

## Supplementary Materials for:

# Population Subdivision in the Gopher Frog (*Rana capito*) across the Fragmented Longleaf Pine-Wiregrass Savanna of the Southeastern USA

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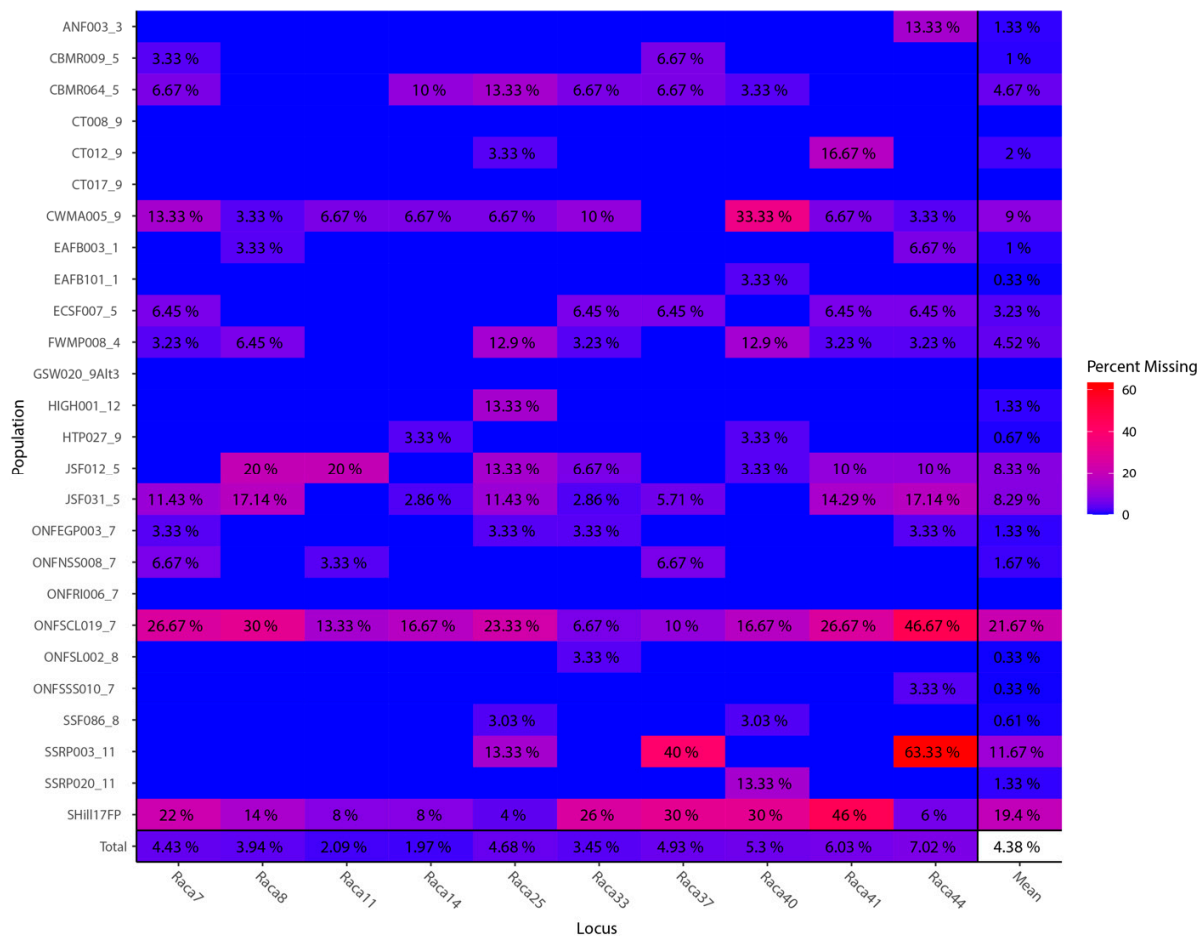
**Table S1. Sampling information. Sites with at least 30 samples are shown in bold (27 sites of 89 total).**

