

# 55

## Constitution of aquacultural stocks : genetic aspects

**B. CHEVASSUS**

*Laboratoire de Génétique des Poissons. I.N.R.A. 78350 JOUY-EN-JOSAS. France*

**Abstract** — *Before reaching of production level justifying an elaborated genetic improvement program, the new aquaculture species deserve a minimal strategy to avoid the consequences of uncontrolled genetic phenomena. Three main aspects will be discussed :*

- *initial constitution of the broodstock : how to collect a sufficient genetic variability without encountering the problems resulting from the mixture of differentiated populations ?,*
- *maintenance of the broodstock : the regular introduction of new animals in the broodstock is a classical way to avoid inbreeding but his cost/benefit ratio has to be discussed in the case of aquaculture. An alternative option (multiple closed gene pools) is proposed to minimize the effect of inbreeding,*
- *selection of the broodstock : mass selection is often feasible selection for tropical species but his efficiency is an object of controversy. We present a modified mass selection procedure (PROSPER), which should maximise the genetic progress.*

Developing new aquacultural species for consumption or restocking generally implies the creation and maintenance of spawner stocks in captivity. The involvement of genetics in this field is generally considered in terms of *improvement of performances* by selection, crossing or various manipulations. On the other hand, the necessity of installing a *genetic stock management* even in the absence of clearly identified improvement objectives is often ignored. We therefore considered that it was necessary to develop this topic in particular by showing how various aspects of genetics may intervene in the creation of aquacultural stocks.

A distinction will be made between two cases :

- sampling from only one source population,
- creation of heterogeneous stocks from several populations.

### ONE SOURCE POPULATION

In many cases stocks should be constituted from only one source population. For instance in the case of particular sanitary guarantees on

one population, the will to save a given population with a view to restocking (the cases of the salmon of the Allier or the sturgeon of the Gironde) or the deliberate choice of a population based on performances proved to be satisfying.

Then it is a question of collecting the largest possible fraction of the population's genetic variability. Empirically, it is obvious that the sample should represent a large number. However, in genetic terms the notion of sample size may be notably different from that of numerical size.

### 1. Definition of genetic size

Usually the introduction is made at the level of fertilized eggs or larvae. If  $N$  is the numerical size of this sample, the share of genetic variance of the sample relative to that of the source population may be estimated by using the following equation (Kimura and Crow, 1963; Lacava and Hughes, 1984) :

$$\frac{V_E}{V_T} = 1 - \frac{1}{2 N_e} \quad (1)$$

where  $N_e$  is the sample's genetic size (or effective number). It is obtained by calculating the effective number of spawners for each sex :

$$N_{em} = \frac{N_m K_m - 2}{K_m - 1 + \frac{V_m}{K_m}} \quad (2)$$

$$N_{ef} = \frac{N_f K_f - 2}{K_f - 1 + \frac{V_f}{K_f}} \quad (3)$$

- $N_{em}$  and  $N_{ef}$  are the effective numbers of male and female spawners,  $N_m$  and  $N_f$  being the real numbers of male and female spawners used.
- $K_m$  and  $K_f$  represent the average number of progeny per male and female spawner. Thus, if the sample includes a total of  $N$  individuals, we obtain

$$K_m = \frac{N}{N_m} \quad (4)$$

$$K_f = \frac{N}{N_f} \quad (5)$$

$V_m$  and  $V_f$  are the variances of these numbers of progeny between male and female spawners.

Finally, the effective number of the sample is obtained by :

$$\frac{4}{N_e} = \frac{1}{N_{em}} + \frac{1}{N_{ef}} \quad (6)$$

Combining the different relations, the equation can also be written :

$$\frac{4}{N_e} = \frac{(K_m + \frac{V_m}{K_m}) + (K_f + \frac{V_f}{K_f}) - 2}{N - 2} \quad (7)$$

## 2. Variation factors

Therefore three parameters are going to play an important role in this equation to maximize  $N_e$  at a given value of  $N$ .

- *The number of spawners used* : for an introduced sample of a given size, this number may in fact vary considerably in aquatic species with a high fertility. A single couple may give rise to a sample of several thousand juveniles. Therefore, it is advisable to attach much importance to this parameter whose role is more essential than that of the numerical size of the introduced sample. Thus, assuming that each spawner contributes to the sample in the same way, we obtain (see further on)  $V/K = 1$ , where :

$$N_e = \frac{4(N-2)}{K_m + K_f} \quad (8)$$

For a sample of 10,000 individuals originating from 4 males and 4 females,  $K_m = K_f = 2,500$ , hence  $N_e \approx 8$  whereas for a sample of only 100 individuals originating from 5 males and 5 females,  $K_m = K_f = 20$ , hence  $N_e = 9.8$ .

- *The sex ratio in the stock of spawners used* : Fig. 1 illustrates this aspect. It seems that if a relatively small number of spawners is sufficient when the sex ratio is equilibrated, an imbalance requires use of a much larger number. Thus, the collected variability is lower with 5 females and 95 males (i.e. 100 spawners) than with 10 females and 10 males, i.e. only 20 spawners (97.2 compared to 97.5 %). Usually, the number of females really constitutes a limiting factor owing either to the difficulty in capturing the animals at the right time or to the will not to reduce too much the reproductive potential of the source population. However, using a large number of males gives only a partial compensation : equation (6) shows that whatever the number  $N_m$  of males used and the sample size, *the genetic size will always be smaller than 4 times the number of females used*.
- *The variance of family size*, in connection with the contribution of each spawner to the constitution of a sample of juveniles. In the theoretical case where all the spawners contribute equally to the constitution of the new generation, the sample constitution can be compared to a random sampling described by the Poisson distribution interestingly characterized by a variance equal to its mean. Equation (8) can thus be used. However, this hypothesis seems little realistic in aquatic species. In particular, the quality

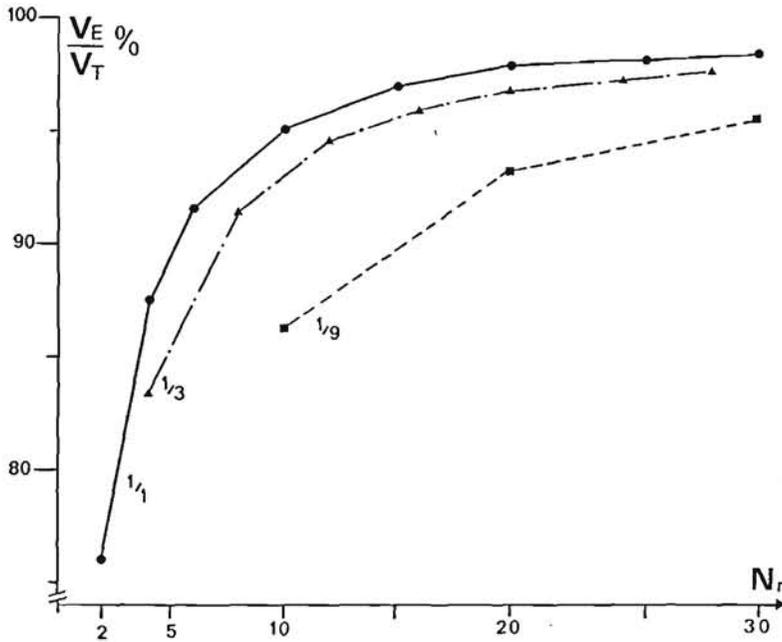


Fig. 1. — Part of the total genetic variance of the initial population (in %) collected in a sample resulting from a group of  $N_r$  spawners with various sex ratios.

