to the time of slaughter. The processes associated with slaughter of fish from capture, through transport, handling, and stunning, are undoubtedly stressful. A number of published studies have examined whether short-term stress affects flesh quality in fish.

There was no difference in the fat content or texture of fillets from rainbow trout killed either by rapid cervical section or asphyxiation⁶. Similar conclusions were reached in a study which examined the effects of three slaughter methods (electroshock, CO_2 narcosis, concussion) on flesh quality in rainbow trout¹. In this case, slaughter method had no significant impact on quality measures (hardness and elasticity, water-holding capacity, bacterial counts) determined during storage. A sensory panel, which evaluated toughness, elasticity, firmness and succulence, could not discriminate between fillets from fish killed by each of the slaughter methods.

It has been suggested that most studies on the effects of harvesting-related stress on fish muscle quality have failed to allow for capture and handling effects and therefore the 'true' condition of 'rested' fish muscle may not have been reported⁷. Using a combination of conditioning, careful handling and anaesthetisation to minimise pre-slaughter stress the tensile strength of chinook salmon (*Oncorhynchus tshawytscha*) white

muscle was shown during storage to be significantly greater in unstressed than in exhaustively exercised/stressed fish. Loss of tensile strength was equated with undesirable 'tenderisation' of the muscle. Based on their findings, these authors emphasised the importance of reducing pre-harvest disturbance in producing high quality fish muscle. This conclusion is supported by the results of other studies.

The rate of onset of rigor mortis (stiffening of the muscle) is an important factor in maximising fillet yield because the yield is reduced if fish are processed while in rigor. Because the detachment of actin and myosin filaments in muscle fibres is ATP-dependent, rigor is observed when depletion of ATP reaches a critical point and the filaments 'engage'. Several studies have reported effects of stress/exercise on the rate of onset of rigor in fish.

In the snapper (*Pagrus auratus*) the depletion of muscle ATP and consequently the onset of rigor was delayed in unstressed fish relative to fish stressed prior to slaughter¹¹. Similarly, the onset of rigor was found to be more rapid in severely exercised chinook salmon white muscle than in unexercised control tissue⁸. In carp (*Cyprinus carpio*) stressed by severe disturbance prior to slaughter the onset of rigor was significantly more rapid (within 1.6 h) than in carp killed in the absence of disturbance (within 21.2 h)¹².

In Atlantic salmon rigor strength was found to be greater in fish stressed prior to slaughter². The onset of rigor is also more rapid in Atlantic salmon which have been stressed prior to slaughter and fillets from fish stressed prior to slaughter were softer than those from unstressed fish and had a lower breaking **strength**^{14,16}.

Conclusions

On balance, what little research has been carried out tends to suggest that pre-slaughter husbandry practices can have measurable effects upon the biochemistry of muscle tissue in fish but that these changes do not necessarily translate into adverse effects on flesh quality. As is the case for mammals and poultry, alterations in the muscle of fish following slaughter appear to be related to the degree to which muscle metabolism is stimulated before death. This, in turn, is directly related to the extent to which the fish are exercised and/or stressed prior to slaughter. The most consistently reported effect of pre-slaughter disturbance is an increase in the rate at which rigor mortis occurs.

An increase in the rate of onset of rigor is most likely to affect fillet yield, rather than flesh quality outright. There appears to be no widespread perception that the quality of farmed fish flesh following slaughter is inadequate or that there is a systematic occurrence of flesh quality problems analogous to those which are evident in other intensive meat production systems.

It seems likely that current approaches to ongrowing and the general nutrition and husbandry of aquacultured fish do not have any adverse effects on post-slaughter condition. However, it may be possible to further improve the quality of the farmed product by reducing the extent to which fish are disturbed/exercised immediately prior to slaughter.

Handling and disturbance cannot be completely avoided but sedation of fish prior to slaughter might be achieved by using one of a number of anaesthetics. In order to be economically viable and practical this approach requires an anaesthetic which does not present residue-related problems and which is affordable. Possible candidates are sodium bicarbonate/acetic acid (a cheaper alternative to gaseous CO₂, narcosis), clove oil, and the food-grade anaesthetic Aqui-S¹⁵.

In conclusion, further research targeted at the relationship between UK farm practices and fillet quality would allow an assessment of the extent to which the existing production of flesh of acceptable quality might be further enhanced.

References

1. Azam, K. *et al.*, 1989. International Journal of Food Science and Technology, 24: 69-79.
2. Berg, T. *et al.*, 1997. Journal of Food Science, 62: 439-446.
3. Black, K.D. and Pickering, A.D., 1998. Biology of Farmed Fish. Sheffield: Sheffield Academic Press. 429 pp.
4. Einen, O. and Thomassen, M. S., 1998. Aquaculture, 166: 85-104.
5. Einen, O. and Thomassen, M. S., 1998. Aquaculture, 169: 37-53.
6. Færgernand, J. *et al.*, 1995. Water Science and Technology, 31: 225-231.
7. Jerrett, A.R. *et al.*, 1996. Journal of Food Science, 61: 527-532.
8. Jerrett, A.R. and Holland, A. J., 1998. Journal of Food Science, 63: 48-52.
9. Johansson, L. and Kiessling, A., 1991. Acta Agriculturae Scandinavica, 41: 207-216.
10. Kieffer, J.D. and Tufts, B., 1998. Fish Physiology and Biochemistry, 19: 239-245.
11. Lowe, T.E. *et al.*, 1993. Journal of Food Science, 58: 770-773, contd. 796.
12. Nakayama, T. *et al.*, 1992. Nippon Suisan Gakkaishi, 58: 1517-1522.
13. Ostenfeld, T. *et al.*, 1995. Water Science and Technology, 31: 233-237.
14. Sigholt, T. *et al.*, 1997. Journal of Food Science, 62: 898-905.
15. Steffly, G.R. and Gingerich, W.H., 1999. Aquaculture Research, 30: 365-372.
16. Thomas, P. M. *et al.*, 1999. Journal of Fish Biology, 54: 1177-1196.
17. Zoccarato, I. *et al.*, 1994. Aquaculture and Fisheries Management, 25: 639-647.

THE AGENT OF PROLIFERATIVE KIDNEY DISEASE (PKD) REQUIRES A BRYOZOAN IN ITS LIFECYCLE - STUDIES AT CEFAS WEYMOUTH ON PKD AND OTHER MYXOZOANS

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Introduction

For a number of years researchers at CEFAS Weymouth have been investigating myxozoan parasites and the impact that they have on wild and farmed fish both in the marine and freshwater environment. World-wide there have been approximately 1400 different species of myxozoans described, mainly from fish, but only relatively few are significant pathogens.

Until 1985, it was thought that the lifecycle of these parasites involved only the fish host. The parasites were believed to be transmitted either directly fish to fish or following a period of maturation in mud for six months

or more before being infective to the fish. In 1985, two American scientists, Drs. Maria Markiw and Ken Wolf, showed that a lesser-known group of parasites called actinosporeans (or actinospores) in oligochaete worms were infective to fish (Figure 1) where they developed into myxospores which they had thought to be completely unrelated parasites. In those pioneering experiments, they showed quite clearly that the myxozoan *Myxobolus cerebralis*, the agent of salmonid whirling disease (WD), could alternate between a fish host and an invertebrate worm host.

In the last 15 years small groups of researchers in Europe, north America and Japan have been examining

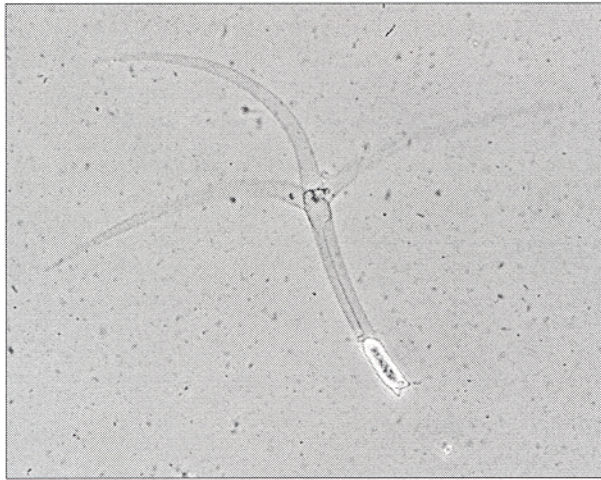


Figure 1. An example of an actinospore after release from an oligochaete host

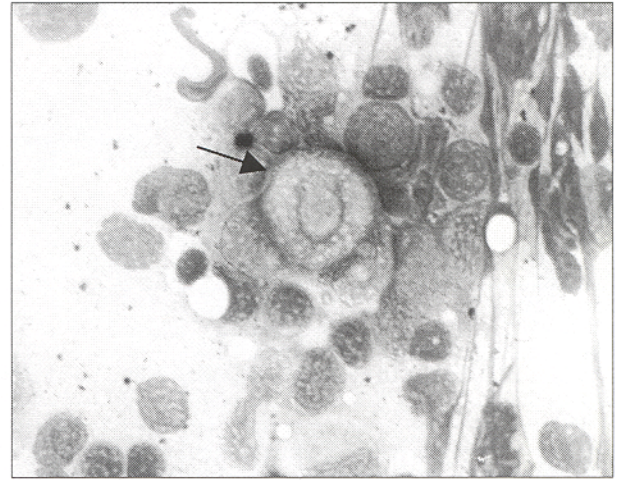


Figure 3. Air-dried and stained PKX cell in a kidney imprint from a PKD affected rainbow trout

the life cycles of these parasites in more detail. The WD lifecycle has now been confirmed and over 20 additional myxozoan lifecycles have been worked out (Figure 2).

Recently at Weymouth we have been working on lifecycles of freshwater myxozoans to try to understand their fundamental transmission requirements. Our interest stems from the known destructive effect of myxozoans on cyprinid fry and to the realisation that the causative agent of Proliferative Kidney Disease (PKD) is also a myxozoan (Figure 3).

The identification of the infective stages in 'traditional' myxozoans (Figure 4) and for PKD is essential in order to understand how infection is transmitted and to develop reliable disease challenges in the laboratory. Once laboratory challenges are available, these allow detailed investigations to proceed to identify factors of use for the development of potential vaccines and therapeutants.



