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# RECRUITMENT OF WILD AND ARTIFICIALLY REARED COD (<u>Gadus morhua</u> L.) TO THE LOCAL SPAWNING STOCK

by

T. Svåsand, K. Jørstad and T.S. Kristiansen

Directorate of Fisheries, Institute of Marine Research Division of Aquaculture, C. Sundtsgt. 37 N-5000 Bergen, Norway

## ABSTRACT

A stock enhancement study of coastal cod was initiated in 1983, when 19,002 artificially reared cod fry were tagged and released in the Austevoll region \_n western Norway. As a part of this program, 3,946 cod fry were released in Osen ( $60^{\circ}$  4'n 5 13'E), a local spawning ground in Austevoll. The impact of the released cod to the wild cod population in the Osen area, was estimated from biological and genetic samples taken of released and wild cod in 1984 and in the spawning season in 1986 and 1987, when the released cod were expected to recruit to the spawning stock.

The released cod were 26%, 36% and 35% of the 1983-yearclass, caught as juvenile cod in 1984 and on the spawning ground in 1986 and 1987, respectively. The released cod seemed to have had the same recruitment pattern to the spawning stock as wild cod of the same yearclass. Comparison of genetic characters between wild and artificially reared cod demonstrated that a rare allele in the enzyme phosphoglucomutase was not present in the broodstock, and subsequently was lost during the artificial production of fry. However, no genetic changes in the other systems investigated were detected, neither when comparing the reared cod fry with the broodstock, nor during the period from release to recruitment to the spawning stock.

#### INTRODUCTION

In 1983 Norwegian scientists succeeded in mass rearing cod in a natural pond in western Norway (Øiestad <u>et al</u>., 1985). One of the aims of the rearing project was to enable enhancement of coastal cod populations, and a release program with artificially produced cod was started in Austevoll in 1983 (Svåsand, 1985).

One purpose of the investigation was to study the impact of released cod on the local cod population from juvenile to recruitment to the spawning stock. The extent to which the released fish will recruit to the local cod populations has earlier been controversial (Solemdal <u>et al.</u>, 1984).

During recent years, the possible negative genetic effects of mass releases of artificially produced juveniles on wild populations have been discussed (Ryman, 1981; FAO, 1982). In accordance with the guidelines of Hynes <u>et al</u>. (1981), the broodstock were collected from the release area and genetic analyses were incorporated in the release program, both during the artificial production of fry and in the period from release in the sea to the recruitment to the spawning stock in the release area.

#### MATERIAL AND METHODS

## Production of cod fry

The broodstock used in 1983 consisted of wild mature cod caught in the Austevoll region. The fish spawned naturally in a spawning pen, and the eggs were collected and incubated until hatching in the laboratory (Huse and Jensen, 1983). About five days after hatching the yolk sac larvae were released in Hyltropollen (Fig. 1) a marine enclosure in Austevoll. A large fraction of the released larvae survived on natural prey until metamorphosis, and from the age of about 55 days the fish were fed dry pellets (Øiestad <u>et al</u>. 1985). 1.

#### <u>Release experiment - 1983</u>

In September - November 1983, 19,002 cod fry, with a mean length

- 2 -

of 17.6 cm and mean weight of 72.8 g, were transferred from Hyltropollen to be used in a release experiment in Austevoll. The 19,002 juveniles were tagged with Floy Anchor Tags and released at several localities in Austevoll (Svåsand, 1985). Of the total, 3,946 tagged cod juveniles were released in Osen, (Svåsand and Kristiansen, 1985), a local spawning ground in Austevoll (Fig. 1).

# Fishing surveys and collection of biological data

Fishing surveys were conducted in Osen in 1984 and on the spawning ground in Osen in 1986 and 1987 (Table 1). The main purpose of these surveys were to estimate the ratio between released and wild cod of the 1983 yearclass in Osen, both in the first year after release (1984) and after recruitment to the spawning stock (1986 and 1987). Biological samples (length, weight, sex, maturity, otoliths, stomachs) were taken from all cod caught in the surveys.

