THE UNIVERSITY OF MANITOBA

EVIDENCE FOR THE EXISTENCE OF SUBPOPULATIONS OF LAKE WHITEFISH, COREGONUS CLUPEAFORMIS (MITCHILL) IN LAKE WINNIPEG

by

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A dissertation submitted to the Faculty of Graduate Studies of the University of Manitoba in partial fulfillment of the requirements of the degree of

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This thesis is dedicated to my grandfather, Gisli Benson, who fished Lake Winnipeg for many years.

ABSTRACT

Lake whitefish in Lake Winnipeg can be differentiated into at least two subpopulations on the basis of significant differences in the frequencies of alleles at the glycerol-3-phosphate dehydrogenase (G-3-PDH) b locus. Samples from a spawning run in the Lake St. Martin-Dauphin River area show an average b^3 frequency of 0.77; while samples of spawners in the remainder of Lake Winnipeg and Little Playgreen Lake show an average of 0.46. As well, biochemical differences (G-3-PDH) indicate that hatchery-reared lake whitefish, obtained from stock in Clearwater Lake and William Lake and planted in Lake Winnipeg at Dauphin River and Grand Rapids, have not contributed significantly to the size of the local whitefish stocks.

On the basis of morphological differences (morphometric measurements and meristic counts) lake whitefish in Lake Winnipeg have been tentatively divided into five subpopulations. Samples from the Lake St. Martin-Dauphin River area are distinct both in G-3-PDH b allele frequencies and in morphological characteristics. Samples from the remainder of Lake Winnipeg, although homogeneous with respect to allele frequencies, can be separated on the basis of morphological differences into four groups spawning at Traverse Bay, Berens-Poplar-Big Black Rivers, Grand Rapids, and Little Playgreen Lake.

ACKNOWLEDGEMENTS

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INTRODUCTION

Identification of subpopulations within species of commercially harvested fishes is necessary to effective fishery management. Subpopulations may differ in their capacity to support harvests and, in order to maintain an optimum yield from all fish stocks, each subpopulation must be managed separately. Management without regard to subpopulation structure may result in under harvest of some stocks and over harvest of others. Further, failure to recognize the existence of subpopulations can result in the complete elimination of vulnerable substocks and subsequent loss of genetic variability within the population in general (Larkin 1977).

The subpopulation can be defined as a fraction of a population that is itself genetically self-sustaining (Marr 1957). This definition is similar to the local population or deme defined by Mayr (1963). The subpopulation is also referred to as a substock (Larkin 1977) or unit stock (Parrish 1964). Subpopulations may have their own characteristics of growth, mortality, recruitment, migration and behavior, more or less independent from one another and, since they sometimes inhabit the same general locality, may be exploited together during part or all of their lifetime (Marr and Sprague 1963).

Marr and Sprague (1963) group methods of studying subpopulations into four categories. They include (1)the study of fish movements as revealed by tagging or marking (2)the study of vital statistics (3)the study of phenotypic qualities (meristic and morphometric characteristics) and (4)genetic characteristics. Tagging experiments are costly in time and effort and are limited to areas where an effective recapture method exists. Vital statistics, according to Marr and Sprague (1963), refer more to a definition of subpopulations than to a tool for studying them. Phenotypic characteristics, although often environmentally modifiable, can be of practical importance in delineating subpopulations provided certain conditions are met during sample collection. The study of genetic characteristics, that is measurable characteristics which bear a direct relationship to the genotype and are not environmentally modifiable, can be a very useful tool in identifying subpopulations. In particular, gene frequency data based on allelic polymorphisms can serve to characterize discrete, reproductively isolated subpopulations (Wilkins 1972).

In order to successfully employ any or all of the above mentioned techniques it is imperative that sampling of specimens be carried out on spawning assemblages. Specimen collections during the remainder of the year could contain mixtures of substocks and results will be less than precise if not impossible to interpret.

Tagging studies by Pollard (1973a) suggest that subpopulations of lake whitefish exist in Lake Winnipeg. Kennedy (1954) refers to the existence of distinct schools of lake whitefish in Lake Winnipeg, and other tagging studies (Anon. 1959, 1960, 1961) tend to support the belief that subpopulations of lake whitefish can be found spawning in certain areas of Lake Winnipeg.

This study deals with the identification of subpopulations of lake whitefish in Lake Winnipeg utilizing biochemical techniques for the analysis of genetically determined characteristics, and phenotypic comparisons using morphometric measurements and meristic counts. As well, lake whitefish from lakes used to supply hatcheries on Lake Winnipeg are compared with indigenous lake whitefish to determine whether hatchery planted fish contribute significantly to the size of indigenous stocks.

METHODS

Specimen Collection

Lake whitefish were collected from spawning assemblages at 7 locations in Lake Winnipeg, one in Lake St. Martin and one in Little Playgreen Lake, during October and November, 1975. Spawning lake whitefish were also collected from Clearwater (Atikameg) Lake and William Lake since spawn collected from these fish are used to supply the hatcheries at Dauphin River and Grand Rapids. All collection locations are shown in Figure 1.

Fish were captured with gill nets, pound nets or beach seines (Table 1). Most specimens were in spawning condition when taken. Males were strongly tuberculate with females somewhat less so. Most males and females had running milt or roe when captured. As soon as possible after capture the specimens were placed in plastic bags, frozen and later shipped to the Freshwater Institute in Winnipeg where they were stored at $-55^{o}C$ until examined.

Biochemical Analyses

Biochemical analyses were carried out on lake whitefish from all locations sampled. Tissue samples were taken after morphometric measurements and meristic counts were completed on each fish. Additionally some specimens not included in the measurement analyses were included in the biochemical analyses.

Figure 1. Locations sampled for spawning lake whitefish during October and November, 1975.

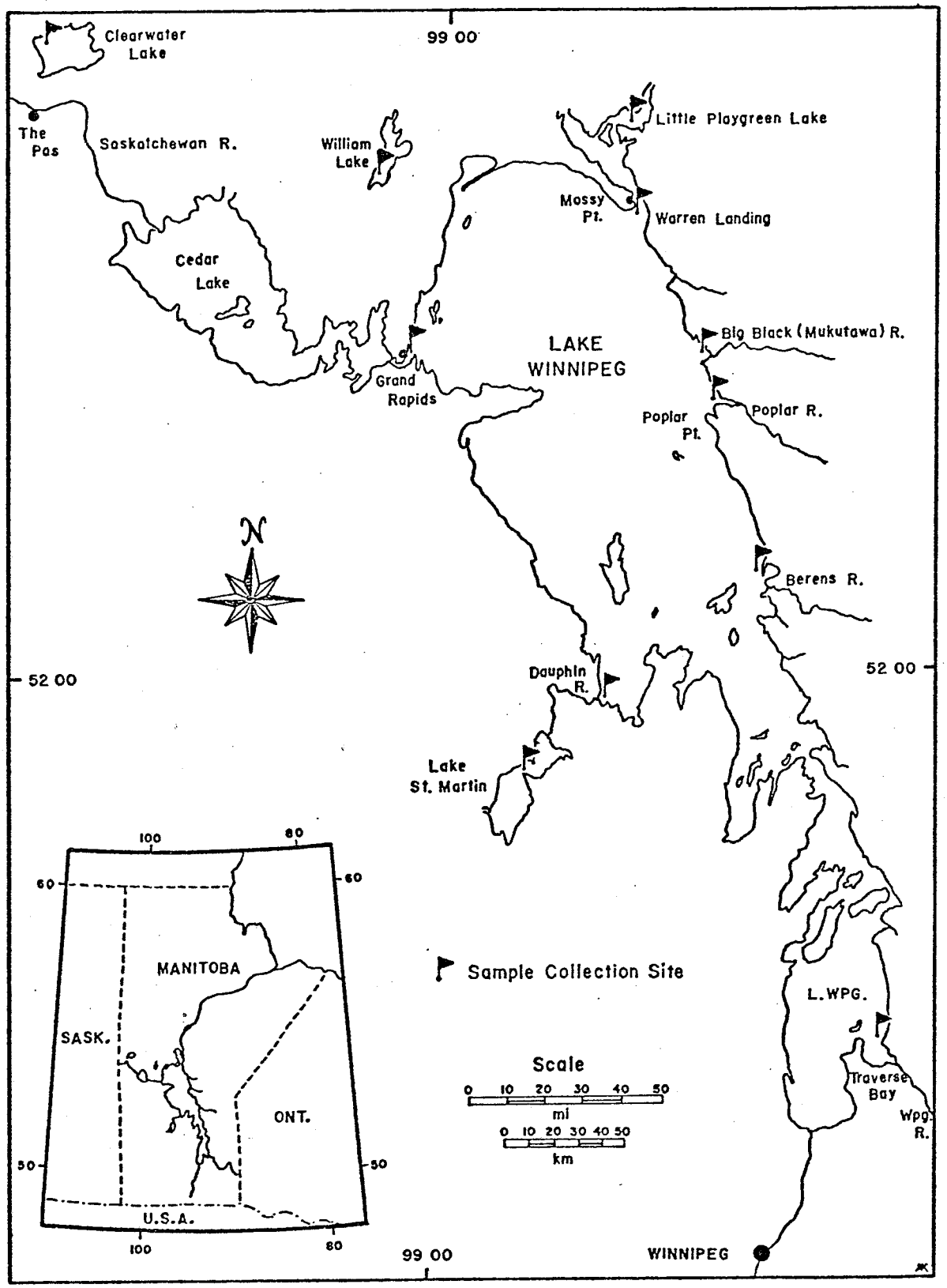

Clearwater Lake
99 00
The Pas
Saskatchewan R.
William Lake
Little Playgreen Lake
Mossy Pt.
Warren Landing
Cedar Lake
Big Black (Mukutawa) R.
LAKE WINNIPEG
Grand Rapids
Poplar Pt.
Poplar R.
N
Berens R.
52 00
Dauphin R.
52 00
Lake St. Martin
100
80
60
60
MANITOBA
SASK.
ONT.
50
50
U.S.A.
100
80
Sample Collection Site
L.WPG.
Scale
0 10 20 30 40 50
mi
0 10 20 30 40 50
km
Traverse Bay
Wpg. R.
99 00
WINNIPEG

Table 1. Catch statistics for whitefish samples, October and November, 1975.

Location	Date of Capture	No. Fish	Gear Used[a]
Traverse Bay (L. Wpg.)	4-12/11/75	48	13.3 cm Gillnet
Berens River (L. Wpg.)	24/10/75	98	9.5 cm Gillnet
Poplar River (L. Wpg.)	25/10/75	99	10.8 cm Gillnet
Big Black River (L. Wpg.)	30/10/75-4/11/75	71	12.7 cm Gillnet
Warren Landing (L. Wpg.)	8,9,10,11/10/75	100	13.3 cm Gillnet
Little Playgreen Lake	17,18/10/75	100	12.1 cm Gillnet
Lake St. Martin	17/10/75	100	Pound Net
Dauphin River (L. Wpg.)	29,30,31/10/75	100	10.8 cm Gillnet
Grand Rapids (L. Wpg.)	31/10/75-1/11/75	205	12.7 cm Gillnet
William Lake	4/10/75	100	Beach Seine
Clearwater Lake	21/10/75	100	Pound Net

[a] Gillnet size is stretched mesh measure.

Tissue Samples

A white muscle sample of approximately 0.5 g was excised from the epaxial muscle bundle just below the dorsal fin. A red muscle sample of similar size was taken from the caudal peduncle along the lateral line. Tissue samples were placed in plastic centrifuge tubes and frozen for later analysis.

Extract Preparation

Tissue extracts for G-3-PDH analysis were prepared by thawing white muscle tissue and macerating it at approximately a 1:1 ratio of tissue to a solution of 300 mg/l nicotinamide adenine dinucleotide (NAD). Extracts for LDH were prepared by thawing red muscle tissue and macerating it at approximately a 1:3 ratio of tissue to a solution of 300 mg/l NAD. In both cases the extracts were spun at 7000 x G and 1°C for 10 minutes in a Fisher Model 59 centrifuge. The extracts were then removed by pipette and frozen until required.

Electrophoresis

Starch gel electrophoresis of tissue extracts was done as described by Tsuyuki *et al.* (1966). Starch gel buffers were 0.00200 M citric acid adjusted to pH 8.0 with tris (hydroxymethyl) amino methane. Electrode buffers were made using 0.0400 M citric acid. NAD was added to starch and bridge buffers to a concentration of 100 mg/l for the

G-3-PDH electrophoresis. G-3-PDH phenotypes were visualized by the methods of Clayton et al. (1973). LDH phenotypes were visualized according to the methods of Clayton and Gee (1969). LDH genotypes were inferred from the genetic model presented by Clayton and Franzin (1970). Nomenclature follows Bailey et al. (1976) and Franzin and Clayton (1977).

Statistical Analyses

G-3-PDH and LDH gene frequencies were calculated for samples from each location. The calculated Castle-Hardy-Weinberg distribution of BB G-3-PDH phenotypes were compared with observed BB G-3-PDH phenotypic distributions to determine homogeneity within samples (Marr and Sprague 1963). G-3-PDH gene frequencies were compared between locations sampled using a maximum likelihood ratio test (Appendix I). Calculations were made using the University of Manitoba IBM Model 370 computer. LDH gene frequencies were not subjected to statistical analysis.

Morphological Analyses

Subsampling Procedure

A subsample of 48 fish was taken from each of the original samples for morphological analyses. Each subsample included representatives of all size classes present in the original samples. Selection of fish involved measuring the

fork length of each fish with a metre stick and tallying the selected sizes as the subsample was drawn. Sex was determined by gross examination of gonads and equal numbers of males and females were included in each subsample where possible (Table 2).

Morphometric Measurements

Prior to examination, specimens were thawed overnight at 1°C. A total of 22 morphometric measurements were carried out on fish from Lake Winnipeg, Lake St. Martin and Little Playgreen Lake. All measurements were taken on the left side of the fish where possible. Measurements were straight line and did not follow body contour.

The morphometric measurements described below and illustrated in Figure 2 were taken using a measuring board graduated to 1 mm.

Adipose fin origin (ADO) - from the snout to the origin of the adipose fin.

Anal fin origin (AO) - the distance from the snout to the anterior face of the anal fin with the fin extended.

Dorsal fin origin (DO) - the distance from the snout to the anterior face of the dorsal fin with the fin extended.

Table 2. Size interval and sex ratio of subsamples (N=48) of lake whitefish used in morphological analyses.

Location	Size Interval (Fork Length in mm)	Sex Ratio (Male:Female)
Traverse Bay (L. Wpg.)	390-540	1.5:1
Berens River (L. Wpg.)	310-470	1:1
Poplar River (L. Wpg.)	320-520	1:1
Big Black River (L. Wpg.)	310-520	1.4:1
Warren Landing (L. Wpg.)	380-550	1:1
Little Playgreen Lake	360-510	2.4:1
Lake St. Martin	300-480	1:1
Dauphin River (L. Wpg.)	320-500	1:1
Grand Rapids (L. Wpg.)	350-540	1:1
William Lake[a]	320-460	1:1
Clearwater Lake[a]	340-430	1:1

[a] Only gill raker number counted on specimens from these lakes.

Figure 2. Measurements taken on lake whitefish using a measuring board. See text for description of measurements and references.