Figure 4. Typical Myxobolus spore isolated from a cyst in dace

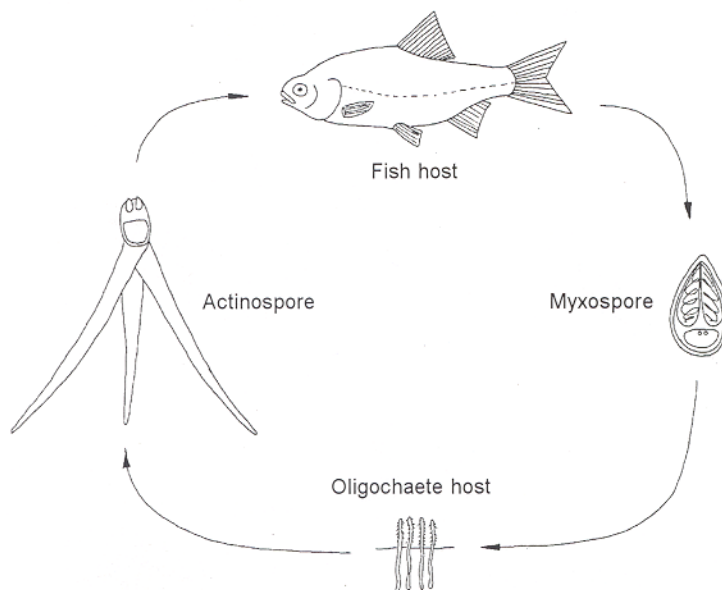


Figure 2. A simplified lifecycle of a typical myxozoan, showing the parasite alternating between an oligochaete and a fish. Note these are not to scale

The purpose of the study was therefore to examine the types, distribution and biology of actinospores from oligochaetes; to link these with their equivalent form in fish and to consider the possibility of other invertebrates being hosts for myxozoans, including the causative agent of PKD.

Oligochaete and fish studies

Over 16,000 oligochaete worms have been collected from five sites in Dorset and Wiltshire over the last three years and returned to the CEFAS Laboratory. The sites were chosen because of their different habitat types and were from areas where PKD is common as well as from sites known to be PKD free. They were maintained over a number of weeks until actinospores were released. These were identified and described and samples kept for electron microscopy and molecular biology studies. Over twenty different actinospore types were identified on the basis of their structure. Samples of fish were also collected at these sites and other reference sites and examined histologically to determine the distribution of myxozoans in fish populations and to try to understand the pathology caused by these parasites (Figure 5).

Samples of actinospores and myxospores were investigated using molecular biology techniques, basically a form of DNA fingerprinting. Two methods were used on the actinospore DNA. The first was to look at the DNA code in areas of that have segments common to all myxozoans and then compare the DNA sequence with other myxozoans to try to match the two stages of the parasite in the fish and oligochaete hosts. Additionally, we used DNA probes that were specific for PKX (designed by Dr Mike Kent and colleagues, USA). These would only work in the presence of stages of the real PKX. It soon became apparent that none of the actinospores that were being released from the oligochaete hosts were PKX.

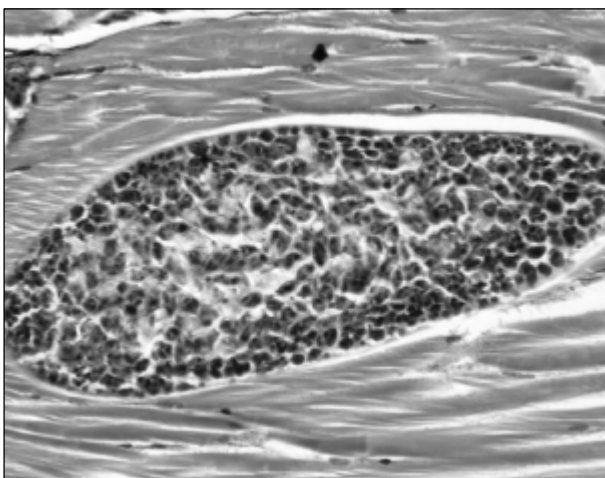


Figure 5. Histological section through the muscle of chub showing a myxozoan cyst within the muscle fibre

PKD studies - the bryozoan link

If PKX was not in oligochaetes, to try to identify where the infective stage of PKD might be when not in the salmonid host we examined over 80 species of invertebrates collected at different times of the year from the rivers feeding farms enzootic for PKD. The animals looked at included *Gammarus*, caddis flies, leeches and snails. We subjected these invertebrates to the PCR test using our PKX specific primers - all were negative.

As part of our general studies on myxozoans, we have been collaborating for several years with Prof. Elizabeth Canning from Imperial College, London on some of the molecular aspects of these parasites. She in turn was collaborating with Dr Beth Okamura from the University of Reading. Dr Okamura is an expert on the biology of a relatively obscure group of invertebrates called bryozoans or 'moss animals'. She was experiencing difficulties in maintaining these animals alive as they were infected with an unidentified parasite. Prof. Canning and Dr Okamura subsequently described the parasite as a new species and genus in the scientific literature and named it *Tetracapsula bryozoides*. The parasite forms large sac-like structures in the body cavity of the bryozoan host. These sacs contain myxozoan spores. When the DNA sequence and the ultrastructure of *Tetracapsula bryozoides* was examined it became apparent that it was different to PKX, but importantly, very closely related to PKX in fish.

Starting in May 1999, samples of bryozoans were collected from the rivers feeding the fish farms. Bryozoans are small, sessile colonial animals often found encrusting on any firm substrate such as submerged tree roots and branches (Figure 6 and 7). Many myxozoans of the same type were observed in the body cavities of the bryozoans collected (Figure 8). We amplified and sequenced the DNA and confirmed that the spores were indeed stages of PKX. At the same time as the sequencing was being carried out, some of these parasites were used in attempts to infect rainbow trout that had not been previously exposed to the parasite.

In July 1999 transmission trials began. Fish which had been reared from eyed eggs at CEFAS Weymouth were exposed to the parasite from bryozoans by several methods. This included injecting spores from bryozoans or infected fish kidney homogenates into the peritoneal cavity, 1 hour bath exposure to disrupted infected bryozoans and finally by exposing the fish for twelve weeks with colonies of infected bryozoans. Subsamples of fish from each of these groups were taken every two weeks and samples examined by kidney imprints and blood smears, by histology, by electron microscopy and by molecular biology (PCR). By the end of ten weeks, the fish that had been exposed to the parasite in the water (bath and exposure to intact bryozoans) and by injection of fish kidney homogenate from naturally infected trout showed the presence of the parasite.

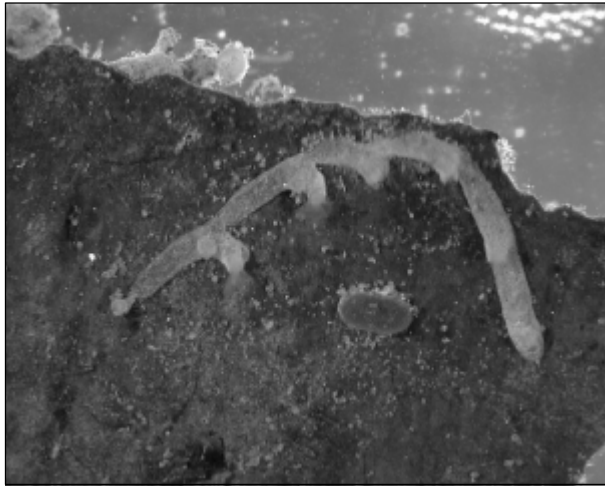


Figure 6. A small bryozoan colony encrusting on a rock

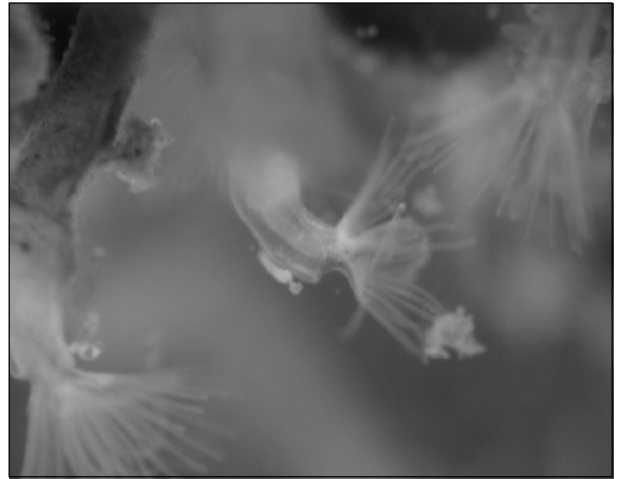


Figure 7. A single bryozoan collected from a site enzootic for PKD. Apparent is the fan-like lophophore used in feeding

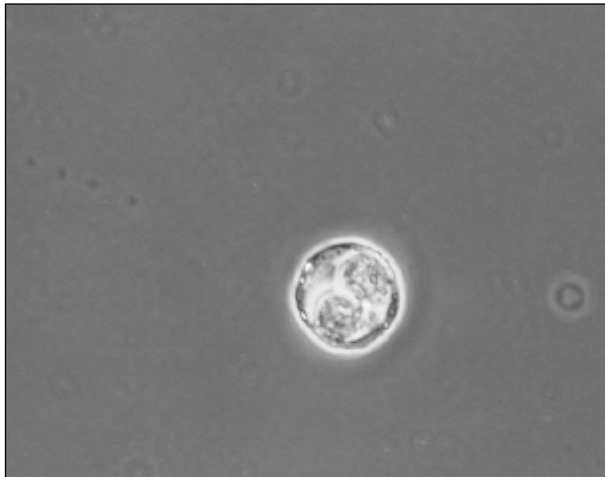


Figure 8. *Tetracapsula bryosalmonae* (PKX) spore floating in the water column after release from the bryozoan host

Parasites were first seen in kidney imprints after four weeks. By ten weeks, clinical signs including swollen kidneys and spleens and exophthalmia (bulging eyes) were present. All controls remained free of the disease.

This positive result was a breakthrough and has provided experimental evidence in support of the molecular biology data that the causative agent of PKD, PKX is indeed a stage of the bryozoan parasite.

Since then, in conjunction with Prof. Canning, Dr Okamura and Dr Curry, the PKX parasite has been formally described and given the scientific name *Tetracapsula bryosalmonae*.

Conclusion/Discussion

One of the major stumbling blocks in the PKD story has been that the parasite in fish doesn't form spores that could be easily isolated. Therefore there has never been a realistic means of infecting fish (or invertebrates) with the parasite. Additionally, it wasn't known where the infective stage of the parasite lived when it wasn't in the rainbow trout. This meant that almost all research depended on the availability of the parasite on the fish farm, limiting work to the summer. We now have something that we can use. We know what one of the alternate hosts is (there still may be others) and we know something of the biology of the bryozoans. This now allows us to carry out controlled experiments in the laboratory in order to understand exactly what is happening during the crucial early part of the disease process. Whilst it should be remembered that we still have a long way to go before we have all the answers, the potential for a vaccine against *T. bryosalmonae* and the possibility of eradicating or reducing the impact of the disease on the farm, and maybe in the wild, becomes a lot closer.

