Genetic analysis of mixed individuals reveals different spawning stocks of the *Tetrapturus pfluegeri* (longbill spearfish) in the Western Atlantic Ocean

Suhaila Karim Khalil Jaser. Rodrigo Rodrigues Domingues. Caio Augusto Perazza. Eric Hallerman. Alexandre Wagner Silva Hilsdorf

Table S1: *In-silico* assessment of segregation of null microsatellite alleles using four different analytic packages.

Locus	Micro-Checker	ML-Null Freq		
		Cervus		FreeNA
	Null Present	*F*(Null)	*F*(Null)	*F*(Null)
Tpf_10	yes	0.2882	0.2840	0.2608
Tpf_12	no	-0.0359	0.0000	0.0000
Tpf_14	yes	0.2685	0.1357	0.1192
Tpf_17	no	-0.0008	0.0000	0.0000
Tpf_18	yes	0.0698	0.0760	0.0759
Tpf_19	no	0.0630	0.0833	0.0500
Tpf_20	yes	0.3935	0.2007	0.1570
Tpf_26	yes	0.4946	0.2227	0.2136
Tpf_35	yes	0.1542	0.0913	0.0679
Tpf_39	yes	0.1297	0.0550	0.0460
Tpf_43	yes	0.1980	0.1637	0.1501
Tpf_23	yes	0.1693	0.1760	0.0285

Table S2. Genetic diversity at 12 microsatellite loci for *Tetrapturus pfluegeri*: *N* (number of samples). *N*a (number of alleles). *Ar* (allelic richness), *H*o (observed heterozygosity). *H*e (expected heterozygosity). u*H*e (unbiased expected heterozygosity = (2*N* / (2*N*-1)) * *H*e). *F* (general inbreeding coefficient). F_{IS} (inbreeding within subpopulations). F_{IT} (Total inbreeding coefficient). F_{ST} (genetic differentiation among subpopulations). *HWE* (*p*-value for departure from Hardy-Weinberg equilibrium). *N*m (equilibrium number of migrants = [1 / *F*st) - 1] / 4). *PIC* (polymorphic index content).

