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REPORT OF THE WORKING GROUP ON THE APPLICATION OF GENETICS IN FISHERIES AND MARICULTURE

Copenhagen, 9-11 March 1994

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1 INTRODUCTION

At the 81st Statutory Meeting in Dublin, September 1993, the former Working Group on Genetics was renamed the Working group on Application of Genetics in Fisheries and Mariculture (WGAGFM, Chairman: J. Mork, Norway). The first meeting of the new Working Group was located at ICES Headquarters in Copenhagen from 9-11 March 1994 (C. R. 1993, 2:27). In its justification for suggesting C.R. 2:27, the Mariculture Committee noted, e.g., that "...the broad range of expertice required will mean that the Working Group will utilize a sub-group format". In cooperation with the Chairman of the Mariculture Committee, the WGAGFM Chairman established a "core" structure of the Working Group during the autumn of 1993. Towards the end of the year a sub-group in qualitative genetics (leader David Thompson, MAFF, UK) and one in quantitative genetics (leader Gerry Friars, Atlantic Salmon Federation, Canada) were established. To secure that the first Working Group meeting could efficiently deal with the Terms of Reference (ToR), a preliminary contact was made with additional scientists in the qualitative and quantitative field. Most of these accepted the invitation to join the respective sub-groups, and to attend the first Working Group meeting in Copenhagen, March 9-11. In a letter of 21. January 1994 from the General Secretary, the national delegates were asked to nominate members of the WGAGFM. By 23. February 1994, thirtyfour members had been appointed (refer to member list in Appendix 6). In a communication from the ICES secretariat of 23. February 1994, all appointed members received a copy of the ToR for the WGAGFM, some background materials and practical information for the meeting, and a letter from the Chairman further indicating the kind of preparation that was desirable for the meeting.

1.1 Attendance

Some members encountered unexpected difficulties and could not attend the WG meeting, which they regretted in letters to the Chairman (this concerns D. Thompson, H. Kincaid, W. Davidson, M.-L. Koljonen, A.R. Childs, W. Villwock, and K. Goryczko).
When it became clear that the leader of the qualitative sub-group, David Thompson (MAFF, UK), was unable to attend, Tom Cross (UCC, Ireland) was asked to serve in his place during the Copenhagen meeting. Tom Cross accepted, and met together with Gerry Friars and Jarle Mork in forehand of the meeting for practical planning of the agenda.

1.2 Working Group substructure and function

Eleven members (incl. the Chairman) attended this first WGAGFM meeting (refer to address list on next page). At the beginning of the meeting, these attendants were ascribed to respective sub-groups:

Qualitative sub-group: **Tom Cross** (const. leader), G. Dahle, A-K Danielsdottir, M. Hansen, K.E. Jørstad, E. Verspoor, F. Volckaert.

Quantitative sub-group: **Gerry Friars** (leader), B. Gjerde, J. Jonasson.

At this first WGAGFM meeting, priority was given to those parts of the ToR which required preparation of materials for other ICES groups. Thus the WG finalized the section for The Study Group on Interactions between Wild, Ranched (Enhanced) and Cultured Salmon (i.e.; section 3.1 plus Appendices 1 and 2 of this report), and the section reviewing the Report of the Study group on Stock Identification Protocols for Finfish and Shellfish Stocks (i.e.; section 3.2 plus Appendix 3 of this report). The material was sent to the respective chairmen of those Study Groups in time before their scheduled meetings in 1994. The other points in the ToR were addressed partly in Copenhagen, and partly by telephone, fax and postal correspondance up to the end of May (only the members attending the Copenhagen meeting were included in this process). This final version also includes national status reports as received after an invitation sent out to all WGAGFM members.

Attendants at the WG meeting 9-11 March 1994 at ICES Headquarters in Copenhagen:

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1.3 A broadened genetic scope for the WGAGFM

The new sub-group format of the WGAGFM reflects the broadening of its function that was recommended by the Mariculture Committee and the Council at the 81st Statutory Meeting. The background for this broadening was the rapid growth of mariculture in marine food production that has already taken place, and the anticipated further increase of this industry.
It is important that this rapidly growing industry is based on firm knowledge and sound principles for genetic management. This applies both to the initiation of breeding programs to increase the production efficiency, disease resistance etc, and to the identification of potential areas of conflict with the traditional use of finfish and shellfish resourses, in order to minimize unwanted effects.

On this background, a significant part of the 1994 Terms of Reference concerns quantitative genetics. Since this may be a novel area for many workers in fisheries biology, the Working Group produced an introductory text on theory and practice of quantitative genetics (refer to the following sections 2, a-i).

2 SELECTION AND GENETIC MODIFICATION IN FINFISH AND SHELLFISH BREEDING PROGRAMS

a) Introduction

Many countries are involved in simple forms of mass selection in fish and shellfish, with little attention to the control of mating and selection systems. These systems need to be considered simultaneously in order to maximize gains. Considerations of these systems appear in the following sections, although references have not been included.
Over the past 25 years, methods of breeding, that have been proven in animal and plant breeding, have been adapted to aquaculture. However the external fertilzation in many fish species allows for unique mating designs and the higher reproductive rate permits higher selection differentials and, hence, scope for high genetic gain. Additionally, fertilization *in vitro* in most species of fish and shellfish allows for the use of complex mating designs. However, attention must be paid to the use of a sufficiently high number of effective parents (refer to section D) to avoid inbreeding and reduction in fitness. This consideration has been seriously violated where many populations of fish have been founded and reproduced with a small number of parents (spawners). Therefore, the use of pedigrees in fish and shellfish breeding programs is more crucial than in any terrestrial farm animal.

b) Objectives of Selection

The objectives of selection in domesticated populations can be defined with a reasonable degree of discreteness. The main objective is to change the average performance of the target traits of the population, in a defined direction, to benefit producers, industry and consumers.
The traits to be considered in the objectives must be measurable, of economic importance and heritable (e.g. growth rate, age at sexual maturity, resistance to disease, product quality, etc.). Experience from plants, terrestrial animals and fish indicate that most traits have some degree of heritability (ratio of genetic to total variance) if measured in an appropriate way and hence it is possible to alter their performance genetically through selection.

c) Establishment of Base Populations

A base population, defined by single (interbreeding) or multiple stocks, must be carefully considered at the beginning of a breeding program. The ultimate goal must be considered in selecting original strains. This base must have a wide array of genotypes (large genetic variation) in order to maximize genetic gains in both the short and long term. This can be achieved by the introduction of several

stocks into the base. Before intense selection is applied, one or two generations of random mating should be conducted to allow the mixing of genes from the original populations if more than one stock is used. This procedure allows a safeguard against the narrowing of the genetic variance in a population.

d) Role of Pedigrees

The genetic relationships among individuals (pedigrees) must be known to control the mating and hence to overcome problems with inbreeding. This also allows the use of information on relatives to increase genetic gain over that possible in mass selection.

e) Breeding Strategies

The choice of a breeding system (inbreeding, crossbreeding or purebreeding) depends primarily on the type and magnitude of genetic variance (additive and/or non-additive) available in the population for the trait(s) of interest. If there is sufficient additive genetic variance, purebreeding (genetic improvement within a population) should be applied. Purebreeding is the selection method that has been proven successful in several species. Estimates of genetic parameters (heritability, genetic correlations and phenotypic correlations) are important in initiating a progam.
If the magnitude of non-additive genetic variance due to dominance or epistasis is of importance, some system of crossbreeding may also need to be implemented. Inbred lines can be used in crossbreeding but experiences in corn and poultry have shown that the crosses of populations, with little inbreeding, have been satisfactory. This result should not be neglected by fish and shellfish breeders. Fish geneticists have probably placed too much emphasis on strain and species crossing programs with little or no attention to selection for additive genetic effects within populations.

f) Systems of Selection

The selection of superior prospective parents can be performed by choosing the individuals with desired phenotype(s) (mass selection). In this system, the size and structure of the population is important in order to control inbreeding. When applying selection, strict control should be kept on the number or offspring per dam and the use of milt from a wide array of sires. A subdivision of the population would allow the use of sires and dams from different subpopulations to avoid inbreeding. Where pedigrees are available to yield information on relatives, the selection of superior families can be accommodated. However, the combination of such information, with that available for the individuals, should yield better response to selection. Family selection can also allow selection for traits that cannot be measured on the live breeding candidate (eg. disease resistance or carcass quality traits) - a feat that cannot be accomplished with mass selection or within-family selection. However, the high fecundity of fish allows the reproduction of the population with a small number of families and, hence, attention must be paid to keeping the number of families to a level that will inhibit serious inbreeding, while attaining reasonable selection intensity. An increase of up to .02 (eg. about 13 single- pair matings), in the inbreeding coefficient per generation, is usually considered acceptable to retain fitness but the critical level may vary between species. In deciding on the numbers of the brood stock, however, the risk of loosing alleles must also be recognized.

g) Genetic Gain

The breeder's goal is genetic gain expressed in monetary units. The rate of progress towards such a goal is a function of the accuracy of selection, genetic and phenotypic variances, genetic and phenotypic covariances between traits, economic weights for each trait, intensity of selection and generation interval. The high reproductive rate in fish is a desirable attribute in attaining highly intensive selection but must be considered hand-in-hand with the avoidance of high degrees of inbreeding. The short generation interval in species like Tilapia (e.g. one year) is much more desirable than that of Atlantic salmon (three or four years). The desirable cost:benefit ratio, accrued during

selection, is enhanced by the high fecundity that allows the rapid spread of improved populations from a small number of breeders.

h) Genotype-Environment Interaction

Genotype-environment interaction is encountered when the superiority of different genotypes is not consistent across different environments. If the reranking is serious, the development of one strain to perform in a series of environments is not practical. The development of unique genotypes for specific environments elevates the cost of breeding programs. Where the environment can be controlled, this problem is not usually serious.

The phenomenon has been noted in aquaculture (eg. salmon, Tilapia, rainbow trout, channel catfish) but the development of specific strains for different environments has not been undertaken because the magnitude of the GxE interaction has been low compared to the total variance of the traits studied. In natural populations of fish, the occurrence of genotype-environment interactions, as manifested by local adaptations, may be of more concern and should be investigated.

i) Other Methods of Genetic Modification

A lot of emphasis has been placed on sterility technology in order to control sexual maturation. All female stocks, sometimes in combination with triploidy, are being considered. In Norway, the aquacultural industry has been reluctant to apply these techniques due to the risk of public reaction and the fact that farmers wish to benefit from the burst of growth experienced at the onset of sexual maturation. The United Kingdom is using triploidy commercially in rainbow trout. The use of all-female populations, in combination with triploidy, is being investigated in Canada and Ireland. Tetraploids are being studied in the United States and France, with a possible view toward producing triploids by crossing tetraploids with diploids.

Transgenics are being studied at the experimental level. Little or no quantitative information on changes in traits genetically or phenotypically correlated to the trait targeted by inserted transgenes is currently available in fish. Growth hormone and antifreeze-protein inserts have been investigated. If such a technology becomes acceptable, its use in domesticated fish will need to be coupled with classical methods of breeding. For example, there is evidence in mice which indicates that the rat growth hormone gene can be lost from selected lines.

The accidental or deliberate release of transgenics or fish genetically modified in another manner (such as in the case of cultured fish), in the wild, is a controversial area. The creation, through selection, of domestic stocks with decreased natural mating behaviour could be one approach to reducing the problem.

3 TERMS OF REFERENCE 1994 (C.R. 2:27, 1993)

The Terms of Reference for the WGAGFM in 1994 consisted of 6 specific points as listed in Appendix 5. These points are addressed in succession in the following sections 3.1 - 3.6.

3.1 "Prepare information for use by the Study Group on Interactions of Wild, Ranched (Enhanced), and Cultured Salmon."

On further request from WGAGFM on what type of information was needed, Alan Youngson (Chairman, "Interaction Study Group") asked the WGAGFM to respond to these three questions:

1. Is more work on interactions required?
2. Is enough work in progress?
3. Will a theme session at the 1995 Statutory Meeting (C.R. 2:62) be worthwhile from the genetics point of view?

In addition to responding specifically to these three questions (sections 3.1.1, 3.1.2, and 3.1.3), the WGAGFM produced information for the "Interaction Group" by reviewing the most recent Study Group report on interactions (refer to Appendix 1), and by preparing a list of current national activities in this area (refer to Appendix 2). Also, preliminary versions of sections 3.3 (on gene flow) and 3.4 (on combined studies) were given to the "Interaction Group".