State	Area	Population	N	Latitude	Longitude
AL	Conecuh National Forest	NPAL	12	31.16	-86.56
AL	Conecuh National Forest	SPAL	17	31.17	-86.54
AL	Conecuh National Forest	YHPAL	2	31.09	-86.55
FL	Allen David Broussard Catfish Creek Preserve	CCP001	1	28.00	-81.48
FL	Allen David Broussard Catfish Creek Preserve	CCP003	4	27.98	-81.49
FL	Allen David Broussard Catfish Creek Preserve	CCP004	8	27.99	-81.50
FL	Allen David Broussard Catfish Creek Preserve	CCP016	6	27.97	-81.47
FL	Allen David Broussard Catfish Creek Preserve	CCP017	8	27.98	-81.48
<b>FL</b>	<b>Apalachicola National Forest</b>	<b>ANF003</b>	<b>30</b>	<b>30.34</b>	<b>-84.32</b>
FL	Apalachicola National Forest	ANFCL002	1	30.26	-84.95
FL	Apalachicola National Forest	ANFCL007	27	30.27	-84.99
FL	Apalachicola National Forest	ANFCL012	3	30.24	-84.99
FL	Big Bend WMA, Spring Creek Unit	BBSCU002	17	29.98	-83.69
FL	Big Bend WMA, Spring Creek Unit	BBSCU022	5	29.98	-83.70
FL	Bluefield Ranch	BR008	3	27.21	-80.62
FL	Buck Lake Conservation Area	BLCA001	2	28.70	-80.90
FL	Buck Lake Conservation Area	BLCA006	7	28.69	-80.90
FL	Camp Blanding Military Reservation	CBMR004	24	29.88	-82.03
<b>FL</b>	<b>Camp Blanding Military Reservation</b>	<b>CBMR009</b>	<b>30</b>	<b>30.00</b>	<b>-81.96</b>
FL	Camp Blanding Military Reservation	CBMR060	6	29.87	-82.04
<b>FL</b>	<b>Camp Blanding Military Reservation</b>	<b>CBMR064</b>	<b>30</b>	<b>29.87</b>	<b>-81.95</b>
FL	Cary State Forest	CSF007	22	30.40	-81.86
FL	Cary State Forest	CSF009	8	30.40	-81.86
<b>FL</b>	<b>Chassahowitzka WMA</b>	<b>CWMA005</b>	<b>30</b>	<b>28.64</b>	<b>-82.58</b>
<b>FL</b>	<b>Eglin Air Force Base</b>	<b>EAFB003</b>	<b>30</b>	<b>30.57</b>	<b>-86.75</b>
FL	Eglin Air Force Base	EAFB008	2	30.48	-86.71
FL	Eglin Air Force Base	EAFB011	7	30.68	-86.46
FL	Eglin Air Force Base	EAFB099	1	30.50	-86.65
<b>FL</b>	<b>Eglin Air Force Base</b>	<b>EAFB101</b>	<b>30</b>	<b>30.67</b>	<b>-86.45</b>
<b>FL</b>	<b>Etoniah Creek State Forest</b>	<b>ECSF007</b>	<b>31</b>	<b>29.79</b>	<b>-81.88</b>
<b>FL</b>	<b>Fort White Mitigation Park WEA</b>	<b>FWMP008</b>	<b>31</b>	<b>29.90</b>	<b>-82.79</b>
<b>FL</b>	<b>Green Swamp West</b>	<b>GSW020</b>	<b>30</b>	<b>28.43</b>	<b>-82.13</b>
<b>FL</b>	<b>Halpata Tastanaki Preserve</b>	<b>HTP027</b>	<b>30</b>	<b>29.02</b>	<b>-82.35</b>
FL	Jennings State Forest	JSF002	1	30.10	-81.92
FL	Jennings State Forest	JSF003	15	30.10	-81.93
FL	Jennings State Forest	JSF004	12	30.15	-81.90

