### Supplementary Methods and results for

# Combining population genomics and forward simulations to investigate stocking impacts: A case study of Muskellunge (*Esox masquinongy*) from the St. Lawrence River basin

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# Fig S1: A simplified representation of the stocking activity in the St. Lawrence water from 1950 until the end of stocking in 1997.

The varying intensity of the arrows depicts the intensity of stocking from the different source in each time periods. The dotted arrow from Pigeon lake reflects an unknown quantity of fishes introduced in the 1950-1965 time period.

The Table below extracted from De Lafontaine (unpublished results) displays the number of fish used for stocking in waters of the St Lawrence. Values in italic present detailed information for each sectors within the St.Lawrence River



Location	Larvae	Frv	Adults	Total
	(< 5 cm)	(5-27 <sup>°</sup> cm)	(> 27 cm)	
St.Lawrence River (SLR):	441,800	593,173	135	1,035,108
Lake St.Francois		77,494	2	77,49
Beauharnois Canal & Soulanges Canal	30,000	11,250		41,250
Lake St.Louis	271,800	168,980	133	440,913
Lake Deux-Montagnes	20,000	81,692		101,692
Des Mille-Isles / Des Prairies Rivers		38,297		38,297
River stretch (downstream Montreal)	120,000	208,160		328,160
Lake St.Pierre		7,300		7,300
Inland lakes & rivers – North of SLR	300,000	74,880		329,880
Inland lakes & rivers – South of SLR		17,825	2	17,827
Tributaries of SLR	112,000	88,908	60	200,968
All	853,800	729,786	197	1,583,108

## Fig S2: Isolation by Distance within the St. Lawrence



## Figure S3: (a) admixture Cross-validation plot obtained for the 643 individuals and different K values. (b) admixture plots for different K values.





b)

a)

Figure S4: Simulated models of divergence with admixture.