3.1.1 Response to question 1

In order to assess or predict genetic effects from interaction between cultured and farmed salmon it is necessary to have information on the

four parameters of interaction:

a) - the effective gene flow in the natural structure of populations
b) - genetic characteristics (incl. magnitude of local adaptation) of wild populations
c) - genetic characteristics of enhanced/cultured salmon
d) - the effective gene flow between enhanced/cultured and wild populations

Assessment of interaction in specific cases will usually require **case-specific** information, while a more general consideration must be based on knowledge of the **range of values** that these four parameters (*a,b,c,d*) can take. Although much information undoubtedly exists, it is scattered in the literature and may be difficult to access. To this end there has not been any systematic survey or review that simultaneously covers all these interaction related parameters in salmon.

a) The amount of gene flow between natural populations sets, so to speak, the evolutionary scene. Without reductions in gene flow between some intraspecific groupings, there would be hardly any substantial or lasting differentiation (or multiple populations at all, for that sake). Estimates of this important parameter should therefore have high priority and be carried out using the best available methodology. Unfortunately, this has not always been the case.
In salmon, published estimates of the natural gene flow between wild populations are diverse. This may not be surprising since in reality, there are probably substantial differences in gene flow between different pairs, or sets, of natural populations. It is important to be aware that the value of gene flow estimated via F_{st} or G_{st} (i.e., the most common method) usually is averaged over many loci and many pairs of populations. Such mean values may not give a realistic description of the population structure (refer to section 3.3.1). In fact they may instead obscure the real variability if they are not

accompanied by adequate measures of their range and variance.
However, part of the variation in estimates of salmon gene flow may also be due to differences in study design, and to how well the assumptions for such estimates are met in various studies (refer to section 3.3). Many gene flow estimates were obtained as by-products in studies that were not properly designed to measure it. This may seriously reduce their usefulness in critical applications such as in the calibration of mathematical models in risk analyses.

The Working Group feels that there is a need for a critical review of the existing litterature and may be for new, carefully designed and properly scaled studies, with a view to obtain the best possible estimates of the actual levels of gene flow within and between hierarchies in the genetic structure of salmon.

b) Wild populations of salmon may be characterized genetically in different ways: the qualitative genetics approach in which the statistics are based on gene frequencies at specific, individual loci, and the quantitative approach in which the mesurements are based on "statistical genes", i.e. the traits under study are affected by many loci and the effect of each locus is unknown.
The qualitative approach can be a rapid one, in which the type of information obtained is related to the number of different genes and their frequencies. The quantitative approach is usually more time consuming. In return, however, it can provide crucial information of a kind that as a rule cannot be obtained by qualitative methods; e.g. on adaptationally relevant effects of differences in gene pools (manifested as genotype-environment interactions which is the same as local adaptations; refer to section 2 h).

The Working Group feels that whenever possible in interaction studies, the genetic characterization of local populations should use both qualitative and quantitative methods. That would give the best result relative to gaining fundamental understanding of the natural structuring and dynamics of Atlantic salmon populations and thus the implications of ecological and genetical interaction with non-native fishes. Since there is a general lack of such combined studies in Atlantic salmon the Working Group recommends that they be initiated (refer also to section 3.4).

c) The genetic characterization of the cultured salmon appears to be a rather neglected aspect in many studies so far. This is unfortunate, since the actual genetic differences between cultured and wild fish is one of the most important parameters of an interaction, and because the genetic composition of the cultured fish is changing due to:

- artificial selection for production traits
- relaxation of natural selection in the culture situation
- random genetic drift in small brood stocks.

At present, some of the significant cultured stocks have been five generations in culture and may have changed substantially from their wild relatives. Both directional artificial selection and genetic drift is expected to result in loss of genetic variability. Hence, for assessing potential effects of genetic interactions between their offspring and wild stocks, a minimum level of information would be to have records of their effective population sizes. Loss of genetic varibility is a problem that may apply to brood stocks in enhancement programs as well. Ideally, genetic monitoring programs for important captive breeding stocks should become a routine cooperation between the industry and the resource management authorities in each country.

The Working Group feels that there is a strong need for an updated genetic characterization of brood stocks used in mariculture, and for a monitoring of genetic changes in those stocks.

d) The effective gene flow between a specific cultured and a specific wild salmon population is addressed in several current experiments. This type of interaction is different from the natural gene flow between wild populations. Therefore the methodology for estimating gene flow is is also different and usually involves some form of genetic marker (refer to section 3.4). It may be anticipated that the gene flow estimates from such experiments will vary substantially since they may be very dependent of the specific regime for the experiment. Sources of variation would include the origin of the cultured immigrants, family differences within immigrants, the specific wild stock used, different degree of neutrality of the genetic marker itself, etc.

In planned, controlled experiments, the importance of an effective and correct experimental design for getting adequate and reliable results from gene flow studies must be stressed. To improve on this, the inclusion of statistical expertice when planning specific experiments should become routine. The working Group also feels that the possibility for ***opportunistic*** *studies of gene flow between cultured and wild salmon has not been utilized satisfactory in the past. For example, cultured fish escapes from rearing-pen wrecks and their subsequent entering of salmon rivers may give excellent possibilities for doing studies that could not be achieved or would not be allowed in controlled experiments. To be able to utilize such situations it is important that there exist genetic base line data on wild populations as well as on the cultured stocks. In general, such base line data should be regarded as a necessary prerequisite for a rational management.*

3.1.2 Response to question 2

The partial list of the many projects currently in progress (Appendix 2) reflects a substantial research effort in many countries on the question of interaction between cultured and wild salmon. If successful, those studies should provide substantial insight into this complex question. The research areas as well as the improvements on experimental designs suggested by the Working Group (above) would come in addition to the listed projects.

There are also some additional areas where specific work will be required. In particular the effects of genetic interactions on genetic fitness in the marine phase and the effects of outbreeding depression (second and later generation mixing) need to be evaluated. Exactly how this might best be done is likely to be easier to identify on the basis of the results of work currently in progress. In general the Working Group feels that agreement of research objectives, good experimental design, coordination of future work and cooperation among different disciplines will be essential for the success.

3.1.3 Response to question 3

C. Res. 1993/2:62 specifically asks the Study Group on Interactions of Wild, Ranched (Enhanced) and reared Salmon to develop preliminary plans for an ANACAT and Mariculture Committee Joint Session for the 1995 Statutory Meeting on "Interactions" for considerations by the Parent Committees at the 1994 Statutory Meeting.

In general it is felt that, even though the results of much experimental work in the area which is currently underway will not be complete in 1995, sufficient results should be available to make a theme session worthwhile. It is also felt that the question of interactions, in particular, is a field where the benefits from ***mathematical modelling and risk analysis*** could be substantial, and that a theme

session on interactions should encourage the presentation of such studies. The Working Group recommends that such a theme session be arranged and is prepared to contribute to its implementation.

3.2 "Review the interim conclusions of the Study Group on Stock Identification Protocols for Finfish and Shellfish Stocks (document C.M.1993/M:3)"

3.2.1 General comments

The WGAGFM regards stock identification as an area in which genetics can make a major contribution to fisheries management. Thus, our comments (1-4 below, and Appendix 3) are designed to increase the utility of the study group document.

1.The working group felt that this report was rather selective and covered only a limited number of areas rather than reviewing the field in general. Either the text should be expanded or explicit reference should be made to the fact that the particular studies are mentioned only as examples.

2. Perhaps the best example of stock discrimination by genetic techniques is the Genetic Stock Identification (GSI) program for Pacific salmon species. While this program is mentioned, it is not discussed in detail.

3. It should be stressed that different principles apply to marine and freshwater/anadromous fish species because of the very different biology of the two groups.

4. The importance of sampling is not stressed. For example, the time the sample is taken relative to the biology of the species is important, as is the sample size.

Refer to Appendix 3 for specific comments on the report on a section by section basis.

3.3 "Review knowledge of the amount of gene flow between specific natural populations as well as between cultured and specific natural populations with a view to proposing future studies."

Introductory to the discussion in the Working Group, the central role of ***gene flow*** in most questions concerning finfish and shellfish population genetics was highlighted. It was pointed out that whether the problem under discussion concerns

- stock identification
- interaction between cultured and wild stocks
- local adaptations
- phylogenetic or speciation processes
- gene spreading from genetically modified organisms
- genetic resources and the precautionary principle
- development of mathematical models for prediction purposes,

knowledge of the historic and/or present amount of gene flow between taxa at various stages of genetic differentiation is usually of paramount importance.

The discussion on this topic considered first the question of selective neutrality for genetic markers which are intended for use in estimating gene flow. It was the view of the group that, with an exception for third codon substitutions (sequencing studies), it is not possible to guarantee selective neutrality for any genetic marker. Not even non-coding DNA would be "safe" in this context because of the possibility for hitch-hiking with genes which are selected. Thus any study using genetic markers should be designed so as to enable evaluation of the actual neutrality of the marker itself.

The discussion on methods of estimating gene flow revealed at least two principal types of need for gene flow estimates, and that the actual choise of methodology for obtaining estimates would be dependent on the need. The two principal type are discussed below (sections 3.3.1 and 3.3.2).

3.3.1 Estimating gene flow within an established population structure

For this type of study both direct and indirect methods are applicable. Direct methods would imply the use of some form of genetic marker, for which changes in frequency away from a base-line value would enable direct estimates of gene flow. In such studies more than one marker should be used in order to enable the detection of potential bias due to selection.

However, indirect methods are most often used in this situation. The most common method is based on the use of F_{st} (or G_{st}) estimates from multilocus studies, and by re-arranging the formulae of those parameters to focus on m, the gene flow (or actually $N_e m$, the absolute number of immigrants per generation). Usually, m is derived from the following basic expression from Sewall Wright's "Island model" of genetic differentiation

$$F_{st}=1/(1+4N_e m),$$

where N_e is the effective population size and m is the proportion of immigrants in each population in each generation. It should be mentioned, however, that this formula is an approximation of the complete expression derived by Sewall Wright, which is

$$F_{st}=(1-m)^2 / (2N_e - (2N_e-1)(1-m)^2)$$

and which preferably should be employed whenever critical studies are undertaken. The estimate of m by using one or another of those two expressions may differ by several percent depending on the true values of m and N_e.
Also of concern in this context is that the values obtained in this way often show substantial variation depending on which locus is used. Hence the need to use **estimates from many loci** should be observed. Another source of variation in this kind of estimate is the **number of individuals in samples** used for estimation of allele frequencies. The relative variance contribution by the number of loci and the number of individuals in samples can be estimated as described in population genetics textbooks. A third variance component is the random genetic drift of allele frequencies in the populations themselves, which in small populations can cause substantial variability between generations. While in theoretical models this variation is levelled out by assuming a large number of populations, practical studies often considers only a few ones and hence gene flow estimates may vary substantially between generations. Therefore, the establisment of base line data and a monitoring of allele frequencies in the populations is necessary in order to assess the stability of the allelele frequencies which are used, e.g., in estimating levels of gene flow.
The F_{st} statistic is a "mean" value under the Island model of genetic differentiation, which assumptions may not be very realistic for salmon populations. They are, e.g., very variable in size, their geographic interdistances are widely different, and the effective gene flow may vary substantially between different sets of them. Therefore, different estimates of F_{st} (and hence the

corresponding value of N_e*m) would be expected, depending on which set of populations is actually included in the study.
Referring to the connection between gene flow and local adaptation (section 3.1.1.a), this implies that the possibility for local adaptation may also show large variability. In very small populations with a substantial immigration, selection coefficients must be very strong in order to establish and maintain a local adaptation. In many instances it may therefore be more realistic to consider **geographical clusters of populations,** rather than the individual ones, to be the real units of adaptation. This may apply, e.g., to situations where several small salmon populations with a substantial gene flow between them inhabit a restricted geographic area.
It should also be observed that the formulae for estimating m via e.g., F_{st}, assumes that an equilibrium has been reached between genetic drift and immigration. If equilibrium has not been reached, the gene flow would tend to be overestimated.
Finally, all available *a priori* information of the actual population structure should be utilized for obtaining relevant samples. Samples containing mixtures from several populations would generally yield overestimates of the actual gene flow within the system.

3.3.2 Estimating gene flow into an established population or population structure

In some situations, for example when assessing the risk that a genetically modified organism (GMO) would be able to enter an existing population and proliferate there, the indirect methods discussed above would not be feasible. Since the organism in question would not be "part of the system", its chance of reproductive success cannot be assessed based on observations or estimates of the natural gene flow in the system. Rather, such situations would call for specific experiments with model organisms that mimic the GMO as good as possible. Again it is the effective gene flow which is important, and hence a genetic marker (rather than counting immigrants) is necessary. Ideally, the marker gene itself should be selectively neutral in order to yield unbiased results (although such neutrality probably cannot be absolutely guaranteed; refer to section 3.3).

3.4 "Propose studies of local adaptations of specific natural populations using combined qualitative (e.g., gene markers) and quantitative (e.g., family studies) genetic approaches."

There is an increasing international awareness of the need for a sustainable use of natural resources. A sustainable utilization of fish stock includes the conservation of genetic resources such as substantial genetic adaptations to local environments. In anticipation of a set of generally accepted criteria for categorization of genetic resources, the Working Group feels that the term "substantial genetic adaptation" must reflect e.g. the number of generations necessary to establish the adaptation, and the amount of genetic change away from the original gene pool. The identification and assessment of such adaptations will probably get increased importance in resource management.

