International Council for
the Exploration of the Sea /
ICES

C.M. 1990 / F:15
Mariculture
Committee

Report
of the Working Group on Genetics, 1990
(by correspondence)

* General Secretary
ICES, Palaegade 2-4
DK-1261 Copenhagen K
Denmark

Table of Contents

(1) Introductory Remarks

According to the adopted resolutions of the 77th Statutory Meeting, The Hague, Netherlands, 5-13 Oct. 1989, the ICES Working Group on Genetics should work in 1990 "..by correspondence and will meet in Tvärminne Station, Finland for 3 days in 1991" (see C. Res. 1989 / 2:38). See the members did.

During the course of updating the activities of 1990, the members of the Working Group provided the chairman with additional informations, including reprints and references on recent scientific material pertinent to the Working Group's tasks. These reprints are cited under pos. 7 / pp. 31 ff. of this paper. They can be obtained from the chairman on request (one set is added to the original copy c/o. General Secretary of ICES). The recommendation is offered to follow conceptionally the approach of the cited contributions drawn from the Bulletin of the American Fisheries Society, Vol. 15 (1) pp. 2 ff.

(2) Reports on Genetics from the Member Countries (mainly on forms, presented in alphabetic order of the member countries)

- Canada (compiled by Richard L. Saunders, Dept. of Fisheries and Oceans, Aquaculture and Invertebrate Fisheries Division, Biological Station St. Andrews: annex 1a and a separate 2nd one by Dr. Jean-Marie Sévigny, Maurice-Lamontagne Institute, Mont-Joli, Quebec: annex 1b).
- Federal Republic of Germany (by Prof. Dr. W. Villwock, Zoologisches Institut und Zoologisches Museum, University of Hamburg: annex 2).
- France (by Prof. Dr. René Guyomard, Laboratoire de génétique des poissons. CRJ-INRAJouy-en Josas: annex 3)
- Norway (twice, one by: Dr. Knut E. Jørstad, Institute of Marine Research, Nordnes / Bergen: annex 4a, 2nd one by: Prof. Dr. Gunnar Naevdal, Institute of Fisheries and Marine Biology, University of Bergen: annex 4b).
- Poland (by Dr. Krzysztof Goryczko, Inland Fisheries Institute, Salmonid Research Laboratory Rutki, Zukowo: annex 5)
- Portugal (by Dr. Ana Maria Teia Dos Santos, National Institute, Fisheries Research, C.I.P.A., Aveiro: annex 6)
- Sweden (by Prof. Dr. Lennart Nyman, Institute of Freshwater Research, Dottingholm: annex 7)
- United Kingdom & Wales (by Dr. David Thompson, MAFF, Dir. Fish. Research, Fisheries Laboratory, Lowestoft: annex 8a).
- United Kingdom, Scotland (by Dr. Alan Youngson, DAFS Marine Laboratory, Torry, Aberdeen: annex 8b)

(3) Brief summary of reports listed above.

The main objects for research remained salmonids, especially the atlantic salmon *(Salmo salar)* and different trout species (see European reports), while Canadian colleagues are also referring to native salmonid species of their New World region (annex 1a). Besides salmonids other species of marine fish families are increasinly investigated (e.g. flatfishes annex 1b, *Sebastes, Gadus, Brosmius, Molva,* annex 4b). The colleagua of Portugal describes investigations on *Merluccius merluccius* and on *Nephrops norvegicus* (annex 6). Dover Sole and Manila Clams are the species of main interest to which colleagues of the United Kingdom reported (annex 8a). Investigations on *Tilapia* species are to be added (annex 2 and 3).

Apart of growthrate and sex control investigations (most European contributions and 1a from Canada) by different means of classic investigation methods or along of somekind of genetic engineering, respectively (e.g. included triploidy, gynogenetic diploids) increasingly mtDNA- and related molecular studies are carried out by specialized working group of different member countries (e.g. annexes 2, 3, 7, 8a). Others are dealing with genetic markers for different purposes, e.g. population discrimination (annexes 2, 3 a.O.) and statements on migrating behavior (Canada, Scandinavian countries). Genetic fingerprinting is tried to introduce for solving special questions (see preliminary Study Group report, pos. 6, pp. 27-31).

All these different activities will be continued for at least the next 12 month, the latter ones will be in the center of interest at the Tvärminne meeting, 1991.

(4) Recommendations

The Mariculture Commitee recommends that the Working Group on Genetics (Chairman: Prof. Dr. Wolfgang Villwock) will meet at Tvärminne-Station, Finland, in the second half of July 1991 for three days, to

a) review report on, and plan research on biochemical (enzyme) markers and other related techniques;

b) evaluate the present trend in "advanced gene technology" in fisheries science and / or aquaculture. Two modern methods need to be adressed in particular: 1) DNA-fingerprinting for specimen discriminations; 2) production of transgenic specimens;

c) describe the risks involved in uncontrolled releases of genetically changed organisms and assess methods to reduce these risks with the aim of assiting the Working Group on Introductions and Transfers of Marine Organisms in its revision of the Code of Practice.