1 2	popA CHD CHP	popB ACH ACH	<b>sxA_avg</b> 0.205 0.198	<b>sxB_avg</b> 0.286 0.284	<b>ss_avg</b> 0.751 0.774	<b>piA_avg</b> 0.0032014 0.0031345	<b>piB_avg</b> 0.0030203 0.0030831	divAB_avg 0.0038205 0.0037389	netdivAB_avg 0.0007097 0.0006301	<b>FST_avg</b> 0.0385870 0.0593632
2 3 4	CHP CHQ	CHD ACH	0.215 0.283	0.216	0.814 0.747	0.0033180 0.0028121	0.0034589 0.0028858	0.0038798 0.0036830	0.0004914	0.0144615 0.0563876
5	CHQ	CHD	0.278	0.152	0.838	0.0030421	0.0033182	0.0032695	0.0000893	-0.0733390
6	CHQ	CHP	0.289	0.168	0.828	0.0030493	0.0031962	0.0036785	0.0005558	0.0507369
7	FRO	ACH	0.236	0.298	0.736	0.0028773	0.0030074	0.0038473	0.0009049	0.0758679
8 9 10	FRO FRO	CHD CHP	0.221 0.229 0.148	0.197 0.201 0.252	0.843 0.853 0.895	0.0031494 0.0031961	0.0034913 0.0033988 0.0031284	0.0034753 0.0038944 0.0032449	0.0001550 0.0005970	-0.0492876 0.0557195
10 11 12	JOS	ACH CHD	0.254 0.263	0.232 0.234 0.163	0.773 0.828	0.0028505 0.0030355	0.0029349 0.0033192	0.0037352 0.0033171	0.0001401 0.0008425 0.0001398	0.0739751 -0.0256134
13 14 15	JOS JOS	CHP CHQ FRO	0.272 0.181 0.228	0.171 0.203 0.140	0.828 0.892 0.909	0.0030589 0.0029813 0.0031623	0.0032047 0.0029800 0.0031127	0.0036815 0.0030750 0.0031743	0.0005497 0.0000944 0.0000368	0.0564989 0.0015817 0.0027899
16	LDM	ACH	0.377	0.082	0.820	0.0030772	0.0026210	0.0031683	0.0003192	0.0715963
17	LDM	CHD	0.438	0.080	0.756	0.0030769	0.0027878	0.0032734	0.0003410	0.0104840
18	LDM	CHP	0.449	0.084	0.749	0.0030705	0.0026623	0.0032138	0.0003474	0.0686423
19 20	LDM LDM	CHQ FRO	0.363	0.116	0.805	0.0030115	0.0024964 0.0024765	0.0031726	0.0004187	0.0619425 0.0780516
21 22 23	LDM LSF LSF	ACH CHD	0.362 0.369 0.453	0.092 0.122 0.144	0.825 0.801 0.683	0.0030576 0.0030488 0.0029540	0.0025319 0.0026764 0.0027575	0.0032187 0.0032729 0.0033641	0.0004240 0.0004103 0.0005084	0.0739248 0.0502516 -0.0337684
24	LSF	CHP	0.447	0.139	0.697	0.0029891	0.0026809	0.0032986	0.0004637	0.0435096
25	LSF	CHQ	0.375	0.175	0.738	0.0028943	0.0024718	0.0032870	0.0006039	0.0752434
26	LSF	FRO	0.457	0.167	0.668	0.0029293	0.0024592	0.0033726	0.0006784	0.0804195
27 28 29	LSF LSF	JOS LDM	0.385 0.162 0.384	0.164 0.208	0.737 0.928 0.820	0.0029221 0.0028330 0.0030694	0.0024924 0.0029214 0.0025979	0.0033140 0.0029724 0.0031718	0.0006067 0.0000952 0.0003381	0.0716988 0.0094015 0.0787662
30 31	LSL	CHD CHP	0.447 0.457	0.078 0.080	0.744 0.747	0.0030409 0.0030573	0.0027388 0.0026379	0.0032828 0.0032174	0.0003929	0.0235034 0.0767192
32	LSL	CHQ	0.397	0.137	0.756	0.0029403	0.0024210	0.0031847	0.0005041	0.0704910
33	LSL	FRO	0.454	0.101	0.726	0.0030126	0.0024393	0.0032709	0.0005450	0.0875128
34	LSL	JOS	0.397	0.114	0.777	0.0029804	0.0024570	0.0032254	0.0005067	0.0792839
35	LSL	LDM	0.172	0.152	0.976	0.0029138	0.0028941	0.0029635	0.0000596	0.0066376
36	LSL	LSF	0.212	0.137	0.949	0.0029542	0.0028438	0.0029270	0.0000280	-0.0007533
37	LSP	ACH	0.387	0.092	0.807	0.0030128	0.0026120	0.0031770	0.0003646	0.0686410
38 39 40	LSP LSP	CHD CHP	0.458 0.452 0.376	0.101 0.094	0.711 0.733	0.0029531	0.0027183 0.0026453	0.0032811 0.0032412	0.0004454 0.0004206	-0.0033252 0.0616424
41 42	LSP	FRO	0.467 0.380	0.126 0.114	0.700	0.0029269 0.0029455	0.0024295 0.0024838	0.0032812 0.0032536	0.0006030 0.0005390	0.0831387 0.0764743
43	LSP	LDM	0.160	0.155	0.979	0.0028656	0.0029154	0.0029548	0.0000644	-0.0076327
44	LSP	LSF	0.178	0.122	0.994	0.0029538	0.0029146	0.0029424	0.0000082	-0.0005377
45	LSP	LSL	0.147	0.160	0.997	0.0028731	0.0029423	0.0029257	0.0000180	0.0015614
46	MAS	ACH	0.232	0.227	0.794	0.0035021	0.0029801	0.0036938	0.0004527	-0.0037864
47	MAS	CHD	0.311	0.235	0.686	0.0034193	0.0030953	0.0038195	0.0005622	0.0540048
48	MAS	CHP	0.316	0.237	0.694	0.0034543	0.0029962	0.0037492	0.0005240	0.0105888
49	MAS	CHQ	0.278	0.308	0.677	0.0032554	0.0026907	0.0036574	0.0006843	-0.0910666
50	MAS	FRO	0.334	0.274	0.651	0.0033667	0.0027397	0.0037931	0.0007399	-0.0279082
51	MAS	JOS	0.274	0.284	0.697	0.0033083	0.0027291	0.0037068	0.0006881	-0.0188995
52 53 54	MAS MAS MAS	LDM LSF	0.085 0.056 0.048	0.372 0.308 0.362	0.818 0.913 0.855	0.0030758 0.0032992 0.0030840	0.0030679 0.0031972 0.0030989	0.0031963 0.0032664 0.0031077	0.0001245 0.0000182 0.0000163	-0.0304527 -0.0275856 0.0165756
55 56	MAS MAU	LSP ACH	0.060	0.355	0.861	0.0031338	0.0030722 0.0030268	0.0031346 0.0038421	0.0000316	-0.0050888 0.0733964
58 59	MAU	CHP CHQ	0.236 0.174	0.220 0.285	0.816	0.0033105 0.0031487	0.0033370 0.0030270	0.0039019 0.0033542	0.0005782	0.0555716 0.0229764
60 61 62	MAU MAU MAU	JOS LDM	0.185 0.160 0.104	0.201 0.263 0.443	0.895 0.853 0.724	0.0034114 0.0031995 0.0026143	0.0032352 0.0030814 0.0030370	0.0035591 0.0033488 0.0032493	0.0002358 0.0002084 0.0004237	0.0239821 0.0257194 0.0693781
63	MAU	LSF	0.157	0.457	0.680	0.0026261	0.0029601	0.0033957	0.0006027	0.0637479
64	MAU	LSL	0.094	0.452	0.734	0.0025900	0.0030238	0.0032872	0.0004803	0.0797389
65	MAU	LSP	0.118	0.460	0.707	0.0025843	0.0029466	0.0032982	0.0005327	0.0708431
66	MAU	MAS	0.262	0.327	0.666	0.0029212	0.0033993	0.0038307	0.0006704	0.0038742
67	MIL	ACH	0.235	0.226	0.798	0.0033860	0.0029771	0.0036553	0.0004738	0.0223787
68	MIL	CHD	0.308	0.224	0.711	0.0033429	0.0031212	0.0037963	0.0005642	0.0573003
69	MIL	CHP	0.311	0.227	0.714	0.0033671	0.0030151	0.0037214	0.0005303	0.0343833
70	MIL	CHQ	0.271	0.299	0.696	0.0031699	0.0027080	0.0036310	0.0006920	-0.0353158
71	MII	FRO	0.324	0.255	0.682	0.0032975	0.0027758	0.0037942	0.0007576	0.0133461
72 73	MIL	JOS LDM	0.264	0.274	0.717	0.0032242	0.0027580	0.0036952	0.0007041	0.0197237 -0.0013795
74 75 76	MIL MIL	LSF LSL	0.077 0.056 0.071	0.364 0.363	0.855 0.847	0.0029799 0.0030091	0.0030954 0.0030502	0.0030894 0.0030962	0.0000665	0.0293529 0.0117672
77	MIL	MAS	0.180	0.173	0.895	0.0035302	0.0036474	0.0036231	0.0000343	0.0013384
78	MIL	MAU	0.318	0.248	0.693	0.0033213	0.0029556	0.0038145	0.0006760	0.0339241