MAFF/BTA FUNDED RESEARCH

RESULTS OF R AND D PROGRAMME ON FISH-EATING BIRDS

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Introduction

In 1995 MAFF, DETR (Department of the Environment Transport and the Regions) and the Environment Agency (EA) commissioned a £1 M programme of research on the problem of fish-eating birds at inland fisheries in England and Wales. In setting up the programme both fishing and bird conservation interests were fully consulted. The overall aim of the research was to improve the current level of information on the behaviour of fish-eating birds, the populations of these birds, the extent of the problem they cause to fisheries and to develop effective management strategies. Five projects were let by competitive tender and these are shown in Table 1 below.

These projects were overseen by a Programme Advisory Group consisting of MAFF, DETR, EA, Scottish Office (now Scottish Executive), Welsh Office (now National Assembly for Wales), JNCC (Joint National Council for Conservation) and the research contractors. The results of the research programme has contributed significantly to understanding the problem of fish-eating birds at inland fisheries and has confirmed the difficulties involved in assessing fish populations and the impact of bird predation upon them.

Dissemination of information

Results from the research were released at annual public seminars to ensure that they were disseminated in an even-handed and balanced manner. The report for the first project on the assessment of the problem, which was of short duration, was published in 1996. Reports for the other projects were launched at the 4th Annual Seminar on Piscivorous Research which was hosted by DETR in London on 30 September, 1999.

Results

The main results from the five projects are summarised below together with details of where copies of the reports can be obtained.

1. Assessment of the problem of fish-eating birds in inland fisheries in England and Wales

This 6 month review at the start of the R&D programme provided a comprehensive evaluation of the scientific literature relating to the extent and significance of the impact of cormorants, goosanders and red-breasted mergansers on inland fisheries in England and Wales

Table 1. Projects let by competitive tender

Sponsor	Project title	Lead contractor	Duration (years)	Total cost (£k)
MAFF	Assessment of the problem of fish-eating birds in inland fisheries in England and Wales	CEFAS (Centre for Environment, Fisheries and Aquaculture Science)	0.5	32
DETR/EA	Population, distribution, movements and survival of fish-eating birds in Great Britain	British Trust for Ornithology (BTO)	3	102
DETR	Feeding behaviour of fish-eating birds in Great Britain	Wildfowl and Wetlands Trust (WWT)	4	210
MAFF	Case studies of the impact of fish-eating birds on inland fisheries in England and Wales	Liverpool John Moores University (LJMU)	3	355*
MAFF	Assessment of the effectiveness of the management measures to control damage by fish-eating birds to inland fisheries in England and Wales	Central Science Laboratory (CSL)	3	332

* Includes a contribution from the Red Scar Angling Club

with due regard for the economic value of the fisheries and conservation of the bird species. CEFAS was the lead contractor of a consortium that also involved Central Science Laboratory (CSL), British Trust for Ornithology (BTO), Agriculture Department Advisory Service (ADAS) and the University of Glasgow.

The review provided an important assessment of current knowledge which brought together the numerous facets of the fish-eating birds problem whilst also demonstrating the complexity of the topic. In addition, the findings influenced the detail of the other four projects in the programme. The report included recommendations for assessing the impact of fish-eating birds at inland fisheries and future research requirements.

Although published in 1996 copies of this report are still available. The full report priced at £10 or a summary report at £7.50 can be purchased by contacting the library at the CEFAS Lowestoft Laboratory, Pakefield Road, Lowestoft, Suffolk NR33 0HT (Tel: 01502 524342).

2. Population, distribution, movements and survival of fish-eating birds in Great Britain

This project addressed the size and distribution of cormorant, goosander and red-breasted merganser populations, movements of these species and the survival rates of cormorants, and made proposals for increased monitoring of populations. The work was undertaken by BTO in collaboration with ADAS, University of Glasgow, CSL and Wildfowl and Wetlands Trust (WWT). Population estimates presented in the report reflect the methods used to record numbers. Therefore, estimates for the breeding season are given as breeding pairs (this figure excludes non-breeders), except for red-breasted mergansers which are based on adult males; winter figures represent total numbers of individual birds.

Cormorant numbers of breeding pairs at inland sites in England rose from sporadic records in 1981 to 1317 pairs in 1996 and include the continental race *sinensis* which now breeds in Britain. The UK wintering population stabilised at 15-16000 birds by 1994/95 after increasing at 5-10% per annum since 1987/88. Although numbers have remained stable on natural waters, coasts/estuaries and rivers the proportion of sites occupied in inland habitats have increased on gravel pits and reservoirs. Survival rates derived from ringing recoveries indicated that between 1965 and 1994 survival for first year birds declined from 55% to 33% and for adults from 85% to 72%.

Goosander numbers have increased in Britain by 10% per annum (13% in Wales) since 1987 with an estimated

6,609 breeding pairs in 1997. Estimated numbers of wintering birds stood at 8,900 in Britain in 1990/91. Ringing data suggest goosanders winter within 95 miles of their breeding sites.

Red-breasted mergansers in Britain and Ireland numbered 2,850 breeding pairs between 1988-1991. Densities on rivers in Britain did not change between 1987-1997 but there has been a southward redistribution of population. Estimates for wintering mergansers during 1987/88 - 1991/92 amounted to 12-13,000 birds (10,000 in Britain). Little is known about the movements of mergansers but the available information suggests that movements are variable during the breeding season and in winter.

Copies of this report, price £40 are available from Publications Sales Centre, Department of the Environment, Transport and the Regions, Unit 21, Goldthorpe Industrial Estate, Goldthorpe, Rotherham S63 9B2 (Tel: 01709 891 318)

3. Feeding behaviour of fish-eating birds in Great Britain

The aim of this project was to assess the feeding behaviour of fish-eating birds with respect to cormorants, goosanders and grey herons. The work was undertaken by WWT in collaboration with the Institute of Terrestrial Ecology and the Institute of Freshwater Ecology.

Grey herons

A literature review of grey herons and their impact on fisheries concluded that, on the basis of available evidence, it was likely that serious damage to fisheries by grey herons occurs only infrequently. The researchers could not find any published scientific studies which attempted to quantify heron predation at fisheries in Britain other than at fish farms. A questionnaire survey of angling clubs in Britain suggested that herons were thought to be a problem throughout Britain, mostly on rivers, but also on standing waters in north-west and south-east England. Reported problems were thought to occur throughout the year and were either stable or increasing in most regions. Impact has been most frequently reported at fish farms and fish ponds, including direct losses through predation and indirect losses through wounding. Caged stock appeared to be more vulnerable to grey heron predation than stock held in ponds, with losses of 7% of standing stock recorded at one cage fish farm in Scotland compared to 0.3-1.0% at other farms in the UK and the rest of Europe. Heron predation has only been quantified in the UK at fish farms. They take mostly small fish (<20 cm), so few of these are likely to be of commercial size or of the size sought by anglers, except in the case of some cyprinids. Predation levels on natural waters, based on European studies, suggest that

herons may take up to 6% of standing stock. It therefore seems unlikely that grey herons in the UK generally cause 'serious damage' to fisheries but may be damaging at individual fisheries.

Cormorants

Studies of feeding behaviour of cormorants at Grafham Water in Cambridgeshire recorded between 3 and 480 birds flock-feeding. This behaviour, which is believed to be more effective than solitary hunting, was frequently observed although reports of its occurrence are few in Great Britain. Comparison of size class frequencies of fish caught in gill-nets with those observed to be eaten by cormorants suggests they selectively feed on small fish.

In assessing feeding success, the researchers considered that cormorants exhibiting wing-shaking and other plumage maintenance, such as preening, after a dive were likely to have swallowed a small fish underwater. Using this assumption considerably affected estimated fish consumption. For example, at Grafham Water almost 70% of the fish caught were accounted for in this way. Fish consumption was also estimated using energetics data derived from radio-tracked birds. Both these data and data from the adjusted fish consumption estimates derived from Grafham, gave a Daily Food Requirement of about 400 g (14 oz) per day per cormorant. This is consistent with previously published estimates but both methods for calculating this figure relies on the assumption that significant numbers of fish are swallowed underwater. An alternative explanation, suggested by the researchers, is that a number of the fish consumed on the surface may have been missed by observers as occasionally the birds were a considerable distance away from them.

The movements of cormorants were investigated by following a small number of birds that had been fitted with either radio or satellite transmitters. Wintering birds were found to travel up to 19 km (12 miles) on a daily basis to feeding sites; which is consistent with published data. Seasonal movements of post-breeding birds of over 200 km (125 miles) were also recorded during summer. Analysis of data collected from 80 inland stillwaters suggested that the presence of cormorants at these sites was best explained by the size and geographical location of the site, the distance to the nearest cormorant roost and the level of water-based disturbance. Fish availability was not found to be a significant factor explaining cormorant presence. However, the researchers concluded that the index of fish production used to address this site characteristic was too imprecise.

Goosanders

Goosander feeding behaviour was studied on the River Wye. Details including numbers and type of fish caught, capture rates, dive and inter-dive times and differences between sexes and between immatures and adults are given in the project report.

Copies of this report, price £40 are available from Publications Sales Centre, Department of the Environment, Transport and the Regions, Unit 21, Goldthorpe Industrial Estate, Goldthorpe, Rotherham S63 9B2 (Tel: 01709 891 318)

4. Case studies of the impact of fish-eating birds on inland fisheries in England and Wales

The main aim of this project was to quantify the short and long-term damage associated with cormorants and goosanders at inland fisheries using a case study approach. The work was undertaken by Liverpool John Moores University in collaboration with Hull International Fisheries Institute. The case study sites for studying cormorants were located at 4 stillwaters (Holme Pierrepont, Nottinghamshire; Colwick Park Trout Lake, Nottinghamshire; Grimsargh Reservoirs, Lancashire and Rye Meads, Hertfordshire) and 2 rivers (Ribble and Trent). Data were collected from October 1995 to April 1998. Goosanders were studied on 3 rivers (Ribble, Hodder and Wye) during the same period. The sites were chosen from regions with perceived bird predation problems which could readily be monitored and had suitable historical data sets.