Sampling Points	Parameters	Microsatellite Loci												Mean	SE
		Tpf10	***Tpf12***	***Tpf14***	***Tpf17***	***Tpf18***	***Tpf19***	***Tpf20***	***Tpf26***	***Tpf35***	***Tpf39***	***Tpf43***	***Tpf_23***		
BRA	***N***	28	28	28	27	28	27	28	28	28	28	28	26	27.667	0.188
	Na	7.000	3.000	5.000	6.000	5.000	4.000	6.000	7.000	3.000	13.000	5.000	2.000	5.500	0.821
	Ar	4.429	1.338	1.651	3.847	3.039	2.705	3.516	2.208	1.717	7.193	3.461	1.039	3.012	0.489
	Ho	0.429	0.286	0.393	0.778	0.607	0.593	0.286	0.179	0.357	0.786	0.571	0.038	0.442	0.067
	He	0.774	0.253	0.394	0.740	0.671	0.630	0.716	0.547	0.418	0.861	0.711	0.038	0.563	0.070
	uHe	0.788	0.257	0.401	0.754	0.683	0.642	0.729	0.557	0.425	0.877	0.724	0.038	0.573	0.071
	F	0.446	-0.131	0.003	-0.051	0.095	0.060	0.601	0.674	0.145	0.087	0.196	-0.020	0.175	0.075
CAR	***N***	99	111	108	103	111	105	98	109	106	103	108	94	104.583	1.554
	Na	9.000	5.000	7.000	11.000	9.000	6.000	8.000	7.000	6.000	15.000	8.000	6.000	8.083	0.793
	Ar	4.776	1.613	2.200	5.020	2.924	3.305	2.998	2.271	1.801	5.195	3.702	1.261	3.089	0.390
	Ho	0.475	0.414	0.296	0.806	0.586	0.619	0.296	0.193	0.311	0.602	0.481	0.149	0.436	0.056
	He	0.791	0.380	0.545	0.801	0.658	0.697	0.666	0.560	0.445	0.808	0.730	0.207	0.607	0.054
	uHe	0.795	0.382	0.548	0.805	0.661	0.701	0.670	0.562	0.447	0.811	0.733	0.208	0.610	0.054
	F	0.400	-0.091	0.457	-0.006	0.110	0.112	0.556	0.656	0.300	0.255	0.340	0.280	0.281	0.064
GOM	***N***	5	5	5	4	5	3	5	5	5	5	5	4	4.667	0.188
	Na	4.000	2.000	3.000	5.000	2.000	2.000	4.000	3.000	2.000	8.000	3.000	1.000	3.250	0.538
	Ar	3.571	1.471	1.852	4.000	1.724	2.000	2.941	2.778	1.923	7.143	2.381	1.000	2.732	0.473
	Ho	0.000	0.400	0.200	0.750	0.200	0.333	0.600	0.400	0.400	0.800	0.200	0.000	0.357	0.075
	He	0.720	0.320	0.460	0.750	0.420	0.500	0.660	0.640	0.480	0.860	0.580	0.000	0.533	0.066
	uHe	0.800	0.356	0.511	0.857	0.467	0.600	0.733	0.711	0.533	0.956	0.644	0.000	0.597	0.074
	F	1.000	-0.250	0.565	0.000	0.524	0.333	0.091	0.375	0.167	0.070	0.655	#N/D	0.321	0.102
Overall	***Fis***	0.605	-0.155	0.365	-0.019	0.204	0.155	0.421	0.559	0.204	0.135	0.380	0.234	0.257	0.064
	Fit	0.622	-0.144	0.386	0.024	0.248	0.186	0.452	0.571	0.217	0.152	0.407	0.262	0.282	0.064
	Fst	0.044	0.010	0.034	0.042	0.056	0.037	0.054	0.027	0.017	0.020	0.044	0.037	0.035	0.004
	HWE	***	NS	***	NS	NS	NS	***	***	*	**	***	ND		
	Nm	5.497	25.032	7.063	5.661	4.234	6.482	4.400	8.853	14.830	12.101	5.455	6.454	8.839	1.734
	PIC	0.767	0.325	0.473	0.767	0.598	0.625	0.643	0.487	0.377	0.815	0.679	0.162		

Table S3. Inferred genetic composition of individuals across clusters and groupings inferred by STRUCTURE and DAPC.

STRUCTURE			DAPC		
St1	St2	St3	Ag1	Ag2	Ag3
oc-159	oc-160	oc-115	oc-45	oc-110	oc-101
oc-144	oc-126	oc-143	oc-296	oc-126	oc-115
oc-151	oc-157	oc-134	oc-201	oc-137	oc-168
oc-147	oc-129	oc-137	oc-231	oc-143	oc-121
oc-142	oc-140	oc-132	oc-39	oc-144	oc-131
oc-148	oc-135	oc-101	oc-183	oc-149	oc-132
oc-149	oc-154	oc-2	oc-254	oc-150	oc-134
oc-155	oc-150	oc-296	oc-298	oc-154	oc-186
oc-156	oc-118	oc-39	oc-299	oc-155	oc-2
oc-121	oc-113	oc-38	oc-304	oc-157	oc-272
oc-133	oc-110	oc-239	oc-215	oc-160	oc-279
oc-131	oc-3	oc-311	oc-251	oc-182	oc-290
oc-158	oc-1	oc-227	oc-239	oc-191	oc-311
oc-161	oc-179	oc-173	oc-241	oc-196	oc-227
oc-168	oc-253	oc-231	oc-243	oc-213	oc-12
oc-6	oc-27	oc-45	oc-245	oc-214	oc-38
oc-252	oc-28	oc-186	oc-247	oc-218	oc-287
oc-36	oc-30	oc-245	oc-259	oc-220	oc-223