Mapping and assessment of natural genetic adaptations must be based on methods and principles from population genetics theory, i.e. the task must be described in terms of the classical evolutionary forces like mutations, genetic drift, gene flow and selection.
While the action of genetic drift increases (non-adaptive) genetic differences between populations, gene flow has the opposite effect. For environmental forces which are uniform for populations, natural selection may be a homogenizing factor while it may increase genetic differences when environmental factors vary between populations. These evolutionary forces may interact in very complex ways. Eventually a situation will be reached where the differentiating and the homogenizing effects neutralize each other in an equilibrium. At this equilibrium the mean fitness of the local population is at its maximum, i.e. at the highest achieveable local adaptation (epistasis disregarded) under the given gene flow regime. The number of generations needed to reach this stage is determined

by the instensity (i.e., the selection mortality each generation) of the natural selection. The necessary factors needed to estimate the speed and thus the magnitude of an adaptation over time would then be:

- mutation rates
- population size (through the effect of random genetic drift)
- gene flow (proportion of non-selected immigrants and their gene frequencies)
- selection coefficients (intensity of selection)

These quantities are important parameters in mathematical models for description and prediction of evolutionary change. Reliable parameter estimates usually require carefully designed and often case-specific studies, in which it may be necessary to combine methods from qualitative and quantitative genetics. Such studies are very scarce in the literature, and should be encouraged. A possible outline is described in the following sections 3.4.1 and 3.4.2.

3.4.1 Methodology and application areas

Adequate methods for assessing **population size** and its variation are available. Qalitative genetic methods exist for direct and indirect assessment of **gene flow**. With respect to **natural selection** it may be concluded, from the enormous mortality commonly observed from egg- to adult stage in most fish species, that there is a large **potential** for natural selection and thus adaptation. However, the actual selection intensity for multilocus, fitness-related traits cannot be assessed without knowledge of the **heritability** of those traits in the actual environment (i.e., how strongly the genotype is reflected in the phenotype on which the environmental factors act).
A list of quantitative genetic traits which probably are related to fitness would include, e.g.:

- Relative fecundity
- Relative egg size
- Time of emergence
- Yolk sack absorbtion
- Survival to first feeding
- Survival to end of first year
- Survival to smoltification
- Extent of precocious maturity
- Disease resistance in fresh water
- Resistance to low or high pH
- Sea abscence pattern
- Growth rate at sea
- Survival at sea
- Disease resistance at sea
- Migration route
- Time of upstream migration
- Susceptibility to angling
- Rate of sexual development
- Time of spawning
- Kelt survival (multiple spawnings)
- Disease resistance as adult in fresh water

Estimating heritability is a field of **quantitative genetics.** A better understanding of the nature and magnitude of local adaptations, and of the potential risks to them posed by certain human activities (like populations bottlenecks and increased gene flow) therefore call for studies which combine methodology from qualitative and quantitative genetics. Until results from such studies are available,

predictions of effects of specific changes in, e.g., immigration regimes are bound to be hypothetical.

3.4.2 Sample outline of a combined study of adaption

In the following, some important aspects to be observed in the design of such combined qualititative and quantitative genetics studies will be outlined. In order to yield parameter estimates that are valid for wild stocks, it is essential that the actual performance tests are carried out under natural conditions.

3.4.2.1 Assessment of genetically effective population size

Usually N_e, the genetically effective population size is not simply the number of spawners counted one year. Most natural populations have overlapping generations, and N_e must be adjusted according to formula I:

I $N_e = N_0 t \cdot l$,

where N_0 is the number of individuals born in a specific generation, t is the mean age at reproduction, and l is an individuals's probability of survival to that age.
Furthermore, N_e is heavily affected by the proportion of male and female spawners (formula II):

II $N_e = 4 \cdot (N_m N_f)/(N_m + N_f)$

where subscripts m and f represent males and females, respectively. Formula II tells that with unequal sex proportions, the effective size is always smaller than the actual size. Likewise, there are formulae which allow adjustment to be made for cyclic changes of populations size when records of such are available (formula III):

III $N_e = n / \Sigma(N_i^{-1})$

where n is the number of generations in the cycle, and N_i is the population size in the i-th generation of the cycle. The formula shows that the effective size is closer to the smaller than to the larger size in the cycle. Finally, N_e is affected by differences in sizes of the offspring groups between families (formula IV):

IV $N_e = 2N / (1 + (V_k/k))$,

where k and V_k are the mean and variance of progeny number per individual. The ratio N_e/N in this respect is about 0.75 in many organisms.

3.4.2.2 Assessment of genetically effective immigration

Immigrants may be less reproductively fit than native individuals (scale 0-1). Due to this, m, the genetically effective immigration may be different from the actually observed proportion that the immigrants constitute of the total spawning group. In assessing the m parameter one should observe the pitfalls and assumptions discussed in 3.3.1 and 3.3.2 (above).

3.4.2.3 Outline of an assessment of genetic parameters for fitness-related traits

Objective: To estimate genetic and environmental variance as well as genotype-environment interaction for fitness-related traits. The genotype-environment interaction on the quantitative context

is a manifestation of genetic adaptation. The present design will require the use of genetic markers (e.g., PCR microsatellites) to establish the full pedigree of the population under study, and hence represents a merging of quantitative and qualitative genetics techniques. It is realized that this approach is costly but it is important that correct procedures be involved from the start. A study of only fresh water stage related traits would be less expensive and could be performed in a much shorter time.
The specific goals of the present study would be to

* *Estimate the contribution of each stock and the families (individuals) within stocks in each river.*
* *Estimate the stock-by-river interaction and the family within stock interaction.*

The following experimental design assumes two rivers, each with a native salmon population.

1) Take a random sample of 50 females from each of two salmon stocks. Mate the females artificially using a nested mating design with one male to two females.

2) Mix all groups at the eyed egg stage and distribute the eggs from the mixed pool into the river-gravel of each river.

3) Sample 10-15 individuals from each full-sib group at the parr and/or smolt stage and identify pedigrees. At this stage one can estimate the parameters for e.g. growth rate and survival in the freshwater stage.

4) Recapture adults, take samples from each fish to identify pedigrees and record traits of interest.

5) Assess of the reproductive success of the returning adults by repeating point 3.

Comments: The design proposed here will involve unequal numbers for the stock, family and river effects. Sufficient numbers to test family and family-by-river effects are more feasible in the use of fresh-water traits than in the data on returned fish. Hence, the estimation of narrow-sense heritability and genetic correlations using sire-components of variance and covariance may be more meaningful in fresh-water traits than in traits measured on returned fish.

3.5 "Report on the status of selective breeding and genetic modifications relative to improving production traits such as growth performance, product quality, disease resistance, etc."

The information in this section is supplementary to that in section 2, which contains a general text on principles and practice in finfish and shellfish breeding. Relevant information is also found in Appendix 4.

3.5.1 Partial list of national Breeding Programs (as reported by WG members of ICES countries. Refer also to Appendix 4).

Norway
Pedigree systems involving two nucleus breeding stations each producing about 150 sib groups (full- and half-sib families) with pen-reared Atlantic salmon and rainbow trout. Selection is based on growth rate, low incidence of grilse, disease resistance and flesh colour. The program is jointly owned by farmers and corporations. The programs were established in the early 1970's and five generations of selection has been performed. Today about 75% of all Atlantic salmon farmed in Norway come from the national breeding programs.

Iceland
Sea ranching of 100 families of Atlantic salmon sib-groups (full- and halfsib families). Selection is for return rate and growth rate. The program was established in 1987. A similar program with an additional 100 families is used for land-based and pen-rearing systems.
A similar program is being initiated for increased growth rate and increased age at maturity in Arctic charr.

Sweden
Work is being done on rainbow trout and Arctic charr using full- and half-sib families.

Finland
About 300 full- and half-sib family work on rainbow trout established in the late 1980's.

Faroe Islands
About 150 full- and half-sib families of Atlantic salmon was initiated in 1991. The program has many similarities with those in Iceland and Norway.

France
Work on brown trout has been reported.

Canada
The program was initiated in the 1970's using wild stocks and became involved in sea-pen rearing in the 1980's. Four control and four selected lines with approximately 50 single-pair matings in each line have been formed in Atlantic salmon. Selection is based on rate of smoltification, low incidence of grilse, market size and resistance to bacterial kidney disease. The improved stocks are released to industry for multiplication each generation. A program using gene probes for the identification of families is under development for salmonids. Some work has been done in Pacific species.

U.S.A.
Catfish: Genetic selection programs are in progress at three locations to enhance growth rate and

disease resistance. Each program uses combined family and individual selection.Other research areas avtively being pursued are sex control methods to produce monosex populations, transgenic methodologies to enhance growth rate and disease resistance, and hybridization (interspecific and intraspecific). Most commercial broodstocks are maintained with random mating or phenotypic assortative mating based on fish size.
Pacific salmonids: Primary effort for Pacific salmonid breeding programs by federal and state egencies are direceted toward maintenance of genetic diversity and preservation of native stocks in the pacific northwest. One coho salmon broodstock is being maintained by a commercial operator using selection for increased growth rate at 18-months-of-age.
Rainbow trout: Federal and state agencies are emphasizing the production of wild broodstocks to manage natuarl fisheroes and to reduce both natural and artificial selection to the extent possible. The National Fish Hatchery program is not conducting any selection programs at this time. State fish agencies have limited selection programs which are typically directed at growth rate, disease resistance, and handling tolerance. The cmmercial trout growers conducy breeding programs primarily to enhance growth, disease resistance, and tolerance to handling stress. The use of sex control methodologies including mmonosex and ploidy are used in varying degrees by the commercial sector.
Striped bass: Most federal and state agencies and commercial growers rely on captured wild adults for broodstock. One commercial organization has established a domestic broodstock that has been in captive culture for two generations. Other breeding work has emphasized production of hybrids of striped bass male by white bass female. Development of breeding programs to support commercial production of striped bass are just beginning in the United States.

Poland
Work on rainbow trout with full-sib families.

3.6 "Evaluate the options for application of genetics research to fisheries and mariculture questions of concern to ICES."

The Working Group feels that, may be more then ever, management of the marine environment with its resources calls for genetic research and advice. The increasing demand for protein in the world can probably not be met by terrestrial agriculture. Since currently only 1% of the human protein harvest comes from the sea, it is anticipated that it is in the aquatic environment that protein production will expand most significantly. This expansion will probably be manifested both as an increased aquacultural production and as the utilization of untraditional finfish and shellfish resources.
In quantitative genetics, genetic advice will be necessary on **breeding programs** for marine species in culture and enhancements activities. In qualitative genetics there will be a continued need for efforts on **stock identification** in traditional fisheries. Also, the question of **mixed stock fisheries** will need further research activity and methodology development. Thus a common neeed in qualitative genetics is the continued efforts in **base-line studies** on the amount and distribution of genetic variability in relevant marine species. One area where **combined quantitative and qualitative genetics studies** are particularly relevant is in studies of **genetic interaction** between cultured and wild fish stocks. Related to the same problem is the need for assessments of reliability and genetic risks connected with various **sterilization techniques** (such as triploidy) when used in sea ranching. Observations that triploid Pacific oysters (*Crassostrea gigas*) showed substantial reversion to mosaics and diploids (in Delaware and Chesapeake Bay, USA) in a relatively short time, made the Working Group on Introductions and Transfers of Marine Organisms (on its meeting in April 1994) suggest that the WGAGFM takes this question under consideration.

A sound management of marine resources would benefit from the ability to predict potential genetic effects of various management actions. Hence it is anticipated that there will be an increased need for **mathematical simulation and -prediction models**. Such models can usually be constructed from existing genetics theory and formulae. Currently, however, their use is restricted due to a common lack of reliable estimates (and variances) of model parameter (population sizes, generation intervals, immigration rates, etc) which is needed for robust and realistic implementations. Carefully designed studies for estimating such model parameters thus deserve high priority.
While substantial work is going on in many of the areas mentioned above, the Working Group will point to one specific area where there is a strong and accumulated need to initiate research and discussions; the question of **genetic effects from selective fishing gear**. Exploring this area is likely to benefit from the joint efforts by qualitative and quantitative geneticists, and the Working Group suggests that this question is given priority in its 1995 Terms of Reference (refer to section 4.4).

4 WGAGFM SELF-EVALUATION OF STRUCTURE AND FUNCTION

4.1 Comments on present WG structure

Many of the current members of the WGAGFM were also members of the former Working Group on Genetics which was discontinued at the 81st Statutory Meeting in Dublin, 1993. In the discussion on WG structure the most common opinion was that the restructuring into separete subgroups for qualitative and quantitative genetics had been timely. The current structure more realistically reflects scientific demands of finfish and shellfish genetics, as well as differences in background/training and focus of interests among WG members. It was felt that the present structure would secure sufficient expertise to deal with all aspects of genetics. At the same time, the subgroup structure provides an efficient working forum among persons with the same scientific background. In this way the discussions in plenary sessions could benefit from a pre-clarification of the qualitative and quantitative aspects of the topics in the respective subgroups, and hence concentrate more on the common and principal aspects.
At this WG meeting this working form functioned well in practice. It is felt, however, that the plenary sessions should not be restricted too much. Being a complement to the informal discussions and personal contacts during the meetings, plenary sessions are very valuable for bringing the two fields of genetics onto a common platform and to merge their respective theories and methodologies, for the benefit of fisheries and mariculture.