ADO - adipose fin origin

AO - anal fin origin

DLAD - dorsal to adipose distance

DO - dorsal fin origin

FL - fork length

HDL - head length

PCL - pectoral fin length

PCO - pectoral fin origin

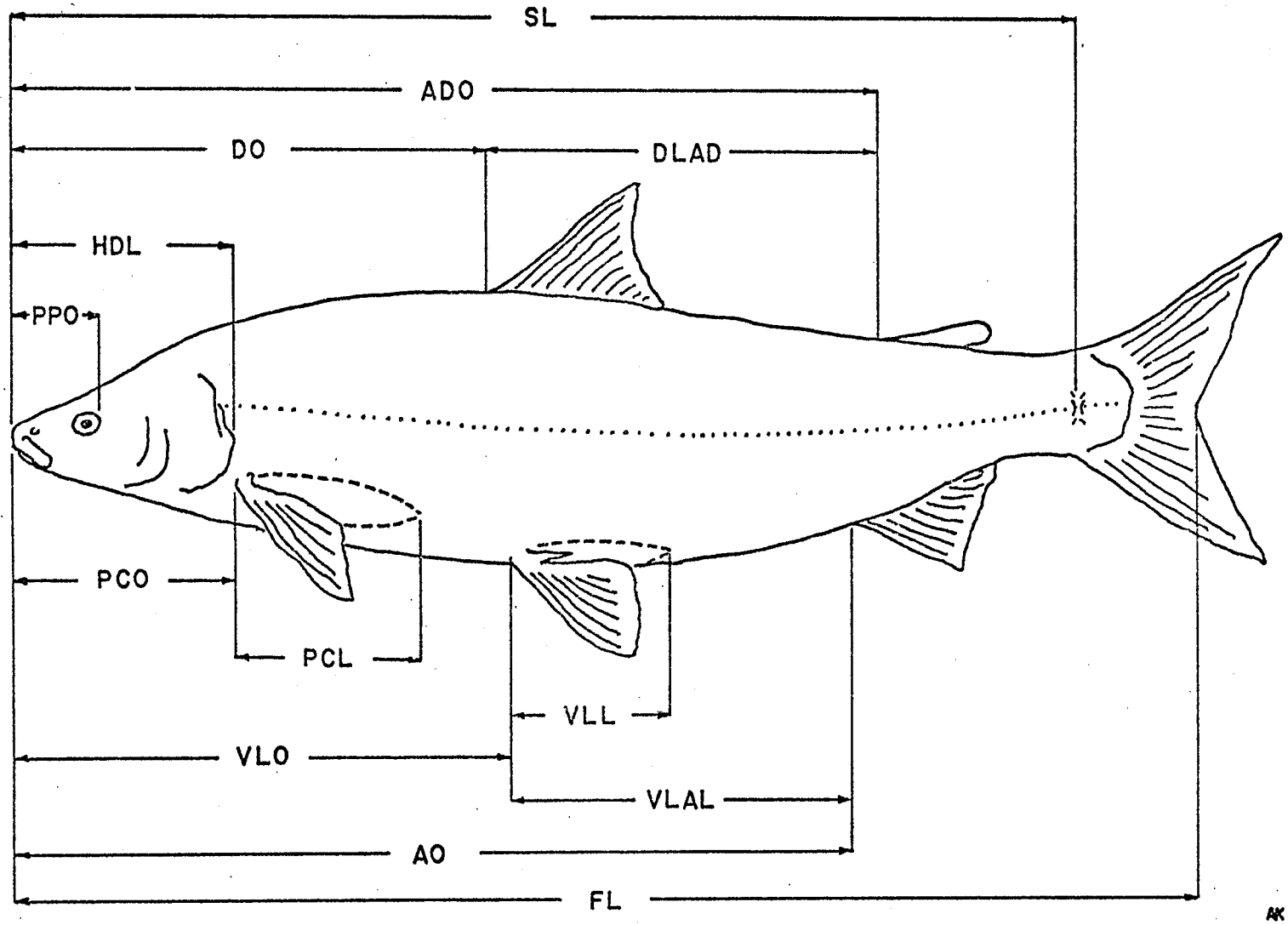
PPO - prepostorbital distance

SL - standard length

VLAL - ventral to anal distance

VLL - pelvic fin length

VLO - pelvic fin origin



AK

Dorsal to adipose distance (DLAD) - the distance from the origin of the dorsal fin to the origin of the adipose fin calculated by subtracting dorsal origin (DO) from adipose origin (ADO).

Fork length (FL) - after Lindsey (1963).

Head length (HDL) - after Lindsey (1963).

Pectoral fin length (PCL) - after Lindsey (1963).

Pectoral fin origin (PCO) - after Lindsey (1963).

Pelvic fin origin (VLO) - after Lindsey (1963).

Pelvic fin length (VLL) - calculated by subtracting pelvic fin origin (VLO) from the distance from the snout to the posterior tip of the longest pelvic fin ray with the fin lying backwards against the body.

Prepostorbital distance (PPO) - after Lindsey (1963).

Standard length (SL) - after Lindsey (1962).

Ventral to anal distance (VLAL) - the distance from the pelvic fin origin to the anal fin origin calculated by subtracting pelvic fin origin (VLO) from anal fin origin (AO).

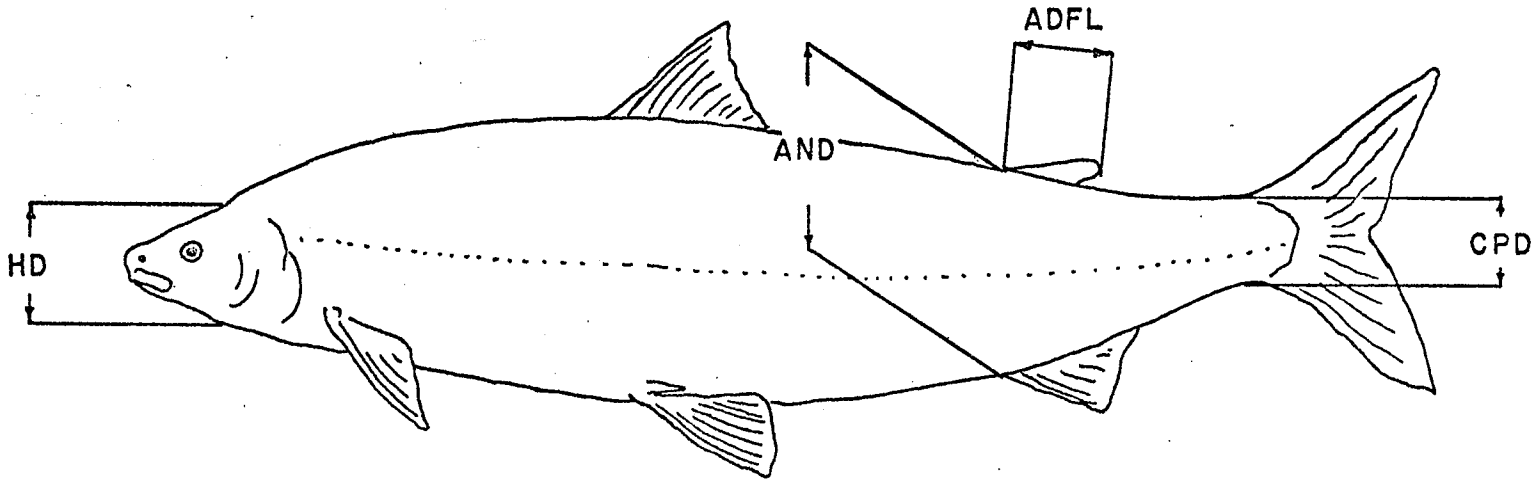
The morphometric measurements described below and illustrated in Figure 3(a,b,c) were made with dial calipers graduated to 0.1 mm.

Figure 3(a,b,c). Measurements taken on lake whitefish using calipers. See text for description of measurements and references.

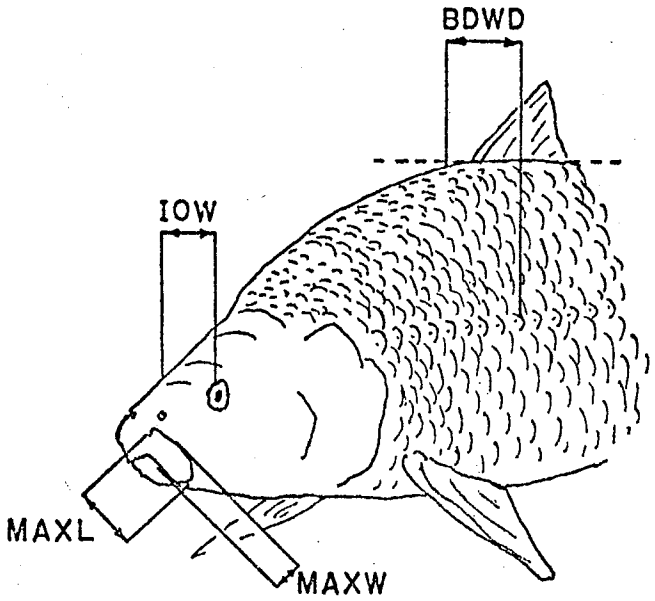
(a) ADFL - adipose fin length
AND - anal depth
CPD - caudal peduncle depth
HD - head depth

(b) BDWD - body width
IOW - interorbital width
MAXL - maxillary length
MAXW - maxillary width

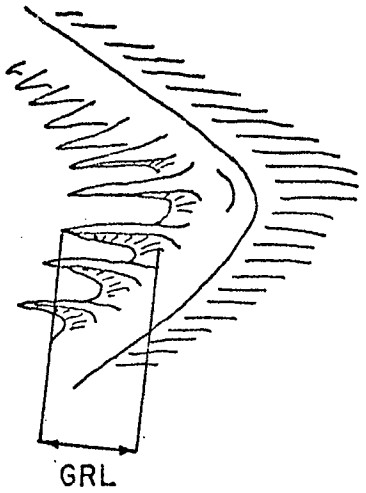
(c) GRL - gill raker length



a.



b.



c.

Adipose fin length (ADFL) - after Lindsey (1962).

Anal depth (AND) - after Loch (1974).

Body width (BDWD) - after Loch (1974).

Caudal peduncle depth (CPD) - after Lindsey (1962).

Gill raker number (GRL) - after Lindsey (1962).

Head depth (HD) - after Lindsey (1963).

Interorbital width (IOW) - after Lindsey (1962).

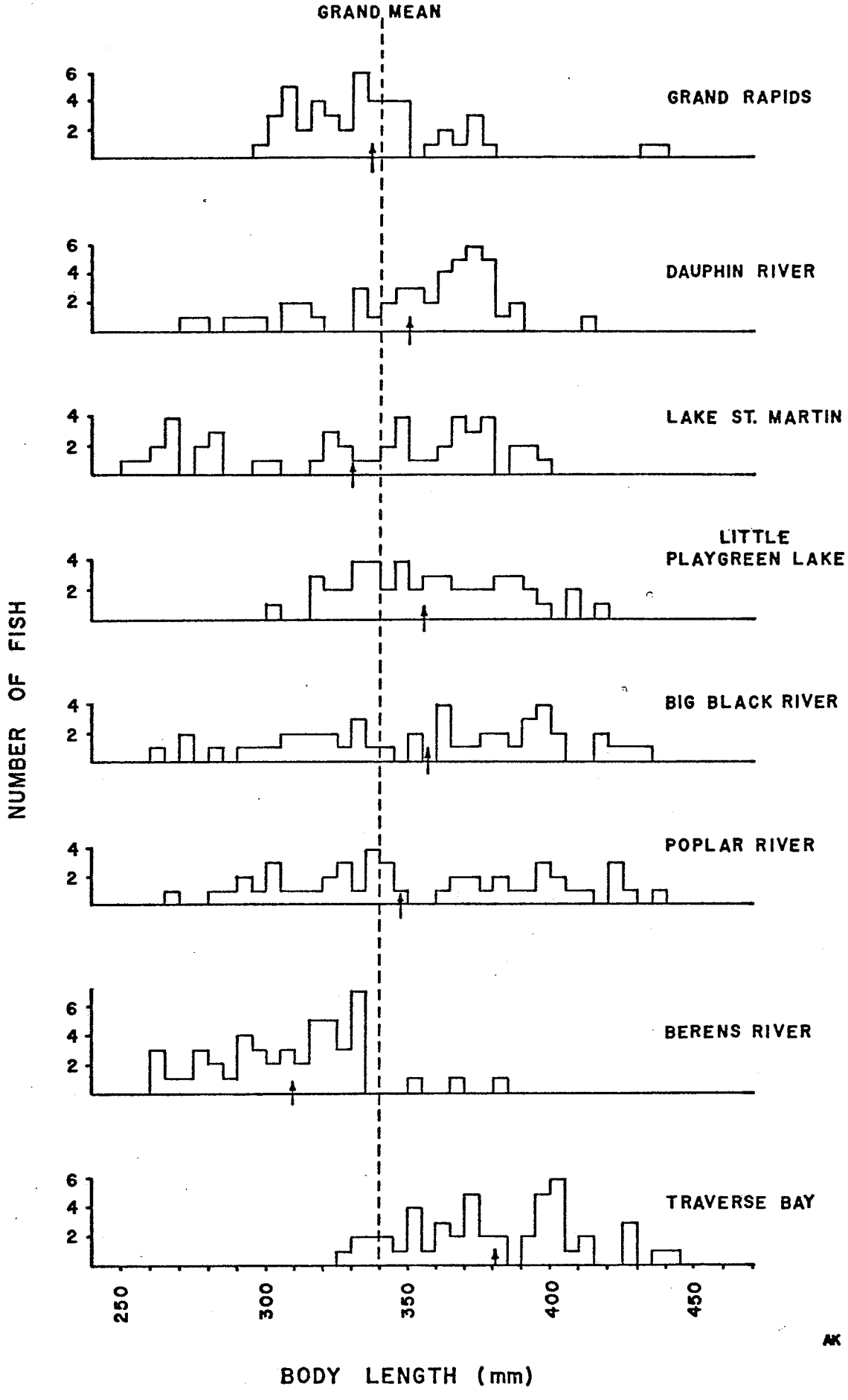
Maxillary length (MAXL) - after Lindsey (1962).

Maxillary width (MAXW) - after Lindsey (1962).

Another variable, body length (BL) was created by subtracting head length (HDL) from fork length (FL).

Statistical analysis. All morphometric data were punched onto computer cards. The data were then $\log_{10}$ transformed and regressed separately for each sample against body length (BL) using Biomedical Computer Program BMDP6D (rev. Aug. 1976) on the University of Manitoba IBM Model 370 computer. The complete data used in this analysis are on file in the library at the Freshwater Institute in Winnipeg. Residuals were printed using the BMDP1R program (rev, Aug. 1976). All morphometric data were then adjusted along the respective regression lines for each sample to correspond to a fish with a body length of 345 mm, using the method described by Lindsey (1963). Body length of 345 mm, the grand mean of all samples, was used since the range of samples from all localities overlapped at this value (Figure 4).

Figure 4. Size distribution of lake whitefish from locations compared using morphometric and meristic analyses. Arrows indicate sample means.


GRAND MEAN
GRAND RAPIDS
DAUPHIN RIVER
LAKE ST. MARTIN
LITTLE PLAYGREEN LAKE
BIG BLACK RIVER
POPLAR RIVER
BERENS RIVER
TRAVERSE BAY
NUMBER OF FISH
BODY LENGTH (mm)
250
300
350
400
450
AK

Adjustments of individual measurements did not involve extrapolation. Adjusted data were compared between localities using the BMDP7M program, (rev. Aug. 1976) stepwise discriminant analysis (Cooley and Lohnes 1971). For examples of the use of discriminant-function analysis see McPhail (1961), Blouw (1976) and Bodaly (1977).

Meristic Counts

Eleven meristic counts were taken on lake whitefish from Lake Winnipeg, Lake St. Martin and Little Playgreen Lake. Counts were made with the naked eye, on the left side of the specimen where possible. Only gill rakers were counted on fish from Clearwater Lake and William Lake. Where scales were missing, scale pockets were counted.

The counts described below are illustrated in Figure 5(a,b,c).

