A genetic test of metapopulation structure in Atlantic salmon (*Salmo salar*) using microsatellites[1]

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Abstract: The principal objective of this study was to describe the pattern of genetic exchange and isolation of Atlantic salmon (*Salmo salar*) populations among geographical regions of the province of Quebec, Canada. Seven riverine populations, associated with three putative regional metapopulations (North Shore, Gaspé Peninsula, and Ungava), were analyzed using microsatellites. Our results did not support the putative metapopulation structure. Significant heterogeneity in allelic frequency was observed among most rivers independently of their location or group subdivision. Interpopulation genetic variance (ϕ_{ST}) indicates less heterogeneity among rivers than χ^2 analysis and was mainly associated with the geographical distance of the most isolated rivers, the Natashquan and the Koksoak. Even with low genetic variance among populations, the overall significant allelic heterogeneity among rivers strongly suggests that each population, whether separated by thousands or tens of kilometres, should be considered and managed as a specific stock.

Résumé : L'objectif principal de cette étude était de décrire le degré d'échange génétique entre des populations de saumon atlantique (*Salmo salar*) appartenant à différentes régions géographiques de la province de Québec. Sept rivières, associées à trois metapopulations régionales hypothétiques (Côte-Nord, péninsule de Gaspé et l'Ungava), ont été analysées à l'aide de microsatellites. Nos résultats ne soutiennent pas une structure composée de metapopulations. Une hétérogénéité significative des fréquences allèliques entre la majorité des rivières indépendamment de leur localisation ou de leur subdivision par région. La variance génétique interpopulation (ϕ_{ST}) suggère une hétérogénéité entre les rivières mais à un degré moindre qu'indiqués par les χ^2 et était associée essentiellement avec la distance géographique des rivières plus isolées, telles que la Natashquan et la Koksoak. Même si on observe de faibles variances génétiques ou structure de population, l'hétérogénéité allèlique significative et générale entre les rivières suggère fortement que les populations, voisines ou séparées par des milliers de kilomètres, soient considérées et gérées comme des stocks spécifiques.

Introduction

Atlantic salmon (*Salmo salar*) are widely distributed in the North Atlantic, from Russia to Portugal in the east and from northern Quebec (Canada) to Connecticut (U.S.A.) in the west (MacCrimmon and Gotts 1979). The recent and gradual colonization of this area occurred after the retreat of the ice cover following the Wisconsinian glaciation (Crossman and McAllister 1986; Schmidt 1986). As such, salmon populations found in 115 rivers of Quebec are thought to be the result of 7000 to 13 000 years of natural selection for local conditions (Power 1981; Fulton and Andrews 1987).

Atlantic salmon are subject to different environmental conditions depending on the latitude and physical setting (48–58°N; Fig. 1) of the natal river. Differences such as mean temperature, length of the growing season, and river discharge create variations among rivers in parameters such as growth, age, and the timing of smoltification and the adult migration. Based on such differences, Power (1981) hypothesized that the province of Quebec contains three distinct regional groups or metapopulations of stocks, a metapopulation being composed of semi-isolated stocks linked by limited gene flow. The level of gene flow is expected to be more important among stocks of one metapopulation than among metapopulations. A stock is defined as an intraspecific group of randomly mating fish exhibiting spatial or temporal integrity (Ihssen et al. 1981). The three groups of stocks proposed by Power (1981) are differentiated by several demographic variables. First, the Ungava Bay stocks show many distinctive characteristics including large, old smolts and maturation cycles that involve a proportion of fish returning to rivers more than 1 year prior to spawning. Secondly, the north shore of the Gulf of St. Lawrence stocks have younger smolt ages and grilse (fish that return to spawn after only 1 year at sea) are almost all males. Finally, the Gaspé and Anticosti stocks have a lower proportion of grilse of both sexes and younger smolt ages.

The gradual variation of the allelic frequency distribution associated with environmental conditions suggests an indirect or direct effect of natural selection on population divergence in Atlantic salmon. In eastern Canada, Vespoor and Jordan (1989) demonstrated that allelic distribution of malic enzyme (Me-2) was not random but associated with temperature. In transferrin, Tf-4 may be associated with the proportion of