1/1 1 male for 1 female  
 1/3 3 males for 1 female  
 1/9 9 males for 1 female

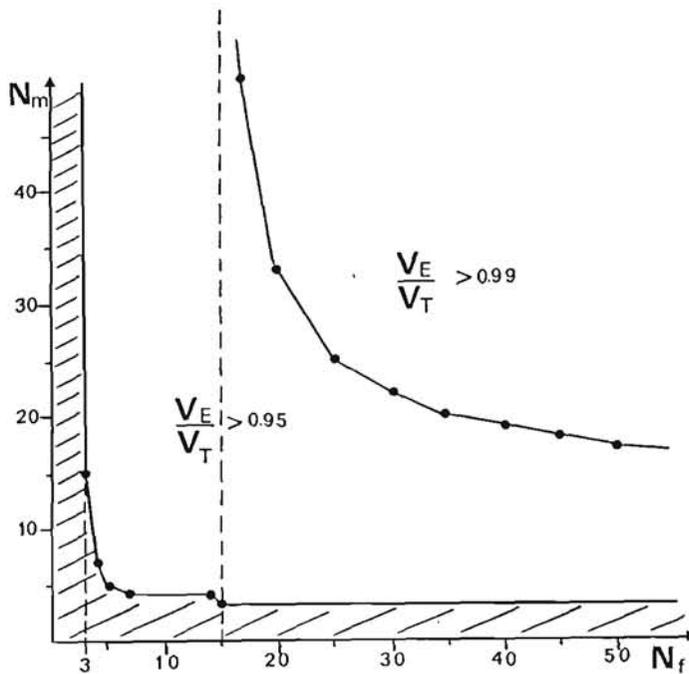


Fig. 2. — Number of male ( $N_m$ ) and female ( $N_f$ ) spawners required to collect at least 95 or 99% of the total genetic variance of the source population: for a number of males larger than 50, the minimal number of females are respectively 3 and 15.

of collected ova may be different from one female to another and fertilization and hatching rates may vary considerably. Thus, the contributions of each female to the sample are different and the ratio V/K may be much higher than 1 which results in a marked decrease in the female effective number (Table 1, case C). In order to minimize this phenomenon, it is therefore desirable to keep the various spawnings separated during the whole period where large variations in survival linked to the female may show up (embryonic development, hatching and vesicular resorption and if possible beginning of exogenous feed intake). At the end of this stage, regrouping is possible by taking in each group :

- equal numbers if each spawning was fertilized by the same number of males,
- numbers proportional to their genetic stock if the spawnings were fertilized by mixtures of sperms from different males

Thus, if three spawnings A, B, and C, fertilized by 5, 10 and 20 males, respectively, are available, the genetic stock of these spawnings ( $N_e \approx 4 N_m / (N_m + 1)$ ) is 3.20, 3.64 and 3.81, respectively. Therefore, for instance 320 alevins originating from spawning A, 364 originating from spawning B and 381 from C will be regrouped.

Tab. 1. — Effect of the variance of family size on the value of the effective number :

$$N_e = \frac{N - 2}{K - 1 + V/K} \quad (\text{see the text})$$

Female	1	2	3	4	5	6	7	8	9	10	N	V	$N_e$
												K	
A	9	5	4	7	6	3	1	4	5	6	50	0.88	9.83
B	5	5	5	5	5	5	5	5	5	5	50	0	12
C	27	25	10	8	4	2	1	1	1	1	80	11.3	4.37
D	27	25	10	8	4	2	—	—	—	—	76	7.57	3.95
E	12	10	10	8	4	2	1	1	1	1	50	3.64	6.28
F	8	8	8	8	4	2	1	1	1	1	42	2.47	7.05

A is the case of a random distribution with the same probability of occurrence for each female

B the effective number is slightly higher in the case of perfect equality of family size

C when the occurrence of the females are very unequal, the effective size can be highly reduced

D, E, F see figure 3

This practise may seem to be paradoxical because it leads to adjusting the sampling on the group having exhibited the lowest survival rate instead of regrouping all the survivors from the various spawnings. This strategy of *truncation from the top* (Table 1, E and F) is, however, to be recommended, as the effective number is much more sensitive to family size variance than to their average number (Fig. 3).

On the other hand, it may be useful to distribute the sampling effort over time. This may allow to collect a larger genetic stock, especially when the number of females constitutes a limiting factor, and to take into account possible « heterogeneities » of the spawner population. All the spawners present at the same place or mature at the same time may represent only a fraction of total variability even if their number is high, especially in migratory populations exhibiting a precise « homing » (coming back to the place of birth). Therefore, sampling should rather be distributed over the spawning period or repeated during several years whether consecutive or not.

### 3. Conclusion

If we go back to equation (1), obviously, by respecting the precautions defined above it is rather easy to collect a large fraction of the genetic variability of the source population (Fig 2.) : if the introduction of 95 %

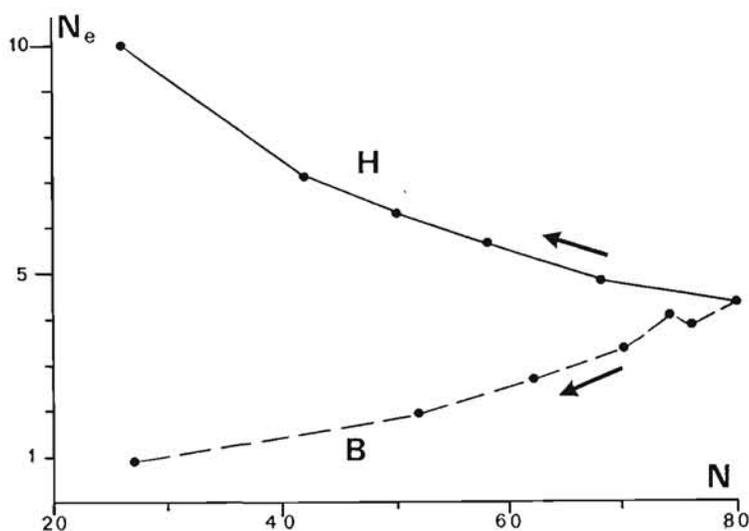


Fig. 3. — Effect of two alternative strategies for equalizing family size. The initial situation is case C (Table 1).

H : reduction of the size of the more numerous families (case E and F). The effective size  $N_e$  increase.

B : elimination of the less numerous families (case D). The effective size  $N_e$  decrease.

initial variability is fixed as a « standard » objective, this end may for instance be attained with a sample of 100 individuals originating from 3 females, each being fertilized by 5 different males. With 15 females also being fertilized by 5 males the loss of variability is only 1 %. Therefore, these principles should be widely disseminated among hatchery managers.

## SEVERAL SOURCE POPULATIONS

It is often possible to identify several natural or cultured populations within a species and a more complex strategy has to be adopted in order to create a spawner stock. Should only one population be chosen (and in this case which one ?) or on the contrary, should a « synthetic » population be created by mixing many of these populations ? In most of the cases, information available on these populations is limited to historical data which are often vague (concerning the origin of culture populations, possible exchanges between populations), or in situ biological observations (growth, morphology, reproduction period) whose interpretation in terms of genetic differentiation is problematic. In this case it is therefore necessary to *determine more accurately the organization of intra- and inter-population genetic variability* in order to define a sampling strategy.

### 1. Description methods

Two types of approaches may be considered.

- The first consists in introducing the different populations in one and the same place in order to undertake the *evaluation of their performances*. Whereas the principle of this method is satisfying it has the disadvantage of being long and expensive : it requires the setting up of specific progeny testing equipment making it possible to raise several groups under similar conditions and a protocol concerning at least two generations in order also to be able to study the crossings between populations.

*[The observation of performance differences between two populations introduced in the same environment can only be interpreted in genetic terms after having evaluated the maternal effects of each population. Therefore, it is generally necessary to create a protocol in two steps : the first (« parallel cultures ») allows to regroup the populations, to carry out a first evaluation of performances and to obtain sexual maturation of the animals under similar conditions; the second (diallel progeny testing) allows to analyse the maternal effects, genetic effects of each population and crossing effects (heterosis) by intra- and inter-population crossings].*

Therefore it is intended for species whose aquacultural development is already large enough to justify such an investment (salmonids, carps...). In the case of poorly developed species, this approach can only be applied a posteriori on a restricted number of selected populations according to other criteria (see further on).