Anal fin ray number (AFRN) - including only principal rays and excluding those anterior rays which were less than three quarters the length of the longest ray. The last two rays arising from a common base were counted as one.

Dorsal fin ray count (DFRC) - after Lindsey (1962).

Gill raker number (GRN) - counted on the entire first left gill arch after it was removed in its entirety from the fish, counting every bony rudiment.

Figure 5(a,b,c). Meristic counts taken on lake whitefish. See text for description of counts and references.

(a) GRN - gill raker number

GRNL - gill raker number, lower limb

GRNU - gill raker number, upper limb

(b) PNCS - caudal peduncle scales

SCAL - scales above the lateral line

SPSC - suprapelvic scale count

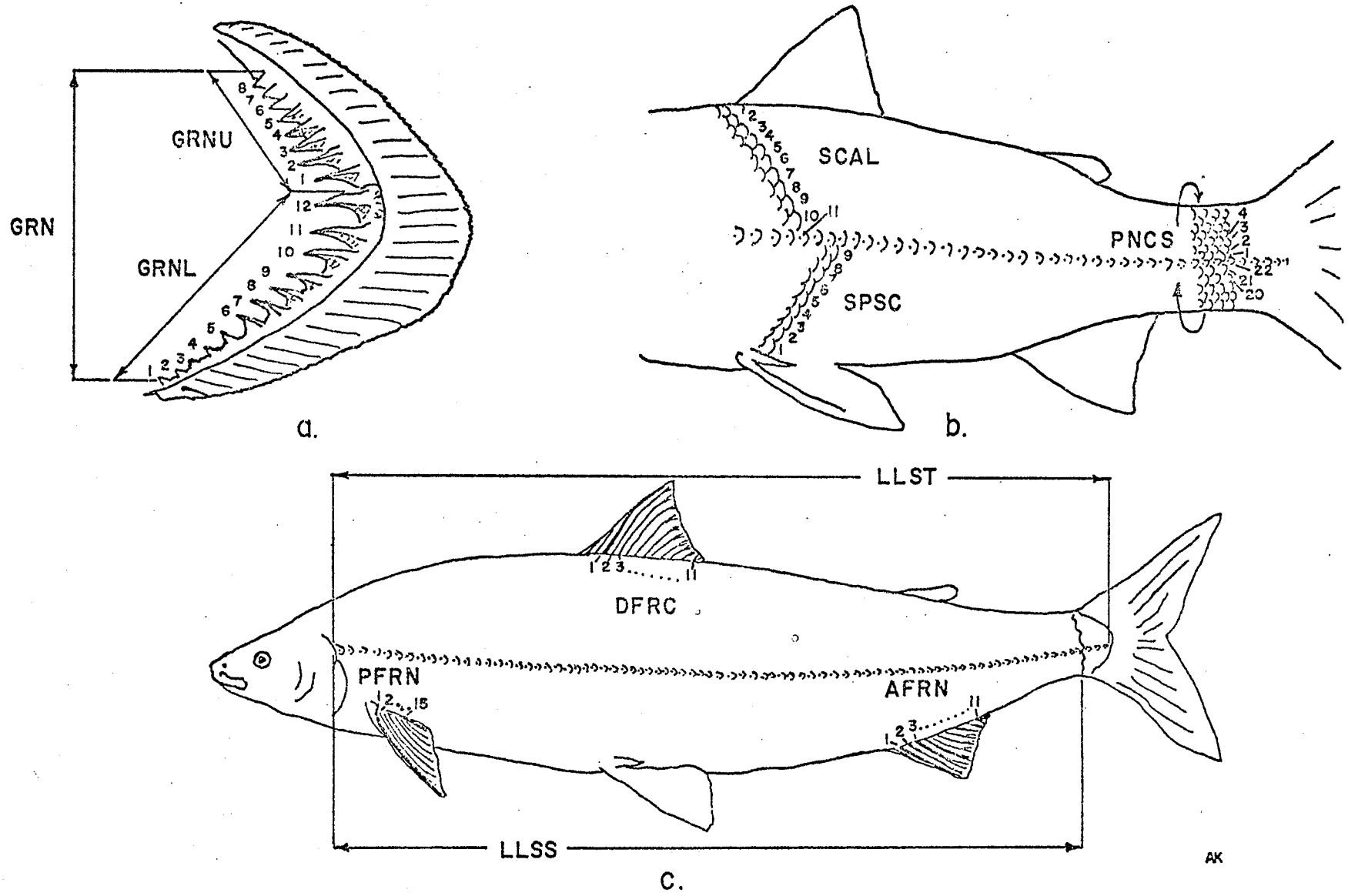
(c) AFRN - anal fin ray number

DFRC - dorsal fin ray count

LLSS - lateral line scales to standard length

LLST - lateral line scales in total

PFRN - pectoral fin ray number


GRN
GRNU
GRNL
a.
SCAL
PNCS
SPSC
b.
LLST
DFRC
PFRN
AFRN
LLSS
AK
c.

Gill raker number, lower limb (GRNL) - those rakers on the lower limb of the first left gill arch, including the raker located at the junction of the upper and lower limbs of the gill arch.

Gill raker number, upper limb (GRNU) - those rakers on the upper limb of the first left gill arch excluding the raker located at the junction of the upper and lower limbs of the gill arch.

Lateral line scales to standard length (LLSS) - starting at the first pored scale touching the pectoral girdle including the last scale anterior to the point marking the end of the standard length.

Lateral line scales in total (LLST) - after Hubbs and Lagler (1964).

Pectoral fin ray number (PFRN) - after Hubbs and Lagler (1964).

Caudal peduncle scale count (PNCS) - after Lindsey (1962).

Scales above the lateral line (SCAL) - after Kliewer (1970).

Suprapelvic scale count (SPSC) - after Lindsey (1962).

Statistical analysis. Meristic data were punched onto computer cards and compared between locations in conjunction with morphometric measurements using BMDP7M, the stepwise discriminant analysis program. Mean gill raker number was compared between some locations using a t-test.

Sexual Dimorphism

Sexual dimorphism was tested in each sample using stepwise discriminant-function analysis with sex rather than location as a grouping variable.

Age and Growth

Scale samples were taken from a position on the left side of each fish just below the origin of the dorsal fin but above the lateral line. Scale ages were determined by Mrs. D. Barnes, an experienced scale reader at the Freshwater Institute in Winnipeg. Scale age was plotted against mean fork length to provide an estimate of growth rates of lake whitefish from the locations sampled.

RESULTS

Biochemical Analyses

Glycerol-3-Phosphate Dehydrogenase (G-3-PDH)

Observed BB G-3-PDH phenotype distributions and expected (Castle-Hardy-Weinberg) distributions for samples of lake whitefish are shown in Table 3. Observed phenotype distributions are in agreement with expected distributions in samples from all locations except Warren Landing. The value calculated for the Warren Landing sample is highly significant ($p<.001$) and indicates the sample is not comprised of fish from a single population. The Warren Landing fish were examined in detail in an attempt to determine whether they represent two or more subpopulations. However, results were inconclusive. Hence, fish from Warren Landing were excluded from further biochemical, morphometric and meristic comparisons with samples from other locations.

Table 4 shows a comparison of calculated G-3-PDH gene frequencies at the a and b loci. The a^2 allele predominates in all samples and no apparent differences in gene frequencies at the a locus exist between locations sampled. Consequently, no statistical analysis was carried out with respect to this locus. Frequencies calculated at the b locus were compared using a maximum likelihood ratio test (Appendix I). No significant differences in G-3-PDH b allele frequencies were found when lake whitefish from Traverse Bay, Berens River, Poplar River, Big Black River, Little Playgreen Lake

Table 3. Observed and expected (Castle-Hardy-Weinberg) BB glycerol-3-phosphate dehydrogenase phenotype distributions for lake whitefish sampled in Lake Winnipeg and some adjacent lakes. Expected numbers in brackets.

Location	No. of Fish and BB G-3-PDH Phenotype						χ^2(3 df)	p
	1,1	1,2	1,3	2,2	2,3	3,3		
Traverse Bay (L. Wpg.)	2(3)	6(7.5)	14(10.5)	4(4.7)	16(13.2)	6(9.2)	3.6	.5>p>.3
Berens River (L. Wpg.)	5(3.8)	6(6.6)	14(15.7)	4(2.8)	12(13.6)	18(16.3)	1.5	.7>p>.5
Poplar River (L. Wpg.)	2(2.6)	10(9.1)	10(9.6)	6(7.8)	19(16.3)	7(8.5)	1.3	.8>p>.7
Big Black River (L. Wpg.)	2(3.1)	7(8.4)	15(11.3)	6(5.7)	16(15.2)	8(10.2)	2.4	.5>p>.3
Warren Landing (L. Wpg.)	3(2.6)	11(3.8)	6(14)	0(1.4)	6(10.3)	25(18.9)	25.2	p<.001
Little Playgreen Lake	3(3.5)	8(7)	13(13)	3(3.5)	13(13)	12(12)	0.3	.99>p>.95
Lake St. Martin	1(0.5)	2(1.1)	6(7.9)	0(0.6)	9(8.7)	32(31.2)	2.3	.7>p>.5
Dauphin River (L. Wpg.)	0(0.9)	0(1.6)	13(9.6)	2(0.8)	8(8.9)	25(26.3)	5.7	.2>p>.1
Grand Rapids (L. Wpg.)	3(3.4)	9(6.6)	12(13.5)	3(3.2)	11(13)	15(13.3)	1.6	.7>p>.5
William Lake	4(1.5)	5(6.7)	4(7.5)	8(7.6)	18(17.2)	11(9.7)	6.8	0.1>p>.05
Clearwater Lake	15(13.8)	8(11)	17(16.5)	2(2.2)	10(6.6)	3(5)	3.8	.3>p>.2

Table 4. Calculated gene frequencies for glycerol-3-phosphate, a and b loci, in samples of lake whitefish from Lake Winnipeg and other lakes.

Location	Gene Frequencies				No. Fish
	a^2	b^1	b^2	b^3	
Traverse Bay (L. Wpg.)	0.927	0.250	0.312	0.438	48
Berens River (L. Wpg.)	0.966	0.254	0.221	0.525	59
Poplar River (L. Wpg.)	0.991	0.222	0.380	0.398	54
Big Black River (L. Wpg.)	0.954	0.241	0.324	0.435	54
Little Playgreen Lake	0.981	0.260	0.260	0.480	52
Lake St. Martin	0.940	0.100	0.110	0.790	50
Dauphin River (L. Wpg.)	0.969	0.135	0.125	0.740	48
Grand Rapids (L. Wpg.)	0.962	0.255	0.245	0.500	53
William Lake	0.990	0.170	0.390	0.440	50
Clearwater Lake	1.000	0.500	0.200	0.300	55

and Grand Rapids were compared. These samples were then pooled and considered homogeneous with respect to G-3-PDH b allele frequencies (Table 5).

No significant difference in G-3-PDH b allele frequencies was found when the Lake St. Martin and Dauphin River samples were compared (Table 5) and these two samples were pooled for subsequent analyses.

Comparison between the pooled Traverse Bay-Berens River-Poplar River-Big Black River-Grand Rapids-Little Plyagreen Lake group and the pooled Lake St. Martin-Dauphin River group revealed a significant difference ($p<.001$) in G-3-PDH b allele frequencies (Table 5).

G-3-PDH b allele frequencies for the sample from Clearwater Lake differ significantly ($p<.001$) from those for the pooled Lake St. Martin-Dauphin River sample, and from the Grand Rapids sample ($p<.005$). William Lake b allele frequencies differ marginally ($p\simeq.05$) from the Grand Rapids sample (Table 5). Figure 6 shows results of pooling locations homogeneous with respect to G-3-PDH b allele frequencies.

Lactate Dehydrogenase (LDH)

Observed LDH phenotype distributions and calculated allele frequencies are shown in Table 6. The William Lake sample was compared only with Grand Rapids since fry raised from William Lake spawn are introduced into Lake Winnipeg at Grand Rapids. Similarly the Clearwater sample was

Table 5. Results of comparisons of G-3-PDH b allele frequencies between samples using a maximum likelihood ratio test. The solid line indicates no significant difference at the .05 level of probability between samples compared. Dashed line indicates a significant difference.

Location	Comparisons Within Lake Winnipeg									Hatchery Lakes		
	1	2	3	4	5	6	7	8	9	1	2	3
Traverse Bay												
Berens River												
Poplar River												
Big Black River												
Little Playgreen L.												
L. St. Martin												
Dauphin River												
Grand Rapids												
William Lake												
Clearwater Lake												
χ^2	2.56	4.70	0.22	0.92	39.70	27.44	1.16	0.78	57.80	5.57	70.00	14.71
Degrees of Freedom	2	2	2	2	2	2	2	2	2	2	2	2
Probability	0.3-0.2	0.1-0.09	.95-.90	0.7-0.5	<.001	<.001	0.7-0.5	0.5-0.3	<.001	≈.05	<.001	<.001
Significance[a]	N.S.	N.S.	N.S.	N.S.	S.D.	S.D.	N.S.	N.S.	S.D.	Mar.	S.D.	S.D.

[a] N.S. = not significant at the .05 level of probability. S.D. = significant difference. Mar. = marginally different (χ^2=5.57;χ^2.05;2 d.f. = 5.99)

Figure 6. Calculated G-3-PDH b allele frequencies of lake whitefish and their division into apparent homogeneous groups. No significant differences ($p<.05$) in gene frequencies exist between locations within the dashed line while significant differences appear between groups outlined by the dashed line.

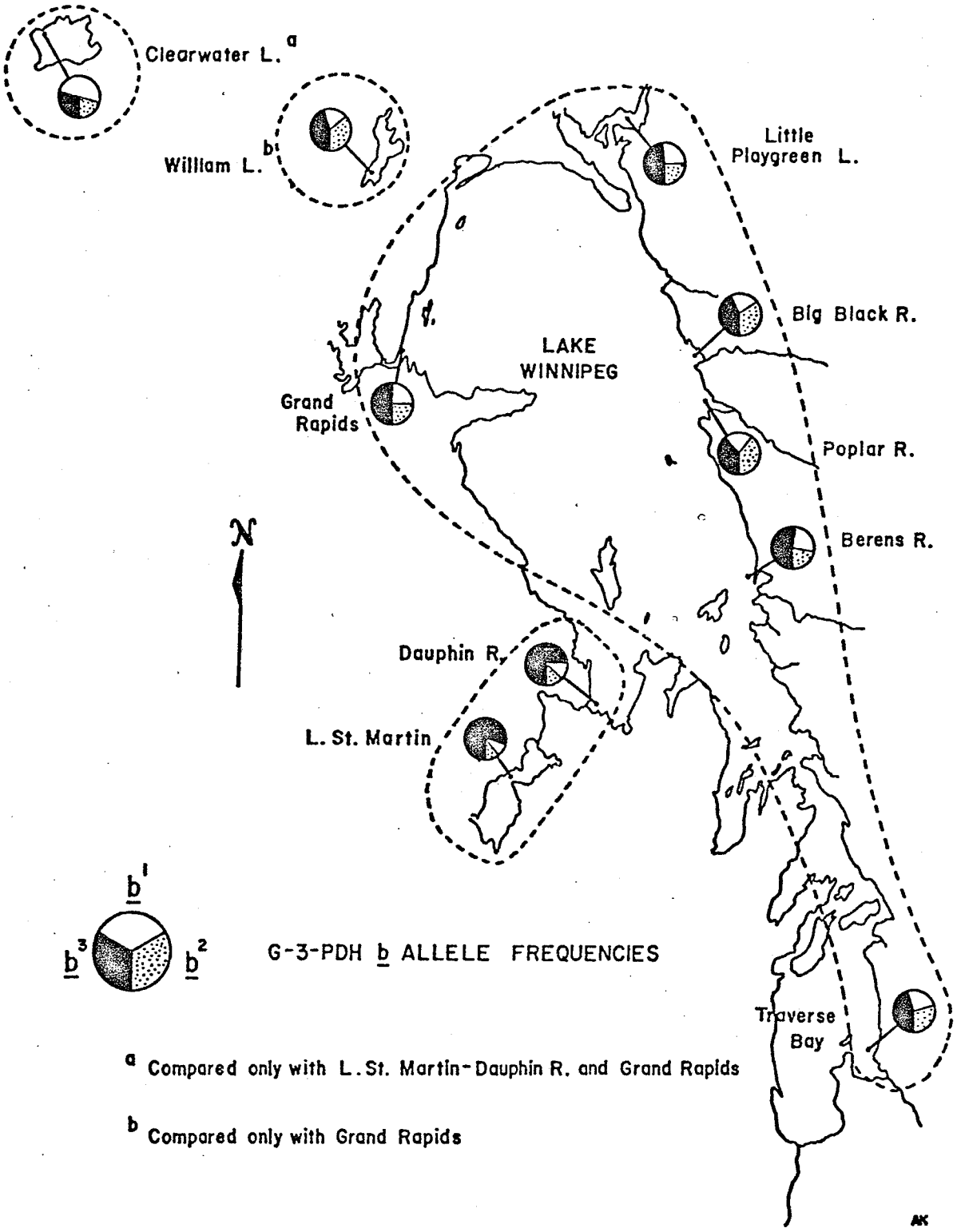

Clearwater L. a
William L. b
Little Playgreen L.
Big Black R.
LAKE WINNIPEG
Grand Rapids
Poplar R.
Berens R.
N
Dauphin R.
L. St. Martin
Traverse Bay
b1
b3
b2
G-3-PDH b ALLELE FREQUENCIES
a Compared only with L. St. Martin-Dauphin R. and Grand Rapids
b Compared only with Grand Rapids
AK

Table 6. Observed LDH Hα phenotype distribution and calculated $\underline{ldhH\alpha^{A}}$ allele frequencies for lake whitefish sampled in Lake Winnipeg and some adjacent lakes.