The project sought to improve on previous impact studies through the use of a Monte Carlo Simulation model which allowed the calculation of confidence limits around estimates of fish consumption by birds. Fish and bird survey data were supplemented by analyses of the stomach contents of shot birds. Impact was estimated in terms of fish consumption by birds in relation to the estimated mass of fish (the standing crop biomass) at a fishery. Predation was modelled using theoretical estimates of the Daily Food Requirement of fish-eating birds and, for a small number of sites, data from direct feeding observations.

The case studies revealed wide variation in fish consumption estimates by cormorants during the course of the project. These ranged from about 4% of the standing crop biomass at Grimsargh to about 57% at Rye Meads. In general, predation was found to be more variable at stillwaters than rivers. The study highlighted the complexity of the issues associated with determining the causes of poor fishery performance. For example, at Holme Pierrepont, the very low catches at the World Angling Championships in 1994 were attributed by anglers to the arrival of large numbers of cormorants at the site during 1993-94. However, data presented by the researchers suggest that whilst cormorants may have played a part in the decline, other factors may have had a more significant impact. Analysis of historical angler catch data revealed that catches, expressed as catches per unit of angler effort, had been declining since the early 1990s. However, since 1994, catches have been rising again despite increasing predation by cormorants. The available data suggests that a series of years of poor

recruitment to the fish population prior to 1994 followed by a succession of good years after 1994 had a major impact on the fishery. At present, it is unclear whether, with increasingly heavy predation by birds, the fishery would be able to withstand another series of poor or even average recruitment years. A variety of environmental factors can affect adversely recruitment, particularly climatic factors.

For goosander broods, the data relating to their impact on fisheries highlighted another problem that affected the impact estimates covered by the project, namely, difficulties associated with the accurate estimation of the size of fish populations. The most extreme example of this problem was the River Wye where it was estimated that goosanders removed 1-7 times the total fish biomass in the river! The researchers suggested that problems relating to the fish surveys undertaken during the study, such as, small sample sizes and difficulties in calibrating fish survey equipment, resulted in an under-estimation of fish biomass. For the Rivers Ribble, Hodder and Wye, 6-20%, 22-60% and 16-97% of juvenile salmon were estimated to have been removed but difficulties were encountered with the fish surveys in all cases.

Overall, the findings from the project suggested that at some sites bird predation may be high enough to cause a decline in the fishery but at others they may not and other factors may be more significant. The researchers conclude that the current study supports the view that the impact of fish-eating birds on inland fisheries is a problem for specific fisheries rather than a general problem. Therefore, no single threshold, above which fish losses are considered to be serious, can be applied to all sites. Based on the finding from the project and also the scientific literature, the researchers consider that sufficient data can be collected to undertake practical impact assessments on a case by case basis; suggested guidance is provided in the project report.

Copies of this report, price £15 are available from MAFF, The Library, Room 320, Nobel House, 17 Smith Square, London SW1P 3JR (Tel: 0171 238 6575)

5. The assessment of the effectiveness of management measures to control damage by fish-eating birds to inland fisheries in England and Wales

The main aim of this project was to investigate how damage to stillwater and river fisheries by fish-eating birds (cormorants and goosanders) could be managed using the techniques that are humane, cost-effective and acceptable in terms of their environmental impact and their effects on recreational activities. CSL was the lead contractor of a consortium that also involved CEFAS, BTO, University of Glasgow and ADAS.

The project commenced with reviews of management practices undertaken in the UK and Europe. Of the 23

techniques examined, only human disturbance was found to be consistently effective at reducing bird numbers at sites. The results from the reviews, together with other information on the management of birds, were assessed in terms of their effectiveness, practicality, acceptability and cost. A number of techniques were identified as worthy of further study and the main results are summarised below:

Shooting as an aid to scaring

A field experiment was established at 13 sites across England and Wales to assess whether shooting could be used to reduce numbers of cormorants present at sites and, in particular, whether shooting to kill enhanced the scaring effect of shooting with blanks. The results showed a 50% mean reduction in cormorant numbers when sites involving shooting to kill (lethal treatment) were combined with those where shooting with blanks (non-lethal treatment) was undertaken. The results did not show a statistically significant difference between the effects of lethal and non-lethal shooting. However, the researchers consider that a number of factors may have affected this result, particularly between-site differences and the timing and duration of shooting. In conclusion, the researchers consider that this single experiment did not resolve the question of whether or not killing enhances the scaring effect of shooting; they have recommended further research to resolve this issue.

Stocking control

A 6-week pilot trial was undertaken at Rye Meads, Hertfordshire to investigate the effect of fish size on cormorant predation. Two lagoons were stocked with 3 sizes of carp (large, medium and small). One lagoon was netted over and the other left open. A 62% mortality of carp was recorded in the open lagoon compared to 4% in the netted lagoon with significantly more large than medium or small fish recovered. Of perhaps more interest, however, was the finding from observations of cormorants feeding which suggested that the number of birds seen to successfully capture fish at the open lagoon could not have accounted for all the 'missing' fish. In contrast, consumption estimates based on the theoretical Daily Food Requirements for cormorants suggested that sufficient quantities could have been consumed by birds visiting the open lagoon to account for the missing fish. The researchers suggest that many of the fish, particularly those in the smaller size classes, could have been consumed underwater. Some nocturnal fishing by herons may also have occurred though this was not observed during the small number of nocturnal observations undertaken.

Fish refuges

The previous trial was repeated but with wire fish refuges placed in each lagoon. A 62% difference in mortality between the open lagoon and the netted lagoon was considered to be the result of bird predation. However, the incidence of wounding of fish from the open lagoon was significantly less in this trial than in the

previous one (41% of large fish compared to 77%). In addition, cormorants spent significantly longer hunting for fish as measured by dive duration times. Once again, feeding observations suggested that cormorants could not account for all of the missing fish whereas Daily Food Requirement calculations suggested the converse. On the basis of this trial, the researchers consider that further work is warranted to optimise the use of this simple and relatively inexpensive technique.

Laser light

The scaring effect of a beam of laser light aimed at cormorants and goosanders was investigated. In pilot trials, the technique successfully reduced numbers of cormorants at one roost but not another (which had alternative roost sites nearby). Goosanders were found to respond to laser light at brighter daylight levels than cormorants, being put to flight at levels up to 8000 lux (a dull day). In contrast, cormorants were put to flight when daylight was between 20 and 1500 lux (dawn and dusk). In 90% of tests, other duck species would remain undisturbed. The use of the laser reduced goosander fishing activity on a 1 mile stretch of river from 129 dives to 23 dives. When birds failed to respond, this was probably because ambient light intensity was too high (at midday). In a second trial on a 3.75 mile stretch of river, the effect of the laser was shown to last less than one day.

Habitat management

Results from pilot trials to assess the use of wires stretched across stock ponds suggested that they could be useful for deterring cormorants provided they were sufficiently closely spaced and an alternative food source was readily available. Roost modification, involving the netting of a tree used as a day roost at one site, was found to be practical and effective in deterring cormorants from using the tree over a 4 month period. However, the technique has a number of limitations, such as its unsightliness in the eyes of the public and

reduced effectiveness if alternative roost sites are available nearby.

Conditioned taste aversion (CTA)

This technique, which involves using a chemical to train animals to avoid specific foods, was investigated in cormorants for the first time. CTA was induced in 5 of the 8 birds used in the study and lasted for 7 months to the end of the trial in some of these birds. The researchers consider that the technique's potential is likely to be limited to put-and-take fisheries where local birds that stay on site can be trained to avoid the species of stocked fish. In addition, further refinement of the technique is considered necessary, particularly with regard to the choice of CTA-inducing chemical. Any candidate CTA agents arising from the research would require regulatory approval before they could be marketed.

Human disturbance

Goosanders on two rivers were surveyed from December to May, over two years. Goosanders were significantly more likely to fly upstream if disturbed, so when censusing goosanders, observers should walk downstream. Goosanders on a river where they are shot under licence became alert to human presence at a greater distance than on an unshot river and at a greater range in April/May than in February/March on the shot river. These differences could be due to differing levels of human disturbance. Activity budget data suggest that goosanders are able to obtain their daily food requirement in only a few hours of foraging per day. Evidence was found that disturbance by anglers reduced goosander numbers on the study rivers. Gas cannons were found to be ineffective on a stretch of river adjacent to crops.

Copies of this report, price £23 are available from MAFF, The Library, Room 320, Nobel House, 17 Smith Square, London SW1P 3JR (Tel: 0171 238 6575)

LINK COMMITMENT TO TROUT R&D PROJECTS NOW £2.2 MILLION

Compiled by Mark James of LINK Aquaculture

The LINK Aquaculture Programme is now committed to the support of 37 projects costing in excess of £10 million with industry contributing £5.3 million (53%) of this sum. Approximately 23% (£1.2 million) of industry's commitment is in cash (12% of total budget), with the balance being made up of a range of in-kind contributions including farm staff time, equipment and stock, etc. The trout industry is now directly involved in eight LINK Aquaculture projects worth £2.2 million. Unlike the salmon sector, the trout industry has no levy

system to raise funds that may be used to support a programme of R&D. For the trout industry, individual farms, often co-ordinated through the British Trout Association, are contributing £238,000 in cash and £950,000 in kind in support of industry priority projects.

Since the last Trout News update on LINK - trout projects, another two project proposals have been approved. Project TRT13 - *Off-flavour problems in farmed trout*, will attempt to identify the organisms that

cause taint in trout and develop appropriate management strategies. Project TRT12 - *Selective improvement in rainbow trout* - will use the latest techniques for selecting broodstock with desirable characteristics for breeding.

The performance of the current LINK programme is to be evaluated in 2000 and plans for a future Programme are under discussion. The progress of current projects will continue to be monitored annually. As usual, the project leaders of the LINK - trout projects have provided brief summaries of the progress in each project. On 31 March 2000, LINK Aquaculture will stage a one-day conference alongside the Aquaculture International Exhibition at the SECC in Glasgow. The Conference will provide a unique opportunity for members of the aquaculture industry to see the fruits of industry-oriented research for themselves.

The Conference has been specifically designed to provide a clear insight into the rationale behind each project. The results will be presented in clear and easily understood format. The schedule of presentations has been structured to allow delegates to take full advantage of both the trade Exhibition and the Conference on the same day. A comprehensive range of posters will also be on display, with the experts on hand to answer your questions.