oc-37	oc-41	oc-287	oc-260	oc-27	oc-240
oc-271	oc-31	oc-240	oc-261	oc-28	oc-250
oc-275	oc-278	oc-247	oc-264	oc-30	oc-113
oc-270	oc-170	oc-243	oc-266	oc-43	oc-118
oc-236	oc-211	oc-299	oc-343	oc-173	oc-129
oc-272	oc-43	oc-290	oc-381	oc-198	oc-135
oc-235	oc-188	oc-241	oc-334	oc-294	oc-140
oc-230	oc-200	oc-254	oc-335	oc-306	oc-179
oc-32	oc-300	oc-8	-	oc-35	oc-194
oc-229	oc-191	oc-16	-	oc-41	oc-203
oc-279	oc-35	oc-298	-	oc-184	oc-204
oc-215	oc-257	oc-251	-	oc-188	oc-219
oc-268	oc-196	oc-282	-	oc-255	oc-1
oc-190	oc-194	oc-207	-	oc-263	oc-3
oc-198	oc-289	oc-238	-	oc-281	oc-8
oc-7	oc-246	oc-381	-	oc-297	oc-31
oc-11	oc-210	oc-261	-	oc-246	oc-200
oc-9	oc-294	oc-203	-	oc-133	oc-253
oc-26	oc-209	oc-259	-	oc-142	oc-270
oc-21	oc-255	oc-260	-	oc-147	oc-278
oc-250	oc-189	oc-204	-	oc-148	oc-295
oc-178	oc-20	oc-267	-	oc-151	oc-20
oc-213	oc-295	oc-223	-	oc-156	oc-257

oc-297	oc-214	oc-266	-	oc-158	oc-268
oc-281	oc-12	oc-264	-	oc-159	oc-289
oc-284	oc-306	oc-183	-	oc-161	oc-300
oc-305	oc-220	oc-304	-	oc-211	oc-207
-	oc-218	oc-343	-	oc-6	oc-209
-	oc-219	oc-334	-	oc-7	oc-210
-	oc-263	oc-335	-	oc-26	oc-238
-	oc-184	oc-201	-	oc-32	-
-	oc-182	-	-	oc-170	-
-	-	-	-	oc-252	-
-	-	-	-	oc-271	-
-	-	-	-	oc-275	-
-	-	-	-	oc-229	-
-	-	-	-	oc-230	-
-	-	-	-	oc-235	-
-	-	-	-	oc-236	-
-	-	-	-	oc-9	-
-	-	-	-	oc-11	-
-	-	-	-	oc-16	-
-	-	-	-	oc-21	-
-	-	-	-	oc-36	-
-	-	-	-	oc-37	-
-	-	-	-	oc-178	-

-	-	-	-	oc-189	-
-	-	-	-	oc-190	-
-	-	-	-	oc-267	-
-	-	-	-	oc-282	-
-	-	-	-	oc-284	-
-	-	-	-	oc-305	-

Number of individuals

STRUCTURE			DAPC		
St1	St2	St3	Ag1	Ag2	Ag3
45	50	49	26	70	48

Table S4. Numbers of observations of mitochondrial partial control region (D-loop) haplotypes among 50 western Atlantic Ocean *Tetrapturus pfluegeri* individuals in each cluster defined by BAPS (Mt1, Mt2,. Mt3). Priv.: number of private haplotypes.

Haplotype	Haplotype observations		
	Mt1	Mt2	Mt3
Hap-1	1	-	-
Hap-2	-	1	-
Hap-3	-	2	-
Hap-4	3	-	-
Hap-5	8	-	-
Hap-6	1	-	-
Hap-7	2	-	-
Hap-8	2	-	-
Hap-9	2	-	-
Hap-10	1	-	-
Hap-11	1	-	-
Hap-12	4	-	-
Hap-13	2	-	-
Hap-14	1	-	-
Hap-15	2	-	-
Hap-16	2	-	-
Hap-17	2	-	-
Hