

P A P E R

ICES 1992



Demersal Fish Committee
C.M. 1992/G:66
Ref. F

STUDIES ON ASSOCIATIONS BETWEEN GENOTYPES AND
GROWTH RATE OF 0-GROUP COD

By

Knut E. Jørstad
Institute of Marine Research
P.O.Box 1872, Nordnes
N-5024 BERGEN - Norway

and

Gunnar Nævdal
Department of Fisheries and Marine Biology
University of Bergen
Bergen High-Technology Center
N-5020 BERGEN - Norway

ABSTRACT

The aim of the present study was to investigate genotype dependent growth rate in young cod. The material of 0-group cod was collected from pond produced cod in western and northern Norway, and analysed for genotype distribution of haemoglobins and several tissue enzymes. The mean growth rate (measured as size at sampling) of the different genotypes were compared by analyses of variance. Associations between growth rate and haemoglobin type were found in one sample, and likewise LDH genotype dependent growth rate was indicated in one sample. The results are discussed in relation to the biological significance of biochemical genetic variation.

INTRODUCTION

Studies on blood types and blood proteins of cod, *Gadus morhua* were conducted in Norway in the years 1962 - 1972 (Frydenberg *et al.* 1965, Møller 1968 and unpublished) for applications on identification of self-sustaining populations of this species. Since 1978 likewise tissue enzymes have been studied by starch gel electrophoresis (Jørstad 1984 and unpublished, Jørstad & Nævdal 1989, Gjøsæter *et al.* 1991). The more recent studies have been conducted as baseline studies in connection with artificial propagation and sea ranching of cod.

Karpov & Novikov (1980) found that the functional properties of the haemoglobin genotypes of cod were differentially influenced by environmental temperature. Likewise Mork, Giskeødegård & Sundnes (1984, a,b) found that the three haemoglobin genotypes possessed different growth rate capacities, and Mork & Sundnes (1985) found different survival of different *Ldh-3* and *Pgi-1* genotypes of wild captured 0-group cod which were kept in captivity. Jørstad (1986) found no genotype dependent growth rate in an extensive sampling programme of cod produced in a marine pond, while Gjøsæter *et al.* (1992) found indication, although not consistently, of genotype dependent growth rate in 0-group cod from the southeast coast of Norway.

The aim of the present study was to investigate potential genotype dependent growth rate in 0-group cod raised in semi-natural ponds for use in sea ranching. The results will be of value for understanding the significance of biochemical genetic variation and for the application of gene frequencies or/and genotype distributions in studies of populations structure in this and other fish species.

MATERIAL AND METHODS

Sampling of blood and tissue have been described by Møller (1968) and Jørstad (1984). For analysis of haemoglobin, the method described by Sick (1965) was applied with modifications (Jørstad 1984). Starch gel electrophoresis was used for analysis of tissue enzymes. After an initial screening of a large number of enzymes to identify polymorphic loci, the following were chosen for routine analyses; lactate dehydrogenase (LDH-3), phosphogluco-mutase (PGM), glucose-6-phosphate dehydrogenase (GPD), phosphoglucose isomerase (PGI-1), and isocitrate dehydrogenase (IDH-2). For the present analysis, the genetic systems with a reasonably high number of specimens within two or more genotype groups within samples were chosen i.e. haemoglobin, LDH and PGI.

An overview of the samples is given in Table 1. All samples were collected in the autumn when the fish were 4-6 months old and chose to the time of release of the fish into the natural environment. Samples 1-3 were collected from cod produced in a pond near Tromsø while the rest of the samples were drawn from cod produced in the pond Parisvannet, located on an island west of Bergen. In 1990 and 1991 two groups of fish were produced in the pond; one offspring of non-selected broodfish, and one offspring of genetically tagged broodfish. The production of broodfish homozygotous for the rare allele *Pgi-1(30/30)* is described by Skaala *et al.* (1990) and Svåsand *et al.* (1990). Offspring of the genetically tagged broodfish (called 6a and 7a) are treated as two groups.

Individual length were recorded for all fish sampled. Length at time of sampling is regarded as a measurement of realized growth rate from hatching to sampling. Mean lengths within frequent genotypes were compared by one-way and two-ways ANOVA statistics.

RESULTS AND DISCUSSION

Gene frequencies calculated from observed genotype distributions are shown in Table 1. Considerable variations between samples were found. Frequencies of the gene *Hb-I(1)* in the range 0.25 -0.40 for coastal cod in northern Norway and in the range 0.50 -0.60 in western Norway are in accordance with earlier findings (Frydenberg *et al.* 1965, Møller 1968, Jørstad and Nævdal 1989), while the frequencies of the more common LDH and PGI genes were found to be very high in some of the present samples compared to earlier findings (Jørstad 1984, Jørstad and Nævdal 1989). Likewise rare genes seemed to be absent or extremely rare in some samples. As expected in the samples 6a and 7a only the PGI gene *Pgi-3(30)* is represented while the other genetic systems are unaffected. However, it is not at all sure that the gene frequencies in pond produced fish are representative for the populations from which the broodfish were drawn, because these frequencies probably are influenced by genetic drift. Although the numbers of spawning cod in the spawning pond are high (Svåsand *et al.* 1990), the actual numbers participating in the spawning the few days the eggs were collected, are unknown and probably low. Thus genetic drift is likely to take place, and too much weight should not be put on the variation between samples.