To secure a place at the conference and ensure a registration pack, contact the LINK Office as soon as possible:

Tel: 01796 472060 ext. 220
Fax: 01796 473523
Email: majames@compuserve.com
<http://www.linkaquaculture.com>

Project progress summaries:

PYCEZE an alternative to Malachite Green – TRT01

*Project Leader: Mr Julian Braidwood
(Vericore Limited)*

Sponsor: MAFF

An application for a marketing authorisation for Pyceze was submitted during 1997. This submission contained information relating to the environmental, pharmaceutical, pharmacological/toxicological residues and efficacy aspects of the product.

An Animal Test Certificate (ATC) was granted in August 1999, which permits field efficacy studies with Pyceze in fish in the UK. Considerable effort has been expended in setting up fish and egg efficacy studies. Investigators and potential trial sites have now been recruited and the first treatments are planned for December 1999 in the UK. A General Exemption for Pyceze has been granted in Norway where some limited field studies have produced very encouraging results in cage treated fish.

The proposed treatment recommendations have previously been established following extensive *in vitro* dose titration, dose confirmation and homogeneity testing of Pyceze.

The Environment Agency (EA) and the Scottish Environment Protection Agency (SEPA) have been approached regarding the use of Bronopol (the active component of Pyceze) in aquaculture. The EA have assessed the ecotoxicological data and calculated a Predicted No Effect Concentration (PNEC) for Bronopol. Discharge concentrations are being calculated for trial sites that have been recruited into the study for reference to the PNEC.

SEPA have completed their assessment of Pyceze and have informally advised that discharge consents can be applied for, and will be considered on a site by site basis once a marketing authorisation is granted. Additional environmental monitoring, including field dispersion studies, have been requested by SEPA to demonstrate the safety of Pyceze administered to fish reared in fresh water loch cages. In addition, two ecotoxicological studies have been performed to determine the effects of Pyceze and Bronopol on the growth of the algal species *Scenedesmus subspicatus*. SEPA have been asked to propose their Environmental Quality Standards (EQS) to enable site discharge calculations to be compared to authorised levels.

Target animal safety of salmonid eggs and fish to Pyceze treatments has been demonstrated in Good Laboratory Practice (GLP) tolerance studies. Pyceze may be a suitable product for other indications, such as Rainbow Trout Fry Syndrome (RTFS), Bacterial Gill Disease and other bacterial infections. Simple, look-see studies are being performed to establish the potential of Pyceze for indications other than fungal infections in fish and eggs.

An application for a Provisional Marketing Authorisation (PMA) has been made and an update on its progress will appear in a later edition of Trout News.

By Dr Richard Hunter

Identification and assessment of chemical control methods for PKD –TRT-04

Project leader: Dr Adams

(Institute of Aquaculture, University of Stirling)

Sponsor: MAFF

Proliferative kidney disease (PKD) is caused by a species of myxosporidean recently named *Tertacapsula bryosalmonae*, formally known as PKX. PKD affects 28 trout farms on different water supplies in England and Wales annually. New sites are reported each year where clinical signs of PKD are observed, and mortalities as high as 50% of infected stocks have been recorded on

some farms on the rivers Test and Avon. At present there is no vaccine against PKD, and effective treatment of early life cycle stages prior to fish infections appears increasingly unlikely with the identification of bryozoa (filter feeding invertebrates) as alternative hosts for the parasite. Chemicals tested in the past, such as Fumagillin, have been linked with toxicity problems in fish and appear to be dependent on a very narrow window for treatment time. Research partners from the Institute of Aquaculture, University of Stirling, and industrial partners from the British Trout Association, Vetrepharm Limited and Aquaculture Vaccines Limited are assessing alternative chemical control methods for PKD.

Results of intensive short-term aquarium trials and small-scale field trials identified five compounds that affect PKD. Methods of administration, dose level and toxicity trials have been performed to select the most efficient treatment strategy. The results from the most recent experimental trials indicate that two compounds in particular have the ability to control PKD. One chemical appears to affect the parasite directly, while the other delays the on-set of infection. These results are encouraging as no toxicity in fish was observed with either of the compounds. Future work will involve large-scale field trials and detailed studies on the mode of action of the compounds.

By Dr Alexandra Adams

Assessment of chemical and potential immunological control methods for *Ichthyophthirius multifiliis*: Chemical methods – TRT06

Project Leader: Rod Wootten

(Institute of Aquaculture, University of Stirling)

Sponsor: MAFF

The LINK Aquaculture project looking at the efficacy of a number of bath and in-feed treatments against the ciliate protozoan whitespot, *Ichthyophthirius multifiliis* (Fouquet, 1876), continues at the Institute of Aquaculture.

Chloramine T has proven 100% effective in vitro against the trophont (visible as a white spot under the epidermis) and theront (the free-swimming infective stage) of *I. multiplies*. Preliminary *in vivo* bath treatments demonstrated significant reduction in the level of trophonts on treated fish probably due to effects on the free-living stages. Field trials to assess the efficacy of Chloramine T using naturally infected farm stock are planned for the coming year.

Pyceze (Vericore) has similarly proven 100% effective in vitro against trophonts and cysts (the external proliferative stage) but has proven only partially

successful in the eradication of theronts at the concentrations tested. Bath treatments of Pyceze showed efficacy in treatments of fish already infected with the parasite and further trials will attempt to refine the number of treatments and the dosage required. Pyceze significantly reduced infections of naive fish brought into contact with previously infected fish and therefore may have potential for use as a prophylactic treatment at times of high infection.

Two in-feed compounds are now being tested which significantly reduced trophont numbers in infected fish. A number of new candidates have been proposed for testing next year together with additional trials to confirm the effective doses of the compounds discussed above.

By Dr Andy Shinn and Dr Rod Wootten

Automated Humane Slaughter of Trout – TRT07

Project Leader: Dr Jeff Lines

(Silsoe Research Institute)

Sponsor: MAFF

This project aims to develop a system for killing trout which is demonstrably humane, maintains current quality standards and which can be automated safely and economically. The work is carried out by Silsoe Research Institute and the University of Bristol and is supported by MAFF, The BTA, five major supermarket chains, GW Aquaculture and the Humane Slaughter Association.

Work on this project began with a survey of a wide range of potential killing methods. Percussion and electrical stunning were found to be the most promising methods. An electrical or mechanical stun applied to the fish head was shown to result in instantaneous loss of consciousness with no detrimental effects on carcass quality. Initially, an electrical stun applied to the whole fish appeared to result in some carcass down grading. However, after further investigations we identified a technique for using high frequency electric current at low voltages which preserves carcass quality. This is likely to be simpler than applying a stun to the fish head since the fish do not need special positioning.

An electric stunning system is now being constructed for testing. This device will be connected to a standard fish pump and will stun the trout as they are pumped through the pipe. This stun will rendered the fish unconscious before they are removed from the water. They can then be pumped into ice slurry following normal industry practise. Extensive testing of this system is planned to ensure that the required quality and welfare standards are maintained. The results of these tests will then be used to specify the design parameters of a pre-prototype device for practical on farm tests.

By Dr Jeff Lines

Development of vaccination methods for RTFS – TRT10

Project Leader: Rachel Rangdale

(CEFAS - Weymouth and Institute of Aquaculture, University of Stirling)

Sponsor: MAFF

Since the last update research at CEFAS has focused on characterisation of antigens of *F. psychrophilum* using immunoblotting techniques, and on further examination of the humoral response to natural infection and immunisation. The results of extensive blotting studies have demonstrated the presence of soluble protein antigens, which are moderately immunogenic in rainbow trout. Periplasmic proteins, extracellular proteases and crude lipopolysaccharides appear less antigenic. Trout antibody raised against live whole cells and soluble protein fractions exhibit bacteriostatis *in vitro*. To determine if this specific antibody confers protection in fry and fingerlings passive immunisation studies are in progress. The application of optimised antibody detection methods has shown that that fish have the capacity to produce specific antibody following IP immunisation with adjuvanted, live, whole cell and soluble proteins. During acute natural infection similar levels of antibody are elicited. However, in serum samples collected from fish at infected farms, showing no overt signs of disease, no antibodies have been detectable. Levels of circulating antibody rapidly decline following immunisation, and are not found from approximately day 48 in laboratory infected trout. Consequently, it is speculated that the duration of the response following exposure to the antigen is relatively short. Immunisation with formalin inactivated dead cells does not induce a measurable antibody response, and this may explain why the initial vaccine trials have been unsuccessful.

Over the last six months research at the Institute of Aquaculture has concentrated on identification of differences in the antigenic profiles of isolates from disparate hosts and geographical locations, and on the effect of culture conditions on antigen expression. The profiles of all bacteria tested so far have been very similar irrespective of the species of isolation, geographical region from which the bacteria originated or media in which bacterial cells are grown in the laboratory. Most of these bacterial isolates have been repeatedly sub-cultured and therefore, may no longer be pathogenic to rainbow trout or represent the pathogens antigenic profile *in vivo*. Thus, a series of laboratory challenges have been performed to passage *F. psychrophilum* through fish. It has been shown that the protein profiles of these re-isolated fish adapted bacteria appear to be very similar to the original laboratory isolates.

An alternative *in vivo* culture method will also be utilised in the next phase of the project as passaging

isolates through fish still requires a period of growth on artificial growth medium and the degree of change in the antigens expressed cannot be determined. Bacteria will be placed in a growth chamber in the peritoneum of fish for a defined period of time and then recovered and analysed. This technique has already been employed successfully with *Renibacterium salmoninarum* and *Photobacterium damsela* subspecies *piscicida* antigens.

By Rachel Rangdale

Development of Vaccination Methods for the Control of BKD – SAL10

Project Leader: Dr Alexandra Adams

(Institute of Aquaculture, University of Stirling)

Sponsor: MAFF

BKD, caused by *Renibacterium salmoninarum* (R.s), is a chronic bacterial disease affecting both wild and cultured salmonid fish. Presently, there is no effective treatment for the condition. Partners from the Institute of Aquaculture, University of Stirling, University of Plymouth, the British Trout Association, the Scottish Salmon Growers Association and Aquaculture Vaccines Limited are developing recombinant and DNA vaccines for the control of BKD. The Plymouth group is preparing the vaccines, while the Stirling scientists are testing vaccine efficacy by challenging fish with BKD under standardised conditions in the aquarium. Three recombinant proteins, identified as possible bacterial virulence factors, have been prepared and the efficacy and immune response of two of the potential vaccines is presently being determined. Preliminary data on the immune response indicates that there appears to be a high antibody response to both of these, and it is anticipated that field-testing will commence next year.