Local fishermen had received information about the release project through an information campaign, and a reward of 25 Nkr. was announced for each tag returned together with information of size, place and date of recapture (Svåsand, 1985).

# Genetic sampling and analysis

Jenetic samples were taken from both wild and recaptured cod in the surveys. To get enough data for comparison of genetical characters, live cod were also bought from local fishermen in the Austevoll region.

Muscle biopsy, and blood samples taken by sterile needles, were taken from live cod. Blood samples and samples of white muscle tissue were also taken from dead fish, but as soon as possible after the fish were caught.

The blood samples were analysed for haemoglobin variants using agar electrophoresis (Sick, 1961). Horizontal starch gel electrophoresis was used to analyse for the polymorphic enzymes phosphoglucose isomerase (PGI), lactate dehydrogenase (LDH), and phospho-

 $\hat{\beta}(x)$ 

glucosemutase (PGM) (Harris and Hopkinson, 1976). Polymorphic enzymes and nomenclature have earlier been described by Jørstad <u>et</u> <u>al</u>. (1980) and Mork <u>et al</u>. (1982). Homogeneity G-test (Sokal and Rohlf, 1969) was used to test variation in allele frequencies.

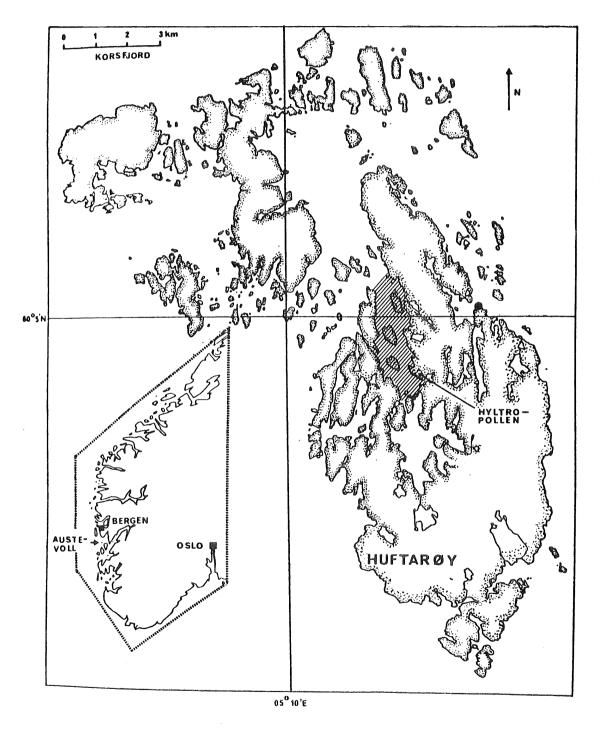


Fig. 1. Map of the release area of juvenile cod in Austevoll in 1983. 3,946 cod fry were released in Osen (hatched area), and 15,056 in other parts of the Austevoll region.

Table 1. Catches of cod and sampling gear used in the fishing surveys in Osen in 1984, 1986 and 1987. The catches are separated in recaptures of cod released in 1983 (rel-83), wild cod of the 1983 yearclass (wild-83), and wild cod of other yearclasses (other wild). Gear is labelled with T:trammel nets, S: single walled gill nets, and stretched mesh length (e.g. T:70 is trammel nets with 70 mm mesh length in inner net).

FISHING PERIOD	SAMPLING G	EAR	CATCHES OF COD							
			N	N	ΣΝ					
	Туре	No.	re1-83	wild-83	other wild					
May - October, 1984	T:70	73	12	24	71					
	T:45	26	1	14	17					
Feb March, 1986	S:140-165	89	2	2	12					
	T:104	67	2	5	19					
	T:45	21	0	0	11					
March, 1987	S:140-165	121	5	7	16					
	S:114-125	65	0	3	7					
	T:104	28	1	1	5					
Sum		490	23	56	158					

#### RESULTS

### Ratio between released and wild cod

In the fishing surveys in 1984 (Table 1), released cod contributed 26% (13 of 51) to the catches from the 1983 yearclass. On the awning ground in 1986 and 1987, the corresponding percentages were 36% (4 of 11) and 35% (6 of 17), respectively. These results indicate an increase in the ratio released/wild cod of the 1983 yearclass in the release area in this period, but the samples were too small for statistical testing.