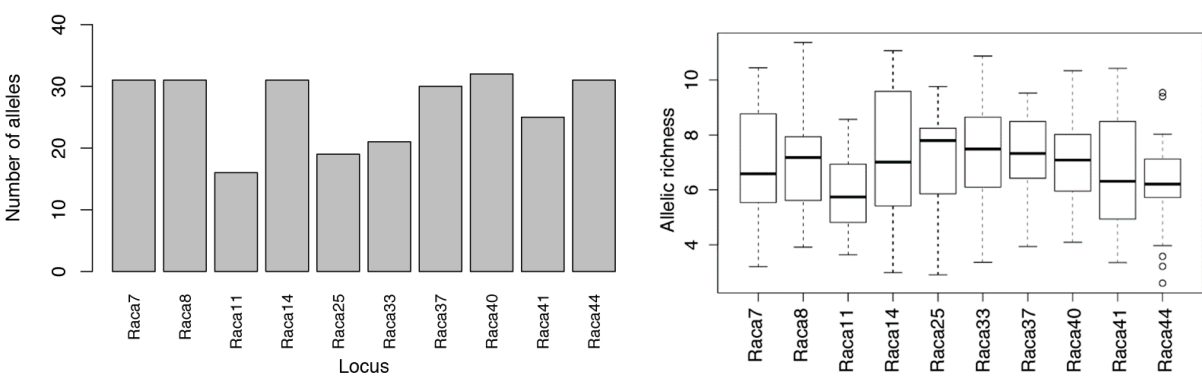
State	Area	Population	N	Latitude	Longitude
FL	Jennings State Forest	JSF010	22	30.13	-81.89
<b>FL</b>	<b>Jennings State Forest</b>	<b>JSF012</b>	<b>30</b>	<b>30.16</b>	<b>-81.93</b>
<b>FL</b>	<b>Jennings State Forest</b>	<b>JSF031</b>	<b>35</b>	<b>30.15</b>	<b>-81.89</b>
FL	Jennings State Forest	JSF039	6	30.15	-81.91
FL	Jonathan Dickinson State Park	JDSP005	5	26.99	-80.11
FL	Jonathan Dickinson State Park	JDSP008	8	27.00	-80.11
FL	Jonathan Dickinson State Park	JDSP011	13	26.99	-80.11
FL	Little Big Econ State Forest	LBESF011	23	28.67	-81.11
FL	Longleaf Flatwoods Reserve	LFR006	16	29.56	-82.20
FL	Well Field Scrub-jay Habitat	LMLW005	22	27.47	-82.20
FL	Ocala National Forest	ONFEGP002	14	29.38	-81.79
<b>FL</b>	<b>Ocala National Forest</b>	<b>ONFEGP003</b>	<b>30</b>	<b>29.38</b>	<b>-81.79</b>
<b>FL</b>	<b>Ocala National Forest</b>	<b>ONFNSS008</b>	<b>30</b>	<b>29.42</b>	<b>-81.75</b>
<b>FL</b>	<b>Ocala National Forest</b>	<b>ONFRI006</b>	<b>32</b>	<b>29.48</b>	<b>-81.81</b>
<b>FL</b>	<b>Ocala National Forest</b>	<b>ONFSCLO19</b>	<b>30</b>	<b>29.17</b>	<b>-81.90</b>
FL	Ocala National Forest	ONFSL001	29	28.98	-81.56
<b>FL</b>	<b>Ocala National Forest</b>	<b>ONFSL002</b>	<b>30</b>	<b>29.06</b>	<b>-81.56</b>
<b>FL</b>	<b>Ocala National Forest</b>	<b>ONFSSS010</b>	<b>30</b>	<b>29.32</b>	<b>-81.75</b>
<b>FL</b>	<b>Private land near Highlands Hammock</b>	<b>HIGH001</b>	<b>30</b>	<b>27.49</b>	<b>-81.51</b>
FL	Private land, Calhoun Co.	CALH001	3	30.44	-85.34
FL	Private land, Suwannee Co.	SUWA002	7	30.09	-82.82
FL	Rock Springs Run State Reserve	RSRSR011	21	28.77	-81.45
<b>FL</b>	<b>Seminole State Forest</b>	<b>SSF086</b>	<b>33</b>	<b>28.99</b>	<b>-81.45</b>
FL	St. Sebastian River Preserve State Park	SSRP003	30	27.83	-80.59
FL	St. Sebastian River Preserve State Park	SSRP011	16	27.84	-80.52
FL	St. Sebastian River Preserve State Park	SSRP013	15	27.79	-80.51
<b>FL</b>	<b>St. Sebastian River Preserve State Park</b>	<b>SSRP020</b>	<b>30</b>	<b>27.84</b>	<b>-80.54</b>
FL	Triple N Ranch	TNR003	19	28.11	-81.02
<b>FL</b>	<b>Withlacoochee State Forest-Croom Tract</b>	<b>CT008</b>	<b>30</b>	<b>28.60</b>	<b>-82.27</b>
<b>FL</b>	<b>Withlacoochee State Forest-Croom Tract</b>	<b>CT012</b>	<b>30</b>	<b>28.61</b>	<b>-82.24</b>
<b>FL</b>	<b>Withlacoochee State Forest-Croom Tract</b>	<b>CT017</b>	<b>30</b>	<b>28.62</b>	<b>-82.23</b>
GA	Fall Line Sandhills WMA	FallLine	25	32.58	-84.27
GA	Fort Benning	FtBenning	12	32.47	-84.67
<b>GA</b>	<b>Fort Stewart</b>	<b>FtStew</b>	<b>47</b>	<b>31.96</b>	<b>-81.77</b>
NC	Croatan National Forest	CROATANP 1	13	34.72	-76.97
NC	Holly Shelter Game Lands	HSGLHolly	7	34.43	-77.67
NC	Holly Shelter Game Lands	HSGLPit	15	34.42	-77.69
NC	MCB Camp Lejeune	CLJGD1	7	34.62	-77.25
NC	MCB Camp Lejeune	CLJHD05	2	34.63	-77.32

State	Area	Population	N	Latitude	Longitude
NC	MCB Camp Lejeune	CLJHE11	6	34.62	-77.30
NC	MCB Camp Lejeune	CLJHP30	2	34.60	-77.30
NC	Military Ocean Terminal Sunny Point	MOTSUP	5	33.99	-78.01
<b>NC</b>	<b>Sandhills Game Lands</b>	<b>SHILL17FP</b>	<b>50</b>	<b>34.99</b>	<b>-79.51</b>
SC	Private Land, Barnwell County	EnerSol	2	33.26	-81.47
SC	Savannah River Site	Bay51	6	33.32	-81.56
SC	Savannah River Site	Bay52	3	33.31	-81.55
SC	Savannah River Site	Bay56	10	33.29	-81.55
SC	Savannah River Site	Bay58	19	33.29	-81.56
SC	Savannah River Site	CraigsPond	1	33.29	-81.48
SC	Savannah River Site	MonaBay	1	33.32	-81.48
SC	Savannah River Site	Sarracenia	25	33.29	-81.49
SC	Savannah River Site	Slough	6	33.31	-81.54
SC	Savannah River Site	ThunderBay	1	33.23	-81.49