Location	LDH Hα Phenotype			Gene Frequency
	AA[a]	AB[b]	BB[c]	(α^{A})
Traverse Bay (L. Wpg.)	23	1	0	0.979
Berens River (L. Wpg.)	19	5	0	0.896
Poplar River (L. Wpg.)	20	4	0	0.917
Big Black River (L. Wpg.)	22	2	0	0.958
Little Playgreen Lake	21	3	0	0.938
Lake St. Martin	18	6	0	0.875
Dauphin River (L. Wpg.)	21	3	0	0.938
Grand Rapids (L. Wpg.)	35	1	0	0.986
William Lake	24	0	0	1.000
Clearwater Lake	23	1	0	0.979

[a] AA, genotype is $\underline{ldh\ H\alpha^{AA}}$
[b] AB, genotype is $\underline{ldh\ H\alpha^{AB}}$
[c] BB, genotype is $\underline{ldh\ H\alpha^{BB}}$

compared only with Grand Rapids and Dauphin River, since fry raised from Clearwater spawn are introduced into Lake Winnipeg at the latter two locations. The gene frequencies do not appear to differ significantly between locations; thus no further work was done with this enzyme.

Genetic Distance

Nei (1971,1972) formulated a method of measuring the genetic distance (D) between populations. For electrophoretic comparisons D is the average number of electrophoretically detectable substitutions per locus between the two populations being compared (Sarich 1977). The genetic distance is defined as $D = -\log_e I$ where I is the normalized identity of genes between populations. This identity measures the proportion of genes that are common in the populations under investigation (Nei 1972). If x and y are the frequencies of the i^{th} allele in populations x and y, then the measure of genetic identity (I) is given by the equation

$$I_{xy} = \frac{\Sigma\, x_i y_i}{(\Sigma x_i^2\ \Sigma y_i^2)^{1/2}}$$

where the summation is over all alleles and all loci compared (Sarich 1977).

Using Nei's method, I and D were calculated using 9 alleles at 5 loci for all pairwise comparisons of lake whitefish samples examined in this study. The results (Appendix III) are in agreement with observations made

during this study based on comparisons of G-3-PDH b allele frequencies using the maximum likelihood ratio test.

Nei (1972), using data calculated for two subspecies of house mouse, Mus musculus musculus and Mus musculus domesticus, found that the average genetic distance between the two subspecies was 0.1714, while D between local populations within subspecies was 0.0137. The average genetic distance calculated for the two subpopulations in Lake Winnipeg shown in Figure 6 was 0.0178, a value which compares favourably with Nei's (1972) observations. The average genetic distance between samples within the Lake St. Martin-Dauphin River subpopulation is 0.0015, while the average D between samples from the remainder of Lake Winnipeg is 0.0022. That is, genetic distance between the lake whitefish subpopulations is ten times as great as D between samples within subpopulations.

Genetic distance between Clearwater Lake fish and the Lake St. Martin-Dauphin River sample is 0.0456. The calculated D between the Clearwater Lake and Grand Rapids samples is 0.0122, and between William Lake and Grand Rapids is 0.0039.

Morphological Analyses

Discriminant-Function Analysis

Results of initial discriminant-function analysis are shown in Table 7. Each sample was considered as a separate group. Overall discrimination between the initial 8 groups resulted in 45.6 percent correct classification of individuals according to their location of capture. Values outlined diagonally in Table 7 represent the number of individuals from each sample size of 48 that were correctly identified as having been captured at that location. Further examination of this table reveals misclassified lake whitefish specimens were, in some cases, most frequently assigned to the location immediately adjacent to where they were actually captured (directly above or below the diagonal line in Table 7). Significant misclassification of individuals between Berens River, Poplar River and Big Black River is evident. A separate discriminant-function analysis involving only these three groups resulted in 54.9 percent correct classification. The samples from these three locations were then pooled. Sixty-four percent of specimens were correctly classified when the sample from Lake St. Martin was compared with Dauphin River, and these two samples were pooled as a result. Samples from Traverse Bay, Grand Rapids and Little Playgreen Lake were left as separate groups, since no tendency for misclassification to adjacent locations was evident.

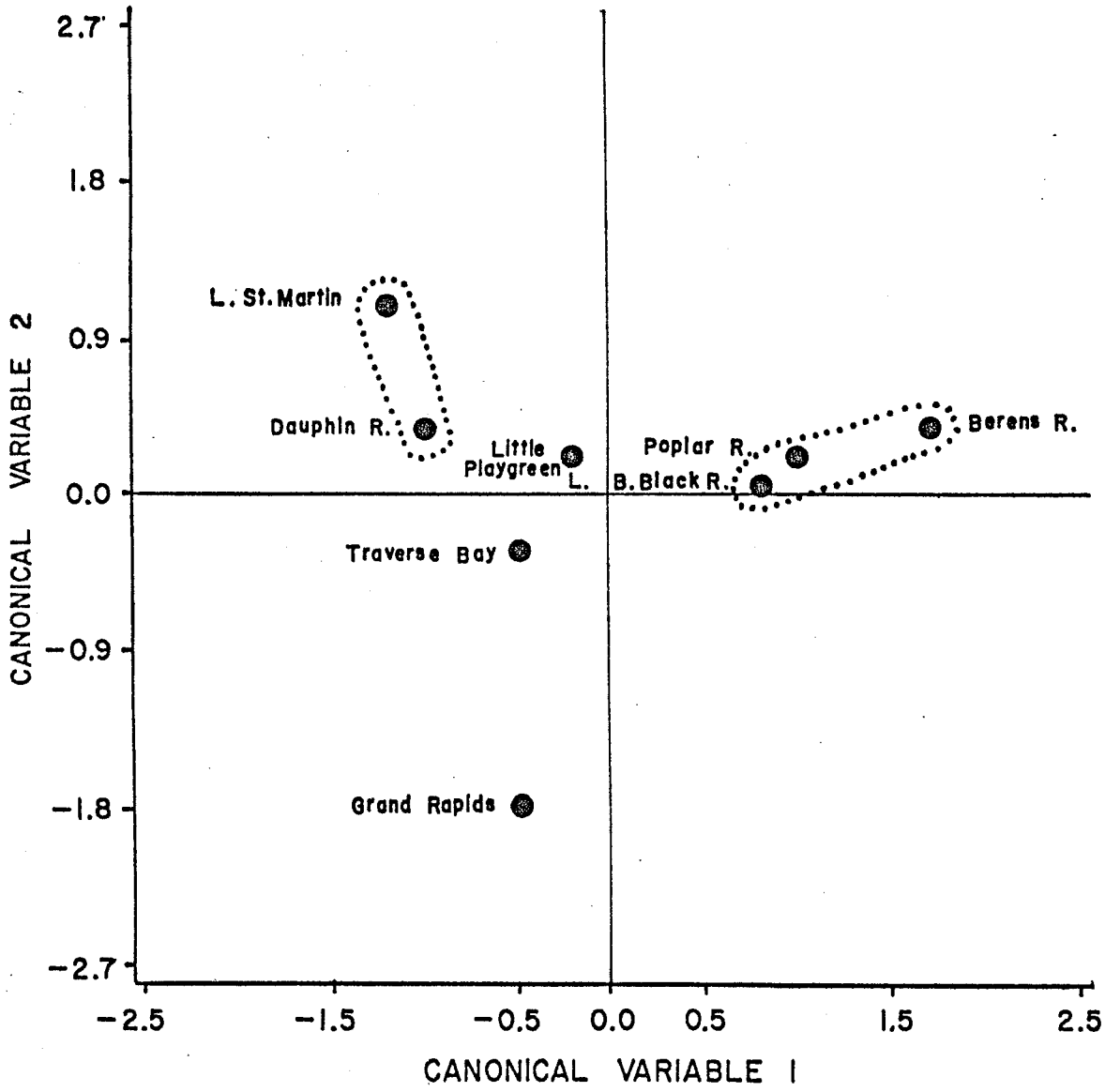
Table 7. Assignment of lake whitefish into groups using discriminant-function analysis of morphological characters. Numbers in the diagonal band represent the number of specimens correctly identified according to location of capture. Dotted line encompasses specimens frequently misclassified as belonging to adjacent locations.

	Group Assigned to Using Discriminant Analysis	Location of Capture								
		Traverse Bay	Berens River	Poplar River	Big Black River	Little Playgreen Lake	Lake St. Martin	Dauphin River	Grand Rapids	
East Side	Traverse Bay	22	1	1	9	4	5	5	5	South→North
	Berens River	2	27	9	9	5	0	1	2	
	Poplar River	2	8	20	10	2	1	1	1	
	Big Black River	6	6	11	13	2	0	2	2	
	Little Playgreen L.	5	5	2	3	20	5	7	6	
West Side	L. St. Martin	3	0	3	1	3	29	12	4	South→North
	Dauphin River	3	0	1	1	9	7	17	1	
	Grand Rapids	5	1	1	2	3	1	3	27	
	Total No. Fish	48	48	48	48	48	48	48	48	

Canonical variables evaluated at group means for each of the initial 8 groups compared are shown in Figure 7. Canonical variable 1 represents the discriminant score of each group, evaluated at the group mean and canonical variable 2, a subset of variables orthogonal to canonical variable 1, contributes to group separation. The similarity of co-ordinates supports pooling the samples from Berens, Poplar and Big Black Rivers; group means all have positive-positive co-ordinates. Group means of Lake St. Martin and Dauphin River samples both have negative-positive co-ordinates. The Little Playgreen Lake sample group mean has negative-positive co-ordinates also, but is geographically well separated from the former locations (Figure 1). Sample group means from Traverse Bay and Grand Rapids both have negative-negative co-ordinates but are geographically well separated (Figure 1).

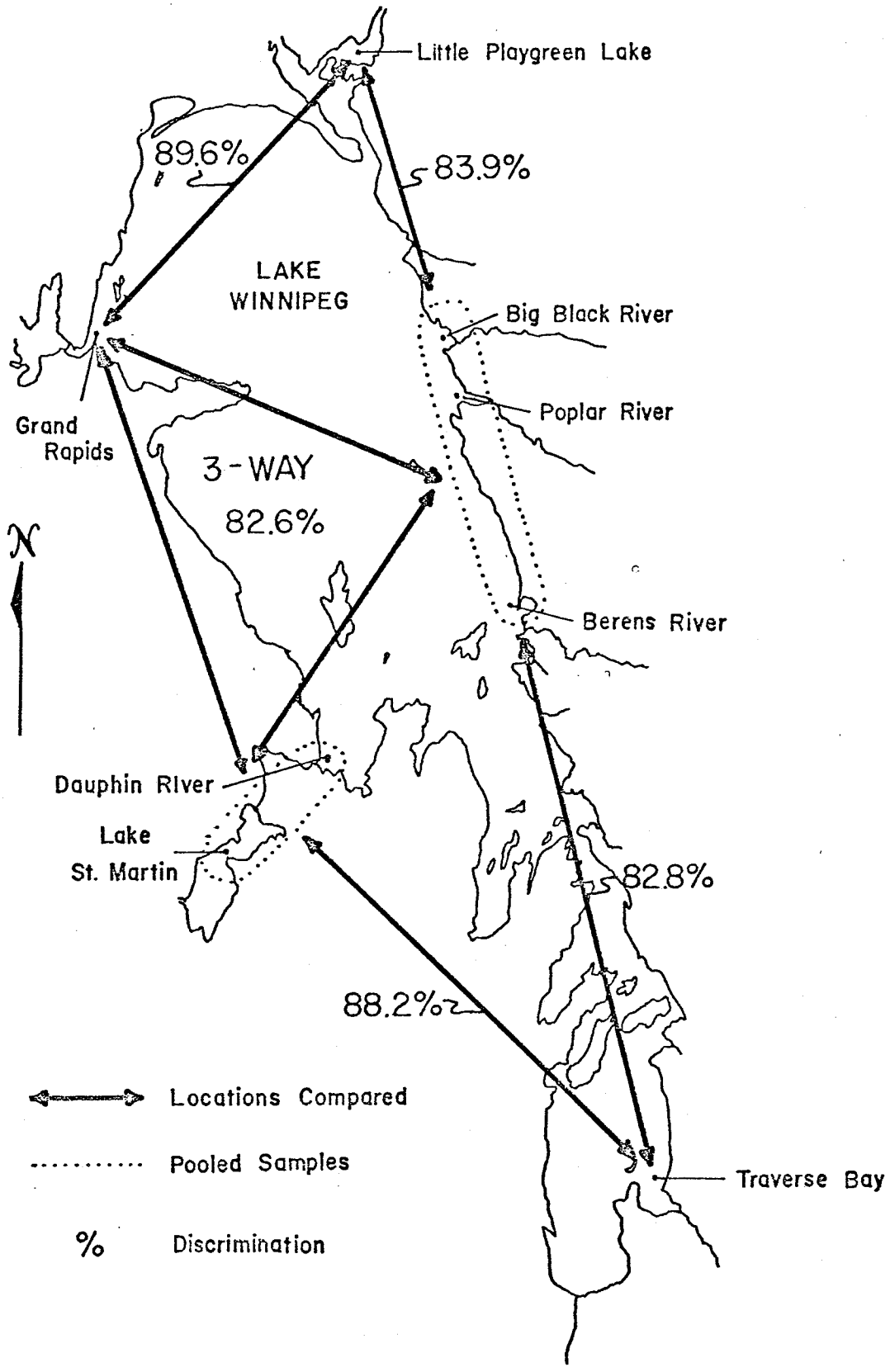
Discriminant-function analysis was repeated using the pooled samples. Eighty-three percent of individuals were correctly classified according to location of capture when the sample from Traverse Bay was compared with the pooled Berens-Poplar-Big Black Rivers sample (Figure 8). Pectoral fin origin, lateral line scales to standard length, pectoral fin length and interorbital width used in combination allowed 81.8 percent correct classification. Addition of adipose fin length and head depth improved discrimination by 1 percent.