79	MSC	ACH	0.246	0.201	0.796	0.0034033	0.0029079	0.0035435	0.0003879	-0.0083523
80	MSC	CHD	0.314	0.201	0.716	0.0033676	0.0030650	0.0037014	0.0004851	0.0497782
81	MSC	CHP	0.327	0.203	0.716	0.0034022	0.0029579	0.0036239	0.0004439	0.0068896
82	MSC	CHQ	0.281	0.267	0.716	0.0032300	0.0026753	0.0035577	0.0006050	-0.0869112
83 84 85	MSC MSC	FRO JOS	0.339 0.267 0.083	0.236 0.244 0.330	0.682 0.739	0.0033245 0.0032845	0.0027180 0.0027287 0.0030775	0.0036754 0.0036172	0.0006542 0.0006106	-0.0271725 -0.0174102
86 87	MSC MSC	LSF	0.093	0.293	0.888	0.0031856	0.0030955	0.0031639	0.0000233	-0.0677176 0.0054298
88 89 90	MSC MSC MSC	MAS MAU	0.065 0.200 0.327	0.319 0.157 0.223	0.892 0.882 0.703	0.0031087 0.0035345 0.0033619	0.0030626 0.0035514 0.0029054	0.0031013 0.0035510 0.0037190	0.0000157 0.0000080 0.0005853	-0.0174391 0.0026766 0.0019818
91	MSC	MIL	0.198	0.163	0.882	0.0035193	0.0034199	0.0035095	0.0000399	0.0013146
92	MSG	ACH	0.267	0.163	0.822	0.0033884	0.0028737	0.0034985	0.0003674	0.0086798
93	MSG	CHD	0.338	0.160	0.741	0.0033551	0.0030209	0.0036101	0.0004221	0.0507400
94	MSG	CHP	0.343	0.169	0.744	0.0033843	0.0029252	0.0035695	0.0004148	0.0232618
95	MSG	CHQ	0.295	0.229	0.741	0.0032092	0.0026436	0.0034625	0.0005361	-0.0548301
96	MSG	FRO	0.360	0.204	0.703	0.0032986	0.0026767	0.0035659	0.0005782	-0.0032537
97 98	MSG MSG MSG	JOS LDM	0.289 0.094	0.202	0.768	0.0032791 0.0030696 0.0031368	0.0027010 0.0030566	0.0035308 0.0031312 0.0031093	0.0005407 0.0000682	0.0022561 -0.0232905
99 100 101	MSG MSG MSG	LSF LSL LSP	0.069	0.271 0.287 0.281	0.918 0.915	0.0031388 0.0030649 0.0030988	0.0030300 0.0030712 0.0030335	0.0030654 0.0030819	-0.0000238 -0.0000027 0.0000158	-0.0042031 -0.0011883 -0.0207967
102	MSG	MAS	0.242	0.134	0.868	0.0034541	0.0034359	0.0034556	0.0000105	0.0066570
103	MSG	MAU	0.351	0.179	0.723	0.0033358	0.0028537	0.0036159	0.0005212	0.0185452
104	MSG	MIL	0.247	0.147	0.858	0.0034321	0.0033060	0.0034205	0.0000514	0.0081986
105	MSG	MSC	0.197	0.143	0.899	0.0034087	0.0033895	0.0033921	-0.0000071	0.0010681
106	MSK	ACH	0.270	0.225	0.781	0.0032230	0.0029289	0.0036098	0.0005339	0.0518805
107	MSK	CHD	0.271	0.147	0.842	0.0034207	0.0033088	0.0036196	0.0002549	0.0163822
108	MSK	CHP	0.291	0.158	0.828	0.0034252	0.0031677	0.0037412	0.0004448	0.0458362
109	MSK	CHQ	0.230	0.217	0.844	0.0032773	0.0028949	0.0034177	0.0003316	0.0028973
110	MSK	FRO	0.256	0.144	0.883	0.0034942	0.0030407	0.0035859	0.0003185	0.0193919
111 112 112	MSK MSK	JOS LDM	0.219 0.128	0.190 0.370	0.871 0.793	0.0033301 0.0028062	0.0029429 0.0029922	0.0034244 0.0031498	0.0002879 0.0002507	0.0230240 0.0240020
113 114 115	MSK MSK	LSL	0.125	0.383	0.785	0.0027660 0.0027635	0.0029676	0.0031517 0.0031534	0.0002850	0.0342210 0.0195307
116	MSK	MAS	0.291	0.254	0.714	0.0030847	0.0032994	0.0035921	0.0004000	0.0115440
117	MSK	MAU	0.275	0.153	0.857	0.0034626	0.0031809	0.0036626	0.0003408	0.0324395
118	MSK	MIL	0.286	0.248	0.732	0.0031073	0.0032130	0.0035805	0.0004204	0.0328650
119	MSK	MSC	0.257	0.259	0.739	0.0030512	0.0032478	0.0034873	0.0003378	0.0084812
120	MSK	MSG	0.216	0.277	0.774	0.0030343	0.0032546	0.0034346	0.0002901	0.0170921
121	MSV	ACH	0.252	0.183	0.816	0.0035035	0.0029160	0.0036237	0.0004139	-0.0098118
122	MSV	CHD	0.324	0.179	0.730	0.0034622	0.0030534	0.0037307	0.0004729	0.0494927
123	MSV	CHP	0.333	0.183	0.733	0.0034933	0.0029529	0.0036728	0.0004497	0.0045771
124	MSV	CHQ	0.284	0.264	0.718	0.0032888	0.0026644	0.0035678	0.0005911	-0.0927010
125 126 127	MSV MSV MSV	FRO JOS	0.343 0.284 0.093	0.222 0.238 0.325	0.700 0.740 0.867	0.0034098 0.0033522 0.0031365	0.0027146 0.0027074 0.0030600	0.0037001 0.0036251 0.0031944	0.0006378 0.0005953 0.0000962	-0.0325348 -0.0230919 -0.0453730
128 129	MSV MSV	LSF	0.103	0.280	0.899	0.0031505	0.0030913 0.0030985	0.0032053 0.0031280	0.0000222 0.0000023	-0.0800394 -0.0053232
130 131 132	MSV MSV MSV	MAS MAU	0.077 0.210 0.339	0.307 0.141 0.205	0.894 0.893 0.713	0.0031722 0.0036146 0.0034447	0.0030419 0.0035348 0.0028926	0.0031260 0.0035796 0.0037465	0.0000189 0.0000049 0.0005778	-0.0335352 0.0029493 -0.0015552
133	MSV	MIL	0.215	0.147	0.881	0.0035920	0.0033950	0.0035430	0.0000494	0.0011880
134	MSV	MSC	0.177	0.153	0.908	0.0035596	0.0034678	0.0035087	-0.0000050	0.0004545
135	MSV	MSG	0.159	0.191	0.895	0.0034595	0.0033874	0.0034201	-0.0000033	-0.0001268
136	MSV	MSK	0.267	0.249	0.748	0.0033276	0.0030469	0.0035344	0.0003472	0.0071730
137	ONT	ACH	0.181	0.501	0.585	0.0019118	0.0031683	0.0039874	0.0014474	0.1158014
138	ONT	CHD	0.201	0.456	0.595	0.0020056	0.0035256	0.0041565	0.0013909	0.0160322
139	ONT	CHP	0.220	0.472	0.572	0.0019874	0.0033549	0.0042612	0.0015901	0.1188597
140	ONT	CHQ	0.176	0.531	0.572	0.0018658	0.0030039	0.0040302	0.0015954	0.1648240
141	ONT	FRO	0.188	0.468	0.618	0.0020241	0.0032131	0.0043537	0.0017351	0.1594679
142 143 144	ONT ONT	JOS LDM	0.173 0.104 0.110	0.511 0.660	0.588 0.528	0.0019077 0.0015679	0.0030706 0.0030475	0.0040661 0.0032845	0.0015769 0.0009768	0.1451907 0.1722403 0.1755705
145 146	ONT ONT	LSL	0.089	0.658	0.536	0.0015543 0.0015638	0.0030380	0.0032619	0.0009658	0.1817642 0.1758420
147	ONT	MAS	0.185	0.510	0.552	0.0018565	0.0036347	0.0039710	0.0012255	-0.0235603
148	ONT	MAU	0.195	0.459	0.621	0.0020403	0.0034090	0.0042767	0.0015521	0.1319314
149	ONT	MIL	0.175	0.506	0.585	0.0018975	0.0035714	0.0039542	0.0012197	0.0293597
150	ONT	MSC	0.173	0.532	0.541	0.0017992	0.0034972	0.0037861	0.0011379	-0.0187260
151	ONT	MSG	0.156	0.556	0.543	0.0017470	0.0034102	0.0036747	0.0010961	0.0263817
152	ONT	MSK	0.159	0.520	0.606	0.0018945	0.0034497	0.0039241	0.0012520	0.1038830
153	ONT	MSV	0.166	0.539	0.556	0.0018019	0.0035813	0.0038445	0.0011529	-0.0210538
154	TRA	ACH	0.169	0.409	0.690	0.0016868	0.0031948	0.0038071	0.0013664	0.0615567
155	TRA	CHD	0.216	0.381	0.659	0.0017255	0.0034733	0.0042162	0.0016168	-0.0724945
156	TRA	CHP	0.218	0.369	0.686	0.0017696	0.0034118	0.0041634	0.0015728	0.0650849