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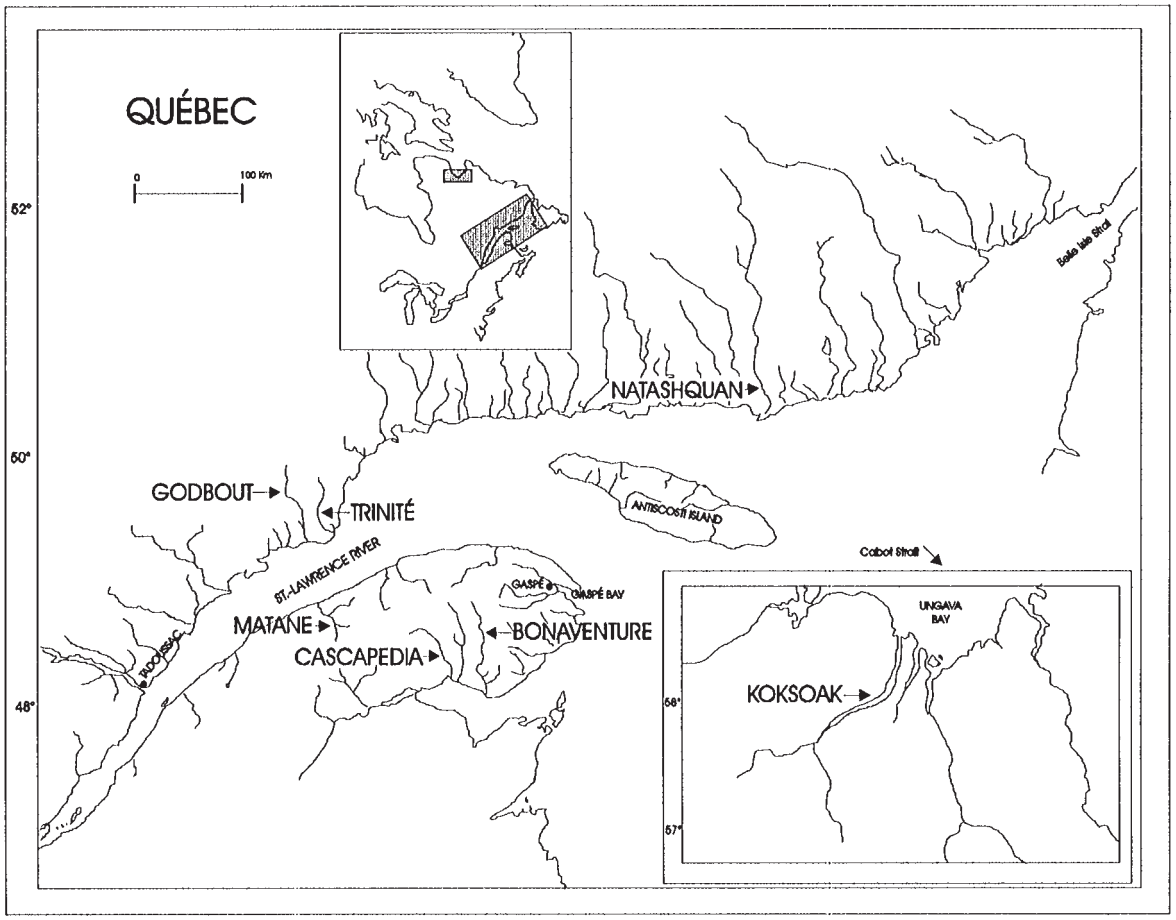
Fig. 1. Location of the rivers studied.



one-sea-winter fish and exhibited a gradual cline, increasing in the northern part of the range of the Atlantic salmon (Vespoor 1986; Payne 1974).

Fishery biologists have long recognized the need to identify differences among stocks and to manage them to conserve local adaptations (Taylor 1991; Carvalho 1993). Several studies of genetic variation in salmon based on neutral polymorphisms in protein loci or mitochondrial DNA (mtDNA) have revealed low levels of genetic diversity. The three major discontinuities found to date exist between salmon from North America, the western Atlantic, and the Baltic sea (Davidson et al. 1989; Althukhov and Salmenkova 1991; Jordan et al. 1992; Stahl 1986). These techniques can also detect significant differences at smaller geographical scales (Hurrell and Price 1993; King et al.1993; Moran et al. 1994; Tessier et al. 1995). In species showing little genetic differentiation with allozymes or mtDNA, the high level of allelic variation revealed by microsatellites make them potentially more useful markers to study population genetic structure (Wright and Bentzen 1994; McConnell et al. 1995*a*, 1995*b*; Angers et al. 1995; Tessier et al.1995). Microsatellites are hypervariable specific regions of genomic DNA composed of a variable number of tandem repetitions (VNTR) of two, three, or four nucleotides, flanked by nonrepetitive DNA (Nakamura et al. 1987; Tautz 1989; Litt and Lutty 1989).

The principal objective in this study was to examine the genetic basis of the three metapopulations proposed by Power (1981) by documenting microsatellite allelic polymorphisms of salmon found in rivers associated with the three putative regional metapopulations.

Materials and methods

A total of seven rivers was sampled. The Cascapedia, Bonaventure, and Matane rivers were associated with the Gaspé group; the North Shore group was represented by the Trinité, Godbout, and Natashquan rivers. The Koksoak is associated with the Ungava group (Fig. 1). Each group, except Ungava, was composed of two neighboring rivers, and one situated approximately 400 km from the others to test intragroup genetic variation at different geographical distances. The mean numbers of adult salmon present in each river between 1987 and 1991, after the fishing season, were 2271, 1751, 2224, 1984, and 1520 for the Bonaventure, Cascapedia, Matane, Godbout, and Trinité rivers, respectively. No estimate is available for the Natashquan and the Koksoak rivers. However, these rivers sustain an annual fishery of 1906 and 2652 adults, respectively, which is double that of the other rivers.

Parr of different age-classes were sampled to reduce the potential effect of genetic differences among cohorts. Specimens were captured with an electrofishing device and preserved at –80°C until genomic DNA was extracted using a standard phenol–chloroform protocol (Sambrook et al. 1989). Population genetic diversity was analyzed on the basis of allelic frequencies at five microsatellite loci. Five pairs of primers, *Ssa202*, *Ssa197*, *Ssa171* (O'Reilly et al. 1996), *SSOSL85* (Slettan et al. 1995), and μ*3* (Presa and Guyomard 1996), were used

Table 1. Observed (H_o) and expected (H_e) heterozygosity for each population and locus.