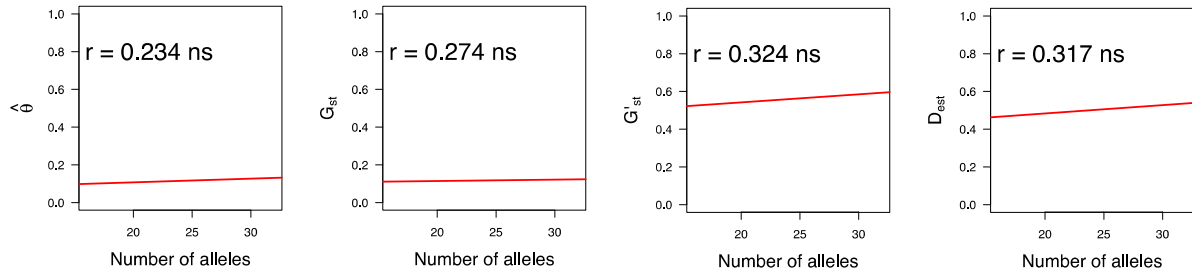




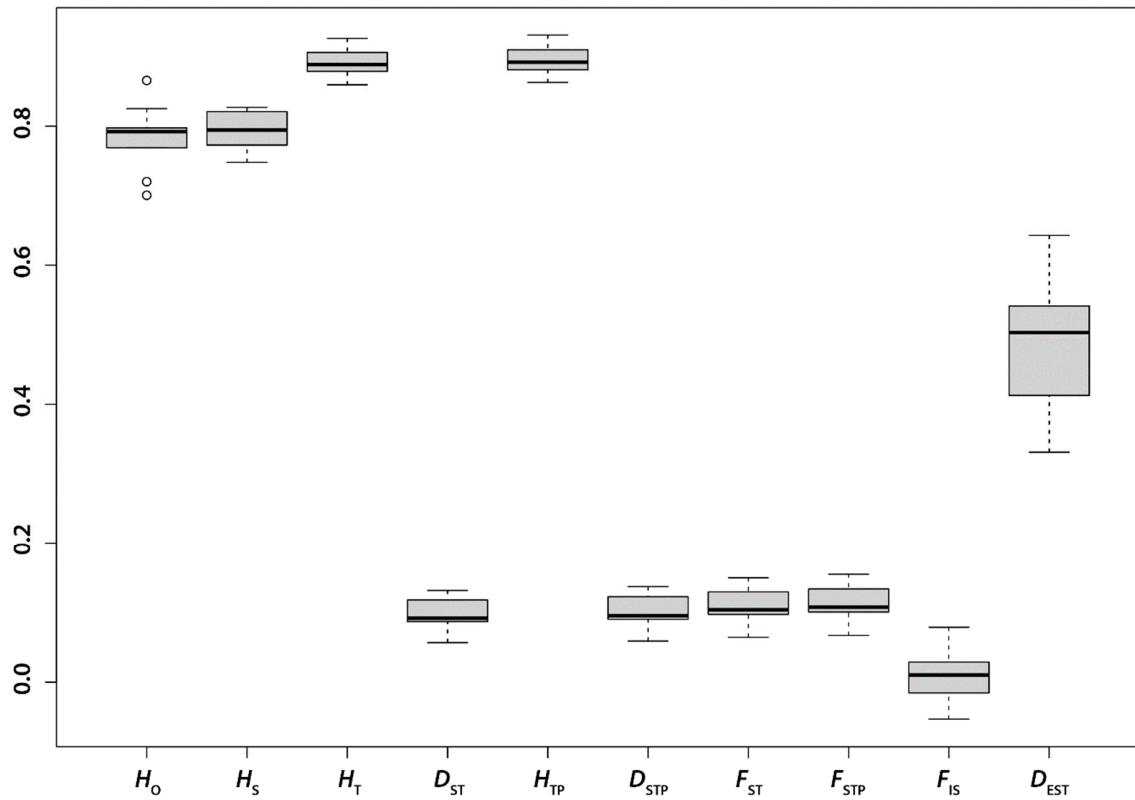
**Figure S2.** Missing data per locus and population for the rangewide dataset consisting of only those populations with at least 30 samples (N = 859). Mean amount of missing data was 4.38%.



**Figure S3.** Number of alleles per locus (left panel) and rarefied allelic counts per locus for populations with at least 30 individuals (right panel).



**Figure S4. Scatterplots of diversity partitioning estimators vs. locus polymorphism for Florida populations with at least 30 samples. Red lines are best-fit lines and  $r$ -values are Pearson product moment correlation coefficients. There is a not a significant correlation between locus polymorphism and diversity partitioning statistics (see [1]).**



**Figure S5. Descriptive statistics averaged over all loci for populations with at least 30 samples in the rangewide dataset. Calculations are based on genotypic (rather than allelic) number and are corrected for levels of heterozygosity. Estimates are observed heterozygosities ( $H_O$ ), within population gene diversities ( $H_S$ ), overall gene diversity ( $H_T$ ), gene diversity among samples ( $D_{ST}$ ), corrected overall gene diversity ( $H_{TP}$ ), corrected gene diversity among samples ( $D_{STP}$ ), fixation index ( $F_{ST}$ ), corrected fixation index ( $F_{STP}$ ), inbreeding coefficient ( $F_{IS}$ ) and Jost's  $D$  ( $D_{EST}$ ).  $F_{ST}$   $F_{STP}$  and  $F_{IS}$  calculated following [2]. Estimates calculated using the *hierfstat* package for R [3].**

