

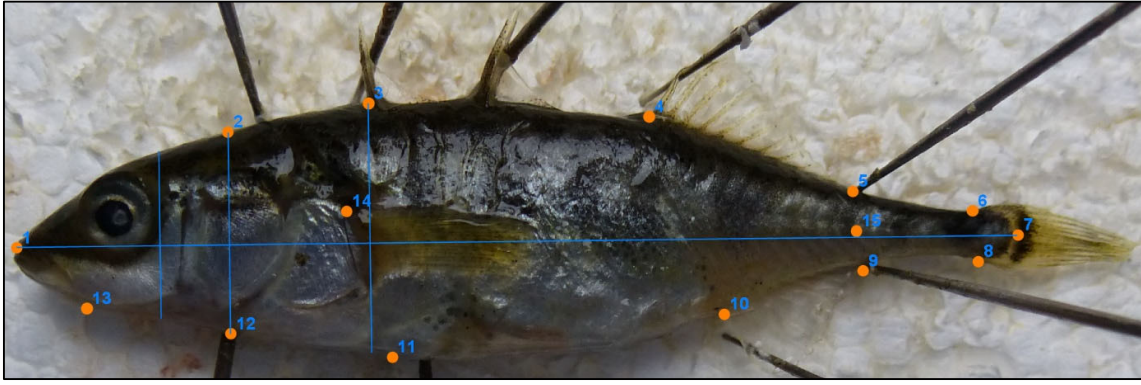


Figure S1. Example of landmarks used for geometric morphometric analysis of body shape, here visualised on the body of a three-spined stickleback individual. For all nine-spined stickleback the same homologous landmarks were used. These 15 landmarks represent a subset of the 19 landmarks that were specifically used for three-spined stickleback in Sharpe *et al.* 2008 ([35] in main paper).

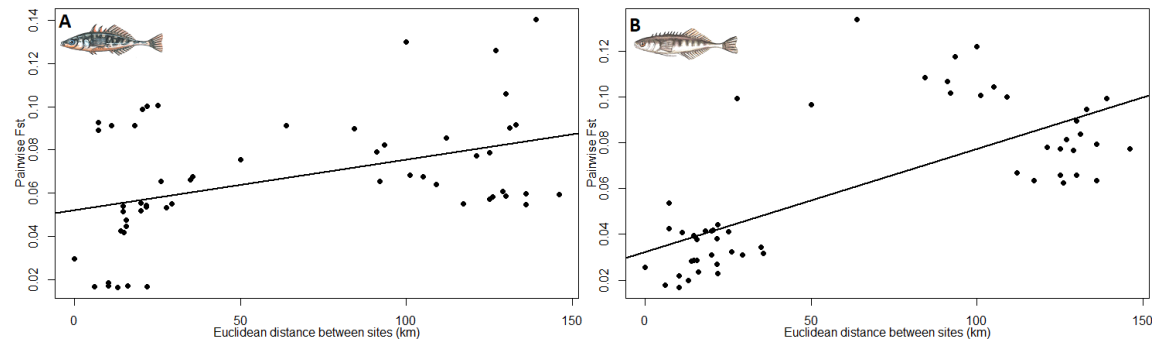


Figure S2. The relationship between Euclidean distances between sites and pairwise F_{ST} in (A) three-spined stickleback ($r = 0.42$, $P = 0.0487$) and (B) nine-spined stickleback ($r = 0.72$, $P = 0.0055$).

