PAPER

ICES 1992

Demersal Fish Committee
C.M. 1992/G:65
Ref. F

GENETIC STUDIES ON RELEASED AND RECAPTURED COD IN A FJORD SYSTEM

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ABSTRACT

The study reported here is part of a more extensive investigation on the effects of mass rearing and release of 0-group cod in fjords and coastal areas in Norway. Each year since 1987 pond produced cod have been liberated in Masfjorden, a small fjord north of Bergen. The released cod as well as the wild cod and the cod recovered in the fjord have been genetically characterized by electrophoretic analyses of haemoglobins and the enzymes LDH, PGI, PGM and GPD. In 1990 and 1991 about half of the released cod consisted of offspring of broodstock homozygotous for a rare allele (*Pgi-1(30)*. This broodstock was produced by crossing preselected heterozygotes for this allele, the homozygotes among the offspring were sorted out on basis of biopsy sampling of muscle tissue, and when matured, used as parents for the released cod.

In general good accordance between gene frequencies of the released and recovered fish, and no indications of directional selection, were found. Genetic tagging seems to be a useful method both for control of survival, growth rate and dispersal of released cod, and also for more long term studies on hybridization and gene introgression between natural and reared populations.

INTRODUCTION

Sea ranching of cod was started at several locations in Norway in the 80-ties. Monitoring of genetic variations in the natural cod in the release areas, of the released material and of the recovered released fish has been carried out for most of the time. One of the release areas is the small fjord Masfjorden north of Bergen, Western Norway. Here extensive sampling has been carried out, and several year-classes have been successively sampled for many years, included both "wild" and released cod. For two years (1990 and 1991) genetically part of the released cod has been genetically tagged. Production of so-called genetically tagged cod are described a.o. by Blom *et al.* and Skaala *et al.* (1990).

The aim of the present paper is to describe the genetic variation in wild, released and recovered cod in Masfjorden and to study possible selection and/or genetic drift through the cod's life cycle. A further aim was to tentatively evaluate use of genetic tags for studies on interactions between endemic and introduced cod and for survival (relative and absolute) of released cod in a fjord system.

MATERIAL AND METHODS

Juveniles for release in Masfjorden were raised in an enclosed pond (small, isolated fjord) called Parisvatnet in Øygarden northwest of Bergen (Blom *et al.* 1990). The main aim of these culture activities is to produce 0-group cod for sea ranching in nearby fjords and coastal areas. The eggs were spawned naturally in spawning pens at the Austevoll Aquaculture Research Station (belonging to the Institute of Marine Research, Bergen) and released in Parisvatnet as 5 days old larvae. The larvae were raised on natural zooplankton until after metamorphosis, and then fed formulated feed for supplement. They were caught by sinking nets in mid-summer and transferred to net pens in the pond for subsequent tagging and release in the autumn. Released fish were always marked by feeding oxytetracycline (Nordeide *et al.* 1992).

Cod in Masfjorden were caught by net fisheries carried out by the Institute of Marine Research regularly each month. Blood and muscle were sampled for genetic analyses. Biological sampling including age determination was always carried out and such data have been put forward to us by the institute.

The methods described by Jørstad (1984) were applied for analyses of haemoglobins and tissue enzymes. The tissue enzymes stained for are those found by initial screening to be most informative about cod (see Jørstad and Nævdal 1989), including lactate dehydrogenase (LDH), phosphoglucomutase (PGM), glucose-6-phosphate dehydrogenase (GPD) and phosphoglucose isomerase (PGI).

Test of accordance between observed distribution of genotypes and expected Hardy-Weinberg distribution, and of material heterogeneity were carried out using standard X^2 and G-tests.

In each of the year 1990 and 1991 the parent fish in one of two spawning pens were genetically tagged, i.e. they were homozygotous for the allele *Pgi-1(30/30)*, a genotype of the enzyme phosphoglucose isomerase (PGI) which is extremely rare in nature. The genetic tagging process has been described by (Skaala *et al.* 1990).

RESULTS AND DISCUSSION

Gene frequencies calculated from observed genotype distributions are shown in Tables 1-3 (released fish) and Tables 4-5 (wild caught fish). The first line in the first tables represent samples of the produced cod before release. The parent fish for all year classes basically represent the same group of fish, coastal cod from the area around Bergen. Similar gene frequencies of all genetic systems, except PGI, should have been expected. Deviation from this expectance probably represent genetic drift because the effective numbers of spawning cod in the spawning pens probably are low, although unknown, and too much emphasize should not be put on this observations. Concerning PGI the gene frequencies naturally are influenced by the fact that about half of the released fish consisted of genetically tagged fish (homozygotes for the *Pgi-1(30)*-allele).

Gene frequencies of the released fish, recovered in different years, by and large were in accordance with corresponding gene frequencies of the samples collected before release, and when tested by standard G-test, no significant deviations were found.

Neither were found any significant differences between year classes of wild caught cod and between samples of the same year-classes collected in different years (Tables 4-5).