Several methods, including antibody based tests (IFAT and ELISA) and molecular tests (PCR), have been developed and optimised for the detection of R.s. in fresh tissue. Immunohistochemistry is also utilised to detect the pathogen in fixed tissue following vaccination and challenge, while an *in situ* hybridisation test to detect pathogen DNA in fixed tissue is presently being developed. Such specific, sensitive tests are useful to establish the presence and location of the pathogen following challenge and to assist in efficacy evaluation.

Studies to identify candidate vaccine antigens, in addition to the three already prepared, are also presently being performed. The Plymouth scientists are preparing fusion proteins from novel recombinant antigens while the Stirling group is investigating the expression of R.s. proteins in bacteria cultured in dialysis bags implanted into fish, and in a variety of cell lines in the laboratory.

By Dr Alexandra Adams

RESEARCH NEWS

The following abstracts of recent research work are taken from papers published in international scientific journals and aquaculture magazines:

1. Induced and synchronised spawning of broodstocks

In a large population of salmon, maturation occurs at different rates and fish spawn at different times. In some cases, spawning within a group of fish may occur over several months. In these fish, it would be a distinct advantage to synchronise and compress the spawning season. In other circumstances, it is desirable to have some fish in a population mature ahead of others. The ability to advance maturation provides greater flexibility in the hatchery and with seawater entry dates of smolts. Also early gametes permit the out-crossing of different strains. With regard to alternate species, several obstacles to research and development could be overcome if fish spawned in a predictable fashion. Under ESC (Health Canada) approval methods have been developed to advance and synchronise maturation in captive broodstock. In controlled studies and trials conducted in British Columbia, New Brunswick and Chile, coho salmon (*Oncorhynchus kisutch*), chinook salmon (*O. tshawytscha*) and Atlantic salmon (*Salmo salar*), rainbow trout (*O. mykiss*) and sablefish (*Anoplopoma fimbria*) were induced to mature using peptide implants. In treated coho salmon, the spawning season was shortened, milt quality was increased and fry reached first feeding earlier. In several trials, this method has proven both effective and safe for humans and fish. The implications of this technology for producers are profound in the development of a broodstock management programme.

Reference

POWELL, J.F., BRACKETT, J. AND BATTAGLIA, J.A., 1998. Induced and synchronised spawning of captive broodstock using Ovaplant and Ovaprim. Bulletin of the Aquaculture Association of Canada, 98(3): 14-18.

2. Factors affecting pigmentation in trout

A number of factors limit the amount of astaxanthin retained in the muscle of trout apart from its concentration in the feed. These include the source of carotenoids, the efficiency of absorption from the gut and the concentration level in the blood. Also because carotenoids are fat soluble compounds the effect of dietary lipid levels on absorption efficiency may also be important. The aim of this study was to examine the

concentration of astaxanthin in the blood of rainbow trout fed diets supplemented with two sources of astaxanthin: that derived from a freshwater unicellular green alga *Haematococcus pluvialis* which is known to accumulate large amounts of the carotenoid astaxanthin and synthetic astaxanthin (CAROPHYLL Pink). These were added at two different dietary lipid levels of 9 and 24%. The relative serum concentrations of astaxanthin were taken as an indication of differences in pigment absorption.

Rainbow trout with an initial mean body weight of 150 g were fed experimental diets supplemented at a rate of 100 mg pigment/kg diet combined with the two different lipid levels (9 and 24%) during 5 days. Astaxanthin concentration in the serum ranged from 5.3 µg/ml to 9.0 µg/ml. Astaxanthin concentration in the serum was higher for fish fed high lipid level diets, independently of the astaxanthin source. No differences in the astaxanthin serum concentration were found for fish fed diets supplemented with either natural or synthetic astaxanthin, respectively 9.0 ±1.9 and 8.4 ±2.4 µg astaxanthin/ml serum, when dietary lipid level was high (24%). On the other hand, there was a higher blood astaxanthin concentration in fish fed diets supplemented with algal biomass (7.0 ±2.4 µg astaxanthin/ml serum) compared to synthetic astaxanthin (5.3 ±2.0 µg astaxanthin/ml serum) when dietary lipid level was low (9%).

Reference

BARBOSA, M.J., MORAIS, R. AND CHOUBERT, G., 1999. Effect of carotenoid source and dietary lipid content on blood astaxanthin concentration in rainbow trout (*Oncorhynchus mykiss*). Aquaculture, 176(3/4): 331-341.

3. Effect of vaccination on growth of trout

This study looked at the effect of single intraperitoneal injection of a commercial vaccine against furunculosis on the growth performance and feed conversion efficiency (FCE) of rainbow trout (n = 490; 90 ±10 g) over a 7 week period. The effect of single vaccine components on growth were also studied to enable a comparison to be made between two different adjuvants. No mortalities were recorded throughout the trial period. Growth performance was significantly impaired by

vaccination with respect to overall specific growth rate in length and weight. Over the duration of the trial, weight loss in vaccinated fish was 8%. It was also determined that the effect of the vaccine upon growth was caused by the inactivated *Aeromonas salmonicida* cell component of the vaccine rather than the adjuvant and/or interactions between the two. No differences were seen in FCE between treatments, although vaccinated fish expressed loss of appetite but normal feeding behaviour was regained by week 4 of the trial. Producers should therefore be especially conscientious of the feeding strategy employed immediately post-vaccination and for a period of up to 4 weeks thereafter if food waste is to be minimised.

Reference

RØNSHOLDT, B. AND McLEAN, E., 1999. The effect of vaccination and vaccine components upon short-term grow and feed conversion efficiency in rainbow trout. *Aquaculture*, 174(3 and 4): 213-221.

4. Genetic variation in commercial size diploid and triploid rainbow and brown trout

This study analysed the genetic variation in growth, carcass yields, morphometric and morphological traits in 16 half-sib diploid and triploid families (from different sires) in rainbow trout and brown trout of commercial size. In France commercial size is defined as 900 g for rainbow trout in freshwater corresponding to 'fillet portion size' and 3 kg for brown trout reared in seawater, corresponding to 'large sized trout'.

At 1 year of age (300 g) for rainbow trout, and 1.5 years (320 g) for brown trout (3 months after transfer to seawater), 20 to 25 fish per family were individually tagged. Individual growth performance was followed for both species until they had attained the commercial size given above. In rainbow trout, a significant genetic variability was observed among families for initial body weight, length and condition factor. Mean daily growth rates for body weight ranged from 0.70% to 0.80% between families but this was not statistically significant. No significant genetic variability in final body weight (900 g) and length was observed. This was associated with an increased variability within families for these parameters and with the expression of a different genetic variability also suggested by a change in ranking between families. In brown trout significant genetic variation in body weight, length and condition factor was observed among families at all stages up to reaching commercial size. After transfer to seawater, a change in ranking of families for body weight was observed. The results obtained suggest that growth performance studies should be performed at a body weight close to commercial size. For 'fillet portion sized' rainbow trout and large sized brown trout. At all stages, body weight, length and condition factor were significantly higher in diploid rainbow trout than in triploids. This was also

observed in brown trout: diploids showed a significantly greater body weight than triploids, but no significant differences were observed in final length at 3 kg. In both species, significant interactions between ploidy and family factors were observed for both body weight and length and condition factor at the initial stage studied but not at slaughtering. This result suggests that selection of diploid male broodstock for growth performance would not be effective for their triploid progeny at 'pan-size' stages but effective at the larger size.

Reference

BONNET, S., HAFFRAY, P., BLANC, J.M., VALLÉE, F., VAUCHEZ, C., FAURÉ, A. AND FAUCONNEAU, B., 1999. Genetic variation in growth parameters until commercial size in diploid and triploid freshwater rainbow trout (*Oncorhynchus mykiss*) and seawater brown trout (*Salmo trutta*). *Aquaculture*, 173(1-4):359-375.

5. New antiviral DNA vaccine against IHNV

A new vaccine has been developed by government laboratories in the US and Canada in association with Clear Springs Foods, the World's largest producer of trout, which provides almost total protection against infectious haematopoietic necrosis virus (IHNV). Unlike most vaccines which contain material that triggers an immune response directly, the new vaccine is DNA based and contains the gene for one of the viral proteins. When taken up by the cells of the fish viral proteins are produced which prime the immune system to attack the virus directly. Both trout fry and salmon smolt injected with as little as a millionth of a gram of vaccine, resulted in no fish deaths when exposed to the virus as against 90% mortality in untreated control fish. It is considered doubtful if the vaccine would be acceptable in Europe given the present bad press over genetically modified organisms even though there is no evidence that the introduced DNA is permanently incorporated into fish chromosomes. Another viral DNA vaccine is currently being developed at a Danish laboratory against viral haemorrhagic septicaemia (VHS) the most important viral disease of trout in Europe.

Reference

THWAITES, T., 1999. A fine kettle of fish. Cutting-edge vaccines for salmon and trout threaten to trigger a new food scare. *New Scientist*, No. 2201 (August 28 issue): page 4.

6. Effect of ration level on growth and swimming performance

A complex relationship exists in fish between feeding, growth and other elements of fitness and a trade-off between growth rate and swimming capacity, for

example, has been reported for several species. Under certain conditions individual fish may maximise either growth rate or swimming performance, but not both. This study evaluated the roles of body size, morphology, haematocrit and feeding in the development of this trade-off and to determine the range of rations over which it occurs.

Aerobic swimming performance, food consumption per meal (by X-ray radiography), specific growth rate, haematocrit and fin condition were monitored in individual juvenile rainbow trout held in groups over a 9 week period and fed daily group rations (dry food) of 2.0, 1.5, 1.0 and 0.5% body weight per day. Specific growth rate declined and competition increased at lower ration levels as reflected by greater fin damage, reduced haematocrits and condition factors and at 0.5% ration level a substantial increase in the coefficient of variation was seen among individual meals. Aerobic swimming performance also declined at lower ration levels. However there was no relationship between swimming performance and haematocrit, fin damage, or condition factor in any of the ration groups. A negative correlation was found between swimming performance and specific growth rate in individual trout fed a group ration of 2.0% body weight per day but a positive correlation was seen at 0.5% body weight per day and no relationship at 1.0 and 1.5% body weight per day. There was a positive relationship between individual specific growth rate and food consumption only among fish fed a ration of 2.0% body weight per day. A significant negative relationship between swimming performance and individual food consumption was also found among fish in this ration group. These results indicate there is a trade-off between aerobic swimming performance and growth rate that occurs only over a narrow but biologically relevant range of food intakes. This trade-off appears to result from costs associated with feeding and digestion.