157	TRA	CHQ	0.152	0.436	0.688	0.0016582	0.0030556	0.0040456	0.0016887	0.1628309
158	TRA	FRO	0.212	0.399	0.678	0.0017563	0.0031856	0.0043245	0.0018535	0.1400106
159	TRA	JOS	0.159	0.420	0.692	0.0016729	0.0030865	0.0040783	0.0016986	0.1178408
160	TRA	LDM	0.066	0.564	0.653	0.0014059	0.0031252	0.0032673	0.0010017	0.1734010
161	TRA	LSF	0.090	0.549	0.642	0.0014402	0.0031112	0.0033746	0.0010989	0.1766408
162 163 164	TRA TRA	LSL LSP MAS	0.079 0.069 0.198	0.585 0.559 0.433	0.623 0.648 0.619	0.0013731 0.0014034 0.0016088	0.0030728 0.0030585 0.0035912	0.0032412 0.0032545 0.0038682	0.0010182 0.0010236 0.0012682	0.1709042 0.1736674 -0.1216717
165 166	TRA TRA	MAU	0.212 0.186	0.386	0.684	0.0017564 0.0016265	0.0033601 0.0035138	0.0042796	0.0017214	0.1014230 -0.0447828
. 37 168 169	TRA TRA	MSG MSK	0.151	0.477 0.440	0.632 0.683	0.0015394 0.0016482	0.0034504 0.0034390	0.0036216	0.0011267	-0.0639497 0.0406647
170 171 172	TRA TRA TRE	ONT ACH	0.165 0.392 0.217	0.466 0.291 0.333	0.632 0.571 0.711	0.0015729 0.0018908 0.0028569	0.0035921 0.0021664 0.0030491	0.0037725 0.0045289 0.0039097	0.0011899 0.0025003 0.0009567	-0.1241551 0.1912753 0.0806607
173	TRE	CHD	0.202	0.248	0.812	0.0031242	0.0035497	0.0035055	0.0001685	-0.0552367
174	TRE	CHP	0.195	0.238	0.834	0.0032015	0.0034841	0.0039974	0.0006546	0.0570675
175	TRE	CHQ	0.125	0.299	0.869	0.0030653	0.0031867	0.0032747	0.0001487	0.0280690
176	TRE	FRO	0.127	0.202	0.967	0.0033726	0.0034563	0.0035186	0.0001042	0.0128397
177	TRE	JOS	0.130	0.284	0.866	0.0030718	0.0031959	0.0032112	0.0000773	0.0108797
178	TRE	LDM	0.100	0.491	0.693	0.0024329	0.0030549	0.0032594	0.0005155	0.0900744
179	TRE	LSF	0.157	0.496	0.639	0.0024243	0.0029527	0.0034000	0.0007115	0.0907540
180	TRE	LSL	0.088	0.503	0.693	0.0024003	0.0030372	0.0032981	0.0005794	0.1003403
181	TRE	LSP	0.116	0.503	0.666	0.0023983	0.0029579	0.0033167	0.0006386	0.0939044
182 183 184	TRE TRE TRE	MAS MAU MII	0.252 0.175 0.249	0.369 0.238 0.368	0.631 0.864	0.0027134 0.0032136	0.0034197 0.0034786 0.0033380	0.0038492 0.0036205 0.0038512	0.0007826 0.0002744 0.0008077	-0.0209169 0.0267870
184 185 186	TRE	MSC MSG	0.249 0.221 0.183	0.388 0.380 0.395	0.646 0.651 0.675	0.0027490 0.0026959 0.0026498	0.0033380 0.0033719 0.0033401	0.0038512 0.0037333 0.0036175	0.0006994	-0.0206856 0.0052278
187 188 189	I RE TRE TRE	MSV ONT	0.135 0.205 0.447	0.312 0.387 0.232	0.838 0.661 0.585	0.0029917 0.0026797 0.0031774	0.0035230 0.0034510 0.0020511	0.0036306 0.0037497 0.0044691	0.0003732 0.0006844 0.0018548	0.0259004 -0.0253675 0.1618313
190	TRE	IRA	0.372	0.252	0.660	0.0031857	0.0017992	0.0044388	0.0019464	0.1402268
191	YAM	ACH	0.169	0.317	0.770	0.0031138	0.0031670	0.0037747	0.0006344	0.0541483
192	YAM	CHD	0.206	0.274	0.754	0.0031968	0.0034555	0.0041369	0.0008107	0.0695933
193	YAM	CHP	0.199	0.278	0.781	0.0032602	0.0033879	0.0040942	0.0007701	0.0652183
194	YAM	CHQ	0.173	0.356	0.741	0.0030286	0.0029935	0.0039307	0.0009196	0.0464905
195	YAM	FRO	0.216	0.301	0.755	0.0032153	0.0031305	0.0041586	0.0009857	0.0701960
196	YAM	JOS	0.176	0.339	0.749	0.0030568	0.0030176	0.0039519	0.0009147	0.0737255
197	YAM	LDM	0.068	0.492	0.717	0.0025963	0.0031043	0.0032756	0.0004254	0.0681979
198	YAM	LSF	0.113	0.487	0.681	0.0026269	0.0030429	0.0033505	0.0005156	0.0202252
199	YAM	LSL	0.058	0.495	0.718	0.0025719	0.0030932	0.0032635	0.0004310	0.0821366
200	YAM	LSP	0.082	0.499	0.697	0.0025747	0.0030175	0.0032706	0.0004744	0.0546913
201	YAM	MAS	0.199	0.345	0.696	0.0029727	0.0035552	0.0038380	0.0005725	0.0498274
201 202 203	YAM YAM	MAU MAU MIL	0.198 0.211 0.182	0.345 0.298 0.339	0.753 0.721	0.0029737 0.0032022 0.0029962	0.0032963 0.0034726	0.0038374	0.0009187 0.0006030	0.0748995 0.0628365
204	YAM	MSC	0.157	0.351	0.722	0.0029295	0.0035012	0.0037346	0.0005192	0.0489057
205	YAM	MSG	0.142	0.378	0.720	0.0028534	0.0034205	0.0036151	0.0004781	0.0590800
206	YAM	MSK	0.156	0.345	0.762	0.0030454	0.0034091	0.0038771	0.0006498	0.0746721
207	YAM	MSV	0.151	0.362	0.724	0.0029080	0.0035661	0.0037564	0.0005193	0.0497268
208	YAM	ONT	0.432	0.246	0.570	0.0033191	0.0020428	0.0043578	0.0016768	0.0640918
209	YAM	TRA	0.348	0.250	0.665	0.0033562	0.0018015	0.0042624	0.0016835	-0.0129744
210	YAM	TRE	0.254	0.275	0.736	0.0032888	0.0031284	0.0042786	0.0010700	0.0703449
211	OUT	ACH	0.144	0.388	0.705	0.0035745	0.0032027	0.0039546	0.0005660	-0.0623304
212	OUT	CHD	0.150	0.332	0.746	0.0037700	0.0036125	0.0040659	0.0003746	0.0195642
213 214 217	OUT OUT	CHP CHQ FPC	0.174 0.135 0.182	0.347 0.425 0.371	0.728 0.702 0.702	0.0037797 0.0035183 0.0037100	0.0034643 0.0030768 0.0032057	0.0041676 0.0037852 0.0040007	0.0005456 0.0004877 0.0005282	-0.0573576 -0.1765928 -0.1314545
216 217	OUT OUT	JOS	0.132	0.402	0.712	0.0035460	0.0031093 0.0031705	0.0033043	0.0004856	-0.0918631 -0.0163542
∠18	OUT	LSF	0.085	0.559	0.632	0.0030140	0.0030957	0.0034125	0.0003576	-0.1647621
219	OUT	LSL	0.039	0.569	0.655	0.0029218	0.0031187	0.0032864	0.0002662	-0.0132900
220	OUT	LSP	0.052	0.557	0.649	0.0029529	0.0030630	0.0033211	0.0003132	-0.0748890
221	OUT	MAS	0.153	0.401	0.671	0.0034641	0.0036618	0.0039643	0.0004013	0.0355293
222	OUT	MAU	0.173	0.363	0.724	0.0037742	0.0034298	0.0040634	0.0004614	-0.0842553
223	OUT	MIL	0.149	0.401	0.677	0.0034781	0.0035582	0.0039182	0.0004000	0.0170616
224	OUT	MSC	0.124	0.413	0.681	0.0033930	0.0035745	0.0038251	0.0003413	0.0392643
225	OUT	MSG	0.099	0.437	0.697	0.0033497	0.0035439	0.0037418	0.0002950	0.0349841
226	OUT	MSK	0.112	0.415	0.728	0.0035323	0.0035112	0.0038394	0.0003176	-0.0187652
227	OUT	MSV	0.118	0.431	0.682	0.0033609	0.0036519	0.0038425	0.0003361	0.0444350
228	OUT	ONT	0.399	0.312	0.535	0.0039337	0.0021328	0.0042946	0.0012613	-0.2859461
220	TRF	OUT	0.354	0.224	0.680	0.0031989	0.0038056	0.0041124	0.0006101	-0.1398444
230	TRA	OUT	0.318	0.307	0.626	0.0018586	0.0039192	0.0044268	0.0015378	-0.4189416
231	YAM	OUT	0.319	0.210	0.701	0.0033912	0.0038403	0.0043382		-0.0035782