Note: These recommendations were already adopted by the last year Statuatory Meeting, The Hague, M. Res. 1989 / 2:38.

With respect to pos. c) both Working Groups mentioned in this paragraph intend to meet together at Tvärminne Station Finland, 1991.

A N N E X E S

International Council for the
Exploration of the Sea

Working Paper:
ICES Working Group
on Genetics

CANADIAN GENETICS STUDIES WITH PARTICULAR REFERENCE TO BIOCHEMICAL MARKERS AND GENETICALLY CHANGED ORGANISMS

Compiled by

Richard L. Saunders
Department of Fisheries and Oceans
Aquaculture and Applied Physiology Division
Biological Station
St. Andrews, N. B. E0G 2X0
Canada

Abstract

This document incorporates material solicited from individuals or groups in Canada conducting studies in genetics with particular reference to biochemical markers, advanced gene technology in fishery science and/or aquaculture, and risks involved in releases of genetically changed organisms.

Ministry of Natural Resources (MNR), Stock Assessment and Genetics Unit, Research Section, Fisheries Branch, Maple, Ontario

Peter E. Ihssen

a) Continuing work on genetic differentiation of Ontario game and forage fish. This year, they will complete work on walleye. Part of the walleye work involved the examination of pond-reared walleye. Will document the effect that small numbers of parents have on the genetic variability of the pond-reared fish.

Have, for the first time, planted genetically (enzyme) marked lake trout embryos in three northern Ontario lakes. The objective is to assess the contribution made by 'marked' stocks of trout to both the fishery and the spawning stock. Concurrently, they will be able to compare the physiology, morphology and growth characteristics of fish planted as embryos to fish reared in hatcheries. Again, this is possible because the fish are genetically marked.

Work is continuing on the genetic differentiation of alewife, yellow perch and brook trout. Have started a project to determine the genetic contribution that hatchery fish have made to existing populations in Algonquin Park.

b) MNR has provided funds to support the development of DNA fingerprinting methods for identifying lake trout. Dr. M. Gross of the University of Toronto is working on this project. Early results are promising. MNR is also funding a project examining mitochondrial DNA in brook trout. The objective is to quantify the amount of genetic differentiation among brook trout stocks in the province and to relate this differentiation to allozyme differences and differences in growth rate. Dr. P. Hebert and R. Danzman at the University of Guelph are working on this project.

Department of Fisheries and Oceans, Freshwater Institute, Winnipeg, Manitoba

Michael A. Giles

Has observed distinct differences in blood-oxygen affinity and metabolic rate in the three strains of Arctic charr (originating from Norway, Labrador, and Nauyuk Lake, NWT) which are currently being developed as broodstock at the FWI Rockwood Experimental Fish Hatchery in Gunton, Manitoba. The differences appear to be related to the complement of hemoglobin isomorphs and, more importantly, to their relative concentration expressed in each strain. Strain crosses have demonstrated that both the hemoglobin phenotypes and the relative rate of expression of each isomorph are heritable. Since hemoglobin-oxygen binding characteristics have a significant influence on the fish's swimming ability and habitat (minimum tolerable oxygen tension and temperature extremes) requirements, he is attempting to develop groups of charr with a range of blood-oxygen equilibrium

characteristics and set up a series of physiological performance tests which would permit him to define the particular environments most suitable for each group. Hopefully, these results would optimize the efforts of rehabilitation and fish farming operations. From past experience with rehabilitation efforts, he has come to think that the physiological suitability of the planted fish to the characteristics of the particular receiving system may be a major determinant in their success or failure.

Department of Biology, University of New Brunswick, Fredericton, New Brunswick

Tillman J. Benfey

Suitability of induced triploidy to produce sterile salmonids for aquaculture.

Sterile fish can be used in aquaculture to meet two objectives: to prevent the deleterious effects that maturation has on marketability and survival of captive fish, and to prevent spawning in the wild should captive fish escape or intentionally be released. At present, induced triploidy appears to be the most effective way to sterilize fish for commercial aquaculture. However, only triploid females are of use for salmonid aquaculture because triploid males go through physiological maturation and produce functional (but aneuploid) sperm. In 1989, a research program was begun at the University of New Brunswick to evaluate the suitability of all-female triploids for the commercial aquaculture of salmonids in Atlantic Canada. This program has two aspects:

Pilot-scale aquaculture of all-female triploid Atlantic salmon - This work is being done in collaboration with the Salmon Genetics Research Program of the Atlantic Salmon Federation (St. Andrews, New Brunswick). Heat shock was used in the autumn of 1989 to induce triploidy in eggs fertilized with sperm from sex-reversed females (i.e., XX males). These fish will be evaluated for growth and survival through the entire production cycle. Other groups of fish were treated with 17 alpha-methyltestosterone in the spring of 1990 to provide a future source of homogametic (X chromosome-bearing) sperm.