Figure 7. Plot of canonical variables evaluated at group means for the 8 locations initially compared for morphological differences using stepwise discriminant-function analysis.


2.7
1.8
0.9
0.0
−0.9
−1.8
−2.7
CANONICAL VARIABLE 2
L. St. Martin
Dauphin R.
Little Playgreen L.
Traverse Bay
Grand Rapids
Poplar R.
B. Black R.
Berens R.
−2.5
−1.5
−0.5
0.0
0.5
1.5
2.5
CANONICAL VARIABLE I

AK

Figure 8. Locations tested for morphological differences using stepwise discriminant-function analysis. Arrows indicate locations compared. Percent correct classification of individuals according to location of capture is included. For characters used see text.


Little Playgreen Lake
89.6%
83.9%
LAKE
WINNIPEG
Big Black River
Poplar River
Grand
Rapids
3-WAY
82.6%
N
Berens River
Dauphin River
Lake
St. Martin
82.8%
88.2%
Locations Compared
Pooled Samples
% Discrimination
Traverse Bay
AK

Discrimination between Traverse Bay fish and the pooled Lake St. Martin-Dauphin River sample allowed 88.2 percent correct classification of individuals as shown in Figure 8. Lateral line scales to standard length, pectoral fin origin, head length and prepostorbital distance in combination allowed 81.3 percent correct classification of individuals. Addition of body width and gill raker number on the lower limb increased discrimination 6.9 percent. Since measurable morphological differences were detected between Traverse Bay fish and those from immediately adjacent locations, no comparisons were made between this and other non-adjacent locations.

Discrimination between fish from Little Playgreen Lake and the pooled Berens-Poplar-Big Black Rivers samples showed 83.9 percent of individuals could be correctly classified according to location of capture (Figure 8). Pectoral fin length, head depth, lateral line scales to standard length and prepostorbital distance in combination provided the discrimination.

Comparison of the Little Playgreen Lake sample and the Grand Rapids sample allowed 85.4 percent discrimination of individuals using lateral line scales in total, caudal peduncle scales, anal fin rays and head depth. Inclusion of gill raker number on the lower limb increased correct classification to 89.6 percent (Figure 8). Since the last two comparisons were with the locations immediately adjacent to Little Playgreen Lake and differences were detected,

further comparisons of this nature were carried out using the Little Playgreen Lake sample.

A three way discrimination between Grand Rapids, the pooled Lake St. Martin-Dauphin River sample, and the pooled Berens-Poplar-Big Black Rivers sampled allowed 82.6 percent correct classification of individuals (Figure 8). Discriminating characters were pectoral fin length, head depth, body width, maxillary length, lateral line scales in total and caudal peduncle scales.

Mean values of the most useful discriminators are shown in Table 8.

Sexual Dimorphism

Using stepwise discriminant-function analysis with males and females as grouping variables, less than 80 percent correct classification into males and females was achieved at all locations except Little Playgreen Lake. Sexual dimorphism was not considered significant at these locations.

A total of 87.5 percent of individuals were correctly classified as males or females when lake whitefish from Little Playgreen Lake were tested for sexual dimorphism. Discriminators were pectoral fin origin, anal fin origin, pectoral fin length and pelvic fin origin. Males had longer mean pectoral fin origin and pectoral fin length and females had longer mean pelvic fin origin and anal fin origin distance.

Table 8. Mean values of most useful discriminators used in discriminant-function analysis, based on measurements adjusted to a fish of 345 mm in body length. Morphometric measurements are expressed in millimeters and transformed to $\log_{10}$. Standard error in brackets.

Location	Discriminator								
	Pectoral Fin Length	Lateral Line Scales	Pectoral Fin Origin	Prepostorbital Length	Body Width	Head Depth	Caudal Peduncle Scales	Head Length	Interorbital Width
North ↑									
Little Playgreen Lake (N=48)	1.8600 (.0038)	73.6 (.42)	1.8415 (.0044)	1.5410 (.0042)	1.7381 (.0030)	1.7708 (.0032)	23.9 (.12)	1.8654 (.0031)	1.3808 (.0029)
Grand Rapids (N=48)	1.8545 (.0040)	79.4 (.49)	1.8264 (.0053)	1.5327 (.0055)	1.7393 (.0038)	1.7870 (.0041)	25.0 (.12)	1.8639 (.0041)	1.3836 (.0030)
Berens-Poplar-Big Black R. (N=144)	1.8266 (.0023)	76.1 (.23)	1.8169 (.0026)	1.5057 (.0029)	1.7318 (.0020)	1.7462 (.0019)	23.8 (.08)	1.8401 (.0020)	1.3666 (.0017)
L. St. Martin-Dauphin (N=96)	1.8712 (.0023)	73.9 (.34)	1.8369 (.0025)	1.5236 (.0030)	1.7119 (.0025)	1.7651 (.0023)	24.1 (.12)	1.8644 (.0021)	1.3775 (.0020)
Traverse Bay (N=48)	1.8616 (.0053)	78.1 (.61)	1.8610 (.0038)	1.5459 (.0052)	1.7224 (.0030)	1.7570 (.0035)	24.0 (.14)	1.8690 (.0038)	1.3736 (.0037)
↓ South									

West ← → East

Most subsamples were comprised equally of males and females (Table 2). However, the sample from Little Playgreen Lake, which showed strong evidence of sexual dimorphism, also had the most unequal sex ratio. In order to test whether results of discrimination between Little Playgreen Lake fish and other locations were based on sex differences the comparisons were repeated, using males compared with males and females compared with females. Males from Little Playgreen Lake compared with males from Grand Rapids showed 93.1 percent correct classification and when compared with males from the pooled Berens-Poplar-Big Black Rivers sample showed 85.5 percent correct classification Little Playgreen Lake females compared with females from the pooled Berens-Poplar-Big Black Rivers sample showed 76.8 percent correct classification and compared with Grand Rapids females showed 81.6 percent correct classification.

Age and Growth

In order to determine the existence of subpopulations of fish within a given water body using morphological comparisons, it is desirable that each sample of fish be composed of a number of age groups. Results of comparisons using single age groups could just reflect (a)annual fluctuations in environmental factors which modify the phenotype, (b)random selection of spawning sites by members of a panmictic population or (c) a combination of the two. Hence, the age structure of samples compared in this study was examined.

Since differences in morphology of whitefish can be a result of differential growth rates, growth rates were compared between samples.

Age distributions within lake whitefish samples examined using discriminant-function analysis are shown in Figure 9. At least 5 age groups are represented in each sample, and 8 age groups in one.

Growth rates calculated for each sample using mean fork length at age are shown in Figure 10. Growth rate of fish 3+ years and older appears to differ little between locations compared.

A comparison of the number of lateral line scales to standard length within each age group was made between the samples from Lake St. Martin-Dauphin River, Grand Rapids and Little Playgreen Lake (Figure 11). The number of lateral line scales appears to be stable between age groups within each sample while the mean number of lateral line scales differs between samples.

Combined Results of Biochemical and Morphological Analyses

Results of the G-3-PDH isozyme analysis are shown superimposed upon results of the morphological analysis (Figure 12). The Lake St. Martin-Dauphin River sample is distinct from other locations on the basis of G-3-PDH *b* allele frequencies and apparently separable from other locations by morphological differences. Other locations compared are homogeneous with respect to G-3-PDH *b* allele frequencies but apparently separable into four groups on the basis of morphological differences.

Figure 9. Age distribution of lake whitefish samples from five locations in or near Lake Winnipeg.

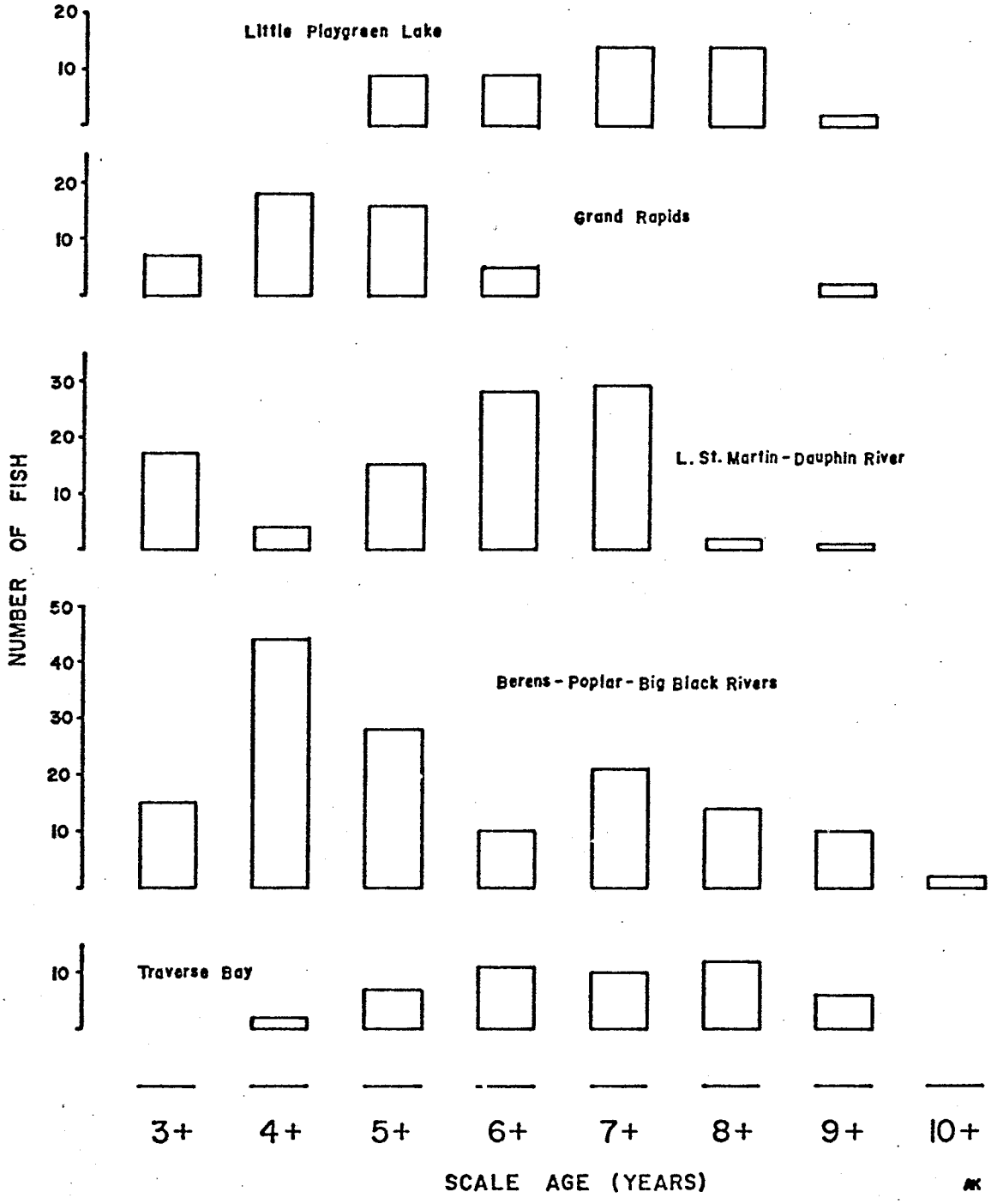

Little Playgreen Lake
Grand Rapids
L. St. Martin - Dauphin River
Berens - Poplar - Big Black Rivers
Traverse Bay
NUMBER OF FISH
SCALE AGE (YEARS)
3+
4+
5+
6+
7+
8+
9+
10+

Figure 10. Growth rates of lake whitefish sampled from five locations in or near Lake Winnipeg.

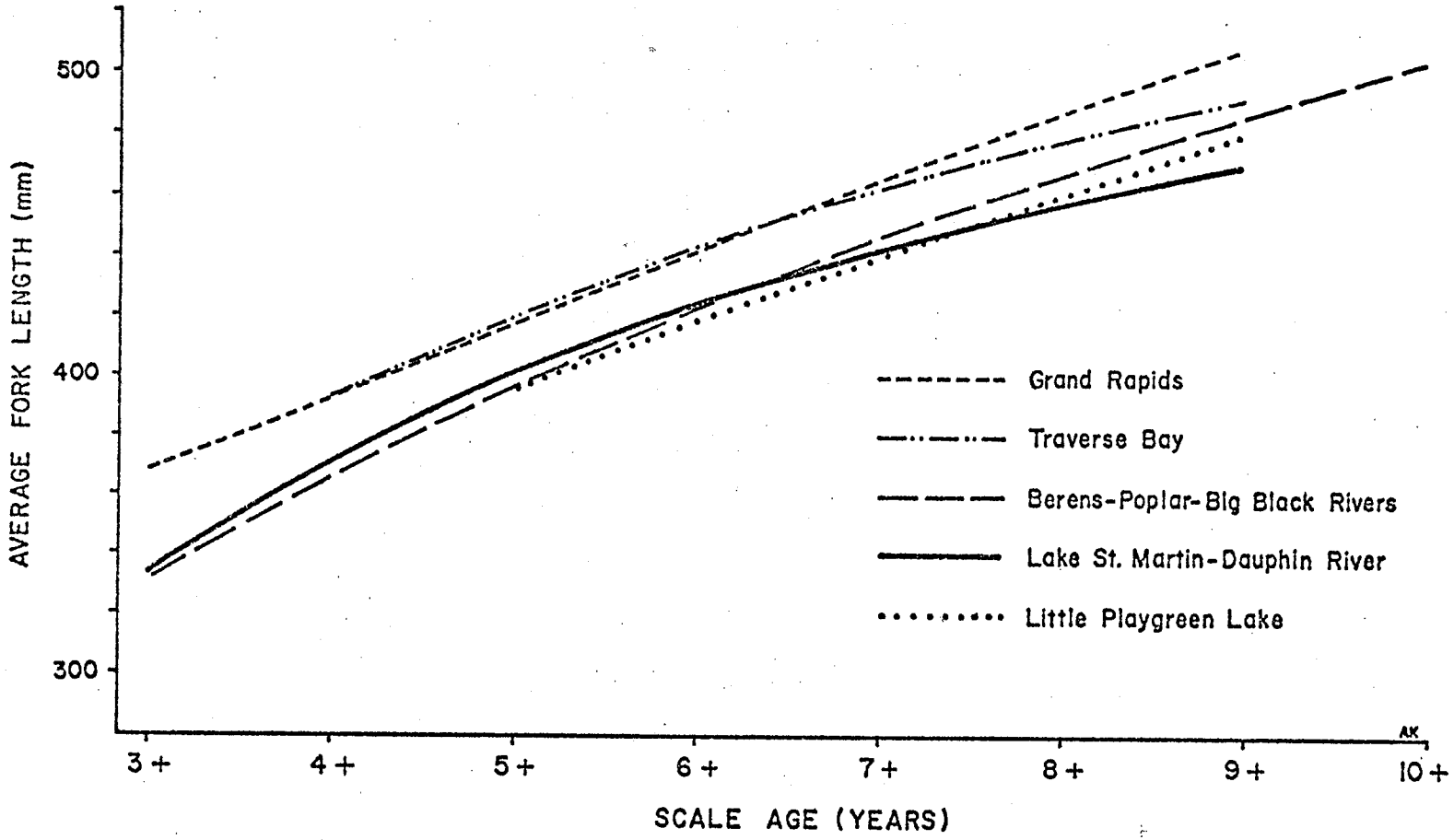

AVERAGE FORK LENGTH (mm)
500
400
300
3+
4+
5+
6+
7+
8+
9+
10+
AK
SCALE AGE (YEARS)
Grand Rapids
Traverse Bay
Berens-Poplar-Big Black Rivers
Lake St. Martin-Dauphin River
Little Playgreen Lake

Figure 11. Lateral line scale counts to standard length by age group for samples of lake whitefish from Lake St. Martin-Dauphin River, Grand Rapids and Little Playgreen Lake.