		Populations[a]						
Locus		Cas	Bon	Mat	Tri	God	Nat	Kok
Ssa202	H_o	0.76	0.82	0.80[b]	0.79	0.89	0.90	0.85
	H_e	0.84	0.91	0.88	0.85	0.87	0.88	0.91
Ssa197	H_o	1.00[c]	0.78	0.86	0.89	0.86	0.85	0.70
	H_e	0.87	0.88	0.85	0.89	0.91	0.84	0.82
Ssa171	H_o	0.72	0.70	0.80[b]	0.93	0.92	0.95	0.73
	H_e	0.84	0.75	0.92	0.95	0.90	0.90	0.91
SSOSL85	H_o	0.75	0.81	0.86	0.84	0.70	0.76	0.83
	H_e	0.77	0.86	0.89	0.89	0.76	0.74	0.85
μ*3*	H_o	0.74	0.61	0.79	0.57	0.67[d]	0.85	0.5
	H_e	0.75	0.73	0.70	0.68	0.75	0.73	0.54

[a]Cas, Cascapedia River; Bon, Bonaventure River; Mat, Matane River; Tri, Trinité River; God, Godbout River; Nat, Natashquan River; Kok, Koksoak River.
[b]Signficant Hardy–Weinberg disequilibrium ($p = 0.034$).
[c]Signficant Hardy–Weinberg disequilibrium ($p = 0.025$).
[d]Signficant Hardy–Weinberg disequilibrium ($p = 0.028$).

to amplify the microsatellite loci using polymerase chain reaction (PCR).

The PCR reactions were performed in 10-μL volumes containing the following: 1μL of buffer (Boerhinger), 200 μM of each dNTP, 0.5 *Taq* unit, 4.5 pmol and 0.5 pmol end-labelled (γ-^{32}P) of one primer, 5 pmol of the other primer, ~60 ng of DNA, and sufficient water to bring the volume to 10 μL for the *Ssa* microsatellites. The conditions for the microsatellite μ*3* were 1.25 μL buffer, 2.5 μM dATP, 75 μM of the three other dNTP, 0.12 μM α-^{35}S-dATP, 0.5 of *Taq* unit, 15 pmol of primers, and sufficient water bring the volume to 12.5 μL. All PCR amplification cycles were preceded by 5 min of denaturation (94°C) and ended with an extension (72°C) of 10 min. The cycles used were 30 s denaturation, 30 s annealing at 58°C, and 30 s extension for the *Ssa* microsatellites and 60 s denaturation, 35 s annealing at 57°C, and 10 s extension for the μ*3* microsatellites. The *SSOSL85* PCR conditions are available in Slettan et al. (1995). The reaction products were resolved on a 6% denaturing polyacrylamide sequencing gel visualized by autoradiography. These microsatellites were selected based on the quality of the amplifications and the ease by which they were scored. The *Ssa* loci were mutltiplexed in the same PCR reaction and on the same gel as described by O'Reilly and Wright (1995). However, we ran the locus *Ssa171* separately in certain cases to avoid overlapping with the locus *Ssa202*. The sizes of alleles of PCR products were determined with a standard M13 sequence (USB, Sequenase version 2.0 DNA sequencing kit). However, our scale differs by +20 base pairs from that used by O'Reilly et al. (1996).

Statistical analysis

Standard measures of genetic variation within samples, including allelic frequency, observed heterozygosity (H_o), expected heterozygosity (H_e), and assessment of Hardy–Weinberg equilibria, were calculated using the GENEPOP program version 1.2 (Raymond and Rousset 1995). The homogeneity of allelic frequencies among samples was evaluated using χ^2 randomization tests (Roff and Bentzen 1989) with 1000 permutations performed by the Monte program of the REAP software package (McElroy et al. 1992).

Gene diversity among all rivers and regions was evaluated by computing ϕ statistics with a test of their significant differences. These significant differences were detected using a random allelic permutation procedure (analysis of molecular variance (AMOVA) computer package; Excoffier et al. 1992; Michalakis and Excoffier 1996). The ϕ statistic differs from Wright's *F* statistic (Wright 1951) by the incorporation of molecular information based on the differences in the number of repeats among microsatellites estimated by squared Euclidean distances between pairwise alleles. It therefore incorporates the nature of the principal mutational mode of microsatellites, best described by a stepwise mutation model (Kimura and Otha 1978; Shriver et al. 1993).

Genetic relationships among populations were depicted with a neighbor-joining tree from a matrix of genetic distances (Schriver et al. 1995). Based on allele frequency distribution and the number of repetitions composing the alleles, this estimate assumes a linear relationship of allelic divergence with respect to time.

The effective number of migrants per generation (N_em) was evaluated by replacing F_{ST} by ϕ_{ST} in the following formula: $F_{ST} = 1/(4N_em + 1)$ (Michalakis and Excoffier 1996).

Results

All loci were highly variable as revealed by levels of heterozygosity that ranged between 0.5 to 1. The highest levels of polymorphism were obtained with tetranucleotide microsatellite loci. Eighteen, 21, 29, 12, and 11 alleles were detected at the locus *Ssa202*, *Ssa197*, *Ssa171*, *SSOSL85*, and μ*3*, respectively. The allelic frequencies of loci are presented in the Appendix. Only 4 cases in 35 showed significant departures from Hardy–Weinberg equilibria (Table 1). At the $p = 0.05$ level, we expect to obtain two Hardy–Weinberg disequilibria by chance alone. We observed two deficits of heterozygosity for the loci *Ssa202* and *Ssa171* in the Matane River and at the locus μ*3* for the Godbout River. In the Cascapedia River, the absence of homozygotes at the locus 197 created an excess of heterozygosity. These four cases were only significant at the ($0.01 < p < 0.05$) level, and they were not associated with any particular loci. The excess of heterozygosity at the locus *Ssa197* may be explained by the high number of alleles for this locus (up to 16) relative to the sample size ($n = 28$).

The AMOVA components did not support the hypothesis of three regional metapopulations proposed by Power (1981). Thus, the nested analysis of population structure revealed that the total genetic diversity explained by variance among the three groups varied between 0 and 3.7% depending on the locus (Table 2). This translated into an overall intergroup variance estimate of 1.3%, which was not statistically different from zero.

Table 2. Proportion of gene diversity due to variation among regional groups.

	SSOSL85		μ*3*		*Ssa202*		*Ssa197*		*Ssa171*		All loci,
Group comparison[a]	ϕ_{ST}	*p*	ϕ_{ST}	*p*	ϕ_{ST}	*p*	ϕ_{ST}	*p*	ϕ_{ST}	*p*	ϕ_{ST}
B+C+M vs. G+T+N vs. K	0	0.56	0	0.78	0.04	0.14	0	0.65	0.01	0.32	0.013

[a]B, Bonaventure River; C, Cascapedia River; M, Matane River; G, Godbout River; T, Trinité River; N, Natashquan River; K, Koksoak River.

Table 3. Genetic homogeneity (χ^2), genetic variance (ϕ_{ST}), and number of migrants per generation (N_em) between pairs of populations.

	Ssa171		*Ssa197*		*Ssa202*		μ*3*		*SSOSL85*		Mean	No. of χ^2,	No. of ϕ_{st},	
Population	χ^2 (*p*)	ϕ_{ST}	χ^2 (*p*)	ϕ_{ST}	χ^2 (*p*)	ϕ_{ST}	χ^2 (*p*)	ϕ_{ST}	χ^2 (*p*)	ϕ_{ST}	ϕ_{ST}	*P* < 0.05	*P* < 0.05	N_em
B–C	0.021	0.003	0.466	0.000	<0.001	0.018	0.041	0.000	<0.001	0.000	0.004	4	0	59.3
B–M	0.015	0.014	0.162	0.025	0.007	0.001	<0.001	**0.290**	0.680	0.000	0.066	3	1	3.5
C–M	0.042	0.000	0.001	0.003	0.005	0.005	<0.001	**0.390**	0.001	0.000	0.080	5	1	2.9
Mean		0.006		0.009		0.008		0.227		0.000	0.050			4.8
G–T	0.002	0.000	0.276	0.018	0.015	0.060	0.018	0.112	0.001	0.007	0.039	4	0	6.1
G–N	<0.001	**0.126**	0.065	0.009	0.297	0.059	<0.001	**0.258**	<0.001	**0.277**	0.146	3	3	1.5
T–N	<0.001	0.051	0.286	0.021	0.068	0.000	<0.001	**0.400**	<0.001	**0.168**	0.128	3	2	1.7
Mean		0.059		0.016		0.040		0.257		0.151	0.104			2.1
B–T	<0.001	0.022	0.427	0.042	0.019	0.000	0.304	0.015	0.399	0.000	0.016	2	0	15.6
B–G	0.001	0.000	0.113	0.000	0.001	0.015	0.007	**0.072**	<0.001	0.000	0.017	4	1	14.1
B–N	<0.001	**0.182**	0.022	0.033	0.066	0.000	<0.001	**0.426**	<0.001	**0.283**	0.185	4	3	1.1
C–T	<0.001	0.000	0.206	0.018	0.029	0.000	<0.001	**0.057**	<0.001	0.000	0.015	4	1	16.4
C–G	0.016	0.000	0.407	0.000	0.562	0.019	0.009	**0.060**	0.969	0.000	0.016	2	1	15.6
C–N	<0.001	**0.108**	0.083	0.009	0.058	0.000	<0.001	**0.524**	<0.001	**0.221**	0.172	3	3	1.2
M–T	0.011	0.000	0.007	0.000	0.037	**0.051**	<0.001	**0.235**	0.557	0.000	0.057	4	2	4.1
M–G	0.271	0.000	<0.001	0.004	0.007	0.000	<0.001	**0.232**	<0.001	0.012	0.050	4	1	4.8
M–N	0.003	**0.085**	0.018	0.000	0.432	0.053	0.049	0.038	<0.001	**0.198**	0.075	5	2	3.1
Mean		0.044		0.012		0.015		0.184		0.079	0.067			3.5
K–B	<0.001	**0.162**	0.038	0.000	0.001	**0.148**	<0.001	**0.110**	0.073	0.000	0.084	4	3	2.7
K–C	<0.001	**0.075**	0.010	0.000	<0.001	**0.184**	<0.001	**0.215**	<0.001	0.000	0.095	5	3	2.4
K–M	0.105	0.052	0.096	0.000	0.005	**0.115**	0.012	**0.095**	0.006	0.000	0.052	4	2	4.5
Mean		0.096		0.000		0.149		0.140		0.000	0.077			3.0
K–T	0.002	0.014	0.013	0.012	<0.001	**0.263**	<0.001	0.041	0.065	0.000	0.066	4	1	3.5
K–G	0.012	**0.095**	0.052	0.000	<0.001	0.034	0.002	**0.115**	<0.001	0.000	0.049	4	2	4.9
K–N	0.044	0.000	0.127	0.004	<0.001	**0.264**	0.001	**0.279**	<0.001	**0.266**	0.163	4	3	1.3
Mean		0.036		0.005		0.187		0.145		0.089	0.092			2.5
Overall		0.047		0.009		0.061		0.189		0.068	0.075			3.1