Basic physiology of triploid salmonids - Aside from being sterile, triploids are unique in having larger but fewer cells in all their tissues and organs. Little is known about the effect of this on the basic physiology of triploids, or how it may influence their suitability for aquaculture. Studies of various aspects of the basic physiology of triploids are planned, initially with respect to their stress response and disease resistance.

Biological Sciences Branch, Department of Fisheries and Oceans, Pacific Biological Station, Nanaimo, British Columbia

Doug P. Swain and Brian E. Riddell

Genetic variation between hatchery and wild populations of coho salmon (*Oncorhynchus kisutch*)

Coho salmon have been cultured in British Columbia for approximately 20 years, but the length of culture history in stocks varies between hatcheries as the enhancement program has developed. Research investigates whether genetic differences have developed between hatchery and proximal wild populations, and what is the genetic basis of expression in traits frequently compared between hatchery and wild populations. Four experiments have been conducted to date: 1) comparison of agonistic behavior in newly emerged hatchery and wild juveniles, 2) a preliminary study of the genetic basis to agonistic behavior in a wild population, 3) morphological comparisons of several hatchery and wild populations under natural and controlled environments, and 4) comparison of the genetic correlation structure of morphometric and meristic traits in 2 hatchery and 2 wild populations.

Newly emerged hatchery juveniles were more aggressive than their wild counterparts when compared using mirror simulation tests. Additive variation for agonistic behavior, again measured in mirror simulation tests, was detected but was entirely due to 1 of 12 male parents. If this parent is omitted, then maternal or non-additive effects were the primary determinants of this trait. Analyses of the last 2 studies are ongoing. However, preliminary results indicate that the hatchery population with the longest culture history (approx. 5 generations) has greater additive genetic variation in morphometric and meristic traits than the wild populations. This result would indicate that selection has been relaxed in the hatchery population.

Chris J. Foote and Chris C. Wood

Sympatric divergence of the anadromous and non-anadromous forms of sockeye salmon, *Oncorhynchus nerka*

The anadromous (sockeye salmon) and non-anadromous (kokanee) forms of Oncorhynchus nerka spawn sympatrically yet appear to be genetically distinct in a number of rivers in British Columbia. A biochemical genetic comparison of 23 populations in British Columbia suggests that the two forms have diverged in sympatry, independently in different watersheds. Further comparisons of the early development, growth rate, swimming performance, seawater adaptability, and age at maturation of sockeye, kokanee, and their hybrids, reared under controlled hatchery conditions, confirm that the forms are genetically different. On average, sockeye genotypes promoted faster yolk absorption, faster and less variable growth (within families), increased seawater adaptability, and delayed maturation. In at least some

rivers, the two forms attempt to interbreed. Experiments with genetically marked fish in relatively natural stream enclosures indicate that kokanee males can fertilize sockeye eggs by "sneaking" in on spawning sockeye pairs. However, the resulting hybrid progeny are genetically different in ways that probably affect their fitness. Thus, it is argued that selection against hybrid progeny limits gene flow and promotes genetic differentiation of the sympatric forms.

T. J. Mulligan, L. Lapi, and G. P. Hudson

Mass marking of chinook and coho salmon

This project is studying the potential for mass marking of hatchery salmon by feeding them diets enriched in selected elements. These elements are retained in the calcified tissues so that the marked fish can be identified by chemical analysis of the tissue. Chemical analysis uses an inductively coupled plasma mass spectrometer. Currently treating with the elements strontium, barium, rubidium, and lanthanum. Scales of treated and control fish have been analyzed for mark elements. Elements have been detected for all treatments.

Ocean Sciences Center, Memorial University of Newfoundland, St. John's, Newfoundland

Garth Fletcher

Research at OSC with transgenic salmon is as follows:

1. Antifreeze genes. At present using genes from winter flounder (*Pseudopleuronectes americanus*), ocean pout (*Macrozoarces americanus*), and wolffish (*Anarhichas lupus*).

2. Growth hormone genes. Using a chinook salmon growth hormone gene linked to the promoter from an ocean pout antifreeze gene.

3. Prolactin gene. Expect to use the chinook salmon gene linked with an ocean pout promoter.

The genes are being constructed in Peter Davies' laboratory, Queen's University and Choy Hews' laboratory, University of Toronto. Gene transfers into Atlantic salmon and rainbow trout eggs are being carried out at the marine laboratory (OSC).

The policy with regards to transgenic fish for aquaculture is that the genes should be derived from fish. This is the reason he works with fish promoters.

At the present time, he believes that the best approach to prevent the release of transgenic fish into the wild is to render them sterile. This way, only the broodstock need to be held under strict security. This could be done in a land-based facility.

Atlantic Salmon Federation, Salmon Genetics Research Program (SGRP), St. Andrews, New Brunswick

G. W. Friars and J. K. Bailey

Research in the SGRP pertaining to biochemical markers and genetically changed organisms includes:

a) Cooperative research with M. M. Ferguson, University of Guelph, on enzyme markers and partitioning within and between family covariance of quantitative traits with heterozygosity.

b) Cooperative research with R. W. Doyle and C. M. Herbinger, Dalhousie University, on DNA fingerprinting related to pedigree.

c) Cooperative work with T. J. Benfey, University of New Brunswick, on triploid-induced sterility.