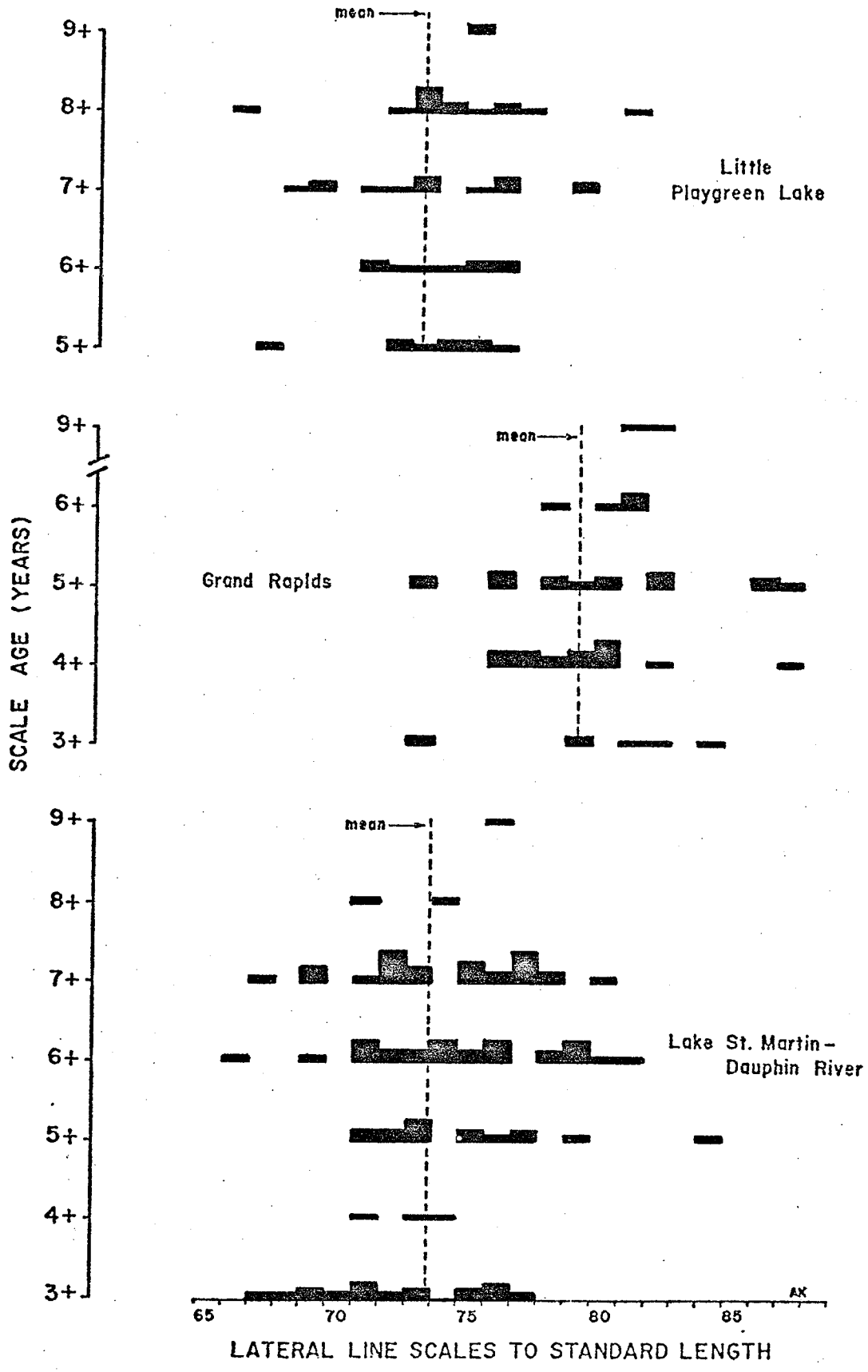
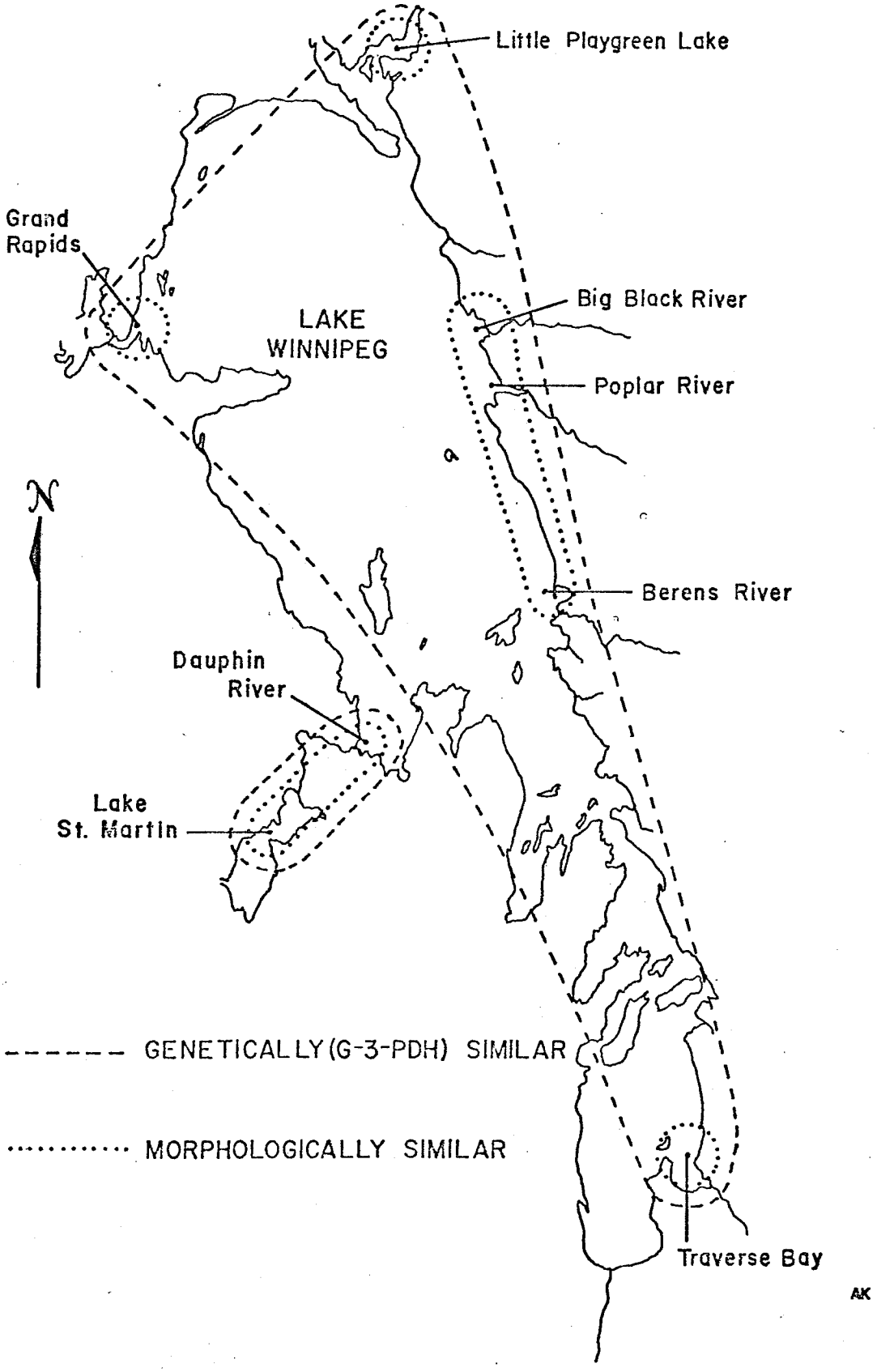

mean
9+
8+
7+
6+
5+
Little
Playgreen Lake
mean
9+
6+
5+
4+
3+
Grand Rapids
SCALE AGE (YEARS)
mean
9+
8+
7+
6+
5+
4+
3+
Lake St. Martin –
Dauphin River
AK
65
70
75
80
85
LATERAL LINE SCALES TO STANDARD LENGTH

Figure 12. Results of lake whitefish BB G-3-PDH isozyme analysis and morphological analysis superimposed on a map of Lake Winnipeg. Differences in G-3-PDH b allele frequencies exist between locations outlined by dashed line but not within dashed line. Morphological differences exist between locations outlined by dotted line but not within dotted line.


Little Playgreen Lake
Grand Rapids
LAKE WINNIPEG
Big Black River
Poplar River
N
Berens River
Dauphin River
Lake St. Martin
GENETICALLY (G-3-PDH) SIMILAR
MORPHOLOGICALLY SIMILAR
Traverse Bay
AK

DISCUSSION

Biochemical Differences Between Subpopulations

Heterogeneity Within Samples

Prior to comparing gene frequencies between locations it is necessary to determine whether each sample is from a homogeneous stock or from a mixture of stocks. As stated in Methods this was accomplished by comparing observed BB G-3-PDH phenotypes with Castle-Hardy-Weinberg expectations.

Results of χ^2 tests involving classes with expected numbers less than 5 should be viewed with caution (Snedecor and Cochran 1967, p. 235). Use of small classes is likely to spuriously inflate χ^2 values. Table 3 shows that even though classes of less than 5 expected individuals did occur all samples produced nonsignificant χ^2 values except the one from Warren Landing. Application of the Yates correction for continuity (Stansfield 1969, p. 142; Snedecor and Cochran 1967, p.212) made no change in the results. Such nonsignificance is interpreted as evidence of homogeneity within all samples with the exception of Warren Landing.

The significant χ^2 value ($p<.001$) of the Warren Landing sample is probably not the result of χ^2 inflation due to small expected numbers in some classes. The expected number of 1, 2 phenotypes is small (3.8) and by itself this class contributes considerably (13.6) to the χ^2 value. If this contribution is subtracted from the

value (25.2) of the sample, the remainder (11.6) is still highly significant ($p<.01$). Hence, the significant deviation of observed from expected BB G-3-PDH phenotypes of the Warren Landing sample probably has biological meaning.

Franzin (1974) found significant deviation of BB G-3-PDH phenotypes from Castle-Hardy-Weinberg expectations when examining lake whitefish from Fox Lake, Yukon Territory. He attributed the heterozygote deficiency to heterogeneity in the sample. The sample was taken by gill net over a twelve hour period and he presumed schools of slightly different genetic composition moving through the area may have contributed to the catch. The deviation of the Warren Landing sample appears to be a result of heterozygote deficiency. Koehn et al. (1971) include possible explanations for deficit of heterozygotes. The so-called "Wahlund Effect", a heterozygote deficiency of the pooled distribution when samples of differing allelic frequencies are mixed, is probably the best explanation for the deviation observed in the Warren Landing sample. This sample was secured over a period of 4 days (Table 1) from a single gill net set off Mossy Point near the outflow to the Nelson River. It was the earliest sample collected from Lake Winnipeg (Table 1) and although attempts were made to capture fish in spawning condition, some in this sample were in prespawning condition when taken. Apparently at least two groups of lake whitefish with different allelic frequencies passed by

the same capture location within 4 days of one another and contributed some of their members to the catch.

These results are interpreted as evidence that different subpopulations of lake whitefish moved through the Warren Landing area during the four day collection period. Hence, the sample was excluded from further analysis.

Heterogeneity Between Samples

Differences in G-3-PDH b allele frequencies indicate at least two subpopulations of lake whitefish reside in Lake Winnipeg. One group spawns in the Lake St. Martin-Dauphin River area while the other group spawns throughout the remainder of Lake Winnipeg (Figure 5).

Postglacial admixture. Franzin (1974) noted differences in lake whitefish G-3-PDH b allele frequencies between lakes sampled across Western Canada and the Yukon Territory. After sampling eight lakes on the North Saskatchewan River system, including north lake Winnipeg he reported differences in b allele frequencies but found no G-3-PDH b^3 allele frequencies in excess of 0.49 in any of these lakes. The subpopulation spawning in Lake St. Martin-Dauphin River is characterized by an average G-3-PDH b^3 allele frequency of 0.77 compared with 0.46 for whitefish in the remainder of Lake Winnipeg. Franzin attributed the observed distributions of gene frequencies to a postglacial admixture of two or more genetically different stocks of

lake whitefish, subjected to mutation and differential selection in widely separated glacial refugia prior to mixing. It is unlikely the Lake St. Martin-Dauphin River subpopulation is a relict population which has retained its distinct G-3-PDH b allele frequencies since deglaciation. Preservation of such a population would seem more likely to have taken place in small isolated headwater lakes where postglacial admixture of stocks would be limited. The relatively high frequency of the G-3-PDH b^3 allele in the Lake St. Martin-Dauphin River subpopulation may be a result of more recent events.

Founder effect and random drift. There is insufficient information regarding the origin of the Lake St. Martin-Dauphin River subpopulation to conclude whether or not Founder effect may have caused the differences noted in G-3-PDH b allele frequencies. According to Lewontin (1974) even very small amounts of gene flow will prevent differentiation between groups as a result of random drift. Other factors may be responsible for the differences in G-3-PDH b alleles observed here.

Selection. Discontinuous variation or polymorphism is often controlled by a few genes or by alleles at a single locus (Mayr 1963). The polymorphism examined during this study occurs as three nondominant alleles designated b^1 b^2 and b^3 at the G-3-PDH b locus. Clarke (1975) states

recent evidence shows natural populations of a wide range of organisms including plants and animals show extreme diversity and widespread protein polymorphism. The "neutralist" view claims polymorphisms are tolerated as they confer neither selective advantage nor disadvantage on the carrier organisms (Clarke 1975). The "selectionist" view states polymorphic genes do affect survival and natural selection acts to maintain polymorphisms. Possibly pleiotropic effects exist where selection acts not on particular isozymes but on some other linked character. Recent evidence suggests selection may in fact act directly on gene products of polymorphic loci (Powell 1971; Clarke 1975).

Although it is beyond the scope of this study to determine how and at what rate selection acts upon lake whitefish G-3-PDH isozymes, certain events in the history of the Lake St. Martin fishery are worth considering in this regard. Poor lake whitefish production appears to be correlated with low water levels in Lake St. Martin four years previous to the catch. Lake whitefish first contribute significantly to the commercial catch in Lake Winnipeg at age 4 (Davidoff et al. 1973). Water levels on Lake St. Martin dropped significantly in 1960 (F. Eggers, pers. comm.) followed by a drastic drop in the 1964 commercial whitefish catch (D.W. Cook, pers. comm.). Water levels rose again in 1966 (F. Eggers, pers. comm.) as did whitefish production in 1970 (D.W. Cook, pers. comm.). During winter 1963, soundings and

water samples on Lake St. Martin revealed little water beneath the ice and zero dissolved oxygen in the shallow northeast end, believed to be a whitefish spawning ground (Anon. (1963).

Low water levels in Lake St. Martin probably kill some postspawners and large numbers of incubating eggs, thus drastically reducing the size of the Lake St. Martin-Dauphin River subpopulation. According to Mayr (1963) rapid changes in population size result in changes in selection pressure. Such changes in selection pressure may be reflected by the G-3-PDH b allele frequencies characterizing the Lake St. Martin-Dauphin River subpopulation.