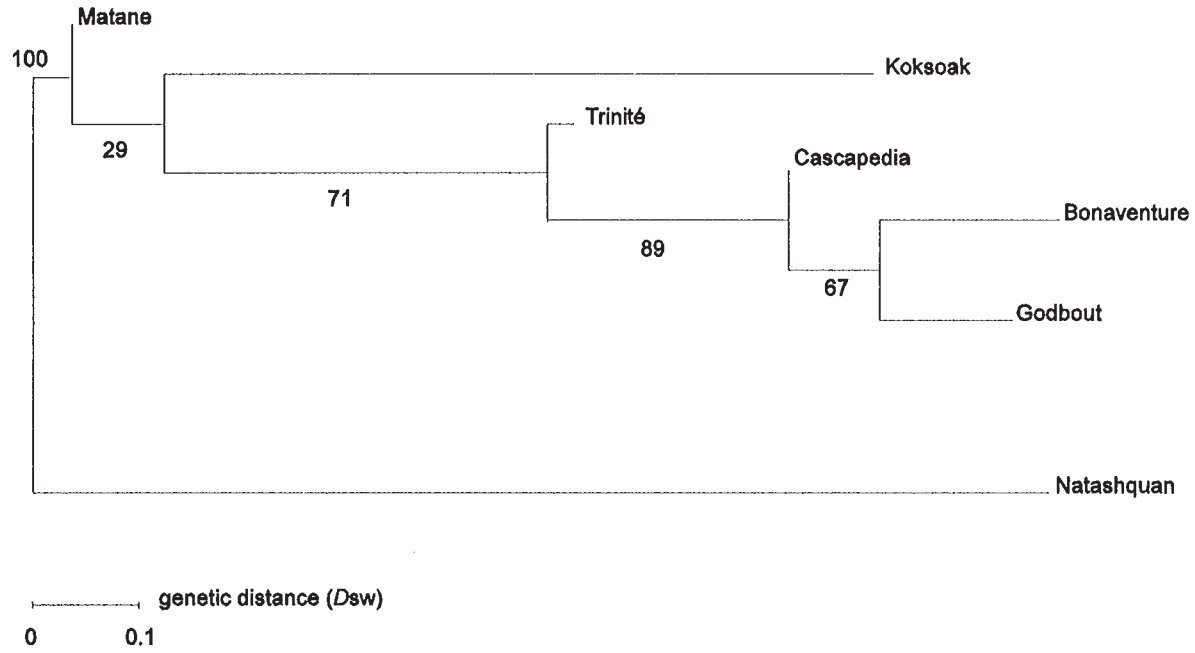
Note: All *p* values for χ^2 less than 0.05 were considered to be significant. The ϕ_{ST} values given in boldface indicate significantly different pairwise comparisons ($p < 0.05$). B, Bonaventure River; C, Cascapedia River; M, Matane River; G, Godbout River; T, Trinité River; N, Natashquan River; K, Koksoak River.

Twenty-one pairwise χ^2 comparisons were done for each locus to examine heterogeneity of allelic frequencies among populations (Table 3). These indicated that salmon from the different rivers compose genetically distinct populations. Seventy-eight of the 105 comparisons were different ($p < 0.05$). Comparisons not significantly different were randomly distributed and do not suggest an influence of subdivision into groups or of the distance separating rivers. With the exception of locus *Ssa197*, all microsatellites detected significant heterogeneity in allelic frequencies between neighboring populations separated by less than 35 km, as in the case of Godbout–Trinité and Bonaventure–Cascapedia. Only two comparisons, Bonaventure–Trinité and Cascapedia–Godbout did not reveal a majority of loci exhibiting significantly different allelic frequencies.

ϕ_{ST} estimates for individual loci were generally low, with a mean value of 0.075 (Table 3). For the same pairwise comparisons as above, we observed only 35 differences ($p < 0.05$). The Natashquan and the Koksoak rivers were involved in 27 of the 35 cases. A majority of loci revealed significant variance for six pairwise comparisons: Koksoak River with the Natashquan, Bonaventure, and Cascapedia rivers and the Natashquan River with the Godbout, Bonaventure, and Cascapedia rivers. Therefore, the ϕ_{ST} estimates indicated genetic heterogeneity among rivers but to a lesser degree than that indicated by χ^2 and mainly associated with the geographical distance of these two most isolated rivers.

The mean of ϕ_{ST} for pairwise comparisons within the region of Gaspé and the North Shore were 0.05 and 0.1, respectively. Within the Gaspé region, no significant difference was detected

Fig. 2. Population phenogram (neighbor-joining method) constructed from Shriver's genetic distance summarizing the genetic relationships among the seven rivers. Bootstrap estimates (percent of 1000 replicates) are given along branches.



between the two neighboring rivers, the Cascapedia and Bonaventure. The most geographicaly isolated river in the two regions, the Matane and Natashquan, respectively, had greater genetic differences (ϕ_{ST}) from the two neighboring rivers of their groups than existed between neighboring rivers themselves. The Natashquan and Matane rivers also showed an important overall genetic divergence with the other rivers. However, the coupling of genetic distance and geographic location seen within regions does not hold true when all rivers and regions are considered together. This is clearly demonstrated with the neighbor-joining population phenogram (Fig. 2). The relative position of the rivers in the tree does not correspond with their geographical proximity, This is especially true for the Koksoak, the most distant river, which was genetically closer than the Natashquan River to the other rivers. However, the Koksoak, Natashquan, and Matane rivers were the most genetically distant from all other rivers. The genetic distance between the Trinité and Godbout rivers was greater than that between Cascapedia and Bonaventure rivers. However, the genetic distance of the Godbout River from the Bonaventure and Cascapedia rivers was lower than between the Godbout and Trinité rivers.

