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A Genetic Stock Structure Study of Dogfish in the Northwest Atlantic

by

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Abstract

Studies of the stock structure of dogfish (Squalus acanthias) using tagging methods suggest that dogfish in the Northwest Atlantic are composed of one stock with an extensive seasonal migration. In this study the amount of genetic differentiation between dogfish from the Gulf of Maine and on the Scotian Shelf was estimated from electrophoretically detectable protein loci expressed in skeletal muscle and liver. Both samples were in Hardy Weinberg equilibrium and showed no genetic differentiation in allelic frequencies. These results support the previous conclusions that there is one stock of dogfish which undergoes large seasonal migrations.

Introduction

The amount of genetic mixing in the population of dogfish in the Northwest Atlantic is currently unresolved, although several studies have been undertaken. For example, Sindermann and Mairs (1961) tried blood typing techniques as an approach to the identification of dogfish sub-populations, but although the study resolved three different alleles in a Gulf of Maine group no further work was done to compare this group with dogfish from other geographical locations.

Several tgging studies have been carried out over most of the species range in order to determine the migration routes and population dynamics of dogfish populations. These studies in general have had low rates of recapture compared to those of commercially important fish species, possibly due to the lack of a significant commercial fishery. Tagging studies from the Pacific show dogfish to be represented by localized (in Puget Sound) as well as far ranging stocks (migrating from Baja California to Japan) (Holland, 1957). More recent work on the Pacific coast, Ketchen (1986) supports Holland's (1957) view about the existence of inshore populations being independent from those off the open coast. Holden distinguished at least three separate stocks in the Northeast Atlantic and both Holden (1967) and Templeman (1954, 1958, 1976) recorded transAtlantic migrations.

Dogfish have been tagged in the Northwest Atlantic by Templeman (1944,

1954, 1976, 1984), Jensen (1961, 1966, 1969), and Shafer (1970). Returns from these tagging studies did not indicate the presence of definitive stocks. Rather they suggested one stock with changes resulting from north-south migrations along the coast of North America related to growth and maturity.

Because of the considerable amount of gene flow suggested by the tagging data, there appears to be a need for genetic data which could delineate genetic relationships and estimate the amount of differentiation of dogfish within the Northwest Atlantic. Such information is most easily obtained through the study of a large number of electrophoretically detectable protein loci. Our objective was to analyse the patterns of genetic variability of several electrophoretic loci in a sample of dogfish from two locations within the species range, in order to provide data on the possible existence of separate stocks within this range.

Materials and Methods

Since the pattern of tag recoveries in NAFO subareas suggests that there are resident groups of dogfish and migratory groups (Jensen, 1969), sampling locations were chosen to reflect any potential separation. Samples were collected during a research cruise in December 1984 from the resident group in the inner Gulf of Maine (comprised of a mixture of mature and immature animals) and an overwintering immature portion of a migratory group from the central Scotian Shelf group: pertinent data for the two samples are given in Table 1. Tissue samples were stored at -40° C before electrophoretic analysis. Extracts of white muscle and liver from each population were analysed by starch gel electrophoresis. Electrophoretic techniques followed those of McGlade et al. (1982) using three buffer systems.

Buffer 1: TRIS EDTA Borate Ph 8.7 (Odense and Leung, 1975). - Gel and Electrode 121 gm TRIS, 12 gm EDTA and 9.2 gm boric acid in 2L; diluted 1:3 with distilled H_2O.

Buffer 2: Amine citrate Ph 6.9 (Clayton and Tretiak, 1972). - Gel 0.002M citric acid, and electrode 0.04M citric acid (M=mol/L) both adjusted to Ph 6.9 with N-3aminopropyl diethanolamine.

Buffer 3: Discontinous TRIS citric acid (Ridgway et al., 1970). - Gel (Ph 8.5) 0.03M TRIS, 0.005M citric acid 0.0006M boric acid. Electrode (Ph 8.5) 0.06M lithium hydroxide, 0.03 M bortic acid.

Gels were visualized according to modifications in the recipes of Shaw and Prasad (1970): allelic nomenclature followed that of Allendorf and Utter (1979), May et al. (1979), and May (1980) in which loci are numbered sequentially from cathode to anode. The most common allele is designated 100 and all others labeled by their migratory ratio to the common allele.

Data were collected for the following enzymes. (Abbreviations and Enzyme Commission numbers in parenthesis); lactate dehydrogenase (LDH 1.1.1.27), malate dehydrogenase (MDH 1.1.1.37), isocitrate dehydrogenase (IDH 1.1.1.42), creatine kinase (2.7.3.2), phosphogluconic dehydrogenase (PGDH 1.1.1.43), alcohol dehydrogenase (ADH 1.1.1.1), glyceraldehyde dehydrogenase (GAP 1.2.1.12), glycerol 3 phosphate dehydrogenase (G3P 1.1.1.8), aspartate amino transferase (AAT 2.6.1.1), esterase (EST 3.1.1.1), phosphoglucose isomerase (PGI 5.3.1.9), phosphoglucomutase (PGM 2.7.5.1), leucine aminopeptidase (LAP 3.4.1.1), and total protein, T.P..

Since the prime purpose of the present study was to look at population structure we chose to target on enzyme loci shown to have variants in other fish species (Kirpichnikov, 1981). Genotypic frequencies for all consistently resolved systems were tested for deviations from Hardy Weinberg proportions using the chi-square (χ^2) method for goodness of fit with 1 degree of freedom: the deviation was considered significant at $P < 0.05$. Becuase we had only one variable locus genetic distances were not calculated.

Results and Discussion

A total of 15 loci were detected from the 13 enzyme systems examined. Although activity occured in all the enzyme systems surveyed with the exception of IDH, LAP, and EST only 7 loci yielded reliable data for scoring. These are presented in Table 2. The only genetic variability was found at the AAT-2 locus; the AAT-1 locus was invariant and assumed to be a supernatant form (Avise and Beardmore, 1977). An alternate allele was observed for the AAT-2 locus, the heterozygote was three banded indicating that AAT in dogfish acts as a dimer, as in other species (Ward and Beardmore, 1977; May, 1980). The observed genotypic proportions with corresponding allele frequencies for the AAT-2 locus have been compiled in Table 3. The observed genetypic proportions did not deviate significanty from Hardy Weinberg equilibrium values; the t-test between samples showed no significant differences ($t_s^2 = 100$; $P = 0.001$). This lack of genetic differentiation between groups of dogfish sampled may be the result of substantial gene flow between groups. However only a small portion of the species range was sampled and so we can only infer that dogfish in the Northwest Atlantic are one large migratory stock.

References

Clayton, J.W. and D.N. Tretiak. 1972. Amine-citrate buffers for pH control in starch gel electrophoresis. J. Fish. Res. Bd. Can. 29: 1169-1172.

Holden, M.J. 1965. The stocks of spurdogs (<u>Squalus acanthias</u> L.) in British waters and their migrations. <u>Fish. Invest., Lond.</u> (2), 24(4): 1-20.

Holden, M.J. 1967. Transatlantic movement of a tagged spurdogfish. Nature, 214(5093): 1140-1141.

Holland, A.A. 1957. Migration and growth of the dogfish shark, Squalus acanthias (Linnaeus), of the Eastern North Pacific. State of Wash., Dept. of Fish., Fish Res. Pap. 2(1): 43-59.

Jensen, A.C. 1966. Life history of the spiny dogfish. Fish. Bull., U.S., 65(3): 527-554.

Jensen, A.C. 1969. Spiny dogfish tagging and migration in North American and Europe. ICNAF Res. Bull., 6: 72-78.

Ketchen, K.S. 1986. The spiny dogfish (Squalus acanthias) in the Northeast Pacific and a history of its utilization. Can. Spec. Publ. Fish. Aquat. Sci. 88: 78 p.

Kirpichnikov, V.S. 1981. Genetic bases of fish selection. Springer-Verlag, Berlin, Heideberg, New York 1981.

May, B.P. 1980. The salmonid genome: Evolutionary restructuring following a tetraploid event. Ph.D. Thesis, Pennsylvania State University, State College, PA. 199 p.

McGlade, J.M., M.C. Annand, and T.J. Kenchington. 1983. Electrophoretic identification of Sebastes and Helicolenus in the northwestern Atlantic. Can. J. Fish. Aquat. Sci. 40: 1861-1870.

Odense, P.H. and T.C. Leung. 1975. Isoelectric focusing on polyacrylamide gel and starch gel electrophoresis of some gadiform fish lactate dehydrogenase isozymes. p. 485-501. In: C.L. Markert [ed.] Isozymes Vol. III. Academic Press, New York, N.Y.

Ridgeway, G.J., S.W. Sherburne, and R.D. Lewis. 1970. Polymorphism in the esterases of Atlantic herring. Trans. Am. Fish. Soc. 99: 147-151.

Shafer, T.C. 1970. Migration and distribution of the spiny dogfish (Squalus acanthias L.) in the western North Atlantic. MS Thesis, Univ. of Rhode Island. 45 p.

Sindermann, C.J. and D.F. Mairs. 1961. A blood group system for spiny dogfish (Squalus acanthias L.). Biol. Bull. 120(3): 401-410.

Templeman, W. 1944. The life history of the spiny dogfish, Squalus acanthias, in the Northwest Atlantic. NAFO Sci. Coun. Studies 3: 47-52.

Templeman, W. 1954. Migrations of spiny dogfish tagged in Newfoundland waters. J. Fish. Res. Bd. can. 11(4): 351-354.

Templeman, W. 1958. Grand Bank tagged dogfish moves to Iceland. Fish. Res. Bd. Canada, Atl. Prog. Rep., No. 70, p. 28-30.

Templeman, W. 1976. Transatlantic migrations of spiny dogfish. J. Fish. Res. Bd. Can., Vol. 33, 1976: 2605-2609.

Templeman, W. 1984. Migrations of spiny dogfish, Squalus acanthias, and recapture. Success from tagging in the Newfoundland area, 1963-65.

Ward, R.D. and J.A. Beardmore. 1977. Protein variation in the plaice, Pleuronectes platessa L. Genet. Res. Camb. 30: 45-62.

Table 1. Sampling data for dogfish used in electrophoretic study.

Location	Lat. and Long.	Date	No. of Fish	Median Length	Gear	Sex Ratio
1) Gulf of Maine	414434 693421		26			
	415598 693510		3			
	413471 691414	Dec/84	2	90 cm	Trawl	76 F
	413726 692677		8	76 cm		23 M
	413709 693508		16			
	424285 685656		27			
	424534 684708		17			
2) Scotian Shelf	431648 620835		48	66 cm		79 F
	433472 613747	Dec/84	56	62 cm	Trawl	25 M

Table 2. Enzyme systems of dogfish scored consistently.

Enzyme	Tissue	No. of Loci Scored	N	Variant alleles	Buffer
LDH	Liver, Muscle	2	196	invariant	I,II
MDH	Muscle	1	196	invariant	I,II
G3P	Muscle	1	196	invariant	I
AAT	Muscle	2	196	one variant allele	I
TP	Muscle	1	196	invariant	I,II

Table 3. Genotypic composition with corresponding allele frequency at AAT-2 locus in dogfish muscle. Test of goodness of fit between observed and expected (in parenthesis) values revealed no significant deviations.

Species	Location	Locus	Genotypic Frequency			N	Allele Frequency	Goodness of fit χ^2
			100/100	100/88	88/88			
Dogfish	Scotian Shelf	AAT-2	25(20)	40(50)	35(30)	100	.45 .55	1.25 (D.F.=1)
Dogfish	Gulf of Maine	AAT-2	26(21)	38(48)	32(27)	96	.47 .53	1.18 (D.F.=1)