Geographic separation and homing. Weinstein and Yerger (1976) consider geographic separation and environmental differences as possible factors contributing to genetic distinction of subpopulations of the spotted seatrout Cynoscion nebulosus (Cuvier), in the Gulf of Mexico and Atlantic coast of Florida. According to Morgan et al. (1973) geographic isolation coupled with a natal homing mechanism may have contributed to detectable differences in gene frequencies of a variety of proteins found in striped bass, Morone saxatilis, in the Upper Chesapeake Bay. Environmental differences may exist between spawning areas utilized by subpopulations of lake whitefish in Lake Winnipeg. The high degree of homing displayed by some salmonids is well documented. Coregonus clupeaformis must home in Lake Winnipeg

since at least two groups of fish with distinct G-3-PDH gene frequencies exist. Homing of lake whitefish would restrict gene flow between subpopulations and tend to perpetuate the discrete characteristics of each group.

Whatever the causative agents, the significant differences in G-3-PDH b allele frequencies detected between subpopulations of lake whitefish in Lake Winnipeg reflect a measure of reproductive isolation and confer biochemical individuality upon each subpopulation.

Morphological Differences Between Subpopulations

Lake whitefish in Lake Winnipeg have been tentatively divided into five subpopulations using stepwise discriminant-function analyses of morphological differences (Appendix II). Differences in seven morphometric measurements and three meristic counts contributed most to discrimination of subpopulations (Table 8).

Morphometric Characteristics of Subpopulations

The Traverse Bay subpopulation is characterized by large body proportions such as pectoral fin origin, pectoral fin length, head depth, interorbital width and head length as well as by a high mean number of lateral line scales. The most distinguishing features of the Berens-Poplar-Big Black Rivers subpopulation are the relatively small size of body measurements such as pectoral fin length, head depth and maxillary length. Lake St. Martin-Dauphin River fish

are characterized by long pectoral fin length and narrow body width. The Grand Rapids subpopulation has the highest mean number of lateral line scales and caudal peduncle scales. Little Playgreen Lake fish are distinguished by large body proportions such as pectoral fin length, prepostorbital distance and head depth. Clinal variation was not evident. No apparent correlation exists between morphological characteristics and latitude (Table 8). In fact, subpopulations most geographically separated had many similar measurements but could be separated from adjacent subpopulations.

Comparison of various morphometric measurements between groups can be complicated by allometric growth (Lindsey 1962). Major growth inflections occur in early development (Martin 1949). Since mature lake whitefish involved in spawning assemblages comprised the samples studied here, growth inflections would already have taken place and in effect all specimens would have been in the same "growth stanza". A growth inflection at sexual maturity known to occur in some fish species (Martin 1949) does not appear to manifest itself in Coregonus clupeaformis (Lindsey 1962).

Bodaly (1977) found many measurements similar to those used in the present study differed significantly between two sympatric lake whitefish populations in various Yukon lakes. He discussed to some extent the significance of environmental modification, particularly growth rate, upon body morphology. He found in some lakes faster growth rate was

associated with smaller body parts while in other lakes the reverse was observed. Various investigators (Martin 1949; Kliewer 1970) found faster growing lake whitefish have smaller heads and fins, shorter snout length and smaller eyes than their slower growing counterparts. Svärdson (1950) made similar observations on European coregonids. Fenderson (1964) studying "dwarf" and "normal" lake whitefish in some Maine lakes found the caudal peduncle depth-to-length ratio differed significantly between two forms of whitefish. He observed that this ratio was more dependent on rate of growth than on absolute size. Martin (1949) explained observations such as these using relative growth analysis. He concluded growth rate affects the body size at which inflections take place in the rate of growth of a body part relative to the body as a whole. Many factors are known to affect growth rates.

Growth rates of lake whitefish vary with temperature (Atton, cited from Qadri 1968), diet (Svärdson 1949, 1950; Qadri 1968), intraspecific competition (Bidgood 1973), interspecific competition (Bidgood 1973, Larkin 1956; Lindström and Nilsson 1962) and rate of exploitation (Miller 1947; Healey 1975). All of these factors could affect whitefish in Lake Winnipeg.

Temperature in Lake Winnipeg varies between different parts of the lake (Brunskill, pers. comm.). Food availability and water chemistry also vary (Rybicki 1966; Pollard 1973b; Kristofferson et al. 1975), as does rate of exploit-

ation of lake whitefish. The Traverse Bay lake whitefish subpopulation has not been commercially exploited since commercial lake whitefish operations were restricted by statute to the northern part of the lake in 1929 (Hewson 1960). Lake whitefish occur as an incidental catch in fisheries for other species carried out in the south basin of the lake (Hewson 1959a, 1959b). Rates of exploitation may vary in the north basin as well. Hewson (1960) points out that after 1954 fishing effort per square mile in a strip of water 12 miles from shore just south of Poplar Point to just northwest of Warren Landing was roughly twelve times as great as on the remainder of the whitefish grounds within 12 miles of shore.

When compared in Figure 10, growth rates of whitefish examined in this study do not appear to differ significantly. However, comparison of growth rates using mean fork length at age is not precise. Calculation of growth rate using regression of fork length on scale radius may provide a more precise measure of growth rate. Edsall (1960) points out growth of lake whitefish in Munising Bay, Lake Superior was greatest in the first year of life and slowed in later years. Differences in growth rates between subpopulations of lake whitefish in Lake Winnipeg may exist during early development before growth inflections occur.

Meristic Characteristics of Subpopulations

Meristic differences useful in discrimination were lateral line scales and caudal peduncle scales. According to Svärdson (1952) the number of scales is environmentally modifiable especially in response to changed temperature conditions. The time for spawning in autumn and warming of the water in spring are particularly important factors. Apparently populations spawned early in autumn have many scales as they hatch early in spring (that is early development is in cold water) while those spawned in late autumn have few scales.

During the present study, time of specimen collection is an indication of spawning time since fish were collected in spawning condition. Grand Rapids fish and Traverse Bay fish were amongst the latest to spawn (Table 1) and observations of high scale counts amongst these subpopulations appear to contradict Svärdson's (1952) observations. The Traverse Bay and Grand Rapids subpopulations, besides having a relatively high mean number of lateral line scales both have in common the fact that they spawn in proximity to large inflowing rivers. The former spawns near the mouth of the Winnipeg River and the latter near the mouth of the Saskatchewan River. Environmental conditions, particularly temperature, may be influenced by these incoming water masses. Possibly these subpopulations spawn in association with cooler river inflow, and this is reflected phenotypically by the number of scales.

Reasons for differences in morphology between subpopulations of lake whitefish in Lake Winnipeg are no doubt complex, but whatever the mechanisms involved, the consequences are present and measureable.

Usefulness of Environmentally Variable Characters

Some researchers tend not to use environmentally modifiable characteristics in subpopulation studies since they do not provide as conclusive evidence of reproductive isolation as genetically based environmentally unmodifiable characteristics do. However, if certain conditions are met, environmentally induced variation can be valuable in identifying subpopulations.

This study shows that there are measurable differences in morphology between samples of lake whitefish taken from different areas in Lake Winnipeg. The differences could be phenotypic, induced by differences in environmental factors within Lake Winnipeg. In order to assign subpopulation status to the different groups it is of paramount importance to determine whether they home to their natal spawning grounds. The biochemical evidence provided by this study shows that lake whitefish do apparently home, and therefore the assignment of subpopulation status to the different groups identified in Figure 8 is justified.

Lindsey (1963) points out that lake whitefish may travel in homogeneous schools, the composition of which may

remain stable for long periods. Kennedy (1954) observed lake whitefish in Lake Winnipeg tagged as a group on the spawning grounds tended to remain together for as long as five years. He reported two lake whitefish tagged during the spawning run at Dauphin River in 1938 were recovered on the same spawning grounds one and two years later. Biochemical evidence presented earlier indicates reproductive isolation between at least two groups of lake whitefish in Lake Winnipeg. These groups must home to their natal spawning grounds. Figure 11 provides further evidence of non-random segregation. Lateral line scales, examined in this figure, are known to be environmentally variable (Svärdson 1952). The figure shows that there are differences in the mean number of lateral line scales between the samples. These may be genetic differences, or environmental variables responsible for modifying the number of lateral line scales differ between the locations compared. Each sample is comprised of fish of different ages and there is apparently little difference in the number of scales per fish between age groups within a sample. This seems to demonstrate that fish within each sample were not subject to year-to-year environmental differences sufficient to significantly alter scale count.

Within such a sample, lake whitefish of different ages are evidently all part of the same subpopulation, and they appear to have been returning, as a group, to spawn once more on their natal spawning grounds.

Morphological Evidence Complements Biochemical Evidence

Northcote et al. (1970) found that differences in lactate dehydrogenase phenotype distributions of stream populations of rainbow trout, Salmo gairdneri, below and above a waterfall were paralleled by frequency differences in meristic characters. Conclusions as to the subpopulation status of the Lake St. Martin-Dauphin River sample based on biochemical evidence were paralleled by morphological evidence (Figure 12). The biochemical differences indicate the Lake St. Martin-Dauphin River subpopulation is genetically distinct and spawns as a discrete unit. The morphological differences, if they are the result of environmental modification, suggest the subpopulation spawns in an environment different from that of other subpopulations.

The similarity of G-3-PDH b allele frequencies calculated for the remainder of Lake Winnipeg apart from Lake St. Martin-Dauphin River does not indicate that these samples are all part of one panmictic population. While differences in a character between groups may be taken as evidence of genetic isolation, similarities have no simple interpretation and certainly do not prove identity. An example is the marked difference in G-3-PDH b allele frequencies observed between Lake St. Martin-Dauphin River and the remainder of Lake Winnipeg yet no difference in G-3-PDH a allele frequencies exist between these two groups (Table 5).

Effect of Hatcheries

Two conservative characteristics (genetically based and not environmentally modifiable) were used to determine if hatchery-reared lake whitefish contribute significantly to native lake whitefish populations in Lake Winnipeg. G-3-PDH isozymes are genetically based (Clayton *et al.* 1973) and gill raker number is not easily modified by the environment (Lindsey 1963).

Lake whitefish spawn has been collected for at least 25 years at Clearwater Lake, incubated at the Dauphin River hatchery and introduced as eyed eggs or fry into Lake Winnipeg near the mouth of the Dauphin River. Since 1967 lake whitefish spawn from Clearwater Lake has been incubated at the Grand Rapids hatchery and introduced as fry into Lake Winnipeg at this location (K.C. Dey, pers. comm.). As well, spawn from William Lake has been used for the last eight years at Grand Rapids.

G-3-PDH *b* allele frequencies differ significantly ($p<.001$) between lake whitefish populations in Clearwater Lake and Lake St. Martin-Dauphin River, and in a manner contradictory to what would be expected if hatchery fish from Clearwater Lake contributed significantly to the size of the Lake St. Martin-Dauphin River subpopulation. The former population has a high frequency (0.50) of the b^1 allele while the latter population has a high frequency (0.77) of the b^3 allele. No significant difference in mean gill raker

number was found between these two locations.

G-3-PDH b allele frequencies also differ significantly ($p<.001$) between lake whitefish populations in Clearwater Lake and Grand Rapids, and marginally ($p\simeq.05$) between William Lake and Grand Rapids. As well Grand Rapids fish and William Lake fish differ significantly ($p<.001$, t-test not shown) in mean gill raker number. The former have a mean gill raker number of 27.73 (N=48) while the latter have a mean of 26.02 (N=48). Evidently the hatchery-reared fish introduced into Lake Winnipeg at Dauphin River and Grand Rapids do not contribute significantly to the native stocks which spawn here. Possibly hatchery-reared fish do contribute some members to native lake whitefish populations but in insufficient numbers to be detected by the techniques utilized in the present study. Individual fish cannot be identified as hatchery-reared or otherwise using G-3-PDH gene frequency comparisons since all three b alleles exist in each of the lakes examined. Similarly individual fish cannot be identified on the basis of gill raker number as considerable overlap in range exists between locations compared.

In certain situations hatcheries are probably useful such as supplementing depleted stocks when population size is small. However, the evidence given here does not offer any support to the belief that hatchery plantings in Lake Winnipeg are producing detectable results.

Implications for Fishery Management

Solutions to the "subpopulation problem" for fishery management require (1)recognition of the presence of subpopulations (2)detection of the location of the spawning grounds of different subpopulations and (3)delineating the geographic range and spawning range of each subpopulation (Marr and Sprague 1963). The present study has, in part, achieved the first two objectives. Collection of vital statistics for subpopulations should be done independently for each stock. Commercial catch samples can provide this data if it can be determined the catch has come from a single subpopulation and not an admixture. Results of tagging studies carried out at Dauphin River (Anon. 1959) and Little Mossy Point (Pollard 1973a) indicate most recoveries were within 45 to 65 miles of the tagging sites although a few individuals travelled as far as 90 miles. Probably the seasonal distributions of some lake whitefish stocks in Lake Winnipeg overlap and admixture occurs during part of the season.

The unique G-3-PDH b allele frequencies of the Lake St. Martin-Dauphin River subpopulation could allow this population to be detected by examining the commercial catch. Electrophoretic analysis of G-3-PDH from a sample (≈50 fish) taken from the commercial catch, and subsequent calculation of phenotype distribution and gene frequencies, could reveal whether the catch included mixed stocks. Disagreement of observed phenotypes from the Castle-Hardy-Weinberg expected

phenotypic distribution would indicate admixture of stocks (Marr and Sprague 1963). If this is the case, the sample should not be used for the collection of vital statistics. However, agreement with expected phenotype distributions does not necessarily indicate the sample is from one subpopulation. Results would depend upon the proportions contributed to the sample by different subpopulations. In fact, in some cases equal contributions by different subpopulations may not produce a significant ($p<.05$) χ^2 statistic at all. In the event the Castle-Hardy-Weinberg equilibrium appears to be satisified, the calculated gene frequency of the sample can be compared first with the gene frequency of one subpopulation identified in this study and then with the other, using the maximum likelihood ratio procedure described in Appendix I. No significant difference between the sample gene frequency and one subpopulation and a significant difference between the sample and the other subpopulation would identify the sample as having come from the former subpopulation and converse results would indicate it belonged to the later. Nonsignificant or significant differences between the sample gene frequency and both subpopulations would probably mean the sample was an admixture in the first place, and it should not be used for the collection of vital statistics.

It is not possible, on the basis of morphological differences noted during this study, to detect mixtures of stocks in a sample of the commercial catch because of the method used to adjust for the body size covariate. Discriminant

functions calculated during the present study were based on measurements adjusted to those of a fish 345 mm in body length and the adjustments were made along regression lines for individual groups and initially individual group membership was known. Lake whitefish of unknown origin could not be identified as to which subpopulation they belong by using discriminant functions calculated during the present study because the adjustment of measurements to those of a fish with body length 345 mm first requires knowledge of group membership in order that the adjustment be made along the proper regression line.

The size covariate could be adjusted for by using a pooled within-group slope as recommended by Thorpe (1976). The pooled within-group slope is an average of slopes of all groups. This technique was utilized by Blouw (1976) and Bodaly (1977). Residuals from the pooled within-group slope were subjected to single-discriminant-function analysis, in which comparisons were made between two groups.

During the present study, measurements which contributed to group separation were observed, for the most part, to have similar slopes but different intercepts. Using a pooled within-group slope method to adjust for size differences, these measurements would provide good discrimination between groups regardless of size. However, slopes of some discriminating measurements differed. The pooled within-group slope technique would then provide variable discriminating power depending on the size of the individuals

to be identified. Where slopes diverged, as size increases discrimination would be poor between small individuals and good between large individuals; where slopes converged, discrimination would be good between small individuals but poor between large individuals.

If the data collected during this study were re-examined and the size covariate discussed above was adjusted for using the pooled within-groups slope method, it might be possible to use the resulting discriminant functions to determine the relative rate of exploitation of different subpopulations. If a sample from the commercial fishery was examined and it was in fact comprised mostly of fish from one subpopulation, results of discriminant function classification would probably indicate this. However 80 percent and not 100 percent discrimination appears possible and one must remember that results would be subject to some error, and at best would provide only a rough indication of relative exploitation.