4.2 Comments on travel funds for WG members

Most of the members expressed their worry about travel money for future Working Group meetings. This has important implications since without the possibility of coming together at least once between the annual Statutory Meetings, the Working Group system will not function. It is felt, therefore, that the securing of travel funds is a problem not only for the individual members and chairmen, but also for ICES and the respective public bodies of the member nations. It appears that some member countries have adopted specific fundings schemes which seem to function well, while other countries seem to have no policy at all regarding support of the Working Group system. The situation is particularly problematic for non-governmental scientists in the Working Groups. Those researchers contribute very significantly to the high scientific standard in ICES, and the the WG system cannot afford to loose them due to lack of national travels funds. The AGFM Working Group feel that the solution to this problem is a political question which should be addressed on the agenda for discussions between ICES and the respective member countries.

4.3 Comments on Terms of Reference for 1994

It was felt that the agenda of this first meeting of the WGAGFM was somewhat specific in that the practical establishing of the group(s) came in addition to the scientific work. It takes, necessarily, some time to establish good working routines in such situations, leaving less time to deal with the scientific questions of special interest to each member. Also, at this meeting, much time was spent on questions that were not raised by the WGAGFM itself. Thus, although the review work posed by the present Terms of Reference was regarded as timely and important, it was time-consuming and did not leave much time for dealing with other important questions of concern to this Working Group. It was concluded, however, that this can be remedied in the future if WGAGFM plays an active role in suggesting and formulating its Terms of Reference (refer to sections 3.6 and 4.4), and if the main annual WG meeting is expanded by at least one day.

4.4. Suggestions for WGAGFM Terms of Reference and meetings in 1995

The Working Group was very pleased with the arrangements and facilities offered by the Secretariat at ICES Headquarters in Copenhagen during its first WG meeting on March 9-11, 1994. The efficiency of the meeting benefitted greatly from the back-up by the very professional staff. It is therefore suggested that also the 1995 WG meeting is located to ICES Headquarters in Copenhagen, preferably at the end of January or the beginning of February, and that it should last for 4-5 days.

Referring to justifications made in section 3.6, the WG suggests that the following topics are included in the 1995 Terms of Reference:

1 - continue the review of knowledge of basic population genetic topics in fisheries and mariculture, with emphasis on a combination of qualitative and quantitative aspects.

2 - review the question of selective fishery with a view to propose studies to identify possible long term genetic effects.

3 - review sterilization techniques (such as triploidy) for use in mariculture and field experiments relative to efficacy and justifications for the techniques, and the risks involved (e.g., relative to reversion to a reproductive state).

4 - prepare updated protocols of fishery and mariculture genetic research in the member countries, and identify areas which can benefit from enhanced international cooperation.

APPENDIX 1

Review of the "Report of the Study Group on Genetic Risks to Atlantic Salmon Stocks" (C.M.1991/M:3).

Prepared by: The Working Group on the Application of Genetics in Fisheries and Mariculture (WGAGFM) during its meeting in Copenhagen, March 9-11, 1994.

For: The Study Group on Interactions on Wild, Ranched (Enhanced), and Cultured Salmon (refer to C.R. 2:27 of the 1993 Statutory Meeting).

WGAGFM reviewed the "Report of the Study Group on Genetic Risks to Atlantic Salmon Stocks" (C.M. 1991/M:3). Our viewpoints are expressed in the following section. The document was reviewed on a section by section basis (actual section headings are in italics below). Where sections are not listed, no changes or additions were put forward.

2.2 Evidence for Genetic Differentiation

2.2.1 Individual Genetic loci

Visual Polymorphisms: The presence of a spot on the tail is a variable character which may be genetically determined and is currently being investigated (Saunders, unpublished). A further potential polymorphism, albinism, is currently being investigated and may be a useful marker for population work (Friars, unpublished). Genetic bases of these variable traits remains to be verified.

Protein Polymorphisms: Considerable new information is available but only some of it is published. However, the new information does not change the generalizations made except to some extent with respect to b). "... within most moderate and large river systems." could be changed to say "...within even some small river systems."

Non-protein coding DNA: A large amount of new work has been done but not by WG people. Work is mostly in progess but from what is known of the results these tend to lead to the same types of generalizations as the allozyme data except that the levels of differentiation generally appear to be higher. The inability to detect differences at some loci does not mean that differences between different spatial or temporal samples do not exist. This needs to be stressed! Estimates can vary depending on which loci are used to assess gene flow. Thus large numbers of loci need to be used to derive accurate estimates. The validity of the approach to estimating gene flow based on levels of observed differentiation depends on the assumption that an equilibrium has been reached between the forces affecting genetic differentiation. If estimates are made prior to equilibrium, the gene flow will be overestimated.
No known update on rRNA gene work.

Mitochondrial Genome
Considerable population data is now available with respect to mtDNA. It is, however, still largely unpublished and not fully analysed. A subjective assessment of the data again points to the same basic pattern of geographical subdivision indicated by the allozyme data.

2.2.2 Chromosome Structure
New work has been done on Q-banding and C-banding and a polymorhism for C-banding has been revealed. Population data is unpublished but shows a level of population differentiation less than for allozymes.

2.2.3 Phenotypic Variation with a Genetic Base
The use of multivariate approaches to stock differentiation as is being carried out by researchers in the Pacific area, needs to be explored with respect to the genus Salmo.

2.3 Estimates of Genetic Exchange among Populations
(Refer to section on gene flow in the 1994 WGAGFM Report)

2.4 Evidence for Adaptive Genetic Differentiation
(Refer to section on combined studies of adaptation in the 1994 WGAGFM Report)
The polygenic basis of traits is an area that needs further research. Studies of epistatic interactions could be productive.

3 Transfer of Stocks

Iceland
Two stock transfers from Norway, in 1981 and 1985, involving several hundred thousand eggs have occurred and are used in land-based, contained farms.

Denmark
Since 1989, approximately 100,000 eggs from each of four European rivers (Corrib and Burrishole, Ireland, the Connon, Scotland and the Atran, Sweden) have been imported annually. These eggs are being used for stocking in a river restoration programme.

3.2 Scotland
Importation of large numbers of Canadian origin eggs from Tasmania has been carried out in 1993.

3.3 Canada
Importation of Land Catch eggs to Maine, involving three year classes, has been carried out. Requests for movement of the eggs into Canada are now being considered. Also increasing numbers of Atlantic salmon are now being reared on the Pacific coast.

4 Genetic Differences between wild and cultured salmon
Within stock variation should also be mentioned, maybe as a separate section after 4.1.1. This is important because stocks are not monotypic entities. In general, both quantitative and qualitative studies show that most species variation is found within stocks and this may in itself be adaptive.

4.1.2 Selective Breeding
Genetic variance for disease resistance is being utilized in breeding programmes.
Genetic interdependence - "Genetic correlations between traits are being considered in many multi-objective selection programmes.

The term "selective breeding" may not be the best one to use here. Breeding encompasses mating and selection systems. Both must be considered simultaneously. The use of "selective breeding" ignores the mating aspect.

5 Biochemical Genetic Techniques

5.1.2 Genetic markers

This section is confusing and inconsistent. Some statements are inaccurate and inappropriate. Two particular samples of the latter are "However, the majority of polymorphisms....are considered neutral or subject to purifying selection...". and "....it is usual to find the majority of all possible alleles in a given population...".

5.2 Methods

5.2.1 Allozymes
A number of new polymorphisms have been detected but the assessment remains essentially the same.

5.2.2 Chromosomes
The limited amount of new work carried out confirms this assessment.

5.2.3 Mitochondrial DNA - restriction enzymes.
This procedure is being superceded by the methods described in 5.2.4. Most variation is revealed using 4-base restriction endonucleases. However, these produce complex fragment patterns which are difficult to screen electrophoretically in a consistent, repeatable manner. Also, it requires high quality intact mtDNA molecules in a highly purified form; a difficult and time consuming procedure.

5.2.4 Mitochondrial DNA - PCR analysis
The use of this method has expanded through the development of new primer pairs for various regions of the mtDNA genome. This is particularly true with respect to restriction digestion of PCR amplified sections of mtDNA using 4-base restriction endonucleases which reveal the most variation. However, unpublished results for Atlantic salmon suggest that the numbers of haplotypes is still expanding, increasing the scope for finding population differences and using the variation to mark experimental stock groups. Further variation have been found in the 16sRNA/ND1 and ND5/ND6 parts of the mtDNA genome. Continental stock differences have been found in the 16sRNA/ND1 region, in addition to those previously identified for cytochrome B, as have major differences between southern and northern European stocks. Differences in haplotype frequencies have also been found been regional stocks and between farm and wild stocks in some situations.
Numbers of samples which can be typed per day range between 40 and 60.

5.2.5 Minisatellite probes - fingerprinting
No new substantive developments with respect to salmon. Most effort is now directed at the use of single locus mini- or micro- satellite probes.

5.2.6 Minisatellite probes - single locus probes

Substantive developments have occurred with respect to the number of loci for which probes have now been developed. The new probes developed reveal a range of levels of heterozygosity and population differentiation. Individual family identification in many experimental contexts is now feasible though the proceedures are time intensive. Realistic numbers of fish which can be analysed per laboratory worker is 40 per week (based on need to use 6-7 probes) for fingerprinting.

5.2.7 Additional techniques

Micro-satellite probes. These are like minisatellite probes but relate to shorter repeat sequences. They are also found to be highly variable in most cases. DNA probes have been developed but by sequencing these shorter pieces of DNA, primers have also been developed for a number of micro-satellite DNA regions. These allow PCR amplification to be used in the detection of FLP's. A large number of microsatellite probes have been developed in Leicester (T. Burke, Department of Zoology, University), Belfast (A. Ferguson, Queen's University) and Halifax (R. Doyle, Marine Gene Probe Laboratory, Dalhousie University). PCR based detection of variation has just started. Little of the microsatellite work in Atlantic salmon has yet been published.

FLP's of single copy nDNA. Primers are now available to amplify regions of the DNA coding for structural genes. This allows detection of FLP's, as well as RFLP's, and variation by direct sequencing of amplified fragment. Studies of other species look promising. Application of this work to Atlantic salmon is only just starting.

5.3 Conclusions

The new developments mean that there is now no methodological impediment to the search for molecular markers. However, the exact number of regions which could now be screened and the extent to which these nuclear DNA regions offer useful variation for studies remains to be fully assessed.

7 Design of experimental studies

No comments since the actual designs used are often dictated by the available stocks and molecular markers as well as other biological and logistical constraints.

9 Conclusions

The commercial use of triploid salmon has not progressed at all in the past 2 years. This is largely due to the public perception of the use of genetically altered animals as a source of food and, from the point of view of farmers, the absence of the prematuration growth spurt. Studies of the potential ecological interactions of triploid escapes with wild salmon are needed.

APPENDIX 2

National Status Reports - Summary of Research Relevant to Genetic Interactions of Native and Non-native Salmonids.

Prepared by: The Working Group on the Application of Genetics in Fisheries and Mariculture (WGAGFM) during its meeting in Copenhagen, March 9-11, 1994.

For: The Study Group on Interactions of Wild, Ranched (Enhanced), and Cultured Salmon.

Canada - (Reported by Gerry Friars)

LABORATORY/RESEARCHER: Atlantic Salmon Federation and University of New Brunswick, J. Anderson and T. Dilworth
SPECIES: Atlantic salmon
PROJECT FUNDING: Atlantic Fisheries Adjustment Program
OBJECTIVE: to evaluate the incidence of escaped aquacultural and wild fish ascending the Magaguadavic River.
DESIGN: monitoring of ascending fish
METHODOLOGY: sonic tags used to track individual fish on spawning grounds. Discrimination of cultured and wild fish using scales, fins and DNA analysis (in conjunction with Dalhousie University).
STATUS: ongoing

Denmark - (Reported by Michael Hansen)

Study 1
LABORATORY/RESEARCHER: Univ. of Århus, Inland Fisheries Lab., Silkeborg, & Queens Univ. Belfast. Michael M. Hansen, Volker Loeschcke, Rosaleen Hynes.
SPECIES: Brown trout
PROJECT FUNDING: The Danish Natural Science Research Council and the Inland Fisheries Laboratory.
OBJECTIVE: Determine the origin of the trout in the re-populated Karup river system.
DESIGN: Compare genetic characteristics of samples from the main river, four tributaries, another river and a hatchery strain.
METHODOLOGY: mtDNA RFLP analysis, involving Southern Blotting and hybridisation to a brown trout mtDNA probe. All fish were screened with 7 restriction endonucleases (2 six-, 2 five-, and 3 four-base cutters).
STATUS: Project initiated Jan. 1992. One paper on results submitted. The project is continuing to address further questions. Methodology is changed to PCR. RFLP analysis (2 restriction enzymes) of PCR amplified ND-1 segment distinguish the four most informative haplotypes.
COMMENTS: Hatchery strain haplotype frequencies significantly different from all other samples. The genetic contribution of the stocked hatchery trout very small or absent. Present population probably descendants of an indigenous population.