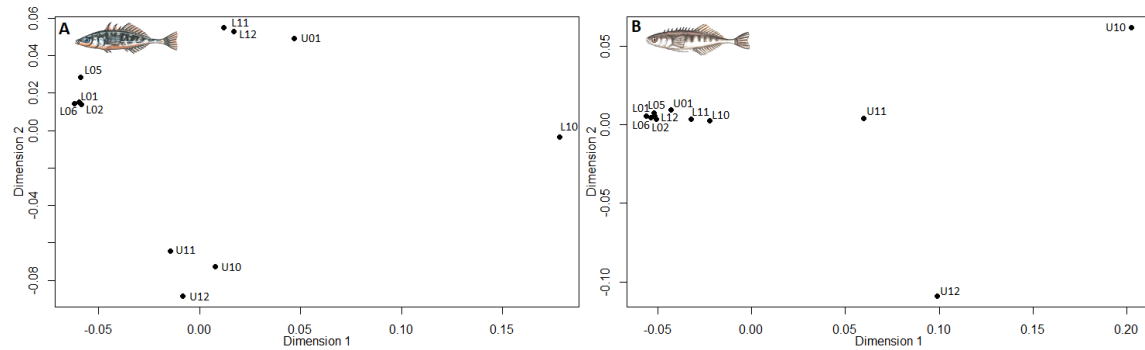


Figure S3. Two-dimensional classical multidimensional scaling of pairwise F_{ST} values in (A) three-spined stickleback and (B) nine-spined stickleback.

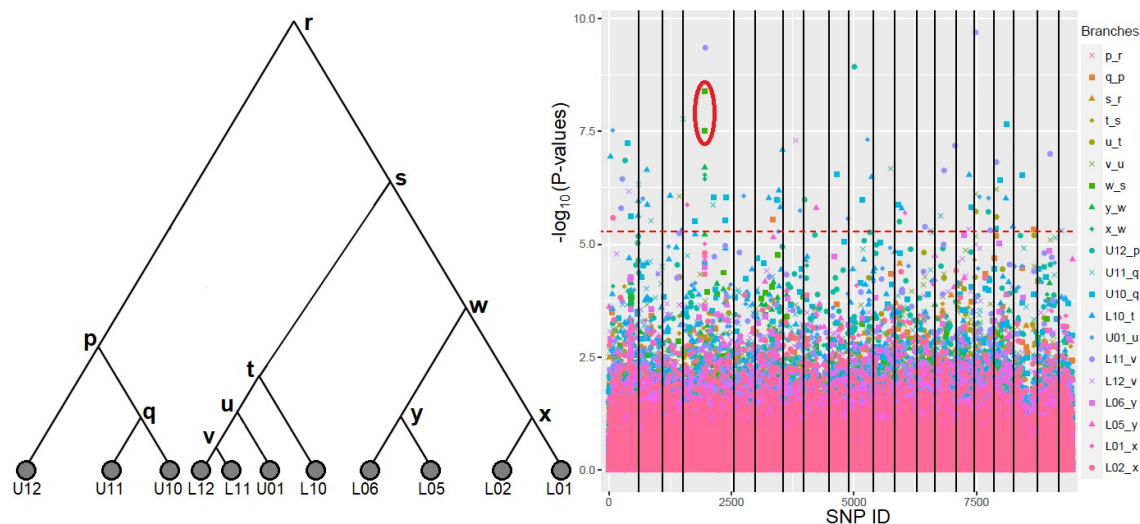


Figure S4. GRoSS v1.0 outlier analysis in three-spined stickleback SNPs along the full genealogy of the eleven populations resulting in a total of 20 outlier tests for each SNP. The horizontal black lines mark linkage groups in chronological order. All SNPs above the red-dashed line represent outliers. The red circle marks two outlier SNPs identified along the w-s branch on LG IV, representing the root of the split between brackish and freshwater sites.