At present, perhaps the best way to determine if subpopulations intermix during the commercial fishing season is to tag fish on their spawning grounds. Mixtures of tags recovered during the fishery could reveal the extent of intermixing of substocks. If admixture of stocks is revealed, vital statistics should be evaluated by sampling lake whitefish on the spawning grounds, rather than from the commercial fishery.

In summary, results of this study show that subpopulations of lake whitefish do exist in Lake Winnipeg, and some of their spawning locations have been identified. Biochemical differences provide a means for identifying one subpopulation from commercial samples. Future programs, designed to determine the size and seasonal distribution of these subpopulations, can utilize as a starting point the information provided by this study.

1. Lake whitefish spawners were captured at Traverse Bay, Dauphin River, Berens River, Poplar River, Grand Rapids and Warren Landing in Lake Winnipeg, as well as from Lake St. Martin and Little Playgreen Lake. Comparisons were made between samples using biochemical and morphological techniques to determine if subpopulations of this species exist in Lake Winnipeg.

2. Differences in G-3-PDH b allele frequencies provide evidence that at least two subpopulations reside in Lake Winnipeg. One spawns in the Lake St. Martin-Dauphin River area while the other spawns in the remainder of the lake.

3. Lake whitefish in Lake Winnipeg can be separated into five subpopulations spawning at Lake St. Martin-Dauphin River, Traverse Bay, Berens-Poplar-Big Black Rivers, Grand Rapids, and Little Playgreen Lake, based on morphological differences.

4. There is evidence that at least two subpopulations passed by Warren Landing during October, 1975, prior to spawning.

5. Selection, environmental differences within Lake Winnipeg, and natal homing may be responsible for different subpopulations in Lake Winnipeg.

6. Spawners from Clearwater Lake and William Lake were compared with the samples from Dauphin River and Grand Rapids to determine the effect hatchery plantings have on indigenous lake whitefish stocks in Lake Winnipeg. Comparisons involved biochemical techniques and gill raker number. Hatch-

eries at Grand Rapids and Dauphin River obtain spawn from Clearwater Lake and William Lake.

7. Results of this study do not provide evidence that hatchery plantings in Lake Winnipeg at Grand Rapids and Dauphin River are producing detectable results.

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Appendix I. Maximum likelihood ratio test

A widely used point estimate of a parameter is based on the principle of maximum likelihood. If $\hat{\theta}$ is the value of θ in the probability space Ω that maximizes the likelihood function $g(x_1, x_2 \ldots\ldots, x_n; \theta)$, then $\hat{\theta}$ is called the maximum likelihood estimator of θ (Bhat 1972, p. 370). $\hat{\theta}$ is a reasonable estimator of θ, since $g(x_1, x_2, \ldots\ldots x_n; \theta)$ is the sampling distribution for the set of observations $x_1, x_2, \ldots\ldots x_n$ under the model $g(\theta)$. Hence $\hat{\theta}$ is simply the value of θ which makes what was observed most probable, given the model. Maximum likelihood estimators, moreover, have highly desirable properties. They are asymptotically unbiased, efficient and normally distributed (Kendall and Stewart 1967, Vol. 2, Chap. 18). Asymptotic properties are ones that hold as sample size becomes large.

Now, if Ω is a set of values the parameter θ can assume, and ω is a subset of Ω, the maximum likelihood ratio statistic λ is defined as: $\lambda = \frac{L(\omega)}{L(\Omega)}$ when $L(\omega)$ is the likelihood function evaluated at the maximum likelihood value for $\theta \varepsilon \omega$.

The Neyman-Pearson Lemma (Wilks 1962, Sec. 13.2) states that the most powerful unbiased test of H_0: $\theta \varepsilon \omega$ vs H_1: $\theta \varepsilon \Omega$ must be based on the statistic λ. For large sample size n the distribution of the test statistic $T = -2 \ln\lambda$ approaches the χ^2 distribution with d degrees of freedom, given that H_0 is true, where d = the number of parameters fitted for H_1

minus the number of parameters fitted for H_0. Hence, values of T which are much larger than could occur by chance from the χ^2_d distribution indicate that H_0 is unlikely to be true, and that H_1 gives a significantly better description of the observed data.

For a simple 2 allele system, to compare two populations with one paramater, p:

let p = the frequency of the $\underline{b}^1$ allele

and q = the frequency of the $\underline{b}^2$ allele

then for population 1,

Phenotype:	B^1B^1	B^1B^2	B^2B^2	Sum
Relative frequency:	p_1^2	$2p_1q_1$	q_1^2	1
Observed frequency:	n_{11}	n_{12}	n_{13}	N_1

then for population 2;

Phenotype:	B^1B^1	B^1B^2	B^2B^2	Sum
Relative frequency:	p_2^2	$2p_2q_2$	q_2^2	1
Observed frequency:	n_{21}	n_{22}	n_{23}	N_2

To test for homogeneity for p_1 and p_2:

H_0: $p_1 = p_2 = p$

H_1: $p_1 \neq p_2$

If H_0 is true, the maximized likelihood under $H_1, L_1(\hat{p}_1, \hat{p}_2)$ should not be significantly larger than the maximized likelihood under $H_0, L_0(\hat{p})$, where $-2 \ln\lambda \simeq \chi^2_d$ with $\lambda = L_0(\hat{p})/L_1(\hat{p}_1\hat{p}_2)$.

The number of parameters fitted for H_1 is 2, $\hat{p}_1$ and $\hat{p}_2$. The number of parameters fitted for H_0 is 1, p. Therefore, d, the number of degrees of freedom is 2 - 1 = 1 and:

$L_0 = L(p|n_{11}\ n_{12}\ n_{13}).\ L(p|n_{21}\ n_{22}\ n_{23})$

$L_1 = L(p_1|n_{11}\ n_{12}\ n_{13}).\ L(p_2|n_{21}\ n_{22}\ n_{23})$

where $L(p|n_1\ n_2\ n_3) = \frac{N!}{n_1!\ n_2!\ n_3!}\ (p^2)^{n_1}\ (2pq)^{n_2}\ (q^2)^{n_3}$ and the estimate $\hat{p} = \frac{2n_1+n_2}{2(n_1+n_2+n_3)}$. $\hat{p}$ is the value which maximizes L, that is, makes the observed sample most probable, given the model. Notice that in both cases the likelihood is the product of two likelihood expressions, one from each population. Hence the log likelihood will be the sum of the log likelihoods contributed by each population.

For a more complicated 3 allele system the test can be extended using 2 parameters, p and q:

let p = the frequency of the $\underline{b}^1$ allele

let q = the frequency of the $\underline{b}^2$ allele

let r = the frequency of the $\underline{b}^3$ allele

then,

Phenotype:	B^1B^1	B^1B^2	B^1B^3	B^2B^2	B^2B^3	B^3B^3	Sum
Rel. frequency:	p^2	2pq	2pr	q^2	2qr	r^2	1
Obs. frequency	n_1	n_2	n_3	n_4	n_5	n_6	N

and,

$$L(p,q|\vec{n},N) =$$

$$\frac{N!}{n_1!\ n_2!.....n_6!}\ (p^2)^{n_1}(2pq)^{n_2}(2pr)^{n_3}(q^2)^{n_4}(2pr)^{n_5}(r^2)^{n_6}$$

$\hat{p}$ and $\hat{q}$ are found by substituting r = (1-p-q) in L and then finding $\hat{p}$, $\hat{q}$ to satisfy:

$$\frac{\partial}{\partial p} \text{Log } L\ (p,q) = 0$$

$$\frac{\partial}{\partial q} \text{Log } L\ (p,q) = 0$$

The solution to these equations can be shown to be:

$$\hat{p} = \frac{2n_1+n_2+n_3}{2N} = \underline{b}^1$$

$$\hat{q} = \frac{n_2+2n_4+n_5}{2N} = \underline{b}^2$$

$$\hat{r} = 1 - \hat{p} - \hat{q} = \underline{b}^3$$

The maximum likelihood estimate of $\hat{p}$ and $\hat{q}$ are simply the observed frequencies of the allele $\underline{b}^1$ and $\underline{b}^2$ respectively.

Calculation of the statistics involved was done using APL (A Programming Language) on the University of Manitoba IBM Model 370 computer. Following are examples of simple and composite tests of G-3-PDH b allele frequencies between populations compared in this study.

Simple Test

H_0: Lake St. Martin = Dauphin River

H_1: Lake St. Martin ≠ Dauphin River

under H_1; individually estimated frequencies are:

$\underline{b}^1$	$\underline{b}^2$	$\underline{b}^3$	
0.1000	0.1100	0.7900	Lake St. Martin
0.1354	0.1250	0.7396	Dauphin River

Contributions to log likelihood are:

-65.93 for Lake St. Martin

-72.36 for Dauphin River

Total log likelihood (L_1) is (-65.93) + (-72.36) = -138.3

Under H_0; pooled frequency estimates are:

$\underline{b}^1$	$\underline{b}^2$	$\underline{b}^3$	
0.1173	0.1173	0.7653	Pooled

Contributions to log likelihood are:

-66.13 for Lake St. Martin

-72.56 for Dauphin River

Total log likelihood (L_0) is (-66.13) + (-72.56) = -138.7

Now $$\lambda = \frac{L_0}{L_1} = \frac{-138.7}{-138.3}$$

and $-2 \ln\lambda \simeq \chi^2 r$

so $\chi^2 = -2\ (138.7) - (-138.3)$

$\chi^2 = -2\ (-0.4)$

$\chi^2 = \underline{\underline{0.8}}$ with 2 degrees of freedom

For H_1, the parameters $\hat{p}_1$, $\hat{q}_1$, $\hat{p}_2$ and $\hat{q}_2$ were estimated ($\hat{r}_1$ and $\hat{r}_2$ are obtained by subtraction as $p + q + r = 1$).

For H_0, the pooled parameters p and q were estimated.

Degrees of freedom are therefore 4 - 2 = 2.

χ^2 of 0.8 with 2 degrees of freedom has $0.975>p>0.95$, hence no significant difference exists between the two populations.

Composite Test

H_0: Traverse Bay, Berens River, Poplar River, Big Black River, Grand Rapids, Little Playgreen Lake, Lake St. Martin and Dauphin River are all homogeneous.

H_1: Traverse Bay, Berens River, Poplar River, Big Black River, Grand Rapids and Little Playgreen Lake (Group 1) $\neq$ Lake St. Martin and Dauphin River (Group 2).

Under H_1; pooled within group estimated frequencies are:

$\underline{b}^1$	$\underline{b}^2$	$\underline{b}^3$	
0.2469	0.2891	0.4641	Group 1
0.1173	0.1173	0.7653	Group2

Contributions to log likelihood are:

-103.1 for Traverse Bay
-121.8 for Berens River
-117.5 for Poplar River — Group 1
-115.9 for Big Black River
-110.7 for Grand Rapids
-109.7 for Little Playgreen Lake
-66.13 for Lake St. Martin
-72.56 for Dauphin River — Group 2

Total log likelihood (L_1) is [(-103.3) + (-121.8) + (-117.5) + (-115.9) + (-110.7) + (-109.7)] + [(-66.13) + (-72.56)] = -817.3

Under H_0; pooled estimated frequencies are:

$\underline{b}^1$	$\underline{b}^2$	$\underline{b}^3$
0.2165	0.2488	0.5347

Total log likelihood (L_0) is -846.2.

Now $$\lambda = \frac{L_0}{L_1} = \frac{-846.2}{-817.3}$$

and $-2 \ln\lambda \simeq \chi^2_d$

where d = 2

so $$\chi^2_2 = -2\,[(-846.2) - (-817.3)]$$

$$\chi^2_2 = -2\ (-28.9)$$

$$\chi^2_2 = \underline{\underline{57.8}}$$

χ^2 of 57.8 with 2 degrees of freedom has $p<.001$, hence there is a significant difference between these two groups.

Appendix II. Discriminant-function analysis

Discriminant-function analysis has been defined as a multivariate technique for studying the extent to which different populations overlap or diverge from one another (Snedecor and Cochran 1967, p. 414). Morphometric and meristic comparisons made during the present study involve multigroup multivariate analyses to which discriminant-function analysis is tailored.

Morphometric comparisons often involve overall body size as a covariate. During this study ratios were not used to alleviate this problem for reasons described by Atchley et al. (1976). Analysis of covariance is inadequate in this case since it does not provide information concerning differences between specific groups in multigroup comparison. The body size covariate was removed here using the technique described by Lindsey (1963).

The BMDP7M Stepwise Discriminant Analysis program includes in its output test statistics (Wilk's lambda and the F approximation to lambda) used to test group differences. Tests of statistical significance were not used in the present analysis because the objective here was to determine whether morphological measurements could discriminate between groups. A predictive discriminator could not be obtained due to the method of adjustment for the size covariate (see Discussion). Hence a different approach, which gives a measure of discriminatory success, was taken. Included in the BMDP7M program output is a classification table containing

the percentage of cases classified correctly according to original group membership. At the start, group membership of each case is known since location of capture of each specimen was recorded. During comparisons a classification function for each group is compiled based on a subset of variables that maximizes group differences. The value of all classification functions (one for each group) are computed for each case and these values are used to compute a posterior probability. Each case is then assigned to the group in which the value of the posterior probability is maximum. A jack knife classification is included where the classification function is computed with the case omitted from the computations. The function then classifies the left-out case, resulting in a less biased classification since the classification function will obviously produce better results when it is used to classify the same cases that were used to compute it.

The percent of cases classified correctly can be used as a measure of success of the discriminant-function analysis procedure. If a large percentage of cases are assigned to their original group one has reason to conclude that group differences exist and a set of variables that exhibit the differences has been chosen.

A score of 80 percent correct classification was arbitrarily selected during the present analysis as indicative of group differences. It can be argued that no statistical significance level can be assigned such results

and conclusions reached will be of a speculative nature. However, if sufficient differences exist between groups of fish whereby eight of ten fish from each group can be correctly identified according to original group membership, such differences must have a biological basis.

Appendix III. Estimates of genetic distance (D) and genetic identity (I) among samples of lake whitefish from Lake Winnipeg and other lakes. Values above the diagonal are D with standard error in brackets, and values below the diagonal are I.

	Population 1						Population 2		Hatchery Lakes	
	Traverse Bay	Berens River	Poplar River	Big Black River	Little Playgreen L.	Grand Rapids	Dauphin River	L. St. Martin	Clearwater Lake	William Lake
Traverse Bay	xxxx	.0039 (.028)	.0027 (.023)	.0003 (.008)	.0016 (.018)	.0012 (.016)	.0168 (.057)	.0248 (.069)		
Berens R.	.9961	xxxx	.0054 (.033)	.0032 (.025)	.0009 (.013)	.0019 (.020)	.0081 (.040)	.0127 (.050)		
Poplar R.	.9973	.9947	xxxx	.0013 (.016)	.0028 (.024)	.0048 (.031)	.0222 (.066)	.0298 (.076)		
Big Black R.	.9997	.9968	.9987	xxxx	.0010 (.014)	.0014 (.017)	.0167 (.057)	.0244 (.069)		
Little Playgreen L.	.9984	.9991	.9972	.9990	xxxx	.0007 (.012)	.0116 (.048)	.0183 (.060)		
Grand Rapids	.9988	.9981	.9952	.9986	.9993	xxxx	.0104 (.045)	.0178 (.059)	.0122 (.049)	.0039 (.028)
Dauphin R.	.9833	.9919	.9780	.9834	.9884	.9896	xxxx	.0015 (.017)	.0394 (.086)	
L. St. Martin	.9755	.9874	.9707	.9759	.9818	.9823	.9985	xxxx	.0518 (.098)	
Clearwater L.						.9878	.9614	.9495		
William L.						.9961				