Marine Gene Probe Laboratory, Dalhousie University, Halifax, Nova Scotia

Roger W. Doyle

The mandate of the Marine Gene Probe Laboratory (MGPL) is to apply recombinant DNA technology to the fisheries and aquaculture industries of Nova Scotia. The laboratory works in close collaboration with federal and provincial agencies concerned with the fishery as well as with commercial firms.

One of the major objectives of the MGPL is to develop "DNA fingerprinting" techniques that will provide powerful new tools for the resolution of long-standing questions concerning the distribution and migration of scallops, salmonids, cod, and other commercial species.

DNA-based techniques are also being developed for enhancing the power and efficiency of broodstock management and selection programs in aquaculture, and for increasing our basic understanding of quantitative traits such as growth rate, conformation, sexual development, behavior, and disease resistance. The program focuses on research of importance to the development of fisheries and aquaculture in Canada, and emphasizes basic, "non-proprietary" sciences.

Programs include: 1) Genetic Registry of Aquatic Strains and Pedigrees, using DNA fingerprinting and related techniques; 2) biotechnology and selective breeding, including immunology and transgenics; 3) ecological genetic projects in the Ocean Production Enhancement Network.

Memorial University of Newfoundland, Department of Biochemistry, St. John's, Newfoundland

William S. Davidson

Mitochondrial DNA

A restriction map of the Atlantic salmon mitochondrial genome has been constructed and the gene organization relative to this map was determined by a combination of cloning, DNA sequencing and southern blot analysis. As part of this study, a portion of the rainbow trout mtDNA was also cloned and subjected to sequence analysis.

The possibility of using mtDNA for stock identification was investigated by determining the mitochondrial genotypes in sympatric populations of anadromous and nonanadromous salmon. Six of 19 restriction endonucleases revealed variation in at least one of the populations. Seven genotypes were detected in Gambo Pond salmon. Three were common to the anadromous and nonanadromous populations and four were only found in the nonanadromous population. There is a strong indication of restricted gene flow between these populations but no population marker could be found that could be used to give an unambiguous identification of a salmon drawn at random from Gambo Pond.

It has been suggested that BstE II and Dra I give patterns that distinguish the mtDNAs of Atlantic salmon from North America and Europe. However, the "European genotype" was observed in both the anadromous and the nonanadromous Atlantic salmon of Gambo Pond and appears to be present in approximately 20% of the salmon that we have examined from Newfoundland rivers and from the LaHave River in Nova Scotia. We are continuing to survey populations of salmon over the entire range in North America (Maine to Labrador) to determine the distribution of these polymorphisms.

Our initial studies using the polymerase chain reaction and direct sequence analysis procedure have been with the mitochondrial cytochrome b gene and we have concentrated on comparing Atlantic salmon from Europe and North America, and Atlantic salmon and its closest relative, the brown trout. In a 300 nucleotide stretch, approximately 12 nucleotide differences were seen between Atlantic salmon and brown trout (4%). Two positions were variable among the salmon examined but these were not associated with the continent of origin. Other regions of the mitochondrial genome that accumulate mutations more readily than the cytochrome

b gene (e.g., the D loop) are better candidates for this procedure and this is being carried out.

Nuclear genome

Variation in the nuclear genome of Atlantic salmon has been sought using several cloned genes as probes. cDNA probes can detect variation (e.g., vitellogenin) but these are not very informative for population studies. A restriction fragment length polymorphism has been identified in the ribosomal RNA gene complex. Preliminary studies using 30 salmon from Europe and 30 from North America suggest that we have identified a genetic marker that can unambiguously tell the continent of origin of an individual Atlantic salmon. This result has tremendous implications for assessing the composition of the west Greenland high sea fishery. A wider survey is being conducted to determine if the marker holds true. Other repetitive elements are being cloned and will be examined for their value as genetic markers.

Chromosomes and gene mapping

We are collaborating with S. Hartley of the University of Stirling, Scotland and have begun analyzing Atlantic salmon chromosomes. As no one has been able to G band salmon chromosomes, we are trying different approaches. These include: *in situ* hybridization of cloned genomic fragments; pulse field gel electrophoresis and Southern blotting; and the production of somatic cell hybrids using mouse or Drosophila cell lines as the carriers. These studies are designed to give a better understanding of the genetic linkage and the genomic map of Atlantic salmon and this information will be of benefit for selective breeding programs.

Transgenic salmon

The equivalent of mammalian serum albumin was identified in salmonids and the protein was purified from rainbow trout. Characterization of the gene for this major secreted protein should yield a strong, constitutively expressed, liver-specific promoter which could be used to drive the production of foreign genes introduced into salmonids (i.e., in transgenics). This work is being continued in collaboration with C. Hew in Toronto.