Iceland - (Reported by Anna Danielsdottir)

Proposed Study
LABORATORY/RESEARCHER: Marine Research Institute, Reykjavik, A.K. Danielsdottir
SPECIES: Atlantic salmon
PROJECT FUNDING: currently being sought
OBJECTIVE: to assess the genetic interactions between wild and farmed Atlantic salmon in the river.
DESIGN: Assess the genetic structure in the R. Ellidaa in different years and evaluate in relation to levels of farm Atlantic salmon in the river as determined from scale studies.
METHODOLOGY: isozymes, scale analysis
STATUS: proposed work

Ireland - (Reported by Tom Cross)

Study 1
LABORATORY/RESEARCHER: T. Cross, University College, Cork (UCC), with two UK and one Spanish group.
SPECIES: Atlantic salmon
PROJECT FUNDING: EC FAR MA-2-480
OBJECTIVE: to detect molecular markers to distinguish wild and reared Atlantic salmon.
DESIGN: Four index samples from Spanish, Scottish and Irish wild populations and a Norwegian-origin farmed stock typed for variation at the four systems listed below and results compared individually and in combination.
METHODOLOGY: Allozymes (UCC), plus minisatellite nuclear DNA, mitochondrial DNA and chromosomes.
STATUS: Two year project completed January 1993.
COMMENTS: The results of this work have been employed in a number of opportunistic and experimental studies designed here under the EC AIR1-CT92-0719 programme.

Study 2
LABORATORY/RESEARCHER: Salmon Research Agency of Ireland, T. Cross (UCC) and P. McGinnety, plus two UK and two Spanish.partners.
SPECIES: Atlantic salmon
PROJECT FUNDING: EC AIR1-CT92-0719
OBJECTIVE: To study the interaction of wild and reared Atlantic salmon in the field using molecular markers.
DESIGN: Field experiment where farmed and wild Atlantic salmon and their hybrids, typeable as to family using genetic profiling, are introduced as eyed eggs into enclosed stretch of river and relative fitness assessed in terms of perfomance. Also retropective study of Atlantic salmon populations effected by escapes from a hatchery and sea cages, using an array of molecular methods.
METHODOLOGY: Minisatellite DNA SLPs, transcribed genes, mtDNA and allozymes.
STATUS: In progress, 3 years from OCD of February 1993.
COMMENTS: Experimental and opportunistic studies.

Study 3
LABORATORY/RESEARCHER: T. Cross (UCC) and A. Ferguson (QUB, Belfast, UK)

SPECIES: Atlantic salmon
PROJECT FUNDING: Electricity Supply Board (ESB)
OBJECTIVE: to assess the effect of stocking practises on the population genetics of River Shannon Atlantic salmon.
DESIGN: Survey of distribution of genetic variation
METHODOLOGY: Allozymes and minisatellite DNA SLPs.
STATUS: Three year study completed in December 1993.
COMMENTS: Part of larger Atlantic salmon biology study on Shannon where extensive mitigation stocking has been used for over 30 years because of interuption of runs by hydroelectric development.

Study 4
LABORATORY/RESEARCHER: N. Wilkins, University College, Galway (UCG)
SPECIES: Atlantic salmon
PROJECT FUNDING: ESB
OBJECTIVE: Breeding of improved strains for enhancement in the River Shannon
DESIGN: Selection of broodstock of grilse or MSW type to use in stocking different areas.
METHODOLOGY: Directed breeding.
STATUS: Three year study completed in December 1993.

Study 5
LABORATORY/RESEARCHER: N. .Wilkins (UCG)
SPECIES: Atlantic salmon
PROJECT FUNDING: commercial
OBJECTIVE: to produce triploids to prevent farm/wild breeding interactions
METHODOLOGY: triploid induction by heat/pressure shock, study of sea migration of triploids
STATUS: ongoing

Study 6
LABORATORY/RESEARCHER: Mr. P. McGinnety (SRA)
SPECIES: Atlantic salmon
PROJECT FUNDING: Institutional
OBJECTIVE: assess the effects of selection on MEP-2* in culture
METHODOLOGY: allozymes; growth and maturation assessment
STATUS: on-going

Norway - (Reported by Geir Dahle, Knut E. Jørstad and Jarle Mork)

Study 1
LABORATORY/RESEARCHER: The Norwegian Institute of Nature Research (NINA), K. Hindar
SPECIES: Atlantic salmon
PROJECT FUNDING: Norwegian Research Council program: "Environmental Effects of Biotechnology".
OBJECTIVE: Using data on gene flow from cultured fish into natural populations to calibrate mathematical models for predicting the effect of various immigration regimes on the genetic variation within and between natural populations.
DESIGN: A genetically characterized set of Atlantic salmon populations is challenged by increased immigration of genetically tagged farmed Atlantic salmon. Baseline, post-challenge and F1 allele frequencies are compared.

METHODOLOGY: Physical and genetic tagging, electrophoresis, mathematical modelling.
STATUS: Started in 1994.

Study 2
LABORATORY/RESEARCHER: I. Fleming and B. Jonsson, NINA (Ims Salmon Research Station)
SPECIES: Atlantic salmon
PROJECT FUNDING: Norwegian Research Council (Program "Environmental effects of aquaculture" 1992-1994).
OBJECTIVE: Assessment of the reproductive behaviour and success of farmed, ranched and wild Atlantic salmon in the presence and absence of competiton with other types.
DESIGN: Artificial spawning areas at Ims.
METHODOLOGY: Ethological and biological studies
STATUS: Started in 1992, on-going; results will be reported from 1994 and on.

Study 3
LABORATORY/RESEARCHER: I. Fleming, B. Jonsson, K. Hindar, I.B. Mjølnerød, NINA.
SPECIES: Atlantic salmon
PROJECT FUNDING: NINA and The Norwegian Research Council (Program "Environmental effects of aquaculture" 1992-1994).
OBJECTIVE: Assessment of interaction of wild, ranched, and cultured Atlantic salmon
DESIGN: Genetically marked fish are allowed to spawn above the fish trap on the River Imsa.
METHODOLOGY: Carlin tags, biopsy, electrophoresis
STATUS: Started in 1993, results available in 1995.

Study 4
LABORATORY/RESEARCHER: T. G. Heggberget, NINA.
SPECIES: Atlantic salmon
PROJECT FUNDING: The Norwegian Research Council (program "Environmental effects of aquaculture" 1992-1994), The Directorate for Nature Management..
OBJECTIVE: To study the behaviour of escaped farmed Atlantic salmon.
DESIGN: Tracking of farmed fish during upstream spawning migration in Rivers Alta, Namsen, and Imsa.
METHODOLOGY: Radio-tagging and -tracking
STATUS: On-going, results to be reported when available.

Study 5
LABORATORY/RESEARCHER: Inst. of Marine Research Bergen, O. Skaala
SPECIES: Atlantic salmon
PROJECT FUNDING: Norwegian Research Council
OBJECTIVE: Measurement of gene flow from cultivated to wild populations by conducting field experiments using genetic markers.
DESIGN: Identify genetic markers in Atlantic salmon family groups and release genetically tagged Atlantic salmon smolts into a salmon river. Estimates of reproductive success of returning spawners.
METHODOLOGY: Allozyme markers; the genetic composition of the native populations of Atlantic salmon will be determined, spawning population estimates and abundance will be estimated and the genetic composition of offspring populations determined.
STATUS: Initiated in 1992, first estimates of returning, genetically marked fish in 1994 and possibly information on reproductive success in 1995.
COMMENTS: DNA analyses (micro-satellites) will be incorporated; population dynamics parameters are included in study. Results for use in modelling impacts from genetically modified organisms.

Study 6
LABORATORY/RESEARCHER: Inst. of Marine Research, Bergen, O. Skaala
SPECIES: Brown trout
PROJECT FUNDING: Norwegian Research Council
OBJECTIVE: to use trout as a case study of genetic interactions between stocks and estimate gene flow into a native population
DESIGN: Genetically tagged farmed spawners will be released into the experimental river and the genetic make-up of subsequent juveniles monitored at different ages.
METHODOLOGY: Tagging is based on a morphologic/visual marker (fine spotted body) and allozymes. Population dynamics and abundance estimates of year classes will be determined.
STATUS: Initiated in 1989 and introduced genes present in the 1990 year class. Monitoring in year classes is on-going.
COMMENTS: Estimates of reproductive success of farmed trout is about 25% less than local wild spawners. Higher mortality rates detected for offspring (all forms) carrying the introduced marker gene.

Study 7
LABORATORY/RESEARCHER: Univ. of Trondheim, Biological Station. J. Mork.
SPECIES: Indifferent (but e.g., salmon)
PROJECT FUNDING: University of Trondheim
OBJECTIVE: General, interactive PC simulation program for, e.g., prediction and analysis of genetic effects of interaction between cultured and wild populations.
DESIGN: Simultaneous handling of combined genetic effects from random genetic drift, gene flow (immigration), and selection on a genetically pre-characterized set of populations.
METHODOLOGY: Theory, mathematical modelling, computer, Monte Carlo simulations
STATUS: Functional version is being tested
COMMENTS: Practical implementation of the model has revealed a scarcity of high-quality estimates of e.g., effective population sizes, effective gene flow, fitness coefficients etc in the literature.

Scotland - (Reported by Eric Verspoor)

Study 1
LABORATORY/RESEARCHER: SOAFD Marine and Freshwater Fisheries Laboratories, E. Verspoor and collaborators
SPECIES: Atlantic salmon
PROJECT FUNDING: SOAFD and EC AIR1-CT92-0719
OBJECTIVE: to assess whether the documented interbreeding of farm Atlantic salmon, which ascended the River Polla in 1989 and 1990, with the wild stock has resulted in genetic changes to the juvenile populations in the river.
DESIGN: Samples of juvenile Atlantic salmon from two year classes were collected from the lower, middle and upper reaches of the river pre-spawning of farm fish in 1989. The genetic composition of these fish will be compared with post spawning juvenile samples from the same locations taken in 1991 and differences related to the genetic make-up of adult farm Atlantic salmon ascending the river.
METHODOLOGY: Allozymes, RFL analysis of PCR amplified mtDNA, mini- and micro-satellite analysis of nuclear DNA, PCR amplification of structural gene nDNA.
STATUS: Currently underway and due for completion in 1995.
COMMENTS: This represents an opportunistic study.

Study 2
LABORATORY/RESEARCHER: SOAFD Marine and Freshwater Fisheries Laboratories, E. Verspoor and collaborators
SPECIES: Atlantic salmon
PROJECT FUNDING: SOAFD and EC AIR1-CT92-0719
OBJECTIVE: to assess whether genetic differences among stocks are relevant to their biological performance during the juvenile freshwater phase in the wild in ways that relevant to their fitness.
DESIGN: Simultaneously spawned eggs of different regional stocks and their hybrids have been planted out within 48 hrs of fertilization in artificial redds using a random stratified planting strategy in a small experimental river where natural spawning is precluded. Performance parameters such as egg mortality, hatching and emergence timing, developmental state, maturation, growth and smoltification will be compared among groups.
METHODOLOGY: Stock groups will be genetically marked using RFLP's PCR amplified mtDNA and single locus minisatellite fingerprinting.
STATUS: Currently underway and due for completion in 1996.
COMMENTS: This represents an experimental study.

Study 3
LABORATORY/RESEARCHER: SOAFD Marine and Freshwater Laboratories, A. Youngson, J. Webb
SPECIES: Atlantic salmon
PROJECT FUNDING: SOAFD and Atlantic Salmon Trust
OBJECTIVE: to determine the geographical distribution and relative extent of spawning of escaped Farm Atlantic salmon in Scottish rivers.
DESIGN: sampling of eggs in salmon redds in lower reaches of representative rivers across Scotland.
METHODOLOGY: Identification of farm eggs on basis of canthaxanthine; allozyme analysis to detect hybrids and trout eggs.
STATUS: Completed in 1992. Two papers have been produced (below).
COMMENTS: This represents an opportunistic study.

Webb, J.H. et al. 1993. Spawning of escaped farmed Atlantic Atlantic salmon, Salmo salar L., in western and northern Scottish rivers: egg deposition by females. Aquaculture and Fisheries Management 24: 663-670.

Youngson, A.F. et al. 1993. Spawning of escaped farmed Atlantic Atlantic salmon (Salmo salar): hybridization of females with brown trout (Salmo trutta). Can. J. Fish. Aquat. Sci. 50: 1986-1990.

Study 4
LABORATORY/RESEARCHER: SOAFD Marine and Freshwater Laboratories, A. Youngson, J. Webb
SPECIES: Atlantic salmon
PROJECT FUNDING: SOAFD and Atlantic Salmon Trust
OBJECTIVE: to determine the frequency of farm escapes among Atlantic salmon in the coastal Atlantic salmon fisheries in Scotland.
DESIGN: regular sampling of Atlantic salmon taken by four representative coastal net fisheries off Scotland.
METHODOLOGY: Identification of farm fish on basis of body morphology and scale analysis.
STATUS: Started in 1992 and ongoing.
COMMENTS: This represents an opportunistic study.

Study 5
LABORATORY/RESEARCHER: SOAFD Marine and Freshwater Laboratories, E. Verspoor
SPECIES: Atlantic salmon
PROJECT FUNDING: SOAFD
OBJECTIVE: to determine the nature and extent of population structuring of Atlantic salmon in Scotland within and among rivers so as to assess the impact of farm escapes on natural structure.
DESIGN: geographic sampling of within and among river genetic variation and statistical analysis of differentiation.
METHODOLOGY: Allozymes, mtDNA, mathematical modelling.
STATUS: Started in 1989 and ongoing.