The present results have given no indications of selective forces acting upon genotypes of haemoglobins and tissue enzymes of cod. All between samples observed in the present study may be explained as effects of genetic drift due to rather small (but unknown) effective numbers of spawners in the spawning pens, when only eggs from a small part of the spawning period is used for fry productions.

On the other hand the present results show that genetic tagging is a useful method for estimating survival of released cod and for later interaction studies and studies of hybridization and gene introgression. So far it has not been possible to see any effects on the genetically tagged fish disqualifying this method for studies as mentioned above. These studies will be continued as far as possible until both the near term information (survival, recoveries) and possible long term information (hybridization, gene introgression) of the genetic marking experiments have been utilized. Simultaneous liberation of codlings with "normal" PGI-frequencies offer a good control of the effects of genetic marking.

ACKNOWLEDGEMENT

The authors are greatly indebted to the staff of Institute of Marine Research and Department of Fisheries and Marine Biology, University of Bergen, for collecting the samples and conducting the analyses. Likewise we want to thank the Norwegian Fisheries Research Council for financial support.

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Table 1. Gene frequencies of cod released in 1988 and recovered in the years 1989-91.

Sample characteristics	N	Hb-1		Ldh-3			Pgi-1				Gpd			Pgm		
		1	2	70	100	150	30	70	100	150	90	100	120	30	100	150
Before released	286	0.58	0.42	0.38	0.62	-	0.01	0.02	0.76	0.21	0.01	0.97	0.02	0.02	0.98	-
Recovered 1989	140	0.57	0.43	0.36	0.64	-	0.02	0.03	0.71	0.24*	0.02	0.96	0.02	0.03	0.95	-
Recovered 1990	35	0.49	0.51	0.37	0.62	0.02	0.03	0.08	0.68	0.21						
Recovered 1991	30	0.59	0.41	0.44	0.57	-	0.02	0.02	0.76	0.21	-	0.95	0.05	-	1	-

Table 2. Gene frequencies of cod released in 1989 and recovered in the years 1989-91. n.a.: not analysed.

Sample characteristics	N	Hb-1		Ldh-3			Pgi-1				Gpd			Pgm		
		1	2	70	100	150	30	70	100	150	90	100	120	30	100	150
Before released	252	0.47	0.53	0.26	0.74	-	0.01	0.02	0.83	0.13	-	0.94	0.06	0.04	0.94	0.01
Recovered 1989	117	0.44	0.56	0.29	0.72	-	0.01	0.05	0.81	0.14	-	0.94	0.06	0.02	0.98	-
Recovered 1990	370	0.45	0.55	0.27	0.73	-	0.01	0.03	0.82	0.14		n.a.			n.a.	
Recovered 1991	40	0.49	0.51	0.26	0.74	-	0.05	0.07	0.67	0.21	-	0.98	0.02	-	0.99	0.01

Table 3. Gene frequencies of cod released in 1990 and recovered in 1991.

		Hb-1		Ldh-3			Pgi-1					Gpd			Pgm	
Sample characteristics	**N**	**1**	**2**	**70**	**100**	**150**	**30**	**70**	**100**	**150**	**90**	**100**	**120**	**30**	**100**	**150**
Before released	305	0.59	0.41	0.29	0.71	*	0.54	0.01	0.38	0.01	-	0.88	0.12	0.06	0.94	-
Recovered 1991	68	0.52	0.48	0.31	0.69	0.01	0.49	0.01	0.43	0.06	-	0.87	0.13	0.02	0.98	-

Table 4. Gene frequencies of "wild" cod of the 1988 year class catches in 1989, 1990 and 1991. n.a.: not analysed.

		Hb-1		Ldh-3			Pgi-1					Gpd			Pgm	
Sample characteristics	**N**	**1**	**2**	**70**	**100**	**150**	**30**	**70**	**100**	**150**	**90**	**100**	**120**	**30**	**100**	**150**
Catched 1989	130	0.50	0.50	0.29	0.72	-	0.04	0.01	0.73	0.23		n.a.			n.a.	
" 1990	65	0.55	0.45	0.36	0.64	-	0.03	0.01	0.70	0.26		n.a.			n.a.	
" 1991	64	0.44	0.56	0.30	0.70	-	0.03	-	0.71	0.26	0.01	0.98	0.01	-	1	-

Table 5. Gene frequencies of "wild" cod of the 1989 year class catched in 1990 and 1991. n.a.: not analysed.

		Hb-1		Ldh-3			Pgi-1					Gpd			Pgm	
Sample characteristics	**N**	**1**	**2**	**70**	**100**	**150**	**30**	**70**	**100**	**150**	**90**	**100**	**120**	**30**	**100**	**150**
Catched 1990	40	0.49	0.52	0.33	0.67	-	0.01	-	0.78	0.21		n.a.			n.a.	
" 1991	35	0.51	0.49	0.28	0.72	-	0.03	0.03	0.66	0.28	-	0.99	0.01	-	0.99	0.01