Brown trout and Atlantic salmon

The interaction between introduced brown trout and Atlantic salmon in Newfoundland rivers serves as a model system for the potential impact of "genetically distinct, domesticated" farmed salmon on wild salmon. Natural hybrids between these species of *Salmo* have been observed but the fate of the hybrids and their impact on either species is unknown. We have begun to examine these questions through breeding experiments and by conducting a genetic analysis of brown trout populations in Newfoundland.

General

The overall amount of genetic variation in Atlantic salmon as a species is low compared with other salmonids. The majority of detectable genetic variation in the species as a whole may be found in any population. This indicates that it will be very difficult to find genetic markers that are population specific. At present, only two methods show promise in this regard: direct sequence analysis via the polymerase chain reaction of amplified DNA, and DNA fingerprinting. Whereas DNA fingerprinting is used to identify individuals in mammalian systems, it is more likely to reveal populations in salmon.

The inability to find population-specific genetic markers may actually reflect the biology and evolution of Atlantic salmon. Atlantic salmon may all be derived recently from a very small ancestral population. There may not have been enough time for mutations to have arisen and become fixed in different populations. In addition, there may be far more migration (straying) from population to population than has been assumed in the past. This would keep the different alleles in the gene pools of each population. Note that it does not require many individuals per generation to stray for this to happen.

INFORMATION FORM
ON ACTIVITIES OF THE MEMBERS OF THE ICES-WG ON GENETICS

Name and full address of the member signed below:

Dr Jean-Marie Sévigny
Maurice Lamontagne Institute
P.O. Box 1000
Mont-Joli, Québec, Canada
G5H 3Z4
Tel. (418) 775-6636
Fax. (418) 775-6542
Telex. 051-3815

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) Study of the genetic differentiation of Greenland halibut (Reinhardtius hippoglossoides). The aim of this research projet is to assess the genetic structure of Greenland halibut from Atlantic (Newfoundland, Labrador), the Estuary and the Gulf of Saint Lawrence and the Saguenay fjord. Methods used: allozymes and mt-DNA.

b) Study of the impact of physical barriers on gene flow in marine organisms. The aim is to determine to what extent physical barriers such as circulation patterns, the presence of sills can influence gene flow among populations of marine species of the Saint Lawrence system. Multispecific approach is used. Methods: allozymes and mt-DNA.

c) Study of the genetic effects of contaminants on marine organisms. A multispecific approach is used to determine the impact that pollution may have on the genetic diversity of marine organisms of the St Lawrence estuary. Methods: allozymes, mt-DNA.

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

All the projects described in the first section will continue for the next 12 months.

Date: June 6 1990 Name: Jean-Marie Sévigny.....

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

Prof.Dr.Wolfgang Villwock (Chairman)

Zoologisches Institut und Zoologisches Museum
Universität Hamburg,

Martin-Luther-King-Pl. 3
D-2000 Hamburg 13

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) Continuation of 1989 referred research carried out by Dr. M. von LUKOWICZ, Director, Bayrische Landesanstalt für Fischerei, Weilheimer Str. 8a, D-8130 Starnberg (Munich): Raising and feeding experiments on improving growth rate, disease resistance in European carp.

b) Dr. Sabine OBERST, Prof.res Dres. L. REMMRANTZ / W. VILLWOCK (address see above): Erythrocyte plasma membrane studies by means of electrophoretic techniques in European carp.

c) Same Group: Blood-Group investigations on diff. Tilapia species for population and species discrimination by immunbiological studies and investigations on immune response.

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) will be continued

b) will be continued

c) will be continued. Additional studies are deepend to develop a cross-test field kit for spontanous species discrimination under aquaculture field condi tions and for species protection purposes (Dr. Sabine OBERST, Prof. Dr. L. REMMRANTZ).

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date Aug. 1990 Name W. Villwock

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

GUYOMARD René
Laboratoire de génétique des poissons
CRJ - INRA 78350 - F JOUY-en-JOSAS
FRANCE

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) BIOGEOGRAPHY of BROWN TROUT (SALMO TRUTTA L.) USING ISOZYMES and mtDNA

b) GENE DIVERSITY IN OREOCHROMIS NILOTICUS AND RELATIVE SPECIES (ISOZYME ANALYSIS)

c)

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) same as 1 a

b) same as 1 b

c) GENETIC INTERACTIONS WITHIN HYBRID ZONES OF DOMESTICATED and NATIVE STOCKS OF SALMO TRUTTA (ISOZYME and GENOMIC DNA RFLPs)

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date 21/05/90 Name GUYOMARD R.

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

K.E. Jørstad
Institute of Marine Research
P.O. Box 1870, Nordnes
N - 5024 Bergen /Norway

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a)

b)

c)

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a)

b)

c)

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date 15/6-90 Name Kåre Jørstad

15.06.90

Dear prof. Villwock,

I refer to your letter of May this year about collecting information about research activities within the different ICES member countries. As far as I understand you are interested in the different activities carried out in the institutions in which the ICES working group members are working.

Concerning our work at Institute of Marine Research, Department of Aquaculture, these are summerized briefly below.