# Table S2. summary statistics for the 80pb haplotype data obtainedfrom stacks.

sx= number of polymorphic sites private to each population ss= number of shared polymorphic sites pi= nucleotide diversity (Tajima, 1989) divAB = Dxy netDiv = divergence net Fst= between population differentiation

## Table S1: pairwise Weir & CockerhamFst values.

Non significant Fst values are in bold

	ACH	CHD	CHP	CHQ	FRO	JOS	LDM	LSF	LSL	LSP	LSW	MAU	TIN	MSC	MSG	MSK	MSV	PIG	отт	TRA	TRE
CHD	0.250																				
CHP	0.222	0.172																			
CHQ	0.290	0.039	0.198																		
FRO	0.309	0.073	0.207	0.056																	
JOS	0.291	0.063	0.195	0.047	0.013																
LDM	0.124	0.129	0.135	0.168	0.185	0.166															
LSF	0.156	0.193	0.178	0.235	0.252	0.233	0.044														
LSL	0.136	0.158	0.149	0.204	0.216	0.200	0.023	0.009													
LSP	0.144	0.172	0.162	0.216	0.231	0.212	0.030	0.004	0.004												
LSW	0.161	0.192	0.186	0.246	0.264	0.241	0.046	0.004	0.008	0.009											
MAU	0.272	0.082	0.198	0.112	0.100	0.089	0.162	0.225	0.190	0.204	0.229										
TIN	0.174	0.202	0.187	0.252	0.271	0.249	0.055	0.020	0.022	0.022	0.015	0.236									
MSC	0.152	0.175	0.167	0.226	0.243	0.222	0.030	0.006	0.001	0.002	0.004	0.211	0.018								
MSG	0.137	0.153	0.153	0.203	0.218	0.198	0.024	0.009	0.001	0.003	0.007	0.190	0.022	-0.002							
MSK	0.189	0.094	0.153	0.127	0.123	0.112	0.096	0.143	0.113	0.125	0.137	0.128	0.147	0.122	0.103						
MSV	0.152	0.167	0.163	0.219	0.235	0.215	0.034	0.007	0.004	0.003	0.007	0.205	0.022	0.001	0.001	0.121					
PIG	0.482	0.461	0.486	0.473	0.506	0.484	0.354	0.382	0.365	0.371	0.444	0.468	0.443	0.441	0.417	0.410	0.435				
отт	0.194	0.128	0.175	0.181	0.200	0.179	0.083	0.138	0.109	0.122	0.135	0.166	0.140	0.122	0.103	0.111	0.117	0.469			
TRA	0.499	0.551	0.518	0.521	0.555	0.538	0.377	0.397	0.388	0.389	0.487	0.534	0.492	0.479	0.453	0.435	0.477	0.709	0.571		
TRE	0.320	0.079	0.222	0.064	0.048	0.035	0.199	0.265	0.230	0.245	0.276	0.111	0.285	0.257	0.233	0.143	0.250	0.521	0.215	0.567	
YAM	0.232	0.260	0.246	0.295	0.312	0.295	0.164	0.203	0.179	0.191	0.210	0.288	0.222	0.205	0.187	0.216	0.200	0.520	0.240	0.574	0.330

#### Table S3 Description of the major models tested in this study.

Model 1 to 17 described the major model tested without mortality rate. These models were tested with varying population size of the 3 descendent population (N= 400/600/800/1600). Model 18 to 23 are the equivalent of model 9 to 11 with varying mortality rate (column Death rate) and with varying migration rate from LDM to SLR.SRC = Source population used for stocking. Given the shared patterns of ancestry between the population used for stocking (i.e. Chautauqua, Joseph and Tremblant lake) individuals were merged together and modelled as a single unit. Given that no sign of ancestry from Pigeon lake was find in the Saint Lawrence (SLR) or the Lac des Deux-Montagnes (LDM) it was not modelled here. Based on our observation of higher admixture in LDM than in SLR, the migration rate (m) was twice higher in LDM than in SLR.