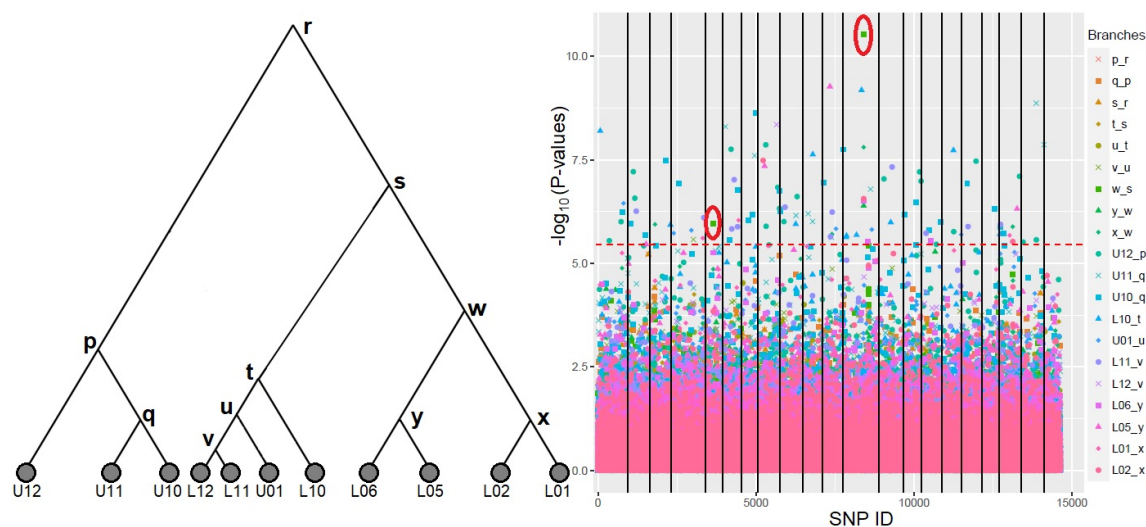


Figure S5. GRoSS v1.0 outlier analysis in nine-spined stickleback SNPs along the full genealogy of the eleven populations resulting in a total of 20 outlier tests for each SNP. The horizontal black lines mark linkage groups in chronological order. All SNPs above the red-dashed line represent outliers. The red circles mark two outlier SNPs identified along the w-s branch on LG V and LG XII, representing the root of the split between brackish and freshwater sites.

Table S1. Pairwise F_{ST} values in three-spined and nine-spined stickleback for eleven sites, with the values for three-spined stickleback above the diagonal and the values for nine-spined stickleback below the diagonal.

	L01	L02	L05	L06	L10	L11	L12	U01	U10	U11	U12
L01		0.017	0.017	0.016	0.100	0.052	0.056	0.066	0.068	0.059	0.060
L02	0.018		0.018	0.017	0.101	0.054	0.054	0.068	0.069	0.058	0.061
L05	0.024	0.022		0.017	0.091	0.045	0.048	0.055	0.064	0.055	0.060
L06	0.020	0.017	0.023		0.099	0.052	0.054	0.065	0.066	0.055	0.057
L10	0.044	0.041	0.041	0.042		0.093	0.089	0.091	0.130	0.126	0.140
L11	0.041	0.038	0.038	0.040	0.054		0.030	0.042	0.082	0.077	0.092
L12	0.031	0.027	0.029	0.029	0.042	0.026		0.043	0.079	0.079	0.090
U01	0.035	0.032	0.031	0.032	0.042	0.039	0.028		0.090	0.086	0.106
U10	0.105	0.101	0.100	0.102	0.122	0.118	0.107	0.109		0.053	0.091
U11	0.066	0.062	0.064	0.064	0.081	0.078	0.066	0.067	0.100		0.076
U12	0.080	0.077	0.077	0.077	0.100	0.095	0.084	0.090	0.134	0.097	

Table S2. The P_{ST} values and the corresponding 95% confidence intervals (CI) of 17 morphological traits in three-spined stickleback ([3s]) and nine-spined stickleback ([9s]) on the full geographical scale LBW-LFW-UFW.