Spain - (Reported by Eric Verspoor)

Study 1
LABORATORY/RESEARCHER: Xunta de Galicia, Lurizan, C. Garcia de Leaniz and collaborators
SPECIES: Atlantic salmon
PROJECT FUNDING: Xuntia de Galicia and EC AIR1-CT92-0719
OBJECTIVE: to assess if genetic differences between native Spanish and non-native northern European Atlantic salmon affect the fitness of the two stock types in the wild in Spanish rivers.
DESIGN: Pure stock groups of Spanish and Scottish Atlantic salmon ova will be planted out at the eyed egg stage in the River Ulla in Spain and various performance parameters compared e.g. egg and juvenile mortality, hatching and emergence timing and condition, smolt production and growth performance.
METHODOLOGY: Non-native stock groups will be artificially selected to fix a RFLP variant detected in a PCR amplified mtDNA fragment which is absent in Spanish stocks.
STATUS: Currently underway and due for completion in 1995.
COMMENTS: This represents an experimental study.

Study 2
LABORATORY/RESEARCHER: Xunta de Galicia, Lurizan, C. Garcia de Leaniz
SPECIES: Atlantic salmon
PROJECT FUNDING: Xuntia de Galicia and EC AIR1-CT92-0719
OBJECTIVE: to assess differences in freshwater peformance and return rate among native Spanish and non-native northern European Atlantic salmon stocks planted out a pre-smolt juveniles in the River Eo.
DESIGN: Pure and hybrid stock groups of Spanish and northern European Atlantic salmon reared in the hatchery to the pre-smolt stage have been released into the river. Mortality and growth are being compared and return rates to the angling fishery will be monitored.
METHODOLOGY: Stock groups will be microtagged and adipose fin clipped. Fish will be monitored in freshwater by electrofishing stocked areas. Returning adults will be monitored with respect to tagged fish caught in subsequent years by anglers.
STATUS: Currently underway and due for completion in 1995.
COMMENTS: This represents an experimental study.

Study 3
LABORATORY/RESEARCHER: University of Oviedo, E. Garcia Vasquez
SPECIES: Atlantic salmon
PROJECT FUNDING: EC AIR1-CT92-0719
OBJECTIVE: to determine if native Spanish stocks have been genetically changed as a result of historical stocking with imported Atlantic salmon ova from northern Europe.
DESIGN: data on population numbers, stocking levels and allozyme variation in Atlantic salmon from stocked and unstocked Atlantic salmon rivers in northern Spain will be assessed in relation to allozyme variation in northern European rivers to determine whether genetic changes have taken place due to stocking.
METHODOLOGY: Analysis of existing data bases.
STATUS: Currently underway and due for completion in 1995.
COMMENTS: This represents an opportunistic study.

APPENDIX 3

Review of the Report of the Study Group on Stock Identification Protocols for Finfish and Shellfish Stocks (ICES C.M.1993/M:3).

Specific comments on the report (Where no comments were considered necessary, the relevant section is not listed).

In section 2.1 it is stated at the end of the first paragraph that gene flow is greater between marine fish populations than between freshwater ones. The Working Group are unaware of experimental results which confirm this statement and suggest that "genetic differentiation" be used instead of "gene flow". We also doubt the next statement that "In principle, it is possible to find unique genetic markers--". We regard this as unlikely in conspecific populations, though unique *genotypes* may occur.

Paragraph 4 of the same section describes stock composition analysis as consisting of three steps, viz. establishment of a baseline, sampling of a mixed fishery and estimate of proportions using a mathematical algorithm. This defines the GSI method, not stock identification in general, which involves only the first of these steps. This paragraph should be modified and perhaps would be better positioned in section 2.2.

The section on allozymes is very brief and selective relative to that on mtDNA which is extremely detailed and specific. In the section on nuclear DNA, we feel that the sections on minisatelite and microsatellite DNA should be expanded. We also suggest that a reference be provided for the use of transposon polymorphism in salmon. Further, we feel that the potential of Randomly Amplified Polymorphic DNA (RAPD) analysis should be cited and that the the rapid expansion of the molecular field, providing extra methods with potential in this area, should be stressed.

Section 2.2 describes just two statistical techniques rather than providing a comprehensive review. The need for more and better statistical techniques should also be highlighted. Most fisheries laboratories have highly competent statisticians. Advice from such experts should be sought in the design and analysis of results from genetic stock discrimination programs.

While acknowledging the utility of the multidisiplinary approach outlined in section 2.3, it should be recognised that different methods define different types of "stock". Some non-genetic methods are strongly influenced by the environment and thus define "management units" which are not necessarily distinct biological populations. In contrast, genetically defined "stocks" will be largely non-interbreeding populations. Thus it is important to use a method of statistical analysis which allows the relative contribution of the various techniques to be quantified. Also, different techniques are more efficent in specific circumstances. This cannot be predicted in advance so some kind of preliminary survey is advisable.

In relation to point 4.1 we feel, contrary to the statement therein, that the topic of fishing as an agent of selection is extremely important and should receive high priority in future ICES deliberations. To highlight our concern, we intend to include this item on the agenda for our next meeting. We also feel that the idea of a stock identification manual is a good one and would be happy to assist with the provision of any genetic advice required.

Appendix 4

National reports on qualitative and quantitative genetic research activities in finfish and shellfish. Refer to Appendix 2 for studies specifically concerning interactions between native and non-native populations of fish.

CANADA

Study 1

LABORATORY/RESEARCHER: C.T. Taggart (Dept. of Fisheries and Oceans, NW Atlantic Fisheries Centre, St. John's, NF) in collaboration with R. Doyle (Marine Gene Probe Laboratory, Dalhousie University), P. Bentzen (University of Washington), and G. Pogson (Cambridge University).
SPECIES: Atlantic cod (*Gadus morhua*)
PROJECT FUNDING: Northern Cod Science Programme (Dept. Fish. Oceans).
OBJECTIVE: Elucidating stock structure of Atlantic cod in the Newfoundland Region (NAFO Division 2J3KLNO, 3M).
DESIGN: Application of nuclear DNA gene probes to using both contemporary tissues (1992-1994) and PCRable historical (1947-1990) preserved tissues.
METHODOLOGY: Nuclear DNA microsatellite gene probes (VNTR's - variable number of tandem repeats) and cDNA gene probes (RFLP's - restriction fragment length polymorphisms).
STATUS: Ongoing research begun in 1992 and scheduled for completion in April 1995.
COMMENTS: Preliminary results of the work show both techniques are capable of revealing fine-scale population structure and have been presented at a variety of national and international symposia. Preliminary results have also been used to design a dedicated one-year IFRP (Interim Funding Research Proposal) partnership research programme to conduct a comparative genetic analysis of stock structure in NW Atlantic cod (Labrador to georges Bank).

Study 2

LABORATORY/RESEARCHER: C.T. Taggart (Dept. of Fisheries and Oceans, NW Atlantic Fisheries Centre, St. John's, NF), and R. Doyle (Marine Gene probe Laboratory, Dalhousie University).
SPECIES: Atlantic cod (*Gadus morhua*).
PROJECT FUNDING: Ocean Production Enhancement Network (OPEN; one of Canada's Networks of Centre of Excellence).
OBJECTIVE: (i) test the hypothesis that in cod larvae, particular groups of siblings of genetically identifiable parentage will show better growth and survival performance relative to other sibling groups;
(ii) to assess the feasability of using genetic "fingerprints" to index hetrozygosity or to develop a set of genetic probes to directly index heterozygosity; (iii) track cohorts of cod larvae in the field and assess the evolving genotypic character of the survivors over a period of ~3 weeks; (iv) to relate the time-varying genotypic character to variations in the biotic/abiotic environment.
DESIGN:
METHODOLOGY: Nuclear DNA microsatellite gene probes (VNTR's - variable number of tandem repeats).
STATUS: Research begun in 1991 and scheduled for completion in September 1994.
COMMENTS: Preliminary results are: 1) the microsatellite gene probes developed by MGPL identified cod pedigrees in the lab (and field) using very small amounts of tissue (larval eyeballs). The high allelic diversity of the probes has proven to be excellent for genetically identifying individuals, families and populations. 2) Larvae from some parents show significantly better growth and survival

performance. The genetic associations between survial, larval weight, and other traits (e.g. lipid condition) are currently beig assessed. 3) Mathematical procedures have been developed which are able to use the extraordinary high microsatellite heterozygosities to detect naturally-spawned family groupings in larval cod populations in the oceanic environment. 4) Early analysis of microsatellite data from individuals from a larval cohort tracked for a period of 20 days in the field revealed consderable spatial heterogeneity and changes through time and some consistencies with laboratory study results that suggest that few of the many larval groups or sub-populations (cohorts) produced annually make significant contributions to recruitment.

Study 3
LABORATORY/RESEARCHER: C.T. Taggart (Canada Dept. of Fisheries and Oceans, NW Atlantic Fisheries Centre, St. John's, NF), R. Doyle, and J. Wright (Marine Gene Probe Laboratory, Dalhousie University), S. Carr (Memorial University of Newfoundland), and K. Zwanenberg (Bedford Institute of Oceanography).
SPECIES: Atlantic cod (*Gadus morhua*)
PROJECT FUNDING: Interim Funding Research Proposal (interim extension of Oceanic Production Enhancement Network - OPEN).
OBJECTIVE: Focus on assessing, in a comparative way, the relative utility of three different genetic techniques to differentiate and characterize the contemporary cod stock/population structure in the NW Atlantic continental shelf regions from Labrador to georges Bank, as well as the Spring/fall spawning components of the eastern Scotian Shelf "stock".
DESIGN:
METHODOLOGY: Nuclear DNA microsatellite gene probes (VNTRs - variable number of tandem repeats), nuclear cDNA gene probes (RFLPs - restriction fragment length polymorphisms), mtDNA cytochhrome-b sequence variations.
STATUS: Research to begin in June 1994 and scheduled for completion in August 1995.
COMMENTS: IFRP proposal has been completed and sample collections are underway. Research programme is expected to begin as scheduled.

DENMARK

Study 1
LABORATORY/RESEARCHER: Lars-Erik Holm, National Institute of Animal Sciences.
SPECIES: Rainbow trout.
PROJECT FUNDING:
OBJECTIVE: Development of microsatellites to be used for identification of hatchery strains and (hopefully) for markers of commercially important traits.
DESIGN:
METHODOLOGY: Screening of library with relevant oligonucleotides, sequencing of positive clones.
STATUS: Ongoing.

Study 2
LABORATORY/RESEARCHER: Einar Nielsen, Dept. of Ecology and Genetics, University of Aarhus.
SPECIES: Atlantic salmon
PROJECT FUNDING:
OBJECTIVE: To determin if the Skjerm River Atlantic salmon population (the only natural salmon

population to have survuved in Denmark) has suffered a severe loss of genetic variation and to determin the genetic relationships to other salmon populations. The project also aims at finding genetic markers to trace the contribution of other salmon stocks being introduced to Denmark.
DESIGN:
METHODOLOGY: RFLP analysis of PCR amplified mtDNA segments, microsatellites.
STATUS: Ongoing.

Study 3
LABORATORY/RESEARCHER: M.M. Hansen, Dept. of Ecology and Genetics, University of Aarhus, and The Inland Fisheries Laboratory, Silkeborg.
SPECIES: Brown trout
PROJECT FUNDING:
OBJECTIVE: Sssessment of genetic differentiation among unstocked brown trout populations and the effects of stocking activity.
DESIGN:
METHODOLOGY: Allozyme electrophoresis, RFLP analysis of PCR amplified mtDNA segments, microsatellites.
STATUS: Ongoing.

Recent Danish publications of relevance are:

Hansen, M.M., Loeschcke, V., Rasmussen, G, and Simonsen, V. 1993. Genetic differentiation among Danish brown trout (Salmo trutta) populations. Hereditas 118, 177-185.

Hansen, M.M., and Loeschcke, V. 1994. Effects of the release of hatchery-reared brown trout to natural trout populations. In: Loeschcke, V., Tomiuk, J., and Jain, S.K. (eds.) Conservation Genetics. Birhause, Basel, pp. 273-289.

FINLAND

Study 1
LABORATORY/RESEARCHER: M.-L. Koljonen, J. Koskiniemi, L. Siitonen, Finnish Game and Fisheries Research Institute, Helsinki.
SPECIES: Baltic salmon, brown trout
PROJECT FUNDING: In house
OBJECTIVE: Mapping genetic resources (concervation and enhancement projects)
DESIGN: Mapping allele frequencies
METHODOLOGY: Allozymes
STATUS: Ongoing

Study 2
LABORATORY/RESEARCHER: M.-L. Koljonen, J. Koskiniemi, L. Siitonen, Finnish Game and Fisheries Research Institute, Helsinki.
SPECIES: Atlantic salmon
PROJECT FUNDING: In house
OBJECTIVE: Genetic stock identification (GSI)
DESIGN:

METHODOLOGY: Allozymes
STATUS: Ongoing

Study 3
LABORATORY/RESEARCHER: K. Elo, Agricultural Center, Dept. of Animal Breeding
SPECIES: Coregonids
PROJECT FUNDING: In house
OBJECTIVE: Species identification, phylogenetic analyses, and genomic variation in Coregonids
DESIGN: Gene frequency mapping
METHODOLOGY: RAPD
STATUS: Ongoing

Study 4
LABORATORY/RESEARCHER: J. Vuorinen, University of Joensuu, Dept. of Biology.
SPECIES: Coregonids
PROJECT FUNDING: In house
OBJECTIVE: Coregonid evolution
DESIGN: Mapping gene frequencies
METHODOLOGY: Electrophoresis
STATUS: Ongoing

Study 5
LABORATORY/RESEARCHER: H. Mölsä, University of Kuopio, Dept. of Physiology.
SPECIES: Rainbow trout
PROJECT FUNDING: In house
OBJECTIVE: Growth hormone gene transfer
DESIGN:
METHODOLOGY:
STATUS: Ongoing

Recent Finnish publications of relevance are:

Bodaly, R.A., Vuorinen, J.A., reist, J.D. and Reshetnikov, Y.S. 1994. Genetic relationships of five Siberian coregonid fishes. J. Fish. Biol. (in press).