Mean lengths of fish of different genotypes within samples are shown in Tables 2-4. These values were tested by one-way ANOVA within samples, and by two-ways ANOVA for all or groups of samples. Concerning haemoglobins significant differences were found in sample No. 4; fish of genotype *Hb-I(2/2)* showing the highest mean length. This is in accordance with the results of Mork *et al.* (1984 a,b), and Nævdal *et al.* (1992) found highest mean lengths of *Hb-I(2/2)* regardless of environmental temperature under experimental conditions. No trend or tendencies were seen among the non-significant differences, and two-ways ANOVA gave no indication of significant interactions between genotype and sample concerning mean lengths.

Likewise only one sample (sample 6) showed significant differences in mean length concerning LDH genotype. No sign of interactions were found and no trend among the non-significant differences, except that the genetically tagged cod (*Pgi-3(30/30)*) were on average smaller in 1990 and larger in 1991 than the rest of the fish raised in the same pond. This, however, are due to differences in age at start of the pond experiment and not associated with genotype (Blom *et al.* 1990).

Thus the present study have shown indication of genotype dependent growth rate in only two cases of totally 27 comparisons. However, the results are not consistent, and the differences are showing up only in a few samples. This is in accordance with earlier findings (Gjøsæter *et al.* 1992) and the biological significance of biochemical genetic variation in fish still seems obscure.

ACKNOWLEDGEMENTS

The authors want to thank Torstein Pedersen, Ole Ingar Paulsen and Solveig Thorkildsen for

sampling and laboratory analyses, and the Norwegian Fisheries Research Council for Financial support.

REFERENCES

- Blom, G., Svåsand, T., Jørstad, K., Otterå, H., Paulsen, O.I. and Holm, J. Chr. 1990. comparative growth and survival of two genetic strains of Atlantic cod (*Gadus morhua* L.) reared through the early life stages in a marine pond in western Norway. *Coun.Meet. Int.Coun.Explor.Sea 1990 (F: 48)*, 19 pp.
- Frydenberg, O., Møller, D., Nævdal, G. and Sick, K. 1965. Haemoglobin polymorphism in Norwegian cod populations. *Hereditas*, 53: 257-271.
- Gjøsæter, J., Jørstad, K. Nævdal, G. and Thorkildsen, S. 1992. Genotype distributions of cod from the Norwegian Skagerrak coast. *Sarsia* 76: 255-259.
- Jørstad, K. 1984. Genetic analyses of cod in northern Norway. In: E. Dahl, D.S. Danielssen, E. Moksness and P. Solemdal (edits), *The Propagation of cod, Gadus morhua, L. Flødevigen rapportser., 1 (1984): 745-760.*
- Jørstad, K. 1986. Growth studies on cod (*Gadus morhua* L.): comparisons between genotypes identified by electrophoresis. EIFAC/FAO Symposium on Selection, Hybridization and Genetic Engineering in Aquaculture of Fish and Shellfish for Consumption and Stocking. *Bordeaux (France), 27-30 May, 1985 (poster).*
- Jørstad, K. and Nævdal, G. 1989. Genetic variation and population structure of cod, *Gadus morhua* L., in some fjords in northern Norway. *J.Fish.Biol., 35 (Supplement A): 245-252.*
- Karpo, L.K. and Novikov, G.G. 1980. The haemoglobin alloforms in cod (*Gadus morhua* L.), their functional characteristics and distribution in the populations (Translated from russian). *Voprosy ichtiologii (T.20), 6 (125): 823-827.*
- Mork, J., Giskeødegård and Sundnes, G. 1984a, The haemoglobin polymorphism in Atlantic cod (*Gadus morhua* L.): Genotype differences in somatic growth and in maturing age in natural populations. In: E. Dahl, D.S. Danielssen, E. Moksness and P. Solemdal (edits), *The Propagation of cod Gadus morhua, L., Flødevigen rapportser., 1 (1984): 721-732.*
- Mork, J., Giskeødegård and Sundnes, G. 1984b. Population genetic studies on cod (*Gadus morhua* L.) by means of the haemoglobin polymorphism; observation in a Norwegian coastal population. *FiskDir. Skr.Ser.HavUnders., 17: 449-471.*
- Mork, J. and Sundnes, G. 1985. 0-group cod (*Gadus morhua*) in captivity: differential survival of certain genotypes. *Helgol. Meeresunters, 39: 63-70.*
- Møller, D. 1968. Genetic diversity in spawning cod along the Norwegian coast. *Hereditas, 60 (1): 1-32.*

Nævdal, G., Folkvord, A., Otterlei E. and Thorkildsen, S. 1992. Growth rate related to genotype of 0-group cod at three environmental temperatures. *Sarsia* 77: 71-73.