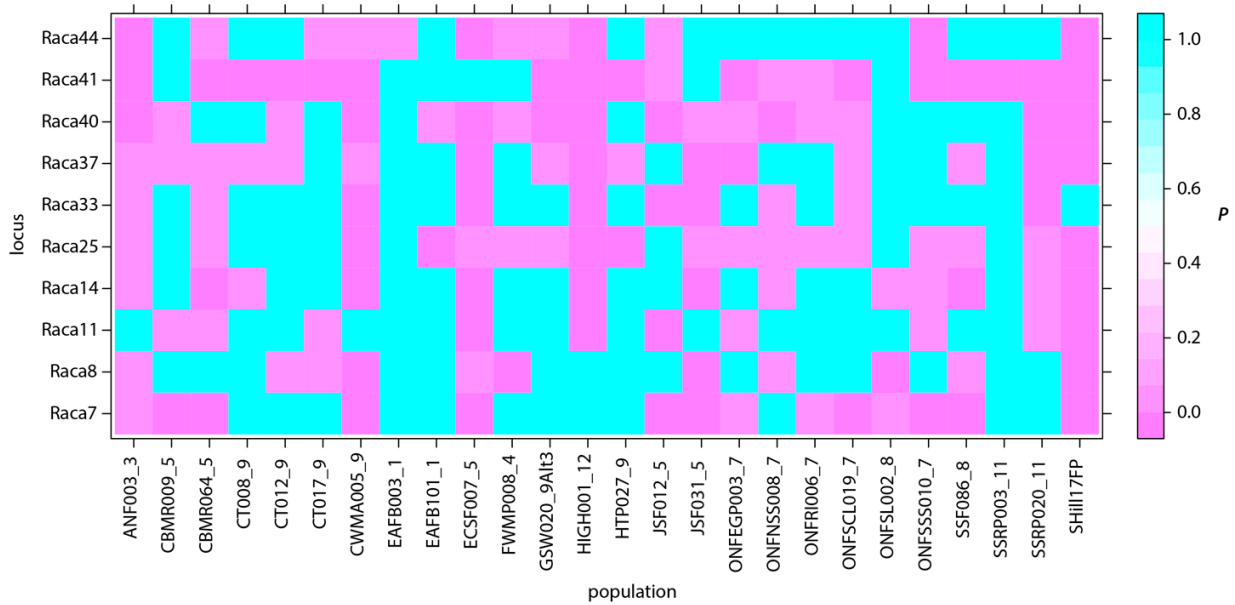
**Table S2. Population differentiation statistics per locus and averaged over all loci for Florida populations with at least 30 samples, including: F-statistic analogs for microsatellites based on allele size ( $Rho_{IS}$   $Rho_{ST}$   $Rho_{IT}$  [4]; single locus estimates following standard ANOVA analogous to [5]; see [6] for formulas), fixation index ( $F_{ST}$ ) based on allele identity [5,7], coefficient of gene differentiation, both in its original form ( $G_{ST}$  [8]) and as standardized by levels of genetic variation ( $G'_{ST}$  [9];  $G''_{ST}$  [10]); and Jost's  $D$  ( $D_{EST}$  [11]).**

Locus	$Rho_{IS}$	$Rho_{ST}$	$Rho_{IT}$	$F_{ST}$	$G_{ST}$	$G'_{ST}$	$G''_{ST}$	$D_{EST}$
<b>Raca7</b>	0.151	0.408	0.498	0.118	0.113	0.574	0.576	0.520
<b>Raca8</b>	-0.116	0.160	0.063	0.093	0.089	0.406	0.408	0.348
<b>Raca11</b>	-0.072	0.173	0.113	0.103	0.100	0.476	0.478	0.417
<b>Raca14</b>	0.106	0.116	0.210	0.134	0.129	0.691	0.693	0.646
<b>Raca25</b>	-0.008	0.144	0.138	0.106	0.102	0.570	0.572	0.522
<b>Raca33</b>	-0.037	0.118	0.085	0.079	0.075	0.452	0.453	0.407
<b>Raca37</b>	0.049	0.141	0.182	0.098	0.094	0.548	0.550	0.501
<b>Raca40</b>	0.373	0.077	0.421	0.060	0.058	0.340	0.342	0.299
<b>Raca41</b>	0.008	0.194	0.201	0.113	0.109	0.514	0.516	0.454
<b>Raca44</b>	0.014	0.081	0.094	0.147	0.150	0.617	0.620	0.550
<b>Overall</b>	0.075	0.219	0.278	0.105	0.102	0.522	0.524	0.468

**Table S3. Private alleles by locus and population for Florida populations.**

Locus	Population	Allele	Locus	Population	Allele	Locus	Population	Allele
<b>Raca7</b>	CBMR009	334	<b>Raca25</b>	CWMA005	364	<b>Raca40</b>	SUWA002	286
<b>Raca7</b>	JSF004	346	<b>Raca25</b>	LBESF011	258	<b>Raca40</b>	SUWA002	346
<b>Raca7</b>	ONFLD001	266	<b>Raca25</b>	ONFLD001	264	<b>Raca41</b>	CWMA005	336
<b>Raca7</b>	ONFEGP002	294	<b>Raca33</b>	BLCA001	277	<b>Raca41</b>	LMLW005	292
<b>Raca7</b>	ONFLD001	430	<b>Raca33</b>	JSF003	215	<b>Raca44</b>	ANF003	333
<b>Raca7</b>	SSRP003	402	<b>Raca33</b>	JSF004	201	<b>Raca44</b>	CBMR004	357
<b>Raca8</b>	HTP027	384	<b>Raca37</b>	CCP003	209	<b>Raca44</b>	CT008	363
<b>Raca8</b>	LBESF011	308	<b>Raca37</b>	CT017	253	<b>Raca44</b>	HTP027	391
<b>Raca8</b>	SUWA002	388	<b>Raca37</b>	FWMP008	157	<b>Raca44</b>	HTP027	405
<b>Raca14</b>	CBMR004	308	<b>Raca37</b>	ONFEGP002	173	<b>Raca44</b>	LBESF011	395
<b>Raca14</b>	EAFB008	310	<b>Raca37</b>	RSRSR011	245	<b>Raca44</b>	LFR006	341
<b>Raca14</b>	ONFEGP002	304	<b>Raca40</b>	CWMA005	382	<b>Raca44</b>	ONFEGP002	377
<b>Raca14</b>	ONFSCL019	212	<b>Raca40</b>	EAFB008	318	<b>Raca44</b>	RSRSR011	389
<b>Raca14</b>	ONFSCL019	222	<b>Raca40</b>	LBESF011	322			
<b>Raca14</b>	SUWA002	290	<b>Raca40</b>	SUWA002	286			