The effective numbers of migrants per generation (N_em) were quite variable depending on the pairwise comparisons (Table 3). The Koksoak and the Natashquan rivers had the lowest level of gene flow with other rivers. The Cascapedia and Bonaventure had the highest score with a N_em value of 59.

Discussion

Our results do not support the hypothesis that the three metapopulations defined by Power (1981) are genetically differentiated. Genetic variance among groups of stocks was low, and allelic heterogeneity between all pairwise populations were mostly significant whatever the group subdivision. The dendrogram confirmed the absence of three metapopulations. The χ^2 analysis revealed significant allelic-frequency heterogeneity between the majority of the rivers whatever their locations. However, the high level of genetic variance observed between the Natashquan and the Cascapedia, Bonaventure, and Godbout rivers may reflect postulated differences in the migration route of salmon entering the Gulf of St. Lawrence. Based on tagging and recapture studies, Belding (1940) postulated that two major stocks of salmon enter the Gulf, one via the Strait of Belle Isle to the north and the other through Cabot Strait to the south (Fig. 1). Fish recaptured in rivers along the North Shore of the Gulf of St. Lawrence east of the Natashquan were tagged to the north of the Strait of Belle Isle, whereas recaptures to the west of the Natashquan were tagged in the vicinity of Cabot Strait. The genetic distinctiveness of the Natashquan population lends some credence to the Belding (1940) hypothesis, but other rivers east of the Natashquan must be analyzed to test it properly. The absence of the three metapopulations described by Power (1981) does not exclude the possibility of such structures occurring elsewhere in North America. Regions such as Newfoundland, Labrador, New Brunswick, Nova Scotia, and New England must be compared before concluding that North America encompasses one or more regional genetic groups (metapopulations).

Even though significant genetic differences were observed among all rivers, the genetic variance or distance among populations of Atlantic salmon was not as great as would be expected based on the apparent homing capabilities of salmon. This low genetic variance may be due to several factors, such as recent colonization, life history of the species, straying, stocking practices, and effective population size. All of these phenomena influence the forces of genetic drift, mutation, and natural selection, which determine the genetic differentiation of populations.

The recent time of colonization, 7000 to 13 000 years ago

(Fulton and Andrews 1987), reduces the contribution of drift to the genetic divergence among populations. This is particularly true in thU2case of the Koksoak River where the genetic distances and variances of the Koksoak from the other rivers were not as great as expected because of its geographical distance and its particular life-history strategy (Power and Power 1987; Robitaille et al. 1986). The time of founding of the Koksoak, about 7000 years ago (Fulton and Andrews 1987), may not be sufficient for genetic divergence to have occurred. The Koksoak harbors a large population and has greater periods of time between generations that act against genetic drift. These characteristics, along with relatively recent population foundation, may have acted to slow genetic differentiation. However, the small sample from the Koksoak River may influence allelic frequencies of this river, and thus, these interpretations may best be considered as preliminary.

The life strategy of the Atlantic salmon is well adapted to counter the problem of a bottleneck, thus reducing stochastic fluctuation of alleles composing a population (genetic drift) (Adkison 1995). The general life history of the Atlantic salmon is complex and can change in relation to environmental conditions. Members of a cohort may grow at different rates, smoltify at different ages, and return to the river after their 2nd to 4th year of sea life. Some of the male parr will develop functional testes (paedogenesis) and will participate in reproduction 2–5 years before anadromous male conspecifics. These precocious parr can also smoltify and thus spawn a second time. Adults may survive after the reproduction and spawn again (multispawners). Individuals from the same cohort can potentially propagate their genome during a period of 6 years. In a variable environment, ecological catastrophe may create a bottleneck or a founder effect. This flexibility, coupled with the recent time of colonization, may be responsible for the relatively low overall genetic variance observed here (7.5%).

Stocking with non-native genetic strains and straying among rivers are major phenomena working against genetic divergence. Historical data are available on stocking from 1900 to the present (Anonymous 1901–1914; Anonymous 1915–1922; Anonymous 1922–1934; Anonymous 1935–1956). Since 1900, two government hatcheries were responsible for stocking salmon parr in the province of Quebec, one situated at Tadoussac and the other at Gaspé (Fig. 1). The Tadoussac hatchery possessed its own pond for retaining reproductive adults. Adult salmon caught locally served as breeding stock. The production of this hatchery was mainly distributed in the salmon rivers of the Saguenay region and the North Shore. However, the Godbout and Natashquan rivers were not stocked. (The Trinité River was stocked from another stock; see below.) The Gaspé hatchery received their eggs for 20 years (1900–1920) from New Brunswick (St. John pound and the Miramichi River). During these years, these New Brunswick fish were used to stock mainly three rivers of the bay of Gaspé (Dartmouth, York, and St.-Jean). Around 500 000 fry per year were stocked in each river. The viability of the fry or parr planted in these rivers is unknown, but there is no reason to think that none of them survived and successfully returned to the river to propagate its genome in the population. After years of potential genetic admixture with a nonlocal stock in the Bay of Gaspé rivers, a pond was built for the Gaspé hatchery, which was supplied with eggs from adults caught directly in the bay. These genetically mixed populations of the bay were used to stock the majority of the salmon rivers of the Gaspé peninsula, including the Bonaventure, Cascapedia, and Matane. The Trinité River was stocked from this mixed stock from 1955 to 1958 and in 1960 with a total of 27 000 smolts.