Elo, K., Vuorinen, J.A., and Niemelä, E. 1994. Genetic resources of Atlantic salmon (*Salmo salar* L.) in Teno and Näätämö Rivers, northernmost Europe. Hereditas 120 (in press).

Elo, K. 1993. Gene flow and concervation of genetic variation in anadromous Atlantic salmon (*Salmo salar* L.). Hereditas 119: 149-159.

Koljonen, M.-L. and Huusko A. 1993. Genetic variation of brown trout stocks in the Koutajoki rivers system. Oulanka Reports 12: 131-143.

Koljonen, M.-L. 1993. Genetic stock composition analyses of Baltic salmon catches. ICES, C.M. 1993/M:28.

Vuorinen, J.A., Bodaly, R.A. and Reist, J.D. 1993. Genetic and morphological differentiation between dwarf and normal size forms of lake whitefish (Coregonus clupeaformis) in Como Lake, Ontario. Can. J. Fish. Aquat. Sci. 50: 210-216.

ICELAND

Study 1
LABORATORY/RESEARCHER: E. Eythorsdottir, Agr. Research Institute, Reykjavik
SPECIES: Arctic charr
PROJECT FUNDING: The National Research Council in Iceland
OBJECTIVE: To determine genetic parametrs, i.e. heritability and genetic correlations of economically important traits of Arctic charr in aquaculture. The results will be utilized in a national breeding program of Arctic charr.
DESIGN: Data collected from charr in the first 2 or 3 year classes of the Arctic charr breeding program. Each year class is made up of 100-120 full sib families, with two or three families per sire. The families are reared for two and a half year from hatching. Data are collected on growth, sexual maturity at different life stages, slaughter weight, visceral fat, flesh coloration and possibly fat content of fish.
METHODOLOGY: Data are analyzed with standard methods in animal breeding.
STATUS: Project started in 1993 and is planned for 4 years.
COMMENTS: The project is a cooperation between the Agricultural Research Institute and the Agricultural School at Holar in North-Iceland which is in charge of the actual breeding program for Arctic charr. The breeding program is funded by the Agricultural Procuctivity Fund in Iceland.

Study 2
LABORATORY/RESEARCHER: A.K. Danielsdottir *et al.*, Marine Research Institute (MRI), Reykjavik
SPECIES: Cod
PROJECT FUNDING: MRI, others pending
OBJECTIVE: Elucidating cod stock structure in Icelandic waters
DESIGN: Mapping of gene frequencies
METHODOLOGY: Allozymes. RAPD and microsatellites under development
STATUS: Sampling started, analyses to start 1994
COMMENTS: Report references on request.

Study 3
LABORATORY/RESEARCHER: E. Arnason, University of Iceland, Dept. of Biology, Reykjavik
SPECIES: Cod, salmon, brown trout
PROJECT FUNDING: In house
OBJECTIVE: Genetic population structure in marine fish species
DESIGN: Mapping of gene frequencies
METHODOLOGY: mtDNA, RAPD and mtDNA cytochrome b sequencing in cod. mtDNA in salmon and trout.
STATUS: Ongoing.
COMMENTS: Report references on request.

Study 4
LABORATORY/RESEARCHER: A.K. Danielsdottir, G. Marteinsdottir, S. Sverrison, S. Gudlaugsdottir, F. Arnason and S. Gudjonsson, Institute of Freshwater Fisheries, Reykjavik.
SPECIES: Atlantic salmon, brown trout, Arctic charr.
PROJECT FUNDING:

OBJECTIVE: Mapping of genetic variation in reared and wild populations of salmon, and in wild populations of brown trout and Arctic charr.
DESIGN: Mapping of gene frequencies.
METHODOLOGY: Allozymes
STATUS: Salmon studies completed but not reported. Brown trout analyses in progress. Arctic charr sampled but not analysed.

IRELAND

Study 1
LABORATORY/RESEARCHER: T. Cross, Univ. College, Cork (U.C.C.) with two U.K groups.
SPECIES: Whiting, *Merlangus merlangus*.
PROJECT FUNDING: EC FAR MA-3.781
OBJECTIVE: To detect DNA markers for use in stock discrimination.
DESIGN: Samples taken from the southern and northern North Sea, Baltic, Norwegian coast and Irish Sea are being investigated for differences at two hypervariable PCRable minisatellite loci.
METHODOLOGY: PCRable minisatellite DNA loci
STATUS: Three-year project to be completed in September 1995.
COMMENTS: Ongoing work. Further minisatellite SLPs will be developed and other areas sampled. Interspecies applicability of probes will be tested (those already developed work well for cod and haddock and show variability).

Study 2
LABORATORY/RESEARCHER: P. McGinnity, Salmon Research Agency of Ireland.
SPECIES: Atalntic salmon.
PROJECT FUNDING: Salmon Research Agency of Ireland.
OBJECTIVE: To determin the impact of Ocean Ranch Atlantic salmon on the natural population of the recipient river, where no action has been taken to prevent the spawning of the ocean anch population.
DESIGN: Two scenario's are being studied where (a) the ocean ranch population has originated from the recipient wild population; (b) there is no relationship between the ocean ranch population and the recipient population.
METHODOLOGY: Allozyme analysis.
STATUS: Ongoing study.

Study 3
LABORATORY/RESEARCHER: R. Powell (Univ. College, Galway) with Oviedo University (Spain) and INRA - Jouy en Josas (France).
SPECIES: Atlantic salmon, brown trout.
PROJECT FUNDING: EC FAR AQ-2.493
OBJECTIVE: To describe molecular markers and assess linkage with quantitative trait loci (QTLs).
DESIGN: Reference samples include wild Irish, Spanish, French, Scottish, Norwegian and Canadian fish; domesticated Spanish line; and laboratory produced inbred lines.
METHODOLOGY: Genetic variation analysis using enzyme loci and defined nuclear genetic microsatellite loci.
STATUS: Three year study for completetion in November 1994.
COMMENTS: Twentyfive Atlantic salmon microsatellite sequencies have been developed to PCR-based essays with the number of alleles defined. Population analysis is currently underway.

Study 4
LABORATORY/RESEARCHER: E.J. Duke, University College Dublin (U.C.D.).
SPECIES: Bream, roach, and bream x roach hybrids.
PROJECT FUNDING:
OBJECTIVE: A biochemical genetic characterization of the bream, roach, and the bream x roach hybrids in the Erne catchment.
DESIGN: Batches of each species and their hybrids were analysed for molecular level and morphological variation. Results were compared on both inter- and intraspecies level.
METHODOLOGY: Isozyme and mtDNA analysis.
STATUS: Ongoing.
COMMENTS: The two species and the hybrid show characteristic isozyme profiles for xanthine oxidase, superoxide dismutase, lactate dehydrogenase and malate dehydrogenase. The mtDNA analysis shows that the hybrid results from female bream x male roach.

NORWAY

Study 1
LABORATORY/RESEARCHER: S. Fevolden, University of Tromsø, and G. Pogson, University of Cambridge.
SPECIES: Atlantic cod
PROJECT FUNDING:
OBJECTIVE: Elucidate the recruitment mechanisms of coastal cod in Norway, and possible interaction with northeast arctic cod.
DESIGN: Studies of juvenile cod before and after settling.
METHODOLOGY: cDNA
STATUS: Ongoing

Study 2
LABORATORY/RESEARCHER: S. Fevolden, University of Tromsø
SPECIES: Iceland scallop (*Chlamys islandica*)
PROJECT FUNDING:
OBJECTIVE: Population structure of the species, including eastern Barents Sea and areas north of Spitzbergen.
DESIGN: Studies on correlation between growth and heterozygosity
METHODOLOGY: Allozymes and mtDNA
STATUS: Ongoing

Study 3
LABORATORY/RESEARCHER: S. Fevolden, University of Tromsø.
SPECIES: Arctic charr (*Salvelinus alpinus*)
PROJECT FUNDING:
OBJECTIVE: Ecological and genetic characterization of landlocked populations.
DESIGN:
METHODOLOGY: Allozymes.
STATUS: Ongoing

Study 4
LABORATORY/RESEARCHER: J. Mork, Biological Station, University of Trondheim.
Collaboration with T. Cross and P. Galvin (University College, Cork, Ireland), and J.E. Eliassen (The Norwegian Institute of Fisheries and Aquaculture, Tromsø).
SPECIES: Cod, haddock, whiting, saithe, blue whiting, Norway pout, capelin, herring.
PROJECT FUNDING: The Norwegian Research Council & The Directorate for Nature Management
OBJECTIVE: Baseline studies of genetic population structures in Norwegian coastal waters
DESIGN: Collection of ~100 specimens from each Norwegian fjord from the Kola peninsula to Aalesund, storing tissue samples at -84 °C, and analysing them using allozymes and various others techniques when such become available. Sample collection during intensive research vessel cruises traversing large portions of the coastline in 6-7 weeks each year 1992-1994.
METHODOLOGY: Allozymes, DNA minisatellites and opportunistic methods
STATUS: Haddock, cod and blue whiting allozyme analyses are *a jour* (2-3000 specimens each). DNA minisatellite analyses ongoing for whiting and (as pilot studies) some other species.
COMMENTS: The genetic studies are coordinated with biological studies performed on the same materials by The Norwegian Institute of Fisheries and Aquaculture under it's Coastal Resource Program. Thus all specimens are biologically characterized (sex, length, age etc). Tissue samples can be made available for researchers with interesting projects.

Study 5
LABORATORY/RESEARCHER: K. Hindar, Norwegian Institute of Nature Research, Trondheim.
SPECIES: Atlantic salmon
PROJECT FUNDING: The Norwegian Research Council, The directorate for Nature Management.
OBJECTIVE: Baseline study of Atlantic salmon population structure in Norway.
DESIGN: Collection of samples from the main Norwegian salmon rivers (<100).
METHODOLOGY: Allozymes and various DNA techniques.
STATUS: Started in 1988, ongoing.

Study 6
LABORATORY/RESEARCHER: K.E. Jørstad, Institute of Marine Research, Bergen
SPECIES: Herring, cod, labridae
PROJECT FUNDING: Institute of Marine Research, The Norwegian Research Council
OBJECTIVE: Genetic population structure and dynamics in marine resource species
DESIGN: Repeated sampling (research vessel) in time-series
METHODOLOGY: Allozymes and various DNA techniques
STATUS: Ongoing

Study 7
LABORATORY/RESEARCHER: G. Dahle, Institute of Marine Research, Bergen
SPECIES: Cod, salmon
PROJECT FUNDING: Institute of Marine Research, The Norwegian Research Council
OBJECTIVE: Development of genetic markers for the study of genetic population structure of marine and andromous species, and specifically for measuring the amount of gene flow between populations
DESIGN: Laboratory and field studies
METHODOLOGY: mtDNA, microsatellites, RAPD
STATUS: Ongoing

Study 8
LABORATORY/RESEARCHER: Ø. Skaala, Institute of Marine Research, Bergen
SPECIES: Atlantic salmon, brown trout
PROJECT FUNDING: The Institute of Marine Research, The Norwegian Research Council
OBJECTIVE: Measuring selection and local adaptations in salmon and trout
DESIGN: Challenging genetically characterized local river stocks with laboratory family groups containing genetic markers, and studying immigrant fitness by gene frequency changes over time (F1, F2 and on) in the local stock.
METHODOLOGY: Allozymes, morphology (finespotted trout)
STATUS: Ongoing

POLAND

Study 1
LABORATORY/RESEARCHER: K. Goryczky, S. Dobosz, K. Kohlman, A. Zyczynski. Inland Fisheries Institute (IFI), Salmonid Lab., Rutki, & Institute of Freshwater Ecology and Inland Fisheries, Berlin, & Warsaw University of Agriculture.
SPECIES: Rainbow trout.
PROJECT FUNDING: Committee of Scientific Research, IFI Statutory activity.
OBJECTIVE: To improve the rainbow trout breeding value.
DESIGN: R.T. family selection; from 4 "lines" an outbred broodstock has been constructed and from these fish 100 families (F1) were produced in 1991. The families ammount was culled 3 times. Finally from 10 selected families the F2 102 (full sibs) families were produced in 1994.
METHODOLOGY: Separate rearing till the end of first summer, then 180 fish from each of the 64 selected families (size exceeding family mean) were tagged (80 PIT and 100 traditional tags). PIT tagged fish were mixed and their growth monitored (two times per year) until sexual maturity at the 3 years age. Traditionally tagged fish were transferred to production farm and analyzed at the time of slaughter. Selection index was based on growth abd survival.
STATUS: Beginning of the F2 breeding cycle.
COMMENTS: Passive Integrated Transponders (PIT) enable presize evaluation of individual fish & family breeding value and facilitated processing.