	N (Size)		m SRC →	m SRC →	m SLR →	m LDM →
name		Death rate	SLR	LDM	LDM	SLR
model01	400/600/800/1600	0	0.000001	0.00002	0.0005	0.001
model02	400/600/800/1600	0	0.00001	0.00002	0.0005	0.001
model03	400/600/800/1600	0	0.0001	0.000015	0.0005	0.001
model04	400/600/800/1600	0	0.001	0.00015	0.0005	0.001
model05	400/600/800/1600	0	0.01	0.0015	0.0005	0.001
model06	400/600/800/1600	0	0.005	0.0075	0.0005	0.001
model07	400/600/800/1600	0	0.0033	0.005	0.0005	0.001
model08	400/600/800/1600	0	0.0025	0.00375	0.0005	0.001
model09	400/600/800/1600	0	0.005	0.0075	0.0003	0.00015
model*10	400/600/800/1600	0	0.0033	0.005	0.0003	0.00015
model/11	400/600/800/1600	0	0.0025	0.00375	0.0003	0.00015
model12	400/600/800/1600	0	0.1	0.15	0.0005	0.001
model13	400/600/800/1600	0	0.05	0.075	0.0005	0.001
model14	400/600/800/1600	0	0.025	0.0375	0.0005	0.001
model15	400/600/800/1600	0	0.15	0.225	0.0005	0.001
model <sup>*</sup> 16	400/600/800/1600	0	0.2	0.3	0.0005	0.001
model <sup>1</sup> 7	400/600/800/1600	0	0.015	0.0025	0.0005	0.001
model <sup>*</sup> 18	400/600/800/1600	50/100/200/400	0.005	0.0075	0.0005	0.001
model 19	400/600/800/1600	50/100/200/400	0.0033	0.005	0.0005	0.001
model20	400/600/800/1600	50/100/200/400	0.0025	0.00375	0.0005	0.001
model21	400/600/800/1600	50/100/200/400	0.005	0.0075	0.0003	0.00015
model22	400/600/800/1600	50/100/200/400	0.0033	0.005	0.0003	0.00015
model23	400/600/800/1600	50/100/200/400	0.0025	0.00375	0.0003	0.00015

#### **Supplementary Methods and Results**

#### 1. Measurement of the symmetry of migration using coalescent simulations and ABC

#### Methods

Migration rate (*m*) was measured using coalescent simulations combined with an ABC procedure for parameter estimation. The objective was to determine if sample sites from the St. Lawrence that displayed weak level of genetic differentiation (e.g. Fst < 0.01) were connected by gene flow or if this was due to populations with large Ne evolving independently (i.e. with very low gene flow).

#### a) model and coalescent simulations

The demographic history consists in a set of two subpopulations exchanging migrants in a stepping stone model. Each subpopulation is of size  $N_{upstream}$  and  $N_{downstream}$  and migrants are exchanged at a rate  $M_{ij} = 4N_0m_{ij}$  at each generation. Here  $m_{ij}$  is the fraction of the subpopulation *i* made of migrants of each generation from sub-population *j*. Migration is modelled randomly and uniformly for each of the upstream and downstream sub-population respectively. Prior for effective population size were uniformly distributed on the interval [0 - 500,000] and sampled independently for each sub-population. Prior for M were sampled initially, randomly on the interval [0-40]. Prior were generated with a modified version of priorgen (Ross-Ibarra et al., 2008), recoded in Python and modified to implement this new model.

Coalescent simulations were performed in msnsam, a modified version of ms allowing for variable sample size among loci (Hudson, 2002; Ross-Ibarra et al., 2008). Here, we set the size of the reference population Nref to 50,000 (the choice of Nref is arbitrary and used as a scaling factor, without impact on the estimated parameter.

We set the mutation rate  $\mu$  to 1x10<sup>-8</sup> bp/generation, following many others (e.g. (Barrio et al., 2016; Rougeux, Bernatchez, & Gagnaire, 2017; Tine et al., 2014). While this choice may seem arbitrary, it cancels out when ratio of demographic parameters (*M* and *N*) are used and is of minimal concern here. Therefore, we set  $\theta = 4N_{ref} \mu L$  to 0.16. Here L is the length of the RAD loci (80 pb).

A first round of ABC parameters estimation indicated that the upper bound of the prior distribution was attained at a value of 40 (see results below) while all other parameters were very well estimated. We run another round using a prior ranging between [0-80]. Since all other parameters were very well estimated, we fixed a narrow prior based on the minimum and maximum value of the posterior distribution from the previous estimation.

#### b) ABC parameters estimation

One million coalescent simulations were generated and associated summary statistics were computed using mscalc (Camille Roux et al., 2011)R for each small sequence. The summary statistics are the same as in (Rougemont & Bernatchez, 2018). These include the mean and standard variations of : the nucleotide diversity  $\pi$  (Tajima, 1989), the net divergence between the two populations (D<sub>A</sub>), the total divergence between populations (D<sub>XY</sub>) (Nei & Kumar, 2000), the between population differentiation (Fst) value, the number of fixed differences between populations (Sf); the number of polymorphic sites private to each population (S*xpop1* and S*xpop2*) and the number of shared polymorphic sites (Ss); and last the Pearson's R2 coefficient of correlation in  $\pi$  between the two populations. These statistics represent a panel of commonly used summary indexes in ABC (Fagundes et al., 2007; Ross-Ibarra et al., 2008; C. Roux, Tsagkogeorga, Bierne, & Galtier, 2013; Camille Roux et al., 2016).

Parameters estimation was performed using the "abc" package (Csilléry, François, & Blum, 2012). We used a logit transformation of the parameters and a tolerance of 0.001. The posterior probabilities of parameter values were then estimated using the neural network procedure with nonlinear regressions of the parameters on the summary statistics using 50 feed-forwards neural networks and 15 hidden layers. All scripts to reproduce the results are available on github.

#### c) Choice of populations

The demographic model was tested on two samples located on the St. Lawrence at two extremities of our sampling range. Namely we chose the Lake St. Pierre as the downstream site and lake St. François as our upstream site. Although more upstream populations were available (TIN, Thousand Islands or LSW, St. Lawrence Lake) these sites were also separated by impassable barriers to gene flow (dams) in the upstream direction at least. Such dams would have certainly obscured our inference.

#### d) Results

#### First round of Parameter estimates:



Figure S5: Posterior distribution of parameter estimates under the demographic model.

Theta 1 = scaled effective population size of the downstream population (Lake St. Pierre) Theta 2 = scaled effective population size of the upstream population (Lake St. François)

Migration rate  $M_{1\leftarrow 2}$  migration from upstream to downstream ( $M_{ij} = 4N_0m_{ij}$ )

Migration rate  $M_{2 \leftarrow 1}$  migration from downstream to upstream.