An extreme example of random founder effects generated by stocking is provided by the Matane River. This river's population was totally rebuilt beginning in 1930 from the Gaspé stock following extirpation of the local stock. This river produced large numbers of salmon after 8 to 9 years of stocking (Anonymous 1935–1956). The absence of competition with native conspecifics may have increased the success of this stocking. The greater genetic distances and variances with the other rivers may be explained by a largely stochastic composition of allele frequencies.

Homing assures that the majority of a stock returns to its natal stream for spawning thereby maintaining and strengthening a more or less discrete genetic pool, which diverges from other stocks as a result of relative isolation (Saunders and Bailey 1978). The accuracy of homing in Atlantic salmon seems to be quite variable depending of the origin of the fish. Stabell (1984), in a review on homing in salmonids, showed that straying rate of native fishes varies normally from 0 to 3.1%. He also summarized data of recovered stocked hatchery fishes. From 10 studies, the straying rate was between 0 and 41.7%. Transplanted fish showed higher straying rates than native fishes, and their return success was lower. Thus, it is plausible that fish transplanted on the Gaspé Peninsula strayed to the other coast of the St. Lawrence river. A greater proportion of straying from the Gaspé populations to the Trinité and Godbout rivers or simply the stocking of the Trinité by the Gaspé stock could explain why their genetic variances and distances were lower relative to the Bonaventure and the Cascapedia rivers than between the latter two rivers. The knowledge of these historical perturbations of the native stocks may be very important knowing that few effective migrants can reduce genetic divergence, particularly among small populations (Stahl 1986; Adkison 1995). The North Shore populations were less stocked than the Gaspé Peninsula stocks. This may explain why neighboring populations such as the Godbout and Trinité rivers have conserved a greater genetic distance and genetic variance than heavily stocked populations, such as the Bonaventure and Cascapedia rivers.

Other studies have used microsatellites to discriminate Atlantic salmon populations. As with protein and mtDNA, clear evidence of discrimination was demonstrated between Canadian and European fish populations (McConnell et al. 1995*a*, 1995*b*). These authors also found significant differences in allele frequencies and (or) genetic variances (χ^2 and G_{ST} analysis respectively) for five populations of Nova Scotia, Canada. The G_{ST} for loci was 0.11 and was comparable with ϕ_{ST} values reported in the present study and F_{ST} reported in others (McConnell et al. 1995*a*, 1995*b*). Sympatric landlocked Atlantic salmon populations of the Saguenay region have highly significant heterogeneity and variances (mean ϕ_{ST} 0.25; Tessier and Bernatchez 1996). This level of genetic variance is similar to values observed among allopatric populations in other freshwater species (Ward et al. 1994). However, the lack of historical stocking data in the areas of these studies complicates the interpretation of the level of genetic variances.

Nevertheless, it is clear that significant allelic heterogeneity (χ^2) between populations is easily obtained with microsatellites even if, as in the present study, heavy stocking occurred.

Molecular genetic data obtained from microsatellites may discriminate the smallest detectable cohesive population even though these genetic divergences are poor indicators of heritable phenotypic adaptation because they are most likely based on neutral polymorphism. As such, we assume such divergence results from reproductive isolation and random drift of allelic frequencies among populations. The presence of significant genetic divergence may, however, be the result of isolation mechanisms related to local adaptation that increases the fitness of native populations and counters the homogenizing effect of gene flow (Taylor 1991). The overlapping generations in Atlantic salmon may help local adaptation (Adkinson 1995). For example, Riddell et al. (1981) demonstrated an adaptive variation in body morphology of juvenile Atlantic salmons for two tributary populations of the Miramichi River. Bielak (1984) found apparently adaptive morphological differences for adult Atlantic salmon between the East and the West branch of the Moisie River.

For the moment, the lack of knowledge about population genetic and phenotypic characters for the majority of salmon rivers imposes the rule of stocking with local stock if only to maintain the remaining intraspecific genetic diversity. Even with low genetic variability among populations, the overall significant allelic heterogeneity among rivers strongly suggests that each population, whether separated by thousands or tens of kilometres, should be considered as an individual management unit (sensu Moritz 1994).

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Appendix. Allele frequencies of the different loci for the seven riverine populations.