In any case it is important to recall that the principles defined in the former paragraph are applied even in the case of a transitory introduction of a population for progeny testing purposes. The sample must exhibit a sufficient variability in order to be representative of the population. It is shown that for a

sample of size  $N$  and effective number  $N_e$ , the precision  $\pm$  (in %) obtained on the genetic value of the original population equals :

$$\sigma = C V \left( \frac{h^2}{N_e} + \frac{2 - h^2}{2N} \right) 1/2$$

$C V$  is the observed variation coefficient of the considered trait,  $h^2$  is the heritability of this trait (proportion of the observed variance linked to the genetic variance between individuals).

Thus, for a trait like growth performance having a variation coefficient of some 30 % and a heritability value of 0.2, at least 100 individuals originating from 5 males and 5 females will be needed to estimate the genetic value with 5 % accuracy, which is a low value. In order to obtain a higher accuracy, the effective number  $N_e$  will generally constitute the limiting factor. It will be necessary to attain at least  $N_e = 60$  (for instance 30 males and 30 females) in order that the accuracy reaches 2 % even by measuring 1000 individuals.

- The second consists in considering traits with a simple genetic determinism which are not sensitive to direct environmental action. This is especially the case of the chromosome number and structure (karyotype) and of protein polymorphism shown by electrophoresis, or more recently of nucleic acid polymorphism. The study of protein polymorphism has been particularly developed in aquatic organisms the last ten years and it seems useful to discuss the results in relation to the constitution of aquacultural stocks.

## 2. Results

Fig. 4 and 5 taken from Guyomard (1989) indicate an example of data obtained from a study on protein polymorphism. Three aspects can be taken into consideration.

- *The differentiation between populations*: the genetic distance between populations can be estimated by various measures where Nei's index is often used (1978). The populations are represented on a dendrogramme (Fig. 4) regrouping them according to their genetic resemblance. In the present example, three big groups can be identified: the first one, which is clearly separated, is made up of two natural Corsican populations; the second one regroups almost all the other Mediterranean populations. The third constitutes the « Atlantic group » in which all hatchery populations are scattered.
- *The genetic intra-population variation* measured by the mean rate of heterozygous individuals for all the genes studied. In this case (Fig. 5) the values are rather spread, ranging from 0 to 10.6 %. The variability is strong for hatchery populations, slightly lower for populations of the Atlantic group and much lower for Mediterranean populations, especially after correction of restocking effects by hatchery populations.

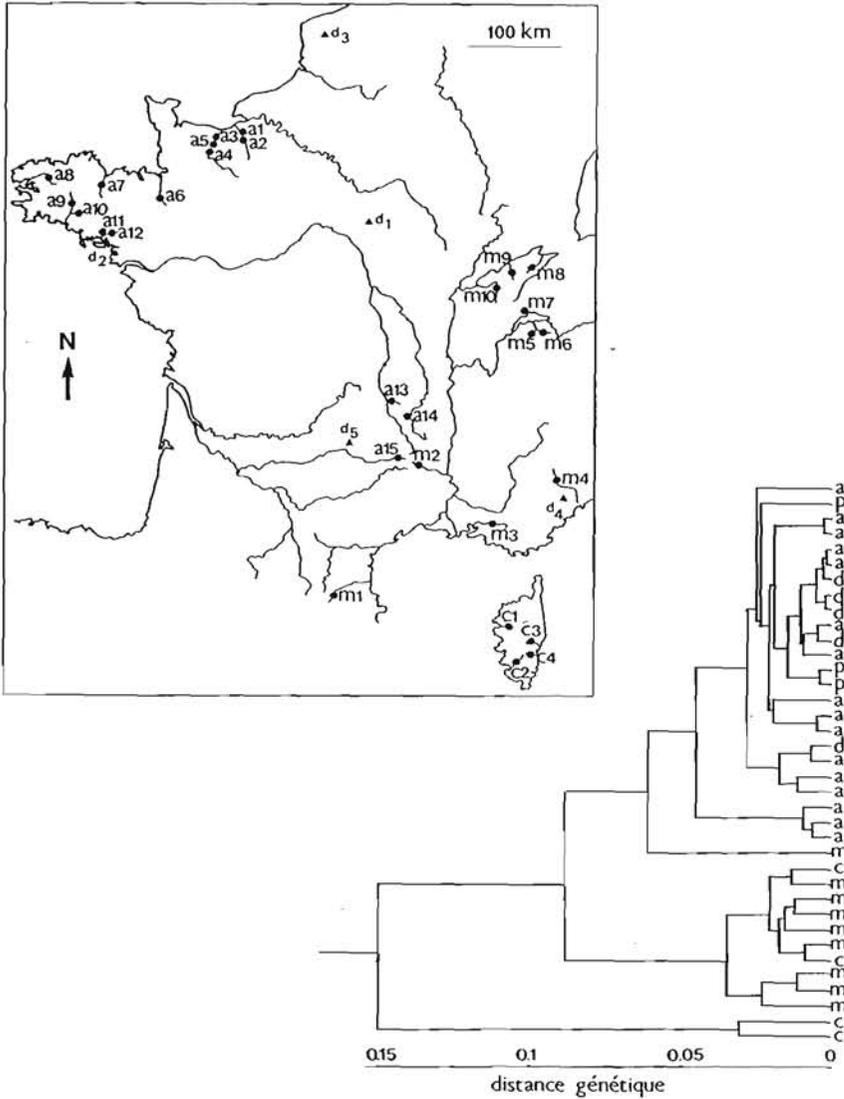
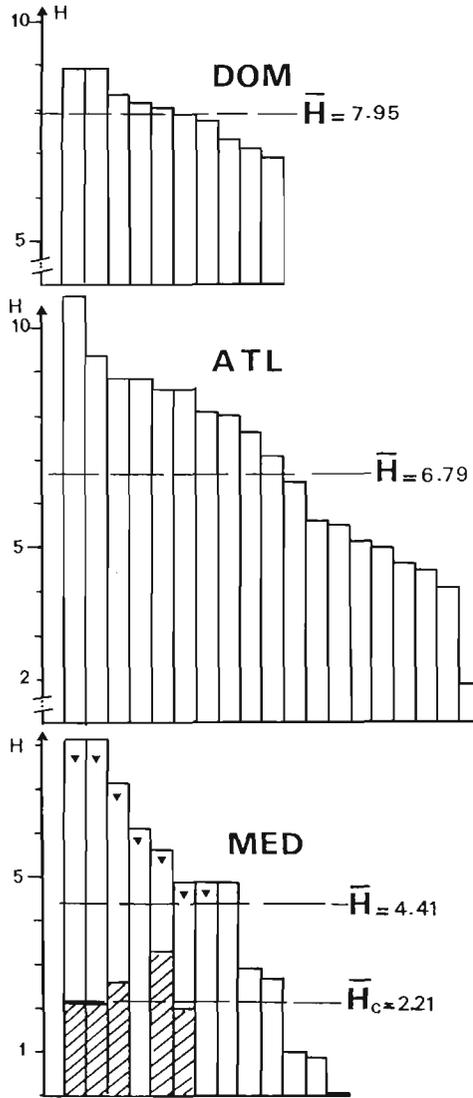


Fig. 4. — Genetic differentiation between french populations of brown trout *Salmo trutta* (from Guyomard, 1989).

- a. wild populations from the Manche and atlantic drainage areas
- m. wild populations from mediterranean drainage area
- d. domestic populations
- c. wild populations from Corsica
- p. population from Polish origin (Baltic sea).

The combination of these two pieces of information makes it possible to give a general opinion on the differentiation rate of the studied populations, namely the relationship between the average genetic inter-population variation and total genetic variation of the species obtained by regrouping all these popu-

lations. In this example, this rate is high, exceeding 50 % for all the 47 populations studied.



**Fig. 5.** — Distribution of heterozygosity (%) among 42 french populations of brown trout *Salmo trutta* (46 enzymatic loci).

DOM 10 domestic populations

ATL 19 wild populations from the Manche and atlantic drainage areas

MED 13 wild populations from the mediterranean drainage area.

— Rivers with a restocking with domestic populations

— Corrected value of heterozygosity to eliminate the effect of restocking.

Fig. 6, 7 and 8 illustrate various situations met in salmonids and allow to draw several general conclusions.