1) Actual research in fish genetics

a. Genotype - environmental interaction in farmed salmon.
In this project salmon sib group have been tested for growth performance in commercial farms which differs in environmental conditions. The results show considerable variation in the salmon production among the farms and differences in the ranking of salmon sib groups.

b. Genetic variation in natural fish populations.
This work have been continued focusing on yearclass variation of different stocks of herring, cod and brown trout. These studies are caaried out by using protein electrophoresis and mitDNA analyses.

c. Genetic studies connected to enhancement of coastal cod
This is a project in cooperation with our institute and Department of Fisheries Biology at the University of Bergen. Genetic analyses have been conducted on the native population in the release areas as well as for the broodstock and the juveniles released. In 1990 we have used genetic tagged broodstock (homozygote for a rare allele in PGI-1) and offspring possessing this marker will be released in the autumn.

d. Genetic interaction between farmed and wild populations - brown trout as a model species.
A morphological genetic marker has been identified in brown trout and a artificial population has been established. Mature fish of this sort (fine spotted) were released/escaped in two locations with natuaral trout populations. Genetic characterization, population size

estimates and spawning behavior studies have been carried out. Sampling and genetic analyses of 0-group trout in the locations this year, will give information about the reproductive success of the farmed trout and the extent of natural crossing with the wild fish.

Project a. is terminated and the different kind of data on growth, maturation, disease conditions and environmental factors will be compared. The other research project are continuing. Project d. this will be considerable increased with regards to a more extended ecological population study.

2. Planned research in fish genetics

a. Genetic variation in immune response in farmed salmon
This is a project in cooperation with Department of Biotechnology, University of Bergen. Salmon sib groups will be tested for immune respone against specific patogens and under controled infection.

b. Genetic studies in connection with salmon sea ranching
Large scale salmon ranching are developing in Norway at present. Genetic studies will include strain performance under ranching condition and studies on possible genetic effects on wild salmon stocks. Genetic analyses including protein electrophoresis and DNA fingerprinting will be important methods to detect gene flow between "artificially ranched" populations and wild stocks.

I hope this information could be of use in preparing the report of the Working group of genetics, and please give a word if a report of genetic studies in Norway is needed.

Your sincerely

Knut E. Jørstad

Information Form on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

Gunnar Nævdal, Institute of Fisheries and Marine Biology, the University of Bergen

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern) (together with Kjell Nedreaas, Institute of Marine Research)

a) Redfishes, genes Sebastes, in the North Atlantic, studied by use of starch gel electrophoresis to confirm species concept, identify young stages (0- and 1-group) to species, and studies on interpopulation variation between areas.

(together with Knut Jørstad, Institute of Marine Research)

b) Cod, studies on genetic effects of sea ranching on local stocks and use of genetic markers to quantify survival of young cod liberated, and interaction between natural and cultured cod (hybridization, gene introgression).

c) Intra- and interpopulation genetic variation in tusk (Brosmius brosme) and ling (Molva molva) with the aim of identifying management units of these two species. Starch gel electrophoresis and isoelectric focusing are being used.

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) Sebastes, if possibly continue with material from Iceland, Greenland and the Northwest Atlantic.

b) continue (until 1992)

c) continue

In addition we conduct research on inter- and intrapopulation variation in shells (oysters and scallops) and marine mammals (harp seal, minke whale etc.)

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date 13 June 1990 Name [signature]

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

Krzysztof Goryczko
Inland Fisheries Institute
Salmonid Research Laboratory
Rutki 83-330 Żukowo POLAND

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) Rainbow trout family selection based on 5 spring spawning strains (second year of realization).

b) Sex control in rainbow trout (limited research) production of phenotypic males of xx genotype (gynogenomes treated with Mt) for fish farms.

c) Polyploidization - The experimental part of programme aimed at evaluation of practical value of "normal", whole female and sterile (triploidized females) rainbow trout was finished in November 1989.

d) Interspecific hybridization among brook, sea, rainbow trout and salmon with and without polyploidization (diploids, triploids and tetraploids) is being realized. The survival, growth rate, kariology and enzyme markers of hybrids are analized.

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

Programmes "a" "b" "d" from item 1 will be continued

b)

c)

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date May 30 90 Name K Goryczko

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

Ana Maria Saldanha Nota Teia dos Santos Mendes Gomes
Centro de Investigação Pesqueira do INIP
Canal das Pirâmides
3800 Aveiro
Portugal

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) Genetic study, by electrophoresis method, of two populations of Nephrops norwegicus(L.) collected on North and South points of the Portuguese coast in order to obtain knowledge that provide us basis for a better management of this fishery.

b) The same of a) but on populations of Merluccius merluccius(L.)

c)

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) To continue (1) a)

b) To continue (1) b) and to try to study the same populations by DNA restriction enzymes method.

c)

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date 30/04/90 Name Ana Maria Teia dos Santos

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

Lennart Nyman
Institute of Freshwater Research
S-170 11 DROTTNINGHOLM, Sweden
phone: Sweden + 8 759 00 40
fax : " + 8 759 03 38

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) effects of hatchery environment and standard rearing methods on allele frequencies at polymorphic loci in Salmo, Salvelinus and Thymallus (starch gel electrophoresis)

b) establishment of a national fish strain registry for salmonid species (population specific data on allele frequencies, growth rate, survival, catchability etc)

c) conservation of genetic resources of fish: a breeding methodology for small populations (analysis of individual parents and their offspring using DNA-fingerprinting

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) (same as above)

b)

c)

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date June 11, 1990 Name .....