	Population						
Size (bp)	Cas	Bon	Mat	Tri	God	Nat	Kok
			Ssa171				
217	0.02	0.02	—	—	0.02	—	—
221	0.07	0.02	0.02	0.05	0.04	—	—
223	—	—	—	0.05	0.05	—	—
225	—	0.02	0.06	0.05	—	—	—
227	—	—	—	0.04	—	0.02	—
229	—	0.04	0.04	0.07	—	—	—
231	—	0.04	0.04	0.02	0.02	0.05	—
235	0.36	0.47	0.15	0.07	0.15	0.14	0.04
237	0.03	0.07	0.10	0.11	0.17	—	0.07
					8		
239	0.12	0.07	0.15	0.02	0.18	0.07	0.11
241	0.05	0.06	0.06	0.04	0.04	—	0.11
243	0.03	0.06	0.08	—	0.07	0.18	0.20
245	0.02	—	—	0.05	0.05	—	0.11
247	0.07	—	0.04	0.09	0.02	0.22	0.07
249	0.02	—	0.08	—	0.02	0.05	0.07
251	0.03	0.07	0.02	0.04	—	0.10	0.14
253	0.09	—	0.04	0.09	—	0.02	0.04
255	0.05	—	—	0.04	0.04	0.02	—
257	0.02	—	0.04	0.04	0.04	—	—
259	—	0.02	0.02	0.07	—	—	—
261	—	—	0.02	0.02	—	—	0.04
263	—	—	0.04	0.02	0.02	—	—
265	0.03	—	—	—	0.02	0.02	—
273	—	—	—	—	—	0.02	—
275	—	—	—	—	—	0.02	—
277	—	—	—	—	—	0.05	—
279	—	0.02	—	—	0.02	—	—
283	—	0.02	—	—	—	—	—
No. of allelles	15	14	17	20	18	15	11
No. of fish	30	27	26	28	27	21	14
			Ssa197				
144	—	—	—	—	0.05	—	—
152	—	0.02	—	—	0.02	—	—
156	—	0.02	—	—	0.02	—	—
160	—	0.02	—	0.02	0.03	—	0.06
164	0.24	0.14	0.05	0.07	0.14	0.02	0.03
168	0.16	0.14	0.13	0.14	0.14	0.21	0.23
172	0.16	0.21	0.12	0.18	0.12	0.14	0.09
176	0.14	0.14	0.16	0.20	0.17	0.29	0.32
180	0.05	0.07	0.26	0.02	0.03	0.05	0.15
184	0.05	0.09	0.15	0.09	0.02	0.10	0.03
188	0.05	0.04	0.03	0.07	0.09	—	—
192	—	0.05	0.02	0.05	—	—	—
196	—	0.02	0.02	—	0.02	0.02	—
200	0.02	—	0.02	0.05	0.07	0.02	—
204	0.02	—	—	0.07	0.02	0.05	—
208	0.05	—	—	0.02	0.03	0.10	—
212	0.02	—	0.02	—	—	—	0.06
216	—	—	—	—	0.03	—	0.03
220	0.02	0.02	—	0.02	—	—	—
224	—	0.02	0.02	—	—	—	—
No. of allelles	13	14	12	13	16	10	9
No. of fish	28	28	30	28	29	21	17

Appendix (*concluded*).

	Population						
Size (bp)	Cas	Bon	Mat	Tri	God	Nat	Kok
			Ssa202				
240	—	—	—	—	—	—	0.08
248	—	0.02	0.02	—	—	—	—
252	0.09	—	0.02	0.06	0.14	0.03	0.04
256	—	0.04	0.03	—	0.05	0.05	0.04
260	—	0.07	0.03	—	0.02	—	0.04
268	—	—	—	—	0.02	—	—
264	0.03	0.05	—	—	0.02	—	—
272	—	—	0.02	—	0.02	—	0.04
276	—	0.02	0.02	—	—	—	0.08
280	0.02	0.02	0.02	0.04	—	—	0.15
284	—	0.02	0.03	0.04	—	—	0.15
288	—	0.04	0.05	0.02	—	0.03	—
292	0.05	0.07	018	0.04	0.04	0.15	0.04
296	0.15	0.07	0.23	0.22	0.16	0.17	—
300	0.21	0.07	0.12	0.17	0.21	0.15	0.04
304	0.26	0.11	0.13	0.06	0.16	0.12	0.22
308	0.14	0.20	0.08	0.27	0.11	0.17	0.08
312	0.02	0.05	—	—	0.05	0.10	—
316	—	0.16	0.02	0.06	0.02	—	—
320	—	—	—	0.02	—	0.03	—
No. of allelles	11	15	15	11	12	10	12
No. of fish	28	28	30	24	28	20	13
			SSOSL85				
182	0.20	0.09	0.07	0.15	0.19	0.10	0.17
184	0.35	0.28	0.18	0.19	0.40	—	0.15
186	0.04	0.11	0.18	0.11	0.02	0.07	—
188	0.04	0.07	0.05	0.08	0.04	0.05	0.23
190	—	0.11	0.12	0.06	0.02	0.02	0.06
192	—	0.02	0.03	—	0.07	—	—
194	0.05	0.07	0.07	0.04	—	0.10	0.12
196	0.02	0.15	0.12	0.11	0.07	0.02	0.21
198	0.05	—	0.07	0.10	0.19	0.48	0.03
200	0.25	0.06	0.10	0.08	—	0.14	0.03
206	—	—	—	—	—	0.02	—
No. of allelles	8	10	11	10	8	9	8
No. of fish	28	27	30	26	27	21	17
			μ3				
172	—	—	—	—	0.11	—	—
196	—	0.04	—	—	—	—	—
200	0.19	0.23	0.02	0.16	0.13	—	—
202	0.23	0.04	0.15	0.02	0.11	0.09	0.18
204	0.42	0.42	0.04	0.45	0.39	0.07	0.18
206	0.10	0.18	0.48	0.32	0.26	0.29	0.64
208	0.02	0.05	0.21	0.05	—	0.43	—
212	—	—	0.04	—	—	—	—
214	—	—	—	—	—	0.05	—
No. of allelles	8	7	7	5	5	6	3
No. of fish	26	28	29	28	27	21	14

[a]Cas, Cascapedia River; Bon, Bonaventure River; Mat, Matane River; Tri, Trinité River; God, Godbout River; Nat, Natashquan River; Kok, Koksoak River.