- The variability within natural populations usually exceeds 50 % of the total variability of the species. Therefore, it is advisable to give up definitively a conception according to which a local population should be made up of identical individuals different from those of another population. On the contrary, with certain exceptions each population represents an important pool of genetic variability.

The variability within hatchery stocks is not systematically lower than that observed in natural populations (Table 2). Some cases of a marked decrease in variability may be observed whereas some stocks may exhibit a high variability possibly linked to their heterogenous origin.

**Tab. 2.** — Comparison of intrapopulation heterozygosity in wild and domestic populations of salmonids

Species		Wild populations	Domestic populations	References
<i>Salmo clarkii</i>	H	2.2 (1, USA)	2.7 (1, USA)	Allendorf and Leary, 1988
<i>Salmo gairdneri</i>	H	31.0 (1, USA)	18.0 (9, USA) (8.6 - 29.5)	in Guyomard, 1981*
	R		31.2 (6, FRANCE) (24.1 - 34.8)	
<i>Salmo salar</i>	H	2.8 (9, SWEDEN)	2.2 (9, SWEDEN)	Stahl, 1983
	R	(2.2 - 3.5)	(1.5 - 3.1)	
	H	21.8 (1, IRELAND)	17.3 (1, IRELAND)	Cross and King, 1983*
<i>Salmo trutta</i>	H	6.79 (19, West EUROPE)	7.94 (5, FRANCE)	Guyomard, 1983
	R	(1.8 - 10.8)		
	H	4.41 (13, South FRANCE)		
	R	(0.0 - 8.2)		

\* heterozygosity is estimated with a set of polymorphic loci and thus overestimated.

The table gives the mean value of heterozygosity (H), the number and origin of the populations and the extremes values observed in those populations.

The genetic differentiations observed between populations do not always confirm the hypothesis based on ecoethological observations. Thus, in salmonids a great attention is often paid to the migratory or sedentary character of the populations or their way of life (lacustrine or river populations) and sometimes so much that various ecotypes are considered as subspecies. The case of the arctic char (*Salvelinus alpinus*) perfectly shows (Fig. 7 B) that these ecological differentiations seem to be genetically minimal as compared to the geographic differentiation between populations of the same ecotype. Inversely, the case of the cutthroat trout (*Salmo clarkii*) illustrates the necessity sometimes of managing ecologically similar populations as different subspecies (Fig. 7 A). The results obtained with the brown trout (Fig. 3) lead to similar conclusions.

— the genetic structure of the species may vary to a wide extent even between similar species (Fig. 8) and therefore a particular study is necessary

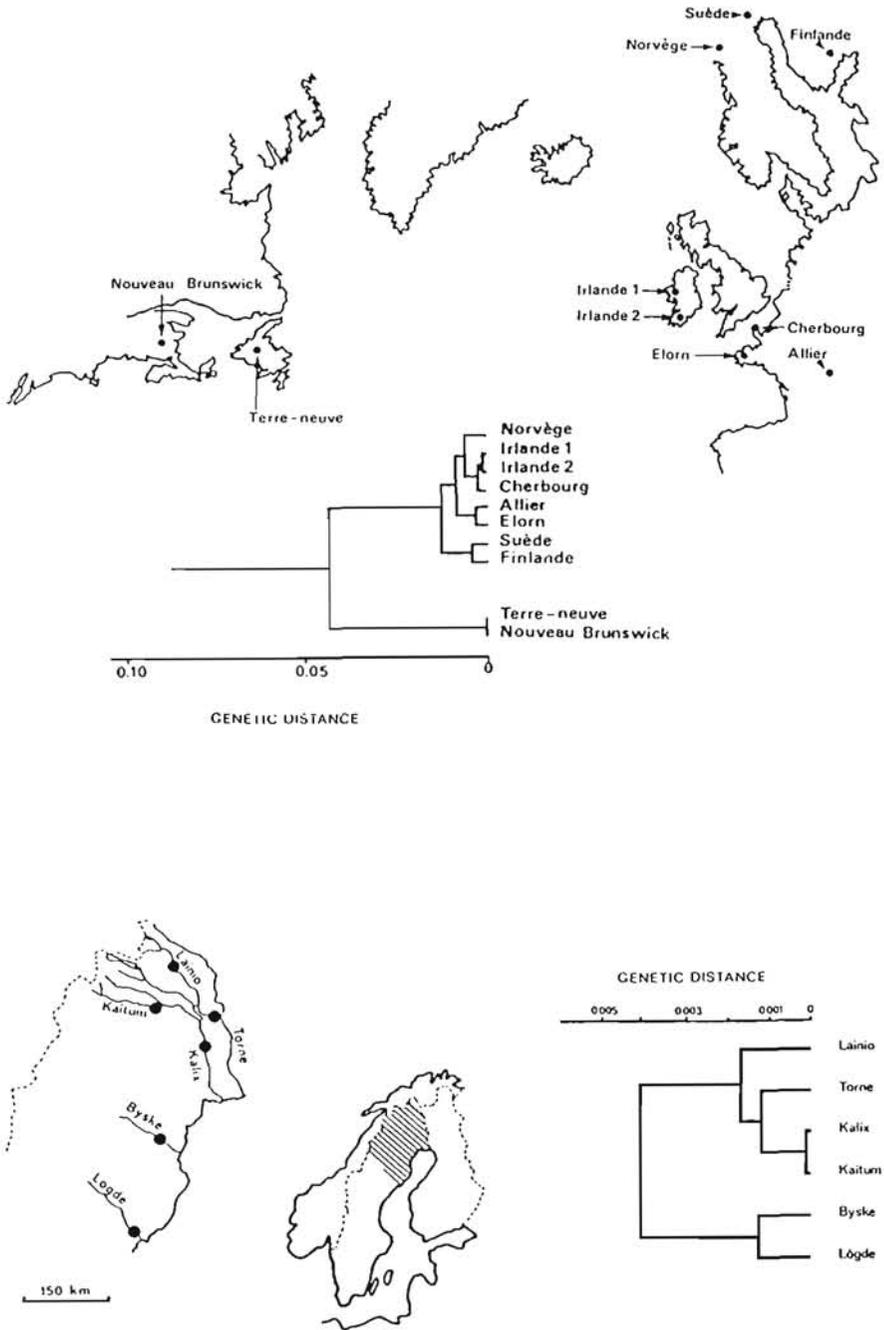
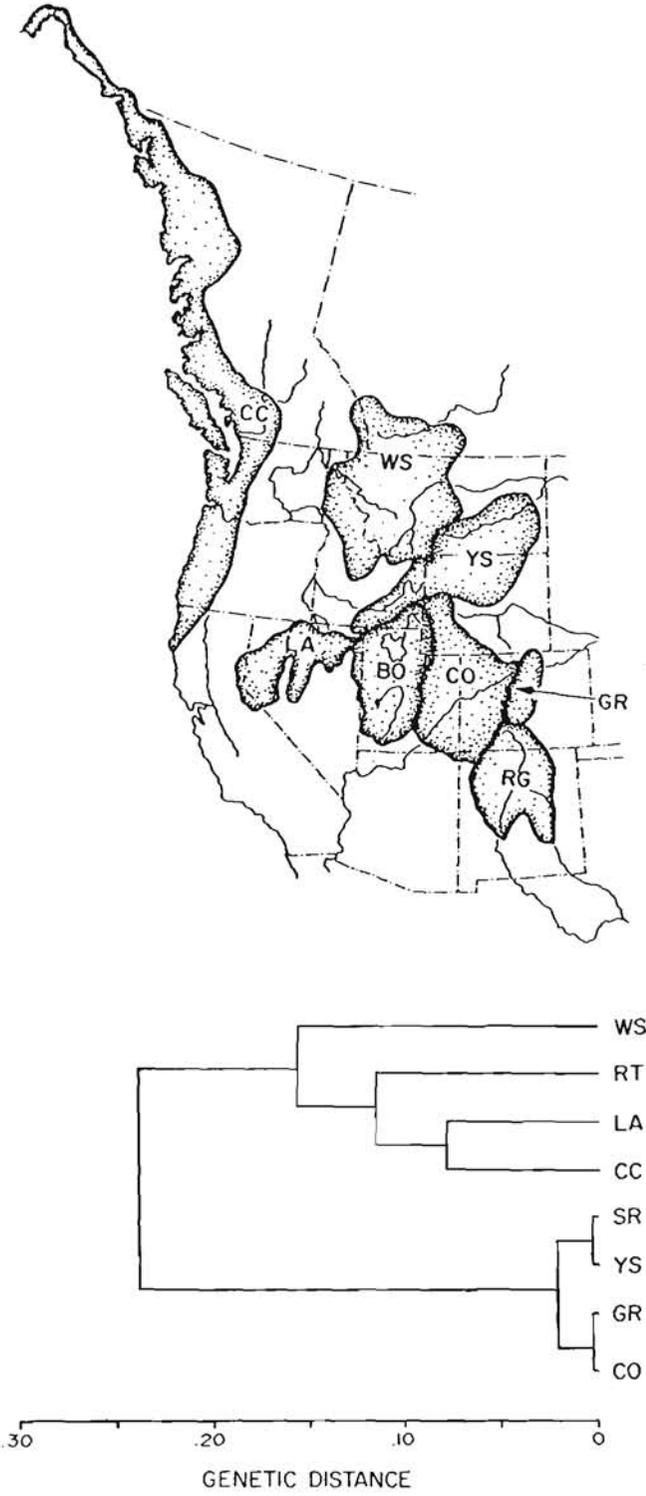