#### Age distribution and maturity.

In the survey in 1986, the catches of mature cod consisted mainly of two to six year old cod, relatively evenly distributed between the age groups (Fig. 2). Mature cod were here defined as cod with maturing, spawning, or spawned gonads. All recaptured released cod (4) and four of seven (57%) of the wild cod from the 1983 yearclass were mature. More than 50% of the wild cod from the 1984 yearclass were mature, and only one immature cod older than three years were caught. In 1987, the 1983 yearclass was more dominant than the year before. All, except one, from both released and wild cod of this yearclass, were mature, and the ratio relased/wild was similar to 1986. In the catches, the relative numbers of one and two years old cod were smaller than the year before, which can be explained by the use of nets with larger mesh lengths this year (Table 1). All except three cod older than two years were mature (Fig. 2).

The mean length at age of released and wild cod was similar, both in the fishing surveys and when compared to the recaptures from local fishermen in the same period (Table 2).

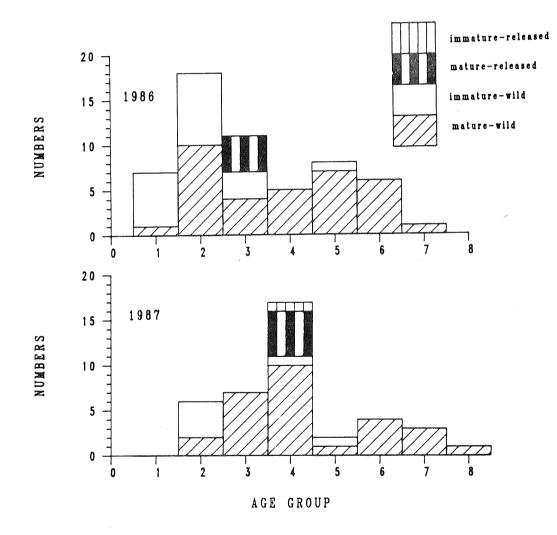


Fig. 2. Age structure in the catches of cod from the spawning ground in Osen in 1986 and 1987. Catches of each yearclass are separated into mature and immature cod.

Table 2. Length distribution of the 1983 yearclass (released and wild cod) caught on the spawning ground in 1986 and 1987 (Fishing surveys) and length distribution of released cod recaptured in the Austevoll region by local fishermen (Recaptures) from January to March the same years.

YEAR	SAMPLING NETHOO	COD TYPE	N	L Min	ENGT <u>Mean</u>	H (mm) Max	(SD)
		1					
1986	Fishing survey	released	4	500	548	590	( 44)
		wild	7	375	504	670	(104)
	Recaptures	released	212	300	532	850	(85)
1987	Fishing survey	released	6	410	650	750	(121)
		wild	11	440	629	700	(80)
	Recaptures	released	79	340	605	900	(110)

# Comparison of genetic characteristics between wild and reared cod

Artificial propagation of fish is likely to include new selection forces, which could result in genetic changes during the production. In enhancement of native stocks, the question is raised if the released fish have the same characteristics as the wild stock.

Table 3 summarizes the results from comparisons between the wild cod stock in the release area, the broodstock, and the offspring relased into the sea in 1983. As shown, rare alleles were `ater detected in the PGI-1 and PGM loci in the samples taken from the local stock. Of these the PGM(150) allele has a very low frequency (0.012), but none of the fish in the broodstock had this allele. Therefore, this allele was missing in the produced cod fry which were later released into the natural environment. No significant differences in allele frequencies were detected for any of the other genetic systems investigated (G-test, p>0.05). Although not significant (p=0.086), a decrease in the PGM(30) frequency was observed.

The allele frequencies estimated for the spawning stock in Osen were also compared to those of juvenile cod in the same area (data not shown), collected outside the spawning season. No significant variation was obtained for any of the genetic systems investigated.

Table 3. Comparison of allele frequencies in samples of the local wild cod stock, the broodstock used for artificial production and the cod fry released into the sea in 1983.

SAMPLE			НЬ1			LDH	-3		F	PGM			F	°GI-1	
	MONTH/YEAR	 N	1	2	 N	70	100	N	30	100	150	N	30	100	150
Osen, wil	ld 2-3/86	120	.579	. 421	118	.352	.648	122	.049	.939	.012	125	.036	.660	.304
Broodstoc	:k 3/83	74	.561	. 439	74	.304	.696	74	.014	.986	-	70	.007	.722	.271
Fry	9/83	191	.558	.442	191	.327	.673	187	.008	.992	-	178	.051	.716	.233

# Genetics of released cod at different ages

After release, the artificially produced fish had to survive in a to look natural environment, and it was therefore of interest genetic changes during the recruitment period. In Table 4, for samples of recaptured cod are compared to broodstock and samples the released cod fry at different ages. The data presented, of however, indicate that no changes have occurred during the period in the sea investigated (p>0.05). It must be pointed out that the number of recaptured fish was low. In addition, the number of low to qive meaningful fish in 1987 was too recaptured comparisons with the other samples shown and were not used in the statistical analyses.

Table 4. Comparison of allele frequencies between broodstock, artificially reared cod fry, and samples of cod recaptured at different time after release into the sea.

SAMPLE				НР1			LDH	-3		ſ	PGM			ł	PGI-1	
	MONT	MONTH/YEAR	N	1	2	N	70	100	N	30	100	150	N	30	100	150
Broodstocl	k	3/83	74	.561	.439	74	.304	.696	74	.014	.986	-	70	.007	.722	.271
Released f	fry	9/83	191	.558	.442	191	. 327	.673	187	.008	. 992	-	178	.051	.716	.233
Recaptured	± 5	-6/84	55	.555	.445	55	.355	.645	55	.064	.936	-	55	.045	.755	.200
Recaptured	d 2	-3/86	57	.535	.465	58	.379	.621	58	.009	.991	-	58	.043	.724	.233

### DISCUSSION

The results from the fishing surveys indicate that the ratio released/wild cod of the 1983 yearclass was at least as high on the spawning ground in 1986 and 1987 as the first year after release. These results are supported by several years studies of released and wild cod in Heimarkspollen (Svåsand and Kristiansen, 1985; Kristiansen, 1987), the small landlocked fjord south of Osen, where no tendency of change in the ratio of released cod in the 1983, 1984 and 1985 yearclass with time, have been found.

Earlier studies of growth and maturity of released and wild cod in Austevoll have shown no significant differences in growth and maturity patterns (Svåsand and Kristiansen, 1985; Kristiansen, 1987), which are in accordance with the investigations on the spawning ground reported here. This indicates that the recruitment patterns of wild and released cod were not significantly different.

Genetic changes during artificial production have earlier been observed in salmonids (Ryman and Ståhl, 1980; Cross and King, 1983; Allendorf and Phelps, 1980) and black sea bream (Tanigushi <u>et al</u>., 1983). Detailed guidelines for artificial production of fish have been proposed by Ryman(1981) and Hynes <u>et al</u>. (1981), pointing out that unwanted genetic changes are usually associated with few parental fish in the broodstock.

Genetic changes from broodstock to offspring are believed to be due to genetic drift, caused by a limited number of fish actively in the spawning at the time when eggs are collected engaged (Tanigushi, 1983; Jørstad, 1986). As mentioned earlier, one rare allele in the PGM system was not present in the selected broodstock used in 1983. This allele, designated 150, is present the natural stock of cod in the area, but was not transferred in to the generation of released fish. The data reported here clearly demonstrate that rare alleles are likely to be lost if specific precautions are not taken. Most important is to use a sufficient large number of broodstock, and to carry out genetic analysis as a control at all stages of artificially production of fish.

The surprisingly high percentage of released cod in the spawning stock (1983 yearclass), may be connected to the use of a broodstock from the local cod population. Except for the loss of the rare PGM(150) allele, no genetic changes were detected neither during the artificial production, nor in the recruitment period in the natural environment. These results suggest that enhancement programs should be based on a broodstock taken from the local populations and a artificial production regime which prevents unintended genetic changes.