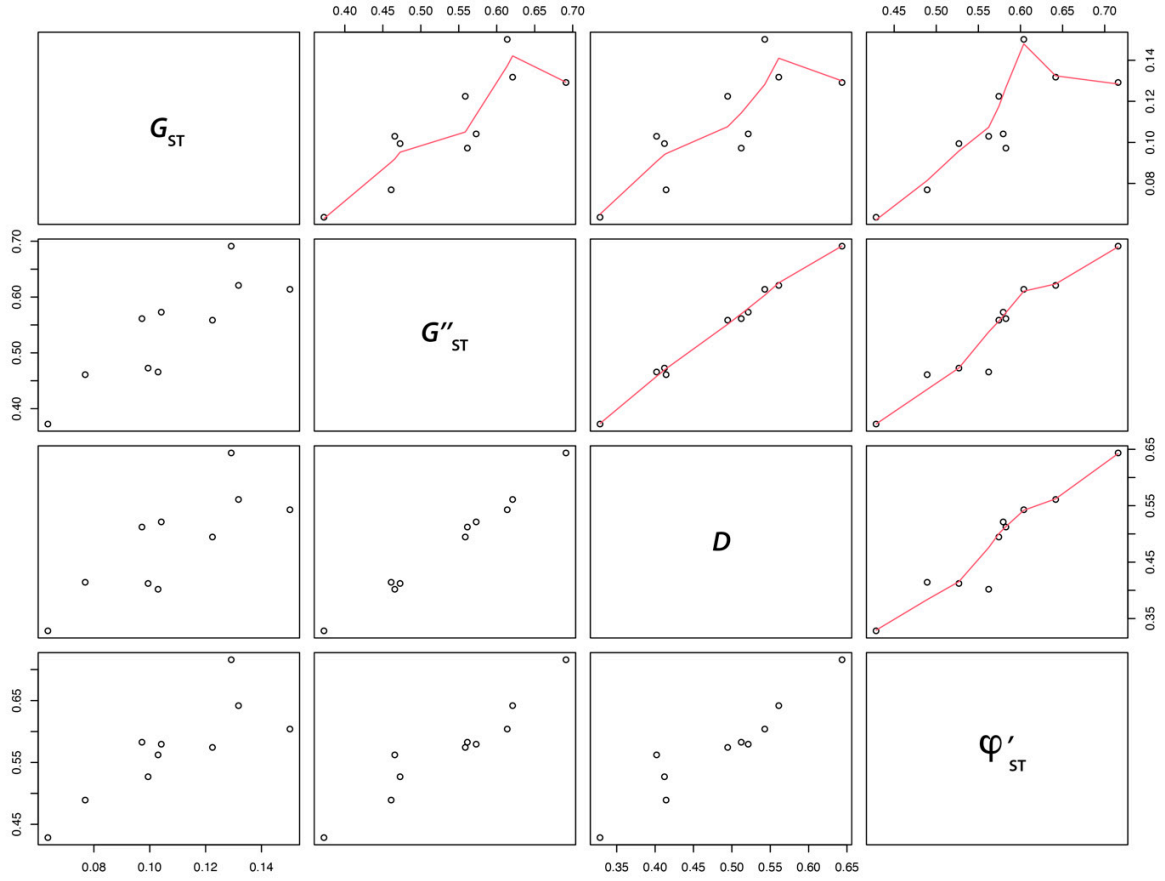




**Figure S6. Significant ( $P < 0.05$ ) deviations from Hardy-Weinberg equilibrium using an exact test based on 1,000 Monte Carlo permutations of alleles [11].**