Fig. 6. — Genetic differentiation between populations of Atlantic Salmon from different european and canadian rivers (above) and within a small Swedish area (below). From Guyomard (1987) and Stahl (1981).



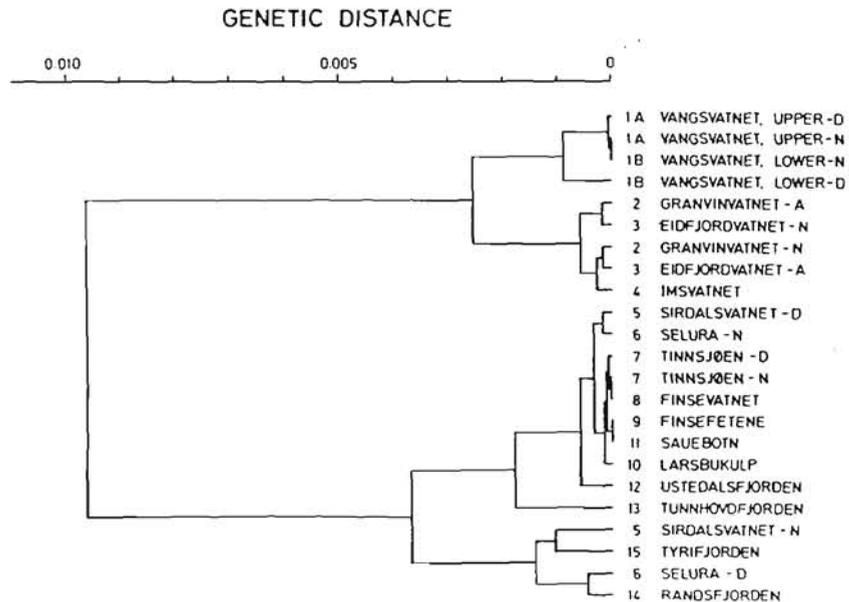


Fig. 7. — Two examples of genetic differentiation between populations of Salmonids.

above : *Salmo clarkii* (cutthroat trout) in the western part of the USA. The level of differentiation is high and the distinction of a new species (WS) is proposed (Allendorf and Leary, 1988).

below : *Salvelinus alpinus* (arctic char) in the south of Norway. The level of differentiation is low, especially between dwarf (d) and normal ecotypes inhabiting the same lake and considered before as « sibling » species. (Mindar et al., 1988).

each time. Thus, among 54 populations of a pacific salmon (*Oncorhynchus keta*) the differentiation rate is only 2 % (98 % intra-population variation). On the other hand, this rate reaches 70 % in cutthroat trout (*Salmo clarkii*) whereas it is only 16 % in a very similar species, the rainbow trout (*Salmo gairdneri*).

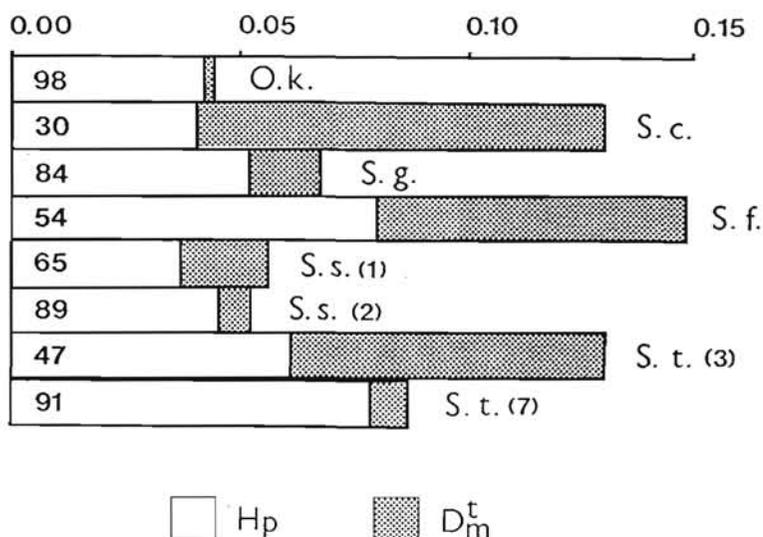


Fig. 8. — Decomposition of the genetic variability of different species of salmonids into two components :

$H_p$  is the mean within population variability,  $D_m^t$  is the mean between populations variability. The left value is the level of differentiation

$$\frac{D_m^t}{D^t + H_p} (\%)$$

OK *Oncorhynchus keta*, 54 populations, 22 loci

Sc *Salmo clarkii*, 24 populations, 35 loci

Sg *Salmo gairdneri*, 38 populations, 16 loci

Sf *Salvelinus fontinalis*

Ss *Salmo salar* (1) 10 populations, 32 loci

(2) 6 atlantic european populations, 32 loci

St *Salmo trutta* (3) 37 european populations, 46 loci

(7) 11

(Guyomard, 1989).

### 3. Interpretation and applications

How can these data be taken into account in the constitution of a spawner stock ? Generally, the advantage of constituting a stock exhibiting a large genetic variability from the beginning is justified by two types of arguments.

- In the short term, there may be a certain relationship between the genetic variability of a stock and its culture performances. Although they are not systematic such relationships were revealed several times in aquacultural species. Some examples will be given hereafter. Thus, a study of the length of time of embryonic development in 6 natural populations of rainbow trout (*Salmo gairdneri*) conducted by Ferguson *et al.* (1985) showed a positive relationship between the average heterozygosity of the strain and its growth rate (Fig. 9). The deviation is approximately 4 days (10%) when the level of heterozygosity increases from 4 to 8%. The same variations were found in this species when the relationship between individual heterozygosity measured by enzymatic markers and early hatching was studied within a population (Danzmann *et al.*, 1984). In the same way in molluscs marked relationships between growth rate and individual heterozygosity have been shown several times (Zouros and Foltz, 1987 a review). Fig. 10 illustrates this phenomenon.