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# REFERENCES

- Allendorf, F.W. and Phelps, S.R., 1980. Loss of genetic variation in a hatchery stock of cutthroat trout. Trans. Am. Fish. Soc., 109:537-543.
- Cross, T.F. and King, J., 1983. Genetic effects of hatchery rearing in Atlantic salmon. Aquaculture, 33:33-40.
- FAO, 1982. Conservation of genetic resources in fish: problems and recommendations. Report of the Expert Consultation on Genetic Resources in Fish, Rome, 9-13 June 1980. FAO Fish. Tech. Pap., 217:1-43.
- Harris, J. and Hopkinson, D.A., 1976. Handbook of Enzyme Electrophoresis in Human Genetics. North-Holland, Amsterdam.
- Huse, I. and Jensen, P., 1983. A simple and inexpensive spawning and egg collection system for fish with pelagic eggs. Aquacult. Eng., 2:165-171.
- Hynes, J.D., Brown, E.B., Helle, J.H., Ryman, N. and Webster, D.A., 1981. Guidelines for the culture of fish stocks for resource management. Can. J. Fish. Aquat. Sci., 38:1867-1876.
- Jørstad, K.E., 1986. Genetic studies connected with artificial propagation of cod (<u>Gadus morhua</u> L.). Aquaculture, 57:227-238.

- Jørstad, K.E., Solberg, T. and Tilseth, S., 1980. Enzyme polymorphism expressed in newly hatched cod larvae and genetic analyses of larvae exposed to hydrocarbons. Coun. Meet. Int. Coun. Explor. Sea, (F:22). (Mimeo.)
- Kristiansen, T.S., 1987. Vekst og ernæring til utsatt oppdrettet torskeyngel og vill torsk (<u>Gadus morhua</u> I.) i Heimarkspollen, Austevoll. Cand. scient. thesis, Departement of Fisheries Biology, University of Bergen, Norway.
- Mork, J., Reuterwall, C., Ryman, N. and Ståhl, G., 1982. Genetic variation in Atlantic cod (<u>Gadus morhua</u> L.): a quantitative estimate from a Norwegian coastal population. Hereditas, 96:55-61.
- Ryman, N. (Editor), 1981. Fish gene pools. Preservation of genetic resources in relation to wild fish stocks. Ecol. Bull., 34:1-111.
- Ryman, N. and Ståhl, G., 1980. Genetic changes in hatchery stocks of brown trout (<u>Salmo trutta</u>). Can. J. Fish. Aquat. Sci., 37:82-87.
- Sick, K., 1961. Haemoglobin polymorphism in fishes. Nature, 192:891-896.
- Sokal, R.R. and Rohlf, F.J., 1969. Biometry. W.H. Freeman and Co., San Francisco, CA, 776pp.
- Solemdal, P., Dahl, E., Danielssen, D.S. and Moksness, E., 1984. The cod hatchery in Flødevigen, background and realities. In: E. Dahl, D.S. Danielssen, E. Moksness and P. Solemdal (Editors), The Propagation of Cod, <u>Gadus morhua</u> L. Flødevigen Rapportser., 1:17-45.
- Svåsand, T., 1985. Preliminary results from tagging and release of artificially reared O-group coastal cod (<u>Gadus morhua</u> L.) in western Norway. Coun. Meet. int. Coun. Explor. Sea 1985 (F:9). (Mimeo.).
- Svåsand, T., and Kristiansen, T., 1985. Release of artificially reared O-group coastal cod (Gadus morhua L.) in a landlocked fjord in western Norway. Coun. Meet. int. Coun. Explor. Sea 1985 (F:10). (Mimeo.).
- Tanigushi, N. Sumantadinata, K. and Iyama, S., 1983. Genetic change in the first and second generation of hatchery stock of black seabream. Aquaculture, 35:309-320.
- øiestad, V., Kvenseth, P.G., and Folkvord, A., 1985. Mass production of Atlantic cod juveniles (<u>Gadus morhua</u>) in a Norwegian saltwater pond. Trans. Am. Fish. Soc.,114:590-595.