Study 2
LABORATORY/RESEARCHER: K. Goryczko, S. Dobosz. Inland Fisheries Institute, Salmonid Research Lab., Rutki.
SPECIES: Sea trout.
PROJECT FUNDING: CSR, FI statutory activity.
OBJECTIVE: Vistula sea trout outbred broodstock as a gene bank.
DESIGN: Freshwater broodstock produced from progeny of representative group of Vistula river ascending fish.
METHODOLOGY: A random sample of 1200 one summer old fish were PIT tagged for monitoring of smoltification, growth, and age of sexual maturation.
STATUS: Ongoing for the third year
COMMENTS: Projects aimed at protection of genetic diversity of economically important sea trout stock.

Study 3
LABORATORY/RESEARCHER: S. Dobosz, K. Goryczko, M. Luczynski. Inland Fisheries Institute, Salmonid Research lab., Rutki, & University of Agriculture and Technology, Dept. of Basic Fishery

Science, Olsztyn.
SPECIES: Rainbow trout.
PROJECT FUNDING: Committee of Scientific Research (KBN), Res. Grant No. 5491910203 and No. 554779102.
OBJECTIVE: To evaluate the magnitude of heterosis resulting from crossing inbred (e.s., gynogenetic) fish of genetically distant strains.
DESIGN: Monitoring of experimental fish survival and growth rate.
METHODOLOGY: Survival and growth in the F1 and F2 gynogenotes and its resiprocal crosses were compared with production trout.
STATUS: Project completed in 1994.
COMMENTS: The heterosis observed merely compensated the growth depress resulting from the artificial gynogenesis.

SPAIN

Study 1
LABORATORY/RESEARCHER: L. Sanchez and P. martinez, Universidad de Santiago de Compostela, Departemento de Genetica. 27002 Lugo, Spain.
SPECIES: Brown trout (*Salmo trutta*).
PROJECT FUNDING: Grant No. XUGA 26106A90
OBJECTIVE: Study of an unusual NOR variation (number) in brown trout detected in a drainage basin from in Galicia (NW Spain). Origin, stability and evolution of this chromosome polymorphism, and assessment of its utility for population screening.
DESIGN: Fourteen crosses with selected individuals are being performed to confirm the M;endelian inheritance of this NOR variation. Several specially polymorphic individuals are being analyzed with Ag- and CMA_3 staining, and with *in situ* hybridization to study the intraindividual variation and NOR regulation patterns. Around 20 populations in the "polymorphic area" will be studied to know the strength and spatial distribution of this polymorphism.
METHODOLOGY: Chromosome studies
STATUS:

Study 2
LABORATORY/RESEARCHER: L. Sanchez and P. martinez, Universidad de Santiago de Compostela, Departemento de Genetica. 27002 Lugo, Spain.
SPECIES: Brown trout (*Salmo trutta*).
PROJECT FUNDING: Grant No. XUGA 26106A90
OBJECTIVE: To study the microgeographical population structure of brown trout in Galicia (NW Spain). Utilization of brown trout as a model for the study of genetic resource management in a species with a large population subdivision. Assessment of the stocking policy carried out from 40 years ago with this species in Galicia.
DESIGN: 40 populations selected according to different criteria like stocking degree, isolation, and different watercourse points will be analyzed in a long drainage basin.
METHODOLOGY: Isozyme (45 loci) and chromosome markers.
STATUS:

Study 3
LABORATORY/RESEARCHER: L. Sanchez and P. martinez, Universidad de Santiago de Compostela, Departemento de Genetica. 27002 Lugo, Spain.

SPECIES: Turbot (*Scophtalmus maximus)*
PROJECT FUNDING: CICYT.MAR91-0807
OBJECTIVE: Assessment of genetic resources in natural and farm populations of turbot (*Scophtalmus maximus);* a species of great interest for aquaculture. Study of chromosome sex determination as a basis for the production of all-female populations in this species.
DESIGN: Four natural turbot populations from Northwestern Spain, and ten farmed turbot populations with different geographic reproductive origins will be analysed for genetic similarities and differences. Cytogenetic analyses of turbot will be carried out to detect possible sex linkage heteromorphism.
METHODOLOGY: Isozymes (45 loci), conventional, fluorochrome, restriction enzyme and replication banding techniques.
STATUS:

SWEDEN

Study 1
LABORATORY/RESEARCHER: H.Jansson, Salmon Research Institute.
SPECIES: Atlantic salmon.
PROJECT FUNDING: The Swedish National Board of Fisheries.
OBJECTIVE: To study genetic variation among Atlantic salmon populations at the Swedish west coast.
DESIGN: Samples of salmon from twelve rivers will be analysed for spatial and temporal genetic variation.
METHODOLOGY: Allozymes.
STATUS: Three year project started in 1994.

Study 2
LABORATORY/RESEARCHER: H.Jansson, Salmon Research Institute.
SPECIES: Atlantic salmon and brown trout.
PROJECT FUNDING: The Swedish National Board of Fisheries, and Power companies.
OBJECTIVE: Genetic monitoring of hatchery stocks to detect possible genetic changes.
DESIGN: The hatchery stocks used for the Swedish compensatory program are sampled and analysed for genetic variation at regular intervals.
METHODOLOGY: Allozymes.
STATUS: Long term study.

Study 3
LABORATORY/RESEARCHER: J.Dannewitz, Department of Genetics, Uppsala University, and H.Jansson, Salmon Research Institute.
SPECIES: Atlantic salmon and brown trout.
PROJECT FUNDING: Internal funds.
OBJECTIVE: To study the extent of hybridization between Atlantic salmon and brown trout in Sweden.

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DESIGN: Spatial and temporal variation in hybrid frequencies are examined. The maternal species of the hybrids is determined.
METHODOLOGY: Allozymes and mitochondrial DNA.
STATUS: Long term study.

Study 4
LABORATORY/RESEARCHER: J.Nilsson and M.Schmitz, Department of Aquaculture, Swedish University of Agricultural Sciences, Umeå. Cooperation with R.Gross, Estonian Institute of Veterinary Science and Animal Breeding, Tartu, Estonia.
SPECIES: Brown trout (anadromous and non-anadromous populations).
PROJECT FUNDING: Swedish Institute, and regional funds.
OBJECTIVE: To find new DNA-markers in trout and to use these markers in studies of population genetic structure and in studies of population mixing.
DESIGN: A number of European sea trout and brown trout populations are screened for variation at DNA level. Microgeographic population genetic structure is studied in River Ammerån, N.Sweden. Effects of introducing migratory trout in a stationary trout population is studied in Låktabäcken creek, N.Sweden.
METHODOLOGY: Variation in single copy genes, microsatellites.
STATUS: Project beginning in 1994, ending in 1997.

Study 5
LABORATORY/RESEARCHER: T.von Schantz and Å.Langefors, Department of Animal Ecology, University of Lund, and B.Widergren, The Wallenberg Laboratory, University of Lund.
SPECIES: Atlantic salmon.
PROJECT FUNDING: The Swedish Council for Forestry and Agricultural Research, and the Swedish Environmental Protection Agency.
OBJECTIVE: To study the genetic variation in MHC and Cytochrome P450s in different salmon populations and to see whether variation in these loci affects disease resistance and the ability to process different persistent organochlorine pollutants.
DESIGN: DNA from three different populations is analysed for typing of genotypes and haplotypes.
METHODOLOGY: RFLP-analyses, DNA sequencing, and DGGE-analyses.
STATUS: The project started in summer 1993. New funding will be applied for by January 1995.

UNITED KINGDOM

Study 1
LABORATORY/RESEARCHER: D. Thompson/D. James, MAFF Fisheries Laboratory, Lowestoft, with one UK (Univ. of East Anglia, Norwich) and one Irish (Univ. College, Cork) group.
SPECIES: Whiting (*Merlangus merlangus*)
PROJECT FUNDING: EC FAR MA.3.781
OBJECTIVE: Investigate the potential of molecular biological methods for stock discrimination in commercially importat species.
DESIGN: Samples of whiting *(Merlamgus merlangus*) are being collected from throughout the geographical range of the species. Using methods given below , the rpesence or absence of any

discernible stock structure will be investigated. Comparisons will be made of the levels of genetic variation identified by the variuos techniques.
METHODOLOGY: Allozymes, mtDNA, mini- and microsatellite DNA.
STATUS:Three year project, started October 1992.
COMMENTS: Development of allozyme, mini- and microsatellite techniques have been carried out and population data is being acquired.

Study 2
LABORATORY/RESEARCHER: D. Thompson/D. James, MAFF, Fisheries Laboratory, Lowestoft.
SPECIES: Atlantic salmon.
PROJECT FUNDING: MAFF.
OBJECTIVE: To investigate the population structure of the Atlantic salmon and the possibility of a genetic control of run-timing in juvenile salmon.
DESIGN: Salmon populations in rivers in Englans and Wales have been sampled over a number of years. Allozyme data is being used to search for geographical and temporal variation. mtDNA variation is being used to test the hypothesis of a relationship between the timing of smolt migration and mtDNA haplotypes.
METHODOLOGY: Allozymes, mtDNA
STATUS: Three year project, commenced 1994.
COMMENTS: Allozyme data are being used to formulate fisheries management policies in the U.K.

Study 3
LABORATORY/RESEARCHER: S.D. Utting & A.R. Child, MAFF Fisheries Lab., Benarth Road, Conwy LL32 8UB, UK.
SPECIES: Manila clam (*Tapes philippinarum*).
PROJECT FUNDING: MAFF in house funding.
OBJECTIVE: Production of sexually sterile bivalve molluscs.
DESIGN: Produce triploids by manipulation of chromosome numbers.
METHODOLOGY: Treat eggs with cytochalasin B and arrest development of polar body formation.
STATUS: Three year project completed in 1994.
COMMENTS: Recent publications are:
Utting, S.D. & Child, A.R. 1994. Genetic manipulation of the Manila clam (*Tapes philippinarum*) using cytochalasin B to induce triploidy. Aquaculture (accepted).
Child, A.R. & Watkins, H.P. 1994. A simple method to identify triploid molluscan bivalves by the measurement of cell nucleus diameter. Aquaculture (accepted).
Allen Jr., S.K., Shpigel, M., Utting, S.D. & Spencer, B.E. 0000. Incidental production of tetraploid Manila clams *Tapes philippinarum* (Adams and Reeve). Submitted to Aquaculture.

Study 4
LABORATORY/RESEARCHER: A.R. Child, MAFF Fisheries Laboratory, Benarth Road, Conwy LL32 8UB, UK.
SPECIES: King scallop (*Pecten maximus*).
PROJECT FUNDING: MAFF in house funding.
OBJECTIVE: Genetic variation in populations of *P. maximus*.
DESIGN: Examine levels of genetic variation in samples of wild stocks in Scotland, Irish Sea and south wet Britain.
METHODOLOGY: mtDNA incorporating PCR amplification of selected fragments and restriction enzyme analysis.
STATUS: Two year project commenced 1994.
COMMENTS: Work due to start in June 1994.

Study 5
LABORATORY/RESEARCHER: A.R. Child, MAFF Fisheries Laboratory, Benarth Road, Conwy LL32 8UB UK, & A.R. Beaumont, UC North Wales. Msc project.
SPECIES: Pacific oyster (*Crassostrea gigas*)
PROJECT FUNDING: MAFF in house funding, and UCNW.
OBJECTIVE: Genetic variation in commercial populations of *C. gigas*.
DESIGN: Examine levels of genetic variation in samples from two commercial populations of Pacific oysters and the offspring of original British introductions at MAFF, Conwy.
METHODOLOGY: Allozyme variation compared with other historical studies.
STATUS: Three months project completed 1993.
COMMENTS: Msc thesis submitted by P. Papageorgiou.

Appendix 5

WGAGFM members (as of February 23., 1994)

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Appendix 6

Terms of reference of the WGAGFM for 1994 (C.R. 2:27, 1993)

2:27

The Working Group on Genetics (Chairman: Dr. J. Mork, Norway) will be renamed the Working Group on Applications of Genetics in Fisheries and Mariculture and will meet at ICES Headquarters from 9-11 March 1994 to:

a) prepare information for use by the Study Group on Interactions of Wild, Ranched (Enhanced) and Cultured Salmon;

b) review the interim conclusions of the Study Group on Stock Identification Protocols for Finfish and Shellfish Stocks;

c) review knowledge on the amount of gene flow between specific natural populations as well as between cultured and specific natural populations with a view to proposing future studies;

d) propose studies on local adaptations of specific natural populations using combined qualitative (e.g., gene markers) and quantitative (e.g., family studies) genetic approaches;

e) report on the status of selective breeding and genetic modifications relative to improving production traits such as growth performance, product quality, disease resistance, etc.;

f) evaluate the options for applications of genetic research to fisheries and mariculture questions of concern to ICES.