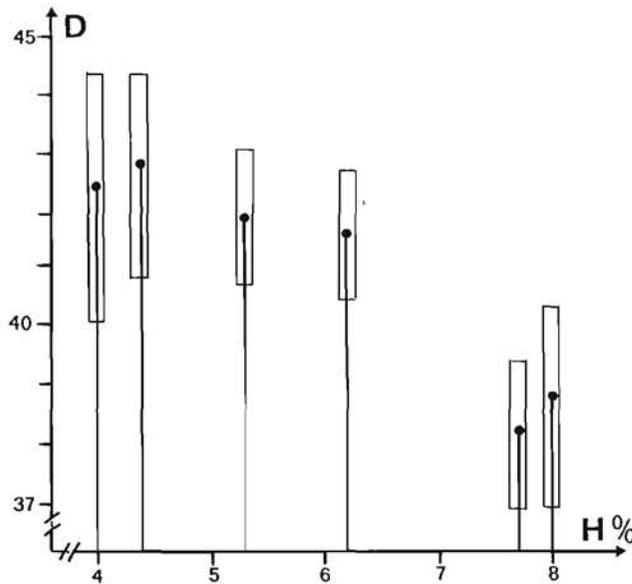
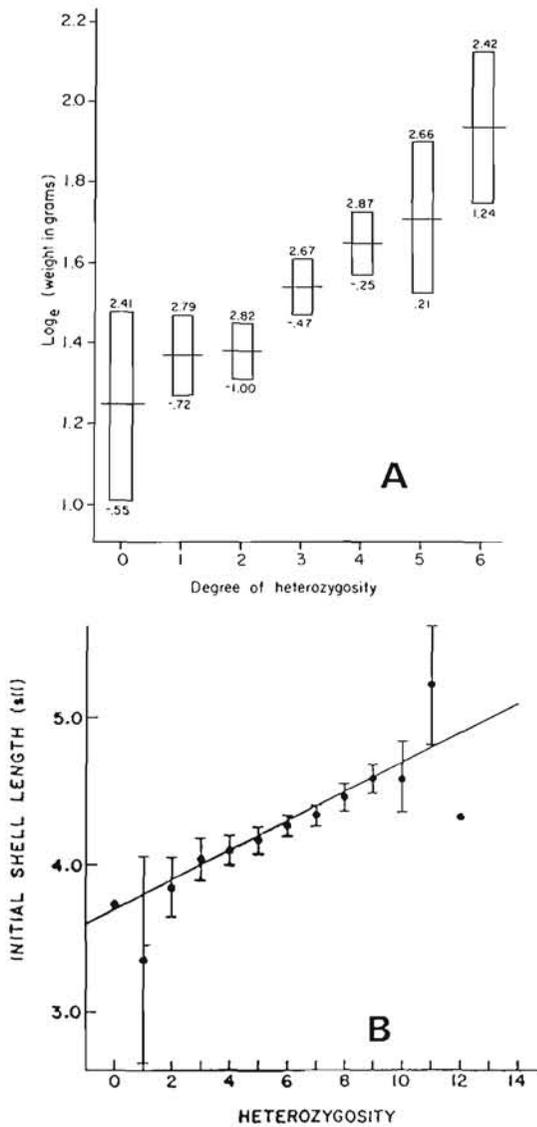


Fig. 9. — Relationship between the duration of embryonic development D (in days) and the mean heterozygosity H in 6 populations of rainbow trout *Salmo gairdneri*. The rectangle gives the standard deviation around the mean (from Ferguson *et al.*, 1985).

The heterozygosity effect may also be revealed on the variability of the trait instead of on its average value. A better « developmental stability » (developmental stability or genetic homeostasis, Lerner, 1954) would result in a lower variance of traits in heterozygous. The already mentioned study of Ferguson *et al.* (1985) (Fig. 9) especially showed that the hatching period was shorter in the most heterozygous populations. In the same way, Mitton (1978) showed in a poeciliidae, *Fundulus heteroclitus*, that



**Fig. 10.** — Two examples of positive relationship between individual heterozygosity (estimated by the number of loci which are heterozygous in a given individual) and growth potential in Molluscs.

A. weight of individuals at 1 year of age in the american oyster *Crassostrea virginica* (Foltz et al., 1983)

b. length of the shell at settlement in *Mulinia lateralis* (Koehn et al., 1988).

the inter-individual variance of meristic characters (number of scales, number of fin rays) was also generally lower in heterozygous individuals. This notion was developed in particular in vertebrates by studies on bilateral dissymmetry of these traits :

based on the counting of even external traits (number of fin rays, branchiospines, scales on the lateral line) it is possible to characterize each individual or each population by a dissymmetry index supposed to measure its « developmental stability » and connect this index with biochemical measures of heterozygosity. Several examples (Fig. 11) illustrate the relevance of this by

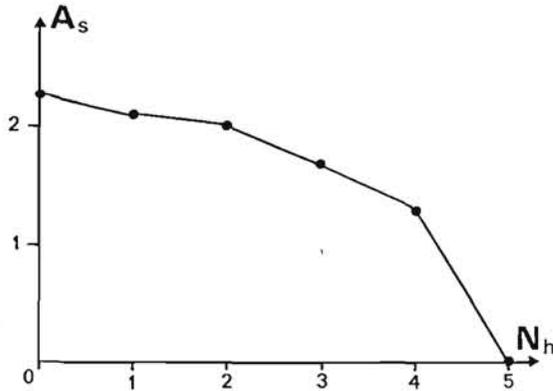


Fig. 11 A. — Individual relationship between number of heterozygous loci  $N_h$  and mean number of asymmetric characters  $A_s$  in a rainbow trout population (from Leary *et al.*, 1983).

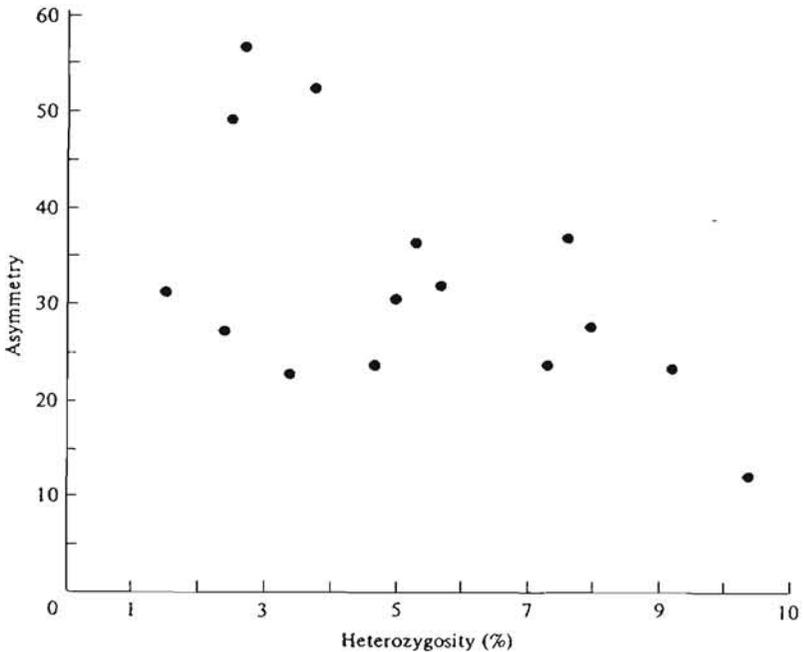


Fig. 11 B. — Relationship between mean heterozygosity and a symmetry coefficient among 14 populations of the lizard *Uta stansburiana* (from Soulé, 1979).