Information Form on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

DR DAVID THOMPSON
MINISTRY OF AGRICULTURE, FISHERIES AND FOOD
DIRECTORATE OF FISHERIES RESEARCH
FISHERIES LABORATORY
PAKEFIELD ROAD
LOWESTOFT
NR33 0HT ENGLAND

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) Salmon Population Genetics: Electrophoretic analyses of protein polymorphisms and a restriction fragment length polymorphism analysis of mitochondrial DNA are being used to identify differences between salmon populations in England and Wales. Comparisons between different environments
b) are being made.

Dover Sole: At Conwy the sex control mechanism of the Dover sole is being determined by studying gynogenetic diploids.

c) Manila Clam: Induced triploidy continues to be studied, again at Conwy.

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) Salmon Population Genetics: Continuation of 1a. Interactions between cultured and wild salmon will be studied in an enhancement exercise using tagged and genetically typed fish.

b) Dover Sole: Sex determination experiments should be completed. Induced triploidy may be attempted.

c) Manila Clam: Project 1c will continue.

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date 30.5.90 Name D. Thompson

Information Form
on activities of the members of the ICES-WG on Genetics.

Name and full address of the member signed below:

A F Youngson
DAFS Marine Laboratory
PO Box 101, Victoria Road
Torry, Aberdeen
AB9 8DB

(1) Actual research in fishgenetics (brief description of used techniques, aims and species of concern)

a) The return of escaped farmed Atlantic salmon to the River Polla in northern Scotland was studied using radiotagging, bankside observation and tissue pigment analysis. The escapes resulted from a single major accident. Farmed escapes tended to spawn later and lower in the river's length than wild fish. Farmed males penetrated the river more than farmed females.

b) The genetic condition of lines of farmed salmon in Scotland was studied using variation at polymorphic protein loci. 0-group juveniles were examined in two years. In general lines differ from representative Scottish populations of wild salmon. Lines differ from the wild populations used to found them. Single named lines differ between years.

c) MEP-2 allele frequencies in juvenile salmon were examined in relation to stream temperature in the tributary populations of the Kyles of Sutherland catchment. Frequencies of the 125 allele in populations at cooler sites exceed those present at warmer sites. Differentiation among the tributaries of the catchment, assessed from allele frequency variation, is consistent with the occurrence of low numbers of strayers only.

d) A mt DNA rfpl for HAE-III has been identified which, on the basis of preliminary study, may prove useful in studying genetic input from farmed salmon of Norwegian origin into wild Scottish populations. Further screening work continues.

(2) Planned research in fishgenetics for the next 12 months (continued and/or new):

a) The final return of escaped farmed salmon to the River Polla will be monitored. Many of the escaped fish were of River Neva stock. If a suitable regionally-based genetic marker can be identified, the genetic consequences for the native Polla stock will be assessed over a number of years.

b) It is intended to construct a study of the comparative performance of genetically tagged wild and farmed juvenile salmon in a natural stream.

c) A three year data set exists for allele frequencies in wild 0-group juvenile salmon in different Scottish populations at index locations. These populations have been shown to be genetically differentiated. The series of data sets will be extended to include the 1990 hatch to assess further the temporal stability of variation among populations.

In case of more than 3 different projects within pos. (1) and/or pos. (2), or, if brief explanations are wanted, please, use backside.

Deadline for return May 31 (latest: June 15), 1990 !

Date 15th May 1990 Name A F Youngson

(6) Preliminary Report of ICES Study Group (see C.Res.1989/2:35)/ pp. 25-27

Department of Agriculture and Fisheries for Scotland
Marine Laboratory PO Box 101 Victoria Road Aberdeen AB9 8DB
Telex 73587 Telephone 0224 876544 ext
Facsimile No: 0224 879156

1

GENETIC STUDIES BY DAFS ON WILD AND FARMED SALMON

Alan Youngson

1. We have quantified genetic protein variation in wild salmon populations in Scottish rivers. Small but significant genetic differences are present among river catchment populations.

2. We have examined the genetic constitution of farmed lines of salmon in Scotland, comparing them with the wild populations from which they were originally derived. The lines showed no marked signs of reduction in genetic diversity but each differed genetically from its original wild source. Overall, lines differed genetically from representative populations of wild salmon.

3. The Me-2 locus shows a geographical cline in the frequency of the alternative alleles at the locus in wild populations of salmon across the species' global range. The 125 allele is common in populations in the northern part of the range: the 100 allele is more common in the south. Differences appear to be associated with latitudinal variation in temperature.

   The same association appears to be present within the salmon populations of the River Dee and in the Kyles of Sutherland rivers. The 125 allele is more common in the colder parts of each catchment and less common in the warmer parts.