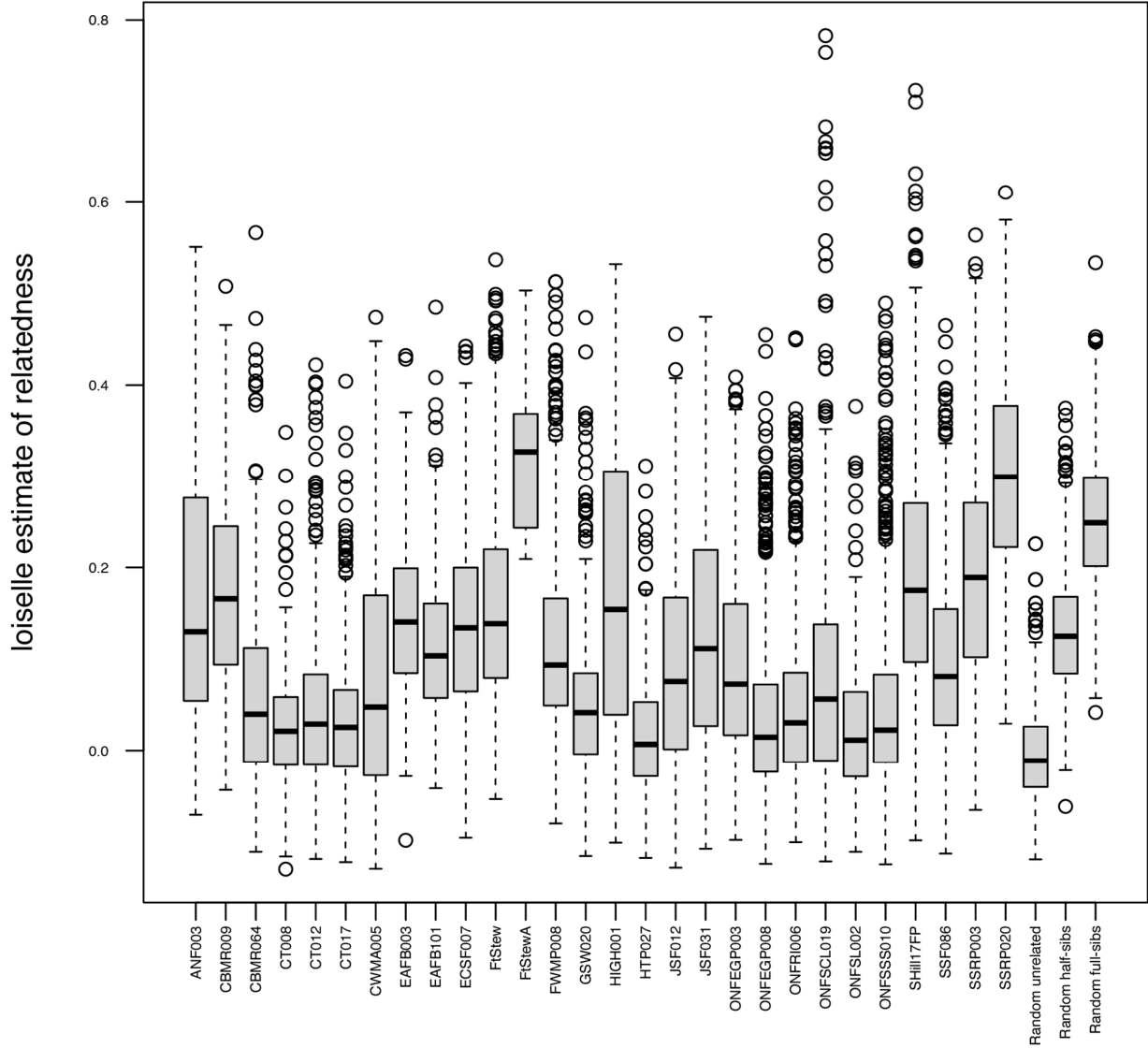
**Table S4. Significance ( $P < 0.05$ ) of heterozygosity-based tests for Hardy Weinberg equilibrium for populations with at least 30 samples. Values are one-sided p-values of  $G_{IS}$  [12] per locus and population. Significant values are shown in bold.**

Population	Raca7	Raca8	Raca11	Raca14	Raca25	Raca33	Raca37	Raca40	Raca41	Raca44	Multi-locus
EAFB003	0.123	0.274	0.065	0.157	0.164	0.569	0.005	0.188	0.477	0.001	<b>0.005</b>
EAFB101	0.541	0.143	0.575	0.190	0.007	0.057	0.006	0.002	0.522	0.019	<b>0.001</b>
ANF003	0.525	0.051	0.238	0.425	0.078	0.101	0.431	0.070	0.045	0.001	<b>0.018</b>
FWMP008	0.109	0.084	0.111	0.336	0.001	0.276	0.013	0.024	0.289	0.002	<b>0.006</b>
JSF012	0.001	0.940	0.964	0.152	0.473	0.113	0.429	0.006	0.554	0.160	<b>0.001</b>
JSF031	0.007	0.043	0.353	0.001	0.742	0.191	0.505	0.001	0.067	0.001	0.990
CBMR009	0.003	0.583	0.026	0.035	0.345	0.232	0.083	0.001	0.296	0.125	0.077
CBMR064	0.001	0.517	0.009	0.001	0.003	0.141	0.219	0.233	0.066	0.582	<b>0.001</b>
ECSF007	0.448	0.357	0.091	0.001	0.403	0.459	0.252	0.001	0.169	0.001	<b>0.001</b>
ONFRI006	0.155	0.392	0.026	0.637	0.040	0.106	0.513	0.311	0.531	0.056	0.419
ONFNSS008	0.216	0.285	0.016	0.243	0.046	0.017	0.551	0.246	0.071	0.064	0.212
ONFEGP003	0.077	0.035	0.009	0.496	0.166	0.093	0.030	0.002	0.045	0.496	<b>0.020</b>
ONFSSS010	0.129	0.215	0.074	0.344	0.002	0.040	0.501	0.002	0.184	0.001	<b>0.023</b>
ONFSL019	0.113	0.003	0.751	0.795	0.033	0.071	0.547	0.270	0.340	0.001	<b>0.001</b>
HTP027	0.466	0.339	0.531	0.490	0.167	0.448	0.038	0.144	0.008	0.078	0.388
ONFSL002	0.552	0.002	0.458	0.486	0.458	0.303	0.326	0.006	0.467	0.610	<b>0.013</b>
SSF086	0.015	0.023	0.404	0.029	0.011	0.445	0.022	0.506	0.045	0.171	<b>0.001</b>
CWMA005	0.001	0.049	0.001	0.284	0.033	0.149	0.014	0.001	0.254	0.174	<b>0.004</b>
CT008	0.461	0.371	0.184	0.004	0.283	0.590	0.498	0.175	0.017	0.231	0.523
CT012	0.522	0.260	0.610	0.002	0.105	0.442	0.592	0.624	0.228	0.403	0.052
CT017	0.296	0.111	0.177	0.032	0.527	0.602	0.262	0.472	0.107	0.347	0.166
GSW020	0.331	0.548	0.100	0.328	0.202	0.485	0.073	0.562	0.383	0.004	0.295
SSRP020	0.246	0.054	0.077	0.233	0.468	0.001	0.007	0.544	0.247	0.045	<b>0.001</b>
SSRP003	0.082	0.195	0.482	0.382	0.470	0.361	0.014	0.190	0.070	0.001	<b>0.001</b>
HIGH001	0.167	0.001	0.003	0.002	0.048	0.405	0.048	0.213	0.008	0.211	0.057