Skaala, Ø., Dahle, G., Jørstad, K.E. and Nævdal, G. 1990. Interactions between natural and farmed fish populations: information from genetic markers. *J.Fish.Biol.* 36: 449-460.

Svåsand, T., Jørstad, K. and Kristiansen, T.S. 1990. Enhancement studies of coastal cod in western Norway. Part I. Recruitment of wild and reared cod to a local spawning stock. *J.Cons.Int.Explor. Mer*, 47: 5-12.

Table 1. Allele frequencies in samples of 0-group cod produced in marine ponds for stocking purposes. 6a and 7a represent samples of genetically tagged cod i.e. offspring of broodstock homozygous for the gene *Pgi-1(30)*.

Sample no.	Locality	Year	Numbers	Hb-I		Ldh-3			Pgi-1			
				1	2	70	100	150	30	70	100	150
1	TROMSØ	1987	140	.31	.69	.39	.60	.01	.02	.01	.63	.34
2	TROMSØ	1988	191	.24	.76	.41	.56	.03	.03	-	.63	.34
3	TROMSØ	1989	172	.24	.76	.36	.61	.03	.02	-	.61	.38
4	PARISVANNET	1988	286	.58	.42	.38	.62	-	.01	.02	.76	.21
5	PARISVANNET	1989	252	.47	.53	.26	.74	-	.01	.02	.83	.13
6a	PARISVANNET	1990	145	.57	.43	.25	.74	-	1.00	-	-	-
6b	PARISVANNET	1990	160	.62	.38	.32	.68	-	.13	.01	.71	.16
7a	PARISVANNET	1991	243	.58	.42	.32	.68	-	1.00	-	-	-
7b	PARISVANNET	1991	162	.55	.45	.43	.57	-	.02	-	.70	.28

Table 2. Mean lengths (in mm) of cod of different haemoglobin types. s: standard error, n: numbers. One sample showing significant genotype dependent length variation is underlined.

Sample no.	Hb-I(1/1)			Hb-I(1/2)			Hb-I(2/2)		
	\bar{I}	s	n	\bar{I}	s	n	\bar{I}	s	n
1	150.9	3.6	14	147.2	1.6	56	145.1	1.9	64
2	131.7	8.7	9	146.3	3.0	72	147.5	2.4	106
3	143.3	5.4	12	147.5	2.3	57	148.2	1.7	97
4	<u>143.2</u>	1.5	94	<u>142.4</u>	1.3	139	<u>150.8</u>	2.3	48
5	162.5	2.8	54	164.2	1.5	127	161.6	2.3	70
6	156.3	2.2	107	156.6	2.1	135	157.5	3.0	53
7	102.8	3.0	110	102.9	1.7	167	100.6	2.6	66

Table 3. Mean lengths (in mm) of cod of different LDH genotypes. s: standard error, n: numbers
 One sample showing significant genotype dependent length variation is underlined

Sample no.	<i>Ldh-3(70/70)</i>			<i>Ldh-3(70/100)</i>			<i>Ldh-3(100/100)</i>		
	\bar{l}	s	n	\bar{l}	s	n	\bar{l}	s	n
1	142.1	3.6	19	146.8	1.5	76	146.7	2.1	48
2	141.4	4.5	33	149.1	3.0	82	146.0	3.1	63
3	147.4	4.1	23	147.6	1.7	77	147.7	2.4	62
4	144.0	2.2	46	142.1	1.3	132	145.9	1.7	107
5	168.5	4.4	23	162.6	1.9	89	162.7	1.6	140
6	<u>172.8</u>	4.6	27	<u>156.8</u>	2.2	120	<u>156.7</u>	1.8	156
7	99.8	3.4	53	101.1	1.6	188	96.0	1.6	165

Table 4. Mean lengths (in mm) of cod of different PGI-genotypes.
 s: standard error, n: numbers

Sample no.	<i>Pgi-1(30/30)</i>			<i>Pgi-1(100/100)</i>			<i>Pgi-1(100/100)</i>			<i>Pgi-1(150/150)</i>		
	\bar{l}	s	n	\bar{l}	s	n	\bar{l}	s	n	\bar{l}	s	n
1	-	-	-	146.6	1.9	53	145.0	1.8	73	149.8	2.7	11
2	-	-	-	144.5	2.7	77	149.1	3.2	77	141.5	5.3	23
3	-	-	-	147.4	2.3	61	147.7	1.8	82	144.1	3.8	23
4	-	-	-	143.0	1.2	161	144.6	1.7	96	140.8	7.4	9
5	-	-	-	162.5	1.2	175	165.6	2.8	53	151.4	4.4	5
6a	155.9	1.8	146	-	-	-	-	-	-	-	-	-
6b	-	-	-	163.6	2.7	83	158.3	4.3	36	122.0	4.0	2
7a	102.8	1.5	243	-	-	-	-	-	-	-	-	-
7b	-	-	-	98.3	2.5	76	88.4	2.1	71	93.4	3.2	9