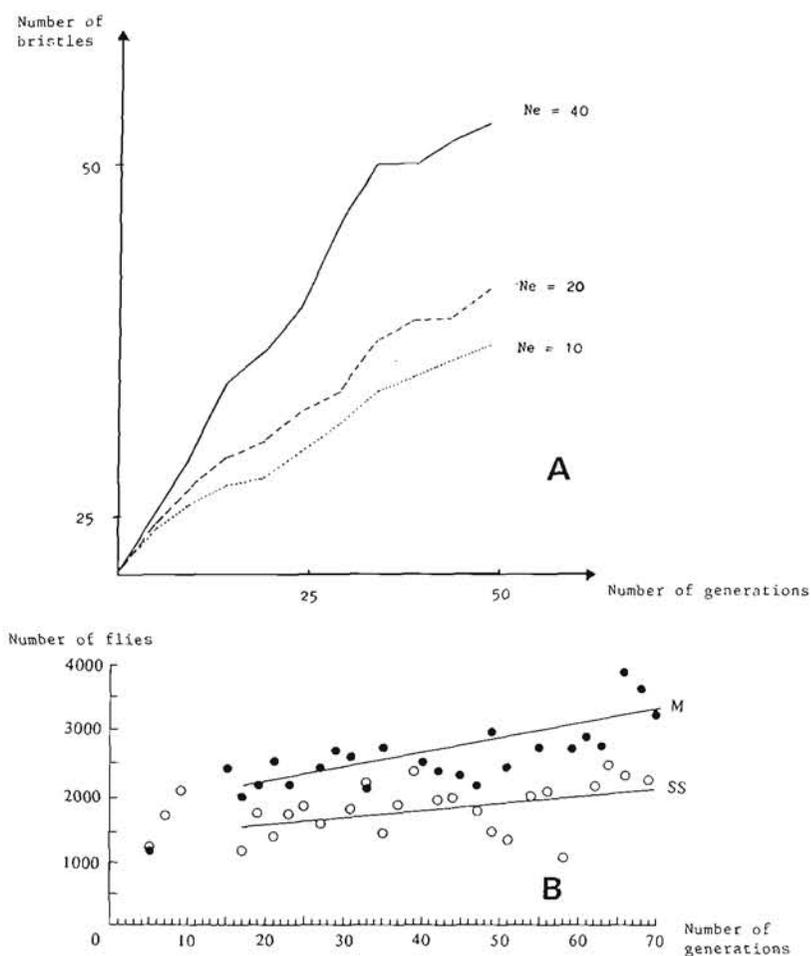
showing an increase in the dissymmetry index when the heterozygosity decreases (Leary et al., 1983, 1984, 1985a).

However, such approaches cannot replace a direct evaluation of performances of individuals or populations : these relationships are not systematic [see especially Mc Andrew *et al.* (1986) in plaice *Pleuronectes platessa*, Beacham and Withler (1985) in pink salmon *Oncorhynchus gorbuscha* for negative results], they only involve some traits and the correlations observed between performance and heterozygosity when they are significant, generally only explain a small part (10 to 30 %) of the total variability of the trait. Therefore, a direct improvement of the traits should not be replaced by methods aiming at maximizing heterozygosity. However, these illustrations seem sufficient to justify *a priori* selection of populations exhibiting a high heterozygosity.

- *In the long term, the response capacity of a population to an environmental change is classically linked to its level of genetic variability.* But domestication, with or without an additional selection of certain traits, constitutes an important « adaptation test » whose final result may partially depend on the population's initial genetic diversity. Carrying out experiments in this field is not possible in the case of aquacultural species but Fig. 12 gives an example of such evolutions in *Drosophila*.

In these conditions the constitution of a stock based on the regrouping of a large number of natural populations appears to be a solution to recommend systematically. However, a considerable modification of this proposal seems to be necessary.

- *In the case of a species with a low differentiation rate,* selection of a population with a high heterozygosity makes it possible to collect a large fraction of the total variability. When heterozygosity varies considerably between populations it is even possible to show that the choice of the population with the highest heterozygosity rate leads to a larger genetic variability than that of a synthetic population regrouping systematically all available populations. Therefore, regrouping two or three of the most heterozygous populations is sufficient. Regrouping live animals is often much more problematic than sampling intended for biochemical analysis. In this approach a progeny testing of performances of some interesting populations may constitute a supplementary selection criterion.
- *In the case of species with a high differentiation rate,* the creation of synthetic populations may have negative consequences in the short or long term which should be evaluated. Beforehand, a comment on the notion of high differentiation rate is probably necessary. As a matter of fact, a simple model known by the name of « molecular clock » (Nei, 1975) makes it possible to relate the genetic distance between two populations and the age of their differentiation. Thus, in the example given in Fig. 6, the average genetic distances between Swedish salmon populations of some 0.3 % are likely to correspond to an isolation of some 15,000 years. Therefore, one should be very prudent towards the possible



**Fig. 12.** — Two examples of relationship between genetic variability and « evolutionary capacity » in *Drosophila*.

**A.** Long term response to selection for the number of sternopleural bristles.  $N_e$  is the effective size of the selected population at each generation.

**B.** Demographic evolution of two experimental populations: SS is a single strain population, M a « synthetic » strain resulting from a mixing of different strains. The average rate of increase in population size is about twice larger in the M population.

(from Ayala, 1965 in Frankel and Soulé, 1981).

disorder created by the regrouping of such populations. However, several examples lead to examine the validity of this approach. In particular experiments aiming at introducing salmonids on the Kerguelen islands conducted for some forty years with a single initial population have led to stocks with the same present differentiation levels (Fig. 13; Guyomard, 1984). Therefore, it seems difficult to concede a major evolutionary significance to differentiation levels of about 1 to 2% unless they are supported,

in the case of some enzyme systems, by marker alleles specific of certain populations.

Thus, in a study on two populations of brown trout coexisting in the same lake and having a relatively low genetic distance (2.5 %), Allendorf *et al.* (1976) were able to show that for an enzyme system (LDH 1) each population had a specific allele (common to all the individuals of one population and absent in the other) which certified the absence of crossings between these two populations. If such a phenomenon should be taken into account in terms of view management of natural populations it should not be interpreted as a genetic incompatibility between the two stocks. Spawner isolation may be the result of various ecoethological phenomena (for instance difference of spawning periods or zones) and only an experimental study would allow to draw conclusions on the possibility of obtaining viable progeny by crossing these populations.

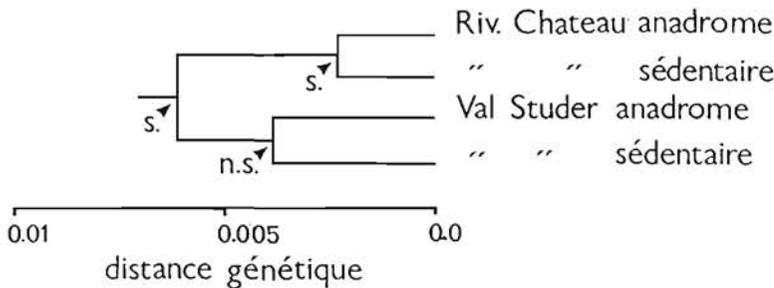


Fig. 13. — Ecological and genetic differentiation between brown trout populations of Kerguelen Islands (Indian Ocean) resulting from the same introduction around 1950.

For higher rates of differentiation (5 to 10 %) there are two possible consequences of the mixing of populations.

- *First generation individuals originating from the crossing between the populations may have a reduced viability.* This is known for interspecific hybrids (see especially Chevassus, 1983). On the other hand, examples in the case of intraspecific crossings are less numerous and not systematic (see in particular Frankel and Soule, 1981, p. 152 for a discussion) and tests carried out in particular between Atlantic and Mediterranean groups of brown trout (differentiation of about 10 %) do not seem to lead to such phenomena. In the same way after restockings in cutthroat trout (Fig. 7), the presence in nature of hybrids between the different populations, and even with a closely related species, rainbow trout, indicates that the viability of these hybrids is not substantially lower even if more accurate measures show slight depressions for some traits (Leary *et al.* 1985; Ferguson *et al.* 1988). Inversely, these crossings may reveal interesting performances, i.e. the heterosis effect (hybrid vigor) in particular illustrated in carp (Wohlfarth *et al.* 1975) and in American catfish (Plomb *et al.*, 1975). Therefore, the relationships between differentiation and performances of crossbreds should be considered according to a

probability curve model (« optimum outcrossing distance », Bateson, 1978) which should be standardized in each case.

- *Second generation individuals may have viability problems even when the first generation reveals to be viable or even more performing than the parental populations.* This phenomenon may in particular be the result of the occurrence of chromosomal abnormalities when the chromosomes of two populations present a certain number of structural differences. In the extreme case first generation individuals may be viable but sterile, and no second generation individual is produced. A gradual disappearance of the less viable individuals as affected by natural or artificial selection can be expected, but some theoretical models illustrate the possibility of establishing a long lasting depression of the fertility of individuals originating from such crossings. Once again examples must be taken outside aquatic species (Frankel and Soule, 1981, p. 152 a review), but they illustrate the necessity of taking this problem into account.