4. In wild salmon, differences in juvenile growth and age at sexual maturity appear to be associated with the different Me-2 genotypes.

5. We are screening populations of wild salmon for genetic variation in mitochondrial DNA structure hoping to extend the range of genetic markers available for population studies. Scottish and Norwegian fish are being analysed in the search for a regional genetic marker which might facilitate the assessment of genetic introgression from imported Norwegian farmed stocks into wild Scottish populations.

6. In 1989 we monitored the behaviour of wild salmon and escaped farmed salmon in a northern Scottish stream. As the result of a major accidental release in a nearby sea loch in February, escapes entered the river together with native fish in summer and autumn. By means of radio-tracking, observation at spawning and pigment analysis we have compared the spawning of the two groups of fish.

   This study is still incomplete. It will continue in 1990 and is being extended now to other river systems which may also have been invaded by farmed escapes.

II

PLANNED STUDY OF THE RELATIVE PERFORMANCE OF NATIVE SALMON AND INTRODUCED SALMON OF FARMED ORIGIN

The studies being performed by DAFS and listed in the second enclosure all relate to the subject of the second part of the ICES resolution. Similar studies are probably underway, elsewhere.

We judge farmed and wild fish to be genetically different by all the measures we have used so far. One of these characters (Me-2) appears to have an associated performance component. We know that escapes from farms return to freshwater to spawn as adults. They have been observed to do so with each other or with wild fish.

A major omission, at least in our own work, is in assessing the relative performance of wild native fish, introduced fish of cultured origin and hybrids in a natural stream. Ideally such a study should examine performance in all its aspects, to the stage of adult return and spawning.

The study will be compromised by any necessity to rear juveniles in tanks to the stage where the different groups can be marked by physical means. The alternative approach is to mark the groups at the outset with a genetic marker, crossing adults of known genotype at some neutral polymorphic locus.

In the past we considered carrying out this experiment in a stream by planting genetically marked eggs from native and non-native adults and their crosses, together. Our intended genetic marker was Me-2 which is expressed in muscle and can be typed in advance of sorting adult broodstock, using biopsy techniques.

However we were forced to abandon this approach when doubt arose as to the neutrality of the variation at the Me-2 locus. No practical alternative marker has become available which would allow us to reconsider this work of this type.

There is a generic experiment which resolves this difficulty but it requires access to paired stream sites.

	Stream I			Stream II		
Group	1	2	3	4	5	6
Genotype	AA	BB	AB	AA	BB	AB
Source	native	farmed or hybrid	native, farmed, or hybrid	farmed or hybrid	native	native, farmed, or hybrid

For any performance character it will be possible to assess the stream effect by comparing Group 3 with Group 6. The source and genotype effects can also be assessed. Further, source-genotype interactions can also be investigated for the homozygote conditions. Performance might be monitored through to adulthood and spawning, assessing finally the proportion of progeny generated by the native and introduced groups of fish.

Three problems attend any experiment of this type. Firstly, to run the experiment through to adulthood might take six years and the results might be devalued in the practical context given the rate at which aquaculture will develop in the interim. On the other hand, the study might generate information of value on juvenile performance, soon after its inception and would be of academic interest in any circumstances.

Secondly, any experiment of this type may be site specific and will also be specific for the source of the introduced fish. However, replication, using different stocks and sites might be considered worthwhile.

Finally, a conflict probably exists in being able to study juvenile and adult performance in the same experiment. A small stream of manageable size in which juvenile performance can be examined will probably not sustain large enough numbers of fish to support destructive sampling of juveniles in addition to generating enough smolts to ensure that useful numbers of adults return to the stream later.

It may therefore be necessary to divide any study into two parts. The juvenile component seems to be practicable and only modestly demanding financially and study of juveniles alone may generate information of worth.

However, the adult component is different. In its proposed form it is probably not practicable. It requires large duplicated stream sites and duplicated traps for smolt capture because genetically tagged fish emerging from the stream must be physically tagged before release. It probably also requires duplicated traps in which adults may be recaptured and sorted according to physical and genetic marks before being allowed to spawn.

Is there a more elegant or more efficient way of performing these studies, perhaps through the use of non-enzymic markers? Is there a less costly or a less demanding option? Is there a more limited option which would generate valid information? Is it possible that studies like these will be performed at all?

(7) Citations

reprints from:

FISHERIES, Bull. Am. Fish. Soc., 15 (1): Jan.-Febr. 1990.

- KAPUSCINSKI, Anne R. and Eric M. HALLERMANN: Transgenic Fish and Public Policy: Anticipating Environmental Immpacts of Transgenic Fish: 2-11.
- HALLERMANN Eric M. and KAPUSCINSKI, Anne R.: Transgenic Fish and Public Policy: Regulatory Concerns: pp. 12-20.
- HALLERMANN Eric M. and KAPUSCINSKI, Anne R.: Transgenic Fish and Public Policy: Patenting of Transgenic Fish: pp.21-24.