All values are scaled (in coalescent units).

The black line represents the prior distribution and the blue line represents the posterior distribution obtained after neural-network regression of the 1,000 retained simulations closest to the observed data.

Given that the maximum value of the prior on  $M_{1\leftarrow 2}$  is reached in this model we performed a new set of 1 million coalescent simulations but using a larger bound on the uniform prior. We used M = [0 - 80]. Since the remaining parameters were very well estimated, we use the [min-max] value of the posterior of N1, N2, M2 in these simulations. This was done in order to reduce the number of "uninformative" simulations generated by too large prior as this first round indicated which of the simulation were informative.

#### Second round of Parameter estimates:

The results for the second round indicated similar estimates for N1 and N2 as in the first round, as expected from their very narrow minimal and maximal posterior distributions. Values for  $M_{2\leftarrow 1}$  was closer to its previously inferred maximal distribution (median = 1.99 [95%CI = 1.88 - 2.00], Figure S6 below).

The value for  $M_{1\leftarrow 2}$  again reached the upper bound of M = 80 [95CI = 76.80 – 80]. This would translate into a high number of migrants (i.e. > 22).

#### e) Discussion

On the first round of parameters estimation, we note that the upper bound of the prior for  $M_{1\leftarrow 2}$  is reached. Given the hypothesized performed above (Nref = 5e4) we obtained m = 0.002 [95%CI = 39.9-40]. A biologically meaningful number is the total number of migrant each generation. The median value of N1 given the posterior distribution is 1.2284\*5e4 = 61,420 [95%CI = 1.0391 - 1.5922]\*5e4.

This give 12.284 migrants each generation in the downstream populations. [95%CI = 10.4 - 16] migrants.

Conversely, in the upstream population we have a median value of  $M_{2\leftarrow 1} = 6.671e-02$  [95%CI = 1.824e-02 – 1.398e-01] and a median of  $N_2 = 1.72*5e4$  [95%CI = 1.5127 – 1.9697]\*5e4.

This translate into 0.028 [95% CI = 0.007 - 0.068] migrants.



*Figure S6:* Parameter estimates under the demographic model using constrained prior on N1, N2 and M<sub>21</sub> based on the previously inferred parameters. Prior on  $M_{1-2}$  was set to twice its previous value.

The second round of parameter estimation also provides unambiguous evidence for higher downstream directed migration than the reverse. Indeed, the ratio  $M_{1\leftarrow 2}/M_{2\leftarrow 1}$  indicates that a 40 times higher downstream directed migration. We therefore suggest that setting the upper bound to higher value would lead to still higher estimates, suggesting that overall, the model may converge to a model of panmixia.

From this inference we conclude that 1) populations are highly connected, 2) connectivity is due to downstream biased dispersal, as expected from theory and 3) the population show only minor deviation from panmixia.

From a conservation perspective, we suggest that maintaining high connectivity among localities is the best way to maintain genetic diversity.

#### 2. Measurement of the divergence time under a model of Strict Isolation

#### a) Motivation

Observed patterns of genetic differentiation (Fst) were strong between several isolated lakes. A closer look at the data indicates low levels of polymorphism, and that most of the variation was still shared between lakes. This raise the trivial question of whether strong differentiation and strong structure are due to a long time of divergence or other processes (bottlenecks, small population size). Addressing this question allows formulating further working hypothesis related to the probabilities of accumulating genetic incompatibilities or to display outbreeding depression or not.

Here, we tackle this question by studying a model of evolutionary divergence between the two lakes initially used for stocking, namely Pigeon lake and Chautauqua. In this case, the lakes are separate by a large distance (see Fig 1) and we make the assumption that there is no ongoing gene flow possible. Therefore, we used a simplified model of strict isolation to estimate the divergence time of these two populations. It is obvious that any initial gene flow should delay the speed of divergence.

In a second analysis, we wanted to verify if the St. Lawrence have diverged more or less at the same time than the two initially compared lakes. To do so we randomly picked sites from within the St. Lawrence (we chose sites with approximately similar numbers of individuals) and compared them to the lakes. We hypothesized that if the St. Lawrence (or its tributaries) diverged at the same time than other lakes, this would provide support for a single divergence event, perhaps due to recolonization of the area after some geological events. Under this scenario, the colonization of the smallest lake by few individuals can explain their lower genetic diversity.

#### b) Model and coalescent simulations

The strict isolation (SI) model was used for coalescent simulations. This model is characterized by the instantaneous split (at time Tsplit) of an ancestral population of size  $N_{anc}$  into two daughter population of size  $N_1$  and  $N_2$ . There is no subsequent gene flow between the population. Population size can vary at the split time so that the daughter population may undergo a bottleneck at this particular moment.

The same procedure as above was used: Prior for effective population size were uniformly distributed on the interval [0 - 1,000,000] individuals. These were sampled independently for each sub-population and for the ancestral populations.

Prior for Tsplit was sampled on the interval [0-6,000,000] generations. Prior were generated with a modified version of priorgen (Ross-Ibarra, 2008), recoded in Python.

Coalescent simulations were performed in msnsam with Nref set to 50,000.

We set the mutation rate  $\mu$  to  $1 \times 10^{-8}$  bp/generation, following many others (e.g. Barrio et al. 2016; Tine et al. 2014; Rougeux et al. 2017). Therefore we set  $\theta = 4N_{ref} \mu L$  to 0.16. Here L is the length of the RAD loci (80 pb). The impact of arbitrary chosen mutation rate is discussed recently in (Brock & Wagner, 2018).

Here, to verify how different mutations rate would affect our conclusions, we also run the simulation pipeline by fixing a mutation rate 5 times lower (i.e.  $\mu = 2x10^{-9}$  bp/generation translating into theta = 0.032) corresponding to the lowest estimates of  $\mu$  in vertebrates, namely the Atlantic herring (Feng et al., 2017). An intermediate mutation rate produce similar results and we provide below only the estimates for these two extremes.

The analysis was replicated 5 times but comparing one randomly chosen site from within the St. Lawrence to either Pigeon lake or Chautauqua lake. This allowed us to test if all populations approximately have split simultaneously.

#### c) ABC parameter estimation

One million coalescent simulations were generated and associated summary statistics were computed using mscalc (Roux et al. 2011) for each small sequence. The summary statistics are the same as in Rougemont & Bernatchez (2018) and are described above. The exact same procedure for parameter estimation was used. All scripts to exactly reproduce the results are available on github.

#### d) Results

#### 1) Comparison between stocking sources: Chautauqua and Pigeon lake

The posterior distribution of parameters was very well differentiated from the prior distribution for the divergence time and ancestral population size parameter, resulting in narrow credible intervals (Fig S7). The descending population sizes were less accurately estimated and we do not interpret them.