Reference

GREGORY, T.R. AND WOOD, C.M., 1999. Interactions between individual feeding behaviour, growth and swimming performance in juvenile rainbow trout (*Oncorhynchus mykiss*) fed different rations. *Canadian Journal of Fisheries and Aquatic Science*, 56(3): 479-486.

7. Assessment of fish waste as a fertiliser

The waste from land based trout farms consists primarily of a soluble fraction from metabolic processes and solids in the form of faeces and uneaten food. This paper focuses on the chemical composition of settleable faecal fish waste from samples collected at 12 commercial rainbow trout farms and compares it with other livestock waste in assessing its suitability as an agricultural fertiliser. The samples from commercial farms averaged 2.83% nitrogen, 2.54% phosphorus,

0.10% potassium, 6.99% calcium and 0.53% magnesium on a dry-weight basis. The mean concentration of various metals was also analysed. These ranged from 0.05 mg/kg for mercury to 1,942 mg/kg for iron. Fresh fish manure has similar levels of nitrogen, phosphorus, calcium and magnesium and lower levels of potassium when compared to manure from beef, dairy cattle, poultry and swine. Fish manure tended to have a higher content of manganese, cadmium, chromium, lead, iron and zinc than most other livestock manures, but had lower levels of arsenic, selenium, cobalt and nickel. The copper content of fish manure was similar to all other livestock manures.

The results from this study indicate that fresh fish manure is essentially similar in its chemical composition to other livestock manures, and should be suitable for use as an agricultural fertiliser. The data presented should help regulatory agencies and farmers to make sensible and pragmatic decisions concerning the appropriate and safe disposal of manure wastes collected from land-based trout farms.

Reference

NAYLOR, S.J., MOCCIA, R.D. AND DURRANT, G.M., 1999. The chemical composition of settleable solid fish waste (manure) from commercial rainbow trout farms in Ontario, Canada. *North American Journal of Aquaculture*, 61(1): 21-26.

8. Demand feeding of rainbow trout

The effects of temperature, dietary energy content and reward level (amount of food received in response to one trigger actuation of the feeder) on the demand feeding activity of rainbow trout was investigated. Fish of initial size 30-120 g were fed in tanks using demand feeders over a period of 60 days. The main parameters recorded were bite activity (number of trigger actuations), growth rate and feed conversion. Bite activity was significantly higher in trout reared at 15°C than those reared at 5°C; consequently, food supply and growth rate were higher in the former group. However, as evaluated by the growth and feed conversion rates, only fish raised at 5°C showed a bite activity that was close to the optimal, whereas the bite activity was too low at 15°C. Groups of fish fed diets with either a low, medium or high gross energy content did not show any significant difference in bite activity or growth rate. This indicates that trout under aquaculture conditions are unable to adjust the bite activity in a way that matches the energy content of the food. The reward level significantly influenced the ability of the trout to meet their demand for food by regulating its supply. Among groups offered either low (0.36 g), medium-sized (0.80 g) or high (1.65 g) reward, the high-reward groups exhibited the highest growth rates. The trout were, to some extent, able to compensate for a low reward by increasing their bite activity. However, at low or medium-sized rewards, fish were unable to

compensate to the degree necessary to meet their food demand.

The author concluded that the best and easiest way to manage a demand feeding system, so that growth and feeding conversion rates are optimised, is to adjust the size of the reward. Taking the water temperature into account, the following recommendations about optimal reward level for rainbow trout are suggested. At 15°C, the reward should be 0.1 g/kg fish per trigger actuation, when reared in tanks of the size of 1 m³ and with a fish stock of 100-200 individuals. For trout reared at 15°C, the size of the reward should increase to 0.2 g/kg fish per trigger actuation. In addition, it may also be necessary to adjust the reward level to match the energy content of the food. Generally, the size of the reward should decrease with increasing energy content.

Reference

ALÄNARÄ, A. 1994. The effect of temperature, dietary energy content and reward level on the demand feeding activity of rainbow trout (*Oncorhynchus mykiss*). *Aquaculture*, 126 (3 and 4): 349-359.

9. Release of alarm substances in parasitised trout

An investigation was made to see if juvenile rainbow trout can detect and respond to substances released by another fish of the same species (conspecific) whilst being infected by cercariae of the trematode parasite (*Diplostomum sp*). It was found that trout did not react to odours of the cercariae alone, indicating they were incapable of detecting the parasites directly. However, when exposed to odours from a conspecific being infected they increased the number of random darts as well as the amount of time spent motionless. These results suggest that even the minor damage inflicted by penetration of the cercariae into the fish's skin was enough to cause the release of alarm substances. Assuming these reactions are adaptive there is some doubt as to how effective these responses would be at avoiding cercariae. In still water, remaining motionless near an infected conspecific may increase the likelihood of contracting parasites, whereas darting away could take the fish to a cercaria-free area. However, in running water, neither response is likely to have an effect on the probability of infection since cercariae are being swept downstream continuously. Further experiments are therefore required to determine if these reactions are of benefit to fish, or a coincidental side-effect.

Reference

POULIN, R., MARCOGLIESE, D.J. AND McLAUGHLIN, J.D. 1999. Skin-penetrating parasites and the release of alarm substances in juvenile rainbow trout. *Journal of Fish Biology*, 55(1): 47-53.

10. Improved growth in all-female triploid trout

In spite of the potential for all-female diploid and triploid stocks in rainbow trout farming there is a lack of published information on their growth and performance in relation to mixed-sex diploid stocks. This paper evaluated growth and production characteristics, in an indoor raceway system, of replicated populations of all-female diploid, all-female triploid (87% triploid) and mixed-sex diploid rainbow trout over a trial period lasting 265 days from an initial size of about 100g through to market size (final weights 568g, 749g and 521g respectively). Growth rates were found to be linear for all groups and were highest in the all-female triploids (2.4g/day), lowest for the mixed-sex diploids (1.6g/day) and intermediate for the all-female diploids (1.8g/day). No significant differences were found among treatments for survival, food, conversion ratio, condition factor, liveweight index, visceral fat and dress-out weight. By day 180 however, the majority of the males in the mixed-sex groups were sexually mature, whereas the females in this group and in the all-female groups were still maturing. Mean gonadosomatic indices at day 180 were 3.1 for the males, 1.1 for females in the mixed-sex groups, 1.9 for the all-female groups and 0.4 for the all-female triploids. It was concluded that monosex female diploid and triploid trout show promise for practical trout farming due to their faster growth and the elimination of males, which mature and lose flesh quality earlier. Farmers should consider all-female triploid production, especially for markets using larger trout.

Reference

SHEEHAN, R.J., SHASTEEN, S.C. AND SURESH, A.V. 1999. Better growth in all-female diploid and triploid rainbow trout. *Transactions of the American Fisheries Society*, 128(3): 491-498.

11. Selection for broodstock development in Arctic char

Arctic char (*Salvelinus alpinus*) is regarded as an excellent candidate for aquaculture. This species grows well at low temperatures and high stocking densities, and secures a higher price than other commercially grown salmonids. However, the size variability exhibited by this species inhibits large scale production. Studies have shown that some individuals may never grow to harvest size and these 'runts' may be genetically programmed for slow growth. Microsatellites are areas of repetitive DNA with repeat units 1 to 4 base pairs in length and an average length of 20 to 300 pairs. Microsatellites may be linked to a genetic locus affecting growth and if an allele is found to be positively correlated with growth it is probably linked to such a gene. Therefore by selecting this allele there is an indirect selection for increased growth. This study was

carried out to test primers designed to amplify microsatellites in other salmonid species to see if they also amplify a homologous site in Arctic char and to determine if there is a correlation between certain alleles and growth rate. Char from the Fraser River strain were grown under identical conditions for 14 months and then weighed when a 10-fold difference in weight between the largest and smallest was found. DNA was then extracted from the largest 5% of the fish and the smallest 5%. This was screened using 74 primer pairs designed for amplification of microsatellite loci in other salmonid species to determine if there is a microsatellite locus which is related to growth. Of these primers, 33 resulted in a monomorphic product, 10 resulted in a polymorphic product, 14 resulted in no product, and 17 have still to be analysed. Of the 10 variable loci, a significant difference between small and large fish was detected at four of the microsatellite loci. If these differences prove to be real and not simply artefacts the potential broodstock will be screened at these loci and crosses set up using parents with known genotypes.

Reference

JOHANSEN, A.D., WILTON, G. AND DAVIDSON, W.S., 1998. Marker assisted selection for Arctic char (*Salvelinus alpinus*) broodstock development. Bulletin of the Aquaculture Association of Canada, 98(2): 30-31.

12. Evaluation of a new fish anaesthetic/sedative

There is a requirement for a fish anaesthetic/sedative in the US for field studies (population estimates, age and growth studies) and in stocking programmes for mild sedation of fish during transport in order to decrease stress related mortalities. AQUI-S™ is a fish anaesthetic/sedative approved in New Zealand and Australia with no withdrawal time. The product contains two ingredients: a synthetic flavouring substance and an adjuvant that may be safely used in food. This paper made a preliminary evaluation of the efficacy and minimum toxic concentration of AQUI-S™ in two size classes of fish - 1+ year old and fry and fingerlings less than 1 year of age in six species of fish important to the US aquaculture industry and including rainbow trout. Efficacy and minimum toxic concentrations was made in 1+ rainbow trout acclimated to water at 7, 12 and 17°C. Most of the tested 1+ fish species were anaesthetised in 3 minutes or less at a nominal concentration of 20 mg L. The minimum toxic concentration was at least 2.5 times the selected efficacious concentration for anaesthesia. In juvenile fish Bluegill, channel catfish and rainbow trout were anaesthetised in less than 4.1 minutes at a nominal concentration of 20 mg L AQUI-S™ while yellow perch and walleye required a concentration of 35 and 25 mg L for similar times of anaesthesia. Recovery times were less than 7.3 minutes for all species in both size classes. In fry and fingerlings, the minimum toxic concentration was at least 1.4 times that for effective anaesthesia.

There appeared to be little relationship between size of fish and concentrations or times to induction, recovery times and minimum toxic concentration. The times required for induction and for recovery were increased in rainbow trout as the acclimation temperature was reduced.

Reference

STEHLY, G.R. AND GINGERICH, W.H., 1999. Evaluation of AQUI-S™ (efficacy and minimum toxic concentration) as a fish anaesthetic/sedative for public aquaculture in the United States. Aquaculture Research, 30(5): 365-372.