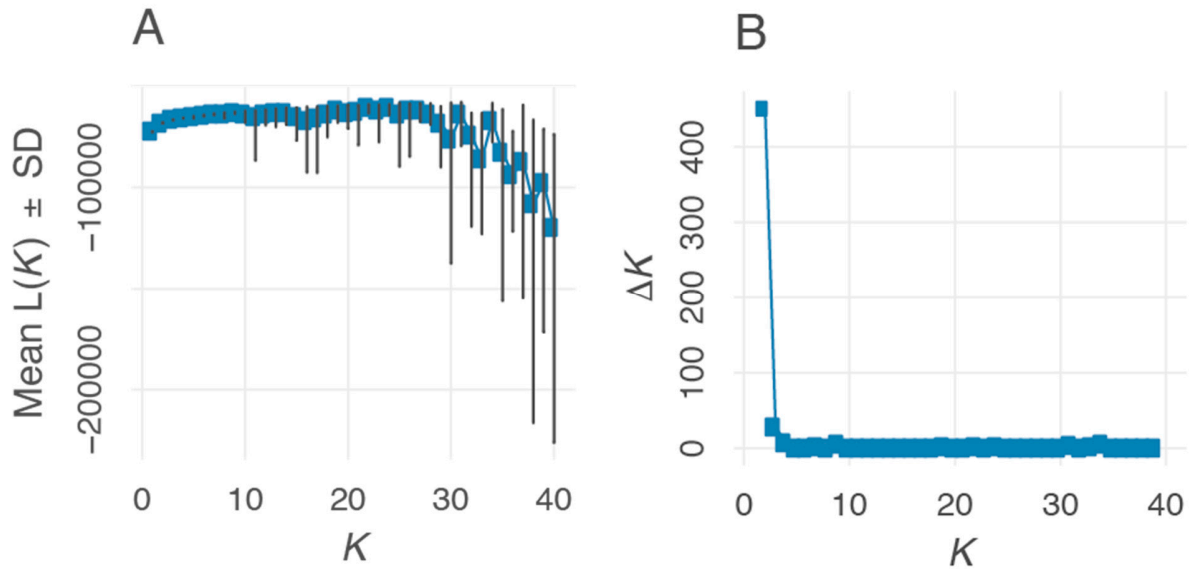


**Figure S7. Comparison of point estimates of population differentiation measures. Coefficient of gene differentiation ( $G_{ST}$ ) both in its original form [7] and as standardized by levels of genetic variation ( $G''_{ST}$  [9]), Jost's  $D$  [10], and  $\phi'_{ST}$  calculated using the *mmod* [13] package for R.**

## Mean relatedness of populations



**Figure S8. Relatedness estimates for ponds with at least 30 samples in the rangewide dataset. Relatedness was calculated using the coancestry estimator of Loiselle et al. [13] implemented in the package *demerelate* [14] for R. Assuming random mating, the estimator is a measure of inbreeding between related individuals, with an expected value of 0.25 for full-siblings and 0.125 for half-siblings. Upper and lower error bars bound the 95% confidence interval around the mean as determined by 1000 simulated pairs for logistic regression to calculate relatedness estimates. Missing values were excluded.**



**Figure S9. Results from Structure analysis of the entire dataset assuming admixture with correlated allele frequencies among populations and using location information as a prior. (A) mean log probability of the data is highest for  $K = 22$ ; (B) the rate of change in the log probability of the data ( $\Delta K$ ) is greatest from 1 to 2 clusters). At  $K=2$ , panhandle and peninsular populations form separate clusters.**

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