## Figure S7: Posterior distribution of parameter estimates under a model of strict isolation between Chautauqua and Pigeon lake.

 $\Theta$ 1 = scaled effective population size of Pigeon lake (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of Chautauqua lake (4N2µ/Nrefµ)

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau$  = Tsplit/(4Nref) scaled divergence time.

Prior distribution of the parameter is displayed in black and the posterior distribution is in blue.

In this case, under the hypothetical mutation rate, we obtain a median divergence time of 22,000 years [95%CI = 10,000 - 45,000] years.

To test the influence of the mutation rate, we run the coalescent simulation with  $\theta ref = 0.032$ . Parameter estimates were not informative for the daughter population but only for the ancestral population and for the divergence time (Figure S8):



*Figure S8:* Estimated divergence time using a 5 times smaller mutation rate in the coalescent simulations.  $\Theta 1$  = scaled effective population size of Pigeon lake (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of Chautauqua lake (4N2 $\mu$ /Nref $\mu$ )

 $\Theta A$  = scaled ancestral effective population size (4N1 $\mu$ /Nref $\mu$ )

 $\tau$  = Tsplit/(4Nref) scaled divergence time.

Prior distribution of the parameter is displayed in black and the posterior distribution is in blue.

In this case, converting the divergence time yield a median Tsplit = 26,000 [7,000 - 70,000] years.

Each analyses using sampling sites from the St. Lawrence yielded similarly very accurate posterior distribution of the split time ( $\tau$ ) with a minimum median value of 10,700 years [CI = 4,550 - 17000] and a maximum value of 19,040 [95%CI = 8,800 - 29,610] years.

The posterior distribution of parameter value for each compared population is provided in the table below. The posterior distribution of the parameter relative to the prior is also provided in the Figures below for each pairs of populations.

**Table S4:** Parameters estimates for each pairs of compared sites. Abbreviation of the sampling site are given in table 1. N1,N2 and Na are the respective effective population size of population 1 population 2 and ancestral population. Tsplit is the time of divergence. 95% Credible Intervals are given in brackets.

pop1	pop2	N1 [95%CI]	N2 [95%CI]	Na [95%CI]	Tsplit [95%CI]
LSF	CHQ	221425 [82840 877540]	210435 [84420 902680]	22955 [13765 173305]	15400 [6500 24300]
ONT	CHQ	72543 [23906 624831]	682064 [459594 974402]	26174 [15772 192508]	22925 [10097 45688]
LSP	CHQ	481555 [259325 956745]	162190 [73725 738125]	19700 [13540 124335]	17500 [8500 25800]
ONT	LSF	198185 [88606 807925]	485464 [259816 945909]	11394 [7359 90519]	17967 [10429 25575]
ONT	MSG	291165 [78978 906731]	883355 [597543 997576]	20516 [13076 144742]	18385 [6904 31455]
LSL	CHQ	415680 [176285 960540]	72280 [25755 631380]	19400 [12390 120585]	19300 [9400 31200]
TRA	LSF	24834 [11879 124296]	690383 [454062 975493]	12240 [9093 57088]	15975 [8285 25267]
MAU	LSF	108610 [38430 798615]	268575 [100410 916210]	30085 [18850 290270]	27200 [12700 42300]
MAU	LSP	66440 [14770 758395]	342890 [107870 963360]	26085 [18380 173160]	13700 [7700 20900]



Figure S9: Estimated divergence time between St. Francis lake and Chautauqua lake.

 $\Theta 1$  = scaled effective population size of St. Francis lake (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of Chautauqua lake (4N2 $\mu$ /Nref $\mu$ )

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau$  = Tsplit/(4Nref) scaled divergence time.



Figure S10: Estimated divergence time between St. Louis lake and Chautauqua lake.

 $\Theta$ 1 = scaled effective population size of St. Louis lake (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of Chautauqua lake (4N2 $\mu$ /Nref $\mu$ )

 $\Theta A$  = scaled ancestral effective population size ( $4N1\mu$ /Nref $\mu$ )

 $\tau$  = Tsplit/(4Nref) scaled divergence time.



Figure S11: Estimated divergence time between St. Pierre lake and Chautauqua lake.

 $\Theta$ 1 = scaled effective population size of St. Pierre lake (4N1µ/NRefµ)

 $\Theta$ 2 = scaled effective population size of Chautauqua lake (4N2 $\mu$ /Nref $\mu$ )

 $\Theta A$  = scaled ancestral effective population size ( $4N1\mu/Nref\mu$ )

 $\tau$  = Tsplit/(4Nref) scaled divergence time.



Figure S12: Estimated divergence time between Pigeon lake and St. Francis lake.

 $\Theta 1$  = scaled effective population size of Pigeon lake ( $4N1\mu/NRef\mu$ )

 $\Theta 2$  = scaled effective population size of St. Francis lake ( $4N2\mu/Nref\mu$ )

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau$  = Tsplit/(4Nref) scaled divergence time.



Figure S13: Estimated divergence time between Pigeon lake and Montreal sorel.

 $\Theta$ 1 = scaled effective population size of Pigeon lake (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of Montreal sorel (4N2µ/Nrefµ)

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau$  = Tsplit/(4Nref) scaled divergence time.





 $\Theta 1$  = scaled effective population size of St. Maurice lake (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of St. Francis lake (4N2 $\mu$ /Nref $\mu$ )

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau = T split/(4 Nref)$  scaled divergence time.



Figure S15: Estimated divergence time between st. Maurice river and St. Pierre lake.

 $\Theta 1$  = scaled effective population size of St. Maurice river (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of St. Pierre lake (4N2µ/Nrefµ)

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau$  = Tsplit/(4Nref) scaled divergence time.



Figure S16: Estimated divergence time between st. Maurice river and St. Pierre lake.

 $\Theta 1$  = scaled effective population size of St. Maurice river (4N1µ/NRefµ)

 $\Theta 2$  = scaled effective population size of St. Pierre ake (4N2µ/Nrefµ)

 $\Theta A$  = scaled ancestral effective population size (4N1µ/Nrefµ)

 $\tau$  = Tsplit/(4Nref) scaled divergence time.

#### e) Discussion & limits

Estimates of divergence time were congruent across each pair of population tested. On the contrary we failed to estimate contemporary population sizes (N1 and N2) in most of the cases.

These estimates are based on assumption regarding the mutation rate that affect more or less our biological estimates. Reassuringly, estimates performed using two different mutation rate still resulted in overlapping 95% credible intervals. Regardless of the mutation rate that affect all our models in the same way, it is likely that all populations have started to diverged simultaneously, for instance at the end of the last glacial event, when the many meters of ice started to melt. At this time, individuals may have been able to invade the water from Quebec, resulting in the establishment (and divergence) of these recent populations.

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