	P_{ST} [3s]	P_{ST} 95% CI [3s]	P_{ST} [9s]	P_{ST} 95% CI [9s]
SL	0.4896	0.4264 - 0.5482	0.268	0.1941 - 0.3412
Plates	0.3359	0.2673 - 0.4018	0.07812	0.02857 - 0.1367
PS	0.3934	0.3256 - 0.4575	0.09732	0.04455 - 0.1573
PP	0.1438	0.08767 - 0.2045	0.0563	0.01504 - 0.1076
DS	0.124	0.07181 - 0.181	0.2305	0.1611 - 0.3016
Eye	0.2622	0.195 - 0.3289	0.079	0.03365 - 0.1324
DF	0.03045	0.001812 - 0.07094	0.01284	0.0006293 - 0.04452
AF	0.07287	0.02886 - 0.1246	0.01312	0.0005333 - 0.04575
Tail	0.1421	0.08579 - 0.2027	0.06391	0.02096 - 0.1159
BD	0.2895	0.2144 - 0.3632	0.06874	0.02107 - 0.126
NLGR	0.1065	0.05428 - 0.165	0.03845	0.002167 - 0.08992
GA	0.04582	0.01109 - 0.09069	0.04088	0.008021 - 0.08922
LGR2	0.2715	0.2044 - 0.3381	0.08127	0.03574 - 0.1362
LGR3	0.2347	0.1695 - 0.3011	0.09468	0.04467 - 0.1534
LGR4	0.2516	0.1853 - 0.3186	0.1665	0.1015 - 0.2357
RW1	0.0707	0.03533 - 0.1136	0.07122	0.03395 - 0.1172
RW2	0.15	0.09864 - 0.2057	0.08888	0.04643 - 0.1391

Table S3. The P_{ST} values and the corresponding 95% confidence intervals (CI) of 17 morphological traits in three-spined stickleback ([3s]) and nine-spined stickleback ([9s]) on the LBW-LFW scale.

	P_{ST} [3s]	P_{ST} 95% CI [3s]	P_{ST} [9s]	P_{ST} 95% CI [9s]
SL	0.4833	0.4041 - 0.5549	0.3111	0.2188 - 0.4008
Plates	0.1837	0.1103 - 0.2602	0.0783	0.01793 - 0.1508
PS	0.2322	0.1534 - 0.3115	0.08885	0.02755 - 0.1626
PP	0.1209	0.05832 - 0.191	0.01105	0.0004783 - 0.04456
DS	0.1209	0.0579 - 0.1912	0.1035	0.03733 - 0.1814
Eye	0.2926	0.2092 - 0.3736	0.02199	0.002171 - 0.06454
DF	0.02472	0.0007702 - 0.07087	0.01377	0.0004991 - 0.05275
AF	0.04779	0.005422 - 0.1039	0.01509	0.0004617 - 0.05702
Tail	0.1462	0.07887 - 0.219	0.01243	0.0005503 - 0.04798
BD	0.3418	0.2485 - 0.4306	0.09314	0.0284 - 0.1699
NLGR	0.04627	0.002088 - 0.1076	0.02486	0.0004155 - 0.08547
GA	0.06528	0.01706 - 0.1259	0.05454	0.00903 - 0.1221
LGR2	0.3024	0.2198 - 0.3831	0.05764	0.01522 - 0.1189
LGR3	0.2626	0.1823 - 0.343	0.0707	0.02052 - 0.1382
LGR4	0.2811	0.1994 - 0.3613	0.1473	0.07069 - 0.2321
RW1	0.05469	0.01961 - 0.1015	0.0598	0.02046 - 0.1131
RW2	0.1138	0.05854 - 0.1778	0.08022	0.03219 - 0.1409

Table S4. The P_{ST} values and the corresponding 95% confidence intervals (CI) of 17 morphological traits in three-spined stickleback ([3s]) and nine-spined stickleback ([9s]) on the LBW-UFW scale.