#### 4. Concrete proposals

Because of the different previously mentioned arguments the manager is thus confronted with a possible contradiction between the advantage of regrouping a large genetic variability and the risk of having to face negative phenomena related to the regrouping of incompatible genetic units. Moreover, the relatively discreet and deferred character of these phenomena (advent in second generation) is likely to make their diagnosis difficult without their impact being negligible. Remaining in the case of limited experimental possibilities it seems advisable to adopt a triple strategy.

- *The constitution of a « reference population »* based on one population or a regrouping of extremely similar populations ( $d < 0.01$ ). The proximity being moreover confirmed by geographical or historical considerations. This or these populations should be selected among those exhibiting a high level of heterozygosity.
- *The constitution of an « experimental synthetic population »* based on a larger selection of slightly different populations.  
The maintenance of these two units will be similar and their survival, growth and fertility performances should be evaluated for at least two or three generations especially if the domestication involves a voluntary selection of certain traits. At the end of this period, it will be possible to eliminate one of the two units in particular if their performances differ markedly. On the other hand, if the performances are similar, conserving the two units, if possible in two different hatcheries, will constitute a precious security and possibly allows to develop original management and improvement strategies.
- *The experimental study of crossings between populations representative of strongly differentiated groups.* Taking into account the necessity of continuing this study to the second generation it can

only involve a restricted number of « couples » and shall be conducted at the same time as the other two operations. It may, however, contribute to detecting the effects of heterosis or to identifying original characteristics for subsequent use.

- 
- Allendorf F.W. and R.F. Leary, 1988.** Conservation and distribution of genetic variation in a polytypic species, the cutthroat trout. *Conservation Biology*, **2** : 170 - 184.
- Allendorf F.W., N. Ryman, A. Stenneck and G. Stahl, 1976.** Genetic variation in Scandinavian brown trout (*Salmo trutta*) : evidence of distinct sympatric populations. *Hereditas*, **83** : 73 - 82.
- Ayala F.J., 1965.** Relative fitness of populations of *Drosophila serrata* and *Drosophila birchii*. *Genetics*, **51** : 527 - 544.
- Bateson P., 1978.** Sexual imprinting and optimal outbreeding. *Nature*, **273** : 659 - 660.
- Beacham T.D. and R.E. Withler, 1985.** Heterozygosity and morphological variability of chum salmon (*Oncorhynchus keta*) in Southern British Columbia. *Heredity*, **54** : 313 - 322.
- Chevassus B., 1983.** Hybridization in fishes. *Aquaculture*, **33** : 245 - 262.
- Cross T.F. and J. King, 1983.** Genetic effects of hatchery rearing in Atlantic salmon. *Aquaculture*, **33** : 33 - 40.
- Danzmann R.G., M.M. Ferguson, F.W. Allendorf and K.L. Knudsen, 1986.** Heterozygosity and developmental rate in strain of rainbow trout (*Salmon gairdneri*). *Evolution*, **40** : 86 - 93.
- Ferguson M.M., R.G. Danzmann and F.W. Allendorf, 1985.** Developmental divergence among hatchery strains of rainbow trout (*Salmon gairdneri*). I. Pure strains. *Can. J. Genet. Cytol.*, **27**, 289 - 297.
- Ferguson M.M., R.G. Danzmann and F.W. Allendorf, 1988.** Developmental success of hybrids between two taxa of salmonid fishes with moderate structural gene divergence. *Canadian Journal of Zoology*, **66** : in press.
- Foltz D.W., G.F. Newkirk and E. Zouros, 1983.** Genetics of growth rate in the American oyster : absence of interactions among enzyme loci. *Aquaculture*, **33** : 157 - 165.
- Frankel O.H. and M.E. Soule, 1981.** Conservation and evolution. Cambridge University Press.
- Guyomard R., 1981.** Electrophoretic variation in four french populations of domesticated rainbow trout (*Salmon gairdneri*). *Can. J. Genetic. Cytol.*, **23** : 33 - 47.
- Guyomard R., 1987.** Differentiation génétique des populations de Saumon Atlantique : revue et interprétation des données électrophorétiques et quantitatives. M. Thibault et R. Billard, ed. Restauration des rivières à Saumons, colloque Bergerac, INRA : 298 - 308.
- Guyomard R., 1989.** Diversité génétique et gestion des populations naturelles de truite commune. Colloque sur la truite commune, Le Paralet (France), Septembre 1988, (sous presse).
- Guyomard R., C. Grevisse, F.X. Oury and P. Davaine, 1984.** Evolution de la variabilité génétique inter- et intrapopulation de populations de salmonidés issues de mêmes pools géniques. *Can. J. Fish. Aquat. Sci.*, **41** : 1024 - 1029.

- Hindar, K., N. Ryman and G. Stahl, 1988.** Genetic differentiation among local populations and morphotypes of arctic charr, *Salvelinus alpinus*. *Bil. J. Linn. Sic.* (in Press).
- Jones L.P., R. Frankham and J.S.F. Barker, 1968.** The effect of population size and selection intensity in selection for quantitative character in *Drosophila*. II. Long term response to selection. *Genet. Res.*, **12** : 249 - 266.
- Kimura M. and J.F. Crow, 1963.** The measurement of effective population number. *Evolution*, **17** : 279 - 288.
- Koehn R.K., W.J. Diehl and T.M. Scott, 1988.** The differential contribution by individual enzymes of glycolysis and protein catabolism to the relationship between heterozygosity and growth rate in the coot clam, *Mulinia lateralis*. *Genetics*, **118** : 121 - 130.
- Lacava J. and J. Hughes, 1984.** Determining minimum viable population levels. *Wild Soc. Bul.*, **12** : 370 - 376.
- Leary R.F., F.W. Allendorf and K.L. Knudsen, 1983.** Development stability and enzyme heterozygosity in rainbow trout. *Nature*, **301** : 71 - 72.
- Leary R.F., F.W. Allendorf and Knudsen, 1984.** Superior developmental stability of enzyme heterozygotes in salmonid fishes. *American Naturalist*, **124** : 540 - 551.
- Leary R.F., F.W. Allendorf and K.L. Knudsen, 1985a.** Developmental instability as an indicator of reduced genetic variation in hatchery trout. *Transactions of the American Fisheries Society*, **114** : 230 - 235.
- Leary R.F., F.W. Allendorf and K.L. Knudsen, 1985b.** Developmental instability and high meristic counts in interspecific hybrids of salmonid fishes. *Evolution*, **39** : 1318 - 1326.
- Lerner J.M., 1954.** Genetic homeostasis. Dover Publications, New York.
- Mc Andrew B.J., R.D. Ward and J.A. Beardmore, 1986.** Growth rate and heterozygosity in the plaice, *Pleuronectes platessa*. *Heredity*, **57** : 171 - 180.
- Mitton J.B., 1978.** Relationship between heterozygosity for enzyme loci and variation of morphological characters in natural populations. *Nature*, **273** : 661-662.
- Nei M., 1975.** Molecular population genetics and evolution. North Holland, Amsterdam and New-York, 228 pp.
- Nei M., 1978.** Estimation of average heterozygosity and genetic distance from a small number of individuals. *Genetics*, **89** : 583 - 590.
- Plum J.A., O.I. Green, R.O. Smitherman and G.B. Pardue, 1975.** Channel catfish virus experiments with different strains of channel catfish. *Trans. Am. Fish Soc.*, **104** : 140 - 143.
- Stahl G., 1981.** Genetic differentiation among natural populations of Atlantic Salmon (*Salmo salar*) in northern Sweden. *Ecol. Bull.* (Stockholm), **34** : 95 - 105.
- Stahl G., 1983.** Differences in the amount and distribution of genetic variation between natural populations and hatchery stocks of Atlantic salmon. *Aquaculture*, **33** : 23 - 32.
- Wolfarth G., R. Moav and G. Hulata, 1975.** Genetic differences between the Chinese and the European races of the common carp. II. Multicharacter variation : a response to the diverse methods of fish cultivation in Europe and China. *Heredity*, **34** : 341 - 350.
- Zouros E. and D.W. Foltz, 1987.** The use of allelic isozyme variation for the study of heterosis. *Isozymes : Current Topics in Biological and Medical Research*, **13** : 1 - 59.