13. Response of resident trout to stocking hatchery-reared brown trout

This study was designed to evaluate the growth, survival and movement of hatchery-reared adult brown trout over short time periods after stocking in a small Austrian stream and to determine if these hatchery fish caused changes in the resident population of brown and rainbow trout. Two strains of hatchery-reared adult brown trout (208-334 mm total length n = 591) were individually marked and released into a limestone stream. The estimated survival after one month (86% n = 508) was comparable to that for resident brown trout and rainbow trout (89% n = 771) but declined to 14% (n = 83) after 8 months compared with 52% (n = 451) for resident trout. The movement of resident trout out of stocked stretches was higher (14%) than from control sites (5%) but the population size in both individual sites and the overall study area was unaffected. The growth of resident brown trout was unaffected by stocking, but rainbow trout showed lower growth rates in stocked versus unstocked stretches both one and 8 months after stocking.

Reference

WEISS, S. AND SCHMUTZ, S., 1999. Response of resident brown trout, *Salmo trutta* L., and rainbow trout, *Oncorhynchus mykiss* (Walbaum), to the stocking of hatchery-reared brown trout. Fisheries Management and Ecology, 6(5): 365-375.

14. Quality characteristics of Danish processed trout

The greater awareness and higher expectations towards product quality of various foods has created a need for establishing similar quality standards in the production and processing of farmed fish. Quality studies to date have mainly been concerned with the affects of different diets on fish without focusing on the quality parameters relative importance. In addition most quality systems have considered Atlantic salmon and are not appropriate for rainbow trout. This study attempts to identify quality characteristics of pan-sized rainbow trout considered

important by Danish processors which might provide fish farmers with practical guidelines for producing a higher quality and potentially more valuable product. The findings showed significant variation with respect to management perception of raw material quality. This prevented establishment of specific guidelines on quality and husbandry methods for trout farmers to achieve product standardisation for the processing industry. Gross morphology was found to be the single most important factor when determining quality of freshly slaughtered trout while skin appearance was recorded as having least impact. Because of the complexity of establishing an effective management system to determine quality control the entire product chain must ultimately be incorporated since retailers have become specification-driven and consumers more quality demanding.

Reference

RØNSHOLDT, B. AND MCLEAN, E., 1999. Quality characteristics of fresh rainbow trout as perceived by the Danish processing industry. *Aquaculture International*, 7(2): 117-127.

15. Behaviour of hatchery-reared and wild brown trout

The aim of this study was to examine if established wild brown trout in a semi-natural stream environment demonstrate a prior-resident competitive advantage over later introductions of both hatchery-reared and wild brown trout and also to determine if hatchery-reared brown trout exhibit relatively higher levels of aggression than simultaneously stocked wild origin fish. It was found that established wild brown trout initiated 44% of the mean aggressive acts whilst hatchery-reared trout initiated 34% and introduced wild trout initiated 22%. There was no significant difference in the number of aggressive acts received by each of the three fish groups. Established wild fish maintained home stations closer to a point source of feed than did both hatchery-reared and introduced wild conspecifics. Established wild fish were the only group to show a positive mean specific growth rate during the trials. Introduced wild fish showed a slightly negative mean specific growth rate, whilst introduced hatchery-reared fish showed a considerable negative mean specific growth rate. These results suggest that established wild brown trout in a semi-natural stream environment display a prior-resident effect over later introductions of hatchery-reared and wild conspecifics. Introduced hatchery-reared fish were more aggressive and exhibited a lower mean specific growth rate than simultaneously stocked wild fish, suggesting that excessive expenditure of energy for unnecessary aggression may contribute to the poor survival of hatchery-reared fish after they are stocked into streams.

Reference

DEVERILL, J.I., ADAMS, C.E. AND BEAN, C.W., 1999. Prior residence, aggression and territory acquisition in

hatchery-reared and wild brown trout. *Journal of Fish Biology*, 55(4): 868-875.

16. Effect of rearing density on growth and stress response in rainbow trout

Several studies have shown that salmonids can be reared at extremely high densities under certain conditions with no adverse impact on growth and survival enabling farmers to increase production. This study evaluated the combined effects of high rearing densities (defined as fish weight per volume of water, g/L) and loading rates (defined as fish weight per unit of water flow, kg/[L.min]) on the growth, food conversion (weight of food fed/weight gained) and stress responses of juvenile rainbow trout. The time course over which fish acclimated to elevated densities and loading rates was also determined. Growth and changes in serum concentrations of cortisol, glucose, and chloride in response to acute stress challenge tests were compared in fish of total length 143-190mm reared at density indices of 2.8-9.3 g/L and loading rates of 0.5 and 0.75 kg/(L.min). A first experiment showed no differences in growth or measures of physiological stress in fish reared for 4 weeks at loading rates of 0.5 or 0.75 kg/(L.min). Fish reared at density indices of 5.6 and 8.4 g/(L.cm) gained significantly less weight than fish at a density index of 2.8 g/(L.cm), and fish reared at 2.8 and 5.6 g/(L.cm) had better food conversion rates than those at 8.4 g/(L.cm). Changes in the serum concentrations of cortisol, glucose and chloride, however, suggested that the reduced growth at elevated densities was not due to physiological stress. In a second experiment cortisol levels were higher in fish 3 days after transfer to net-pens than in fish 10 or 17 days after transfer. There were no differences in the cortisol levels of fish reared at density indices of 3.1, 6.2 and 9.3 g/(L.cm), suggesting that acclimation to the net-pen conditions was not density dependent. Elevations in serum glucose and reductions in serum chloride in fish reared at 3.1 g/(L.cm), however, suggested that fish at low density were stressed, possibly a result of stressful social interactions at low rearing densities. These findings indicate that, at relatively high loading rates, rainbow trout reared at high densities exhibit decreased growth and food conversion rates. Additionally the study provides evidence that high rearing density itself is probably not a chronic stressor in rainbow trout, because fish reared at high densities did not experience changes in physiological measures that would normally be expected under stressful conditions.

Reference

PROCARIONE, L.S. BARRY, T.P. AND MALISON, J.A., 1999. Effects of high rearing densities and loading rates on the growth and stress responses of juvenile rainbow trout. *North American Journal of Aquaculture*, 61(2): 91-96.

17. Effects of electrofishing on movements of brown trout

Much of the knowledge on salmonid ecology is based on electrofishing. Because there is little information on how electrofishing affects the subsequent behaviour of fish, some of the salmonid data may be unreliable. Estimates of population sizes of stream fish based on mark-recapture or removal rely to a large extent on electrofishing as well. The population estimates obtained can be affected by high rates of movement suggested among stream-dwelling fish, regardless of whether the movements are natural or sampling induced. The amount of movement before and after electrofishing a closed population of brown trout was determined by trapping in a small stream during 1994-1996. To assess the influence of movements on mark-recapture estimates and evaluate obtained estimates in relation to removal estimates, four different estimates (removal in summer, removal in autumn, conventional mark-recapture, and mark-recapture considering fish movements) were made for the closed brown trout population. The number of brown trout caught in upstream traps increased following electrofishing. For these fish, mean size was larger and recapture rates were lower compared with the general population in the stream. In all years the conventional mark-recapture method resulted in the highest population estimates and differed from the mark-recapture estimates in which fish movements were accounted for. Mark-recapture estimates accounting for movements did not differ from autumn removal estimates. The results emphasise the importance of minimising disturbances in connection with studies of the population dynamics of stream fish.

Reference

NORDWALL, F., 1999. Movements of brown trout in a small stream: effects of electrofishing and consequences for population estimates. *North American Journal of Fisheries Management*, 19: 462-469.

18. Clove oil as a fish anaesthetic

Many chemicals traditionally used by the aquaculture industry are now being scrutinised both for their cost effectiveness and safety. At present the only fish anaesthetic approved by the U.S. Food and Drug Administration (FDA) for aquaculture used in the United States is tricaine methaine sulfonate (MS - 222) and carbon dioxide both of which have disadvantages in use. This paper examined the safety and efficacy of clove oil as an anaesthetic for juvenile and adult chinook and coho salmon, juvenile rainbow trout and white sturgeon. The median lethal concentrations for a 10 minute exposure were 62 mg/L for chinook, 96 mg/L for coho, 250 mg/L for rainbow trout and 526 mg/L for white sturgeon. A dosage of 25 mg/L was effective in anaesthetising all species for 120 minutes without mortality. The authors concluded that clove oil at this concentration provides rapid immobility, rapid recovery, without toxic effects on

the species studied and was less expensive than existing compounds. The cost to treat 10 L of water amounts to \$0.003 as against \$0.12 for MS-222. Clove oil is already cleared for specific human uses and should therefore pose few mammalian safety problems although the question of tissue residues would need to be addressed.

Reference

TAYLOR, P.W. AND ROBERTS, S.D., 1999. Clove oil: an alternative anaesthetic for aquaculture. *North American Journal of Aquaculture*, 61(2): 150-155.

19. Genetic consequences of fertilisation procedure

In salmonid hatcheries the simultaneous or sequential addition of semen from a number of males to batches of eggs prior to addition of water is a common practice during fertilisation procedures. This study used biochemical genetic variation to determine the proportion of progeny sired by individual male chinook salmon (*Oncorhynchus tshawytscha*) in crosses derived from multiple male parents. For crosses conducted with mixtures of semen, an equal volume of semen from each of three males was pooled and used to fertilise the eggs of an individual female immediately after pooling, and after holding the pooled semen for 15 and 60 minutes before use. Male potency varied in these crosses, with individual males siring between 5 and 88% of the progeny of a cross. Potency differences among males was reduced with the use of pooled semen that had been held for 60 minutes prior to fertilisation. For crosses derived from the sequential addition of semen, samples from three males was added at 45 second intervals to the eggs of a single female. In these crosses, individual males sired between 0 and 94% of the progeny of a single cross. The position occupied by a male in the fertilisation sequence was a significant factor in determining the proportion of progeny that he sired, but differences in male potency were also observed. It was concluded that to maintain genetic variation the addition of semen from more than one male to egg batches should be avoided in hatchery populations. Instead the addition of semen from single males to egg batches pooled from several females should be used to maintain genetic variation within hatcheries. One possibility is to pool eggs from individual females and then subdivide them to form replicate egg batches. Fertilisation of each batch with semen from a different male would prevent loss of all gametes from a single female in the case of an infertile male, and would provide some degree of factorial mating without a requirement for semen pooling.

Reference

WITHLER, R.E. AND BEACHAM, T.D., 1994. Genetic consequences of the simultaneous or sequential addition of semen from multiple males during hatchery spawning of chinook salmon (*Oncorhynchus tshawytscha*). *Aquaculture*, 126(1/2): 11-23.

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