	P_{ST} [3s]	P_{ST} 95% CI [3s]	P_{ST} [9s]	P_{ST} 95% CI [9s]
SL	0.4672	0.3852 - 0.5417	0.1752	0.09392 - 0.2618
Plates	0.4557	0.3712 - 0.5324	0.00917	0.0002633 - 0.04082
PS	0.4752	0.3921 - 0.5506	0.04984	0.006588 - 0.1116
PP	0.1709	0.09565 - 0.2511	0.08504	0.02219 - 0.1617
DS	0.1359	0.06892 - 0.2101	0.2309	0.1414 - 0.3216
Eye	0.09692	0.03959 - 0.1644	0.1267	0.05738 - 0.2053
DF	0.02626	0.0004488 - 0.07839	0.03328	0.001372 - 0.09297
AF	0.09286	0.03211 - 0.1648	0.02972	0.0009508 - 0.08837
Tail	0.1033	0.03977 - 0.1771	0.08769	0.02684 - 0.1612
BD	0.06288	0.01007 - 0.1323	0.02837	0.001176 - 0.08305
NLGR	0.1482	0.07141 - 0.2322	0.04871	0.002025 - 0.1183
GA	0.01746	0.001342 - 0.05522	0.02781	0.003191 - 0.07454
LGR2	0.2575	0.1737 - 0.3405	0.1253	0.05544 - 0.2068
LGR3	0.2324	0.1501 - 0.3155	0.1205	0.05188 - 0.1999
LGR4	0.2662	0.1817 - 0.3501	0.2251	0.1366 - 0.3159
RW1	0.1004	0.04627 - 0.1647	0.08101	0.03254 - 0.1416
RW2	0.1113	0.05668 - 0.1742	0.08829	0.03677 - 0.1518

Table S5. The P_{ST} values and the corresponding 95% confidence intervals (CI) of 17 morphological traits in three-spined stickleback ([3s]) and nine-spined stickleback ([9s]) on the LFW-UFW scale.

	P_{ST} [3s]	P_{ST} 95% CI [3s]	P_{ST} [9s]	P_{ST} 95% CI [9s]
SL	0.5137	0.4363 - 0.5831	0.2874	0.1933 - 0.379
Plates	0.2406	0.1568 - 0.3245	0.1154	0.04257 - 0.1983
PS	0.3326	0.2461 - 0.4154	0.1439	0.06701 - 0.2285
PP	0.04434	0.003879 - 0.1027	0.06092	0.008827 - 0.1288
DS	0.08193	0.02771 - 0.1477	0.2972	0.2032 - 0.3887
Eye	0.3617	0.2742 - 0.4444	0.1005	0.03872 - 0.1728
DF	0.04005	0.001703 - 0.09696	0.007717	0.0003492 - 0.03266
AF	0.01945	0.0005656 - 0.06483	0.007589	0.0002607 - 0.03435
Tail	0.1593	0.0849 - 0.2392	0.07803	0.01874 - 0.1511
BD	0.4124	0.3173 - 0.4995	0.08696	0.02085 - 0.1672
NLGR	0.09442	0.03413 - 0.1653	0.03903	0.001217 - 0.1014
GA	0.05516	0.008992 - 0.1177	0.04339	0.005713 - 0.1058
LGR2	0.1571	0.08411 - 0.2361	0.05893	0.01594 - 0.1192
LGR3	0.106	0.04484 - 0.1774	0.08345	0.02766 - 0.1546
LGR4	0.08734	0.03193 - 0.1547	0.1066	0.04223 - 0.1838
RW1	0.06489	0.02433 - 0.1174	0.08283	0.03224 - 0.1468
RW2	0.2101	0.1359 - 0.287	0.1002	0.04389 - 0.168

Table S6. Genomic position of the outlier SNPs associated with the w-s branch of the population genealogy as detected by GRoSS v1.0 in three-spined stickleback ([3s]) and nine-spined stickleback ([9s]).

	Group	Position	Gene ID	Gene name	Gene description
[3s]	IV	19858133	ENSGACG00000018958	<i>ppara</i>	Peroxisome proliferator-activated receptor alpha a
[3s]	IV	19871462	-	-	-
[9s]	V	6140695	ENSGACT00000003223.1	<i>CDHR1</i>	Cadherin related family member 1
[9s]	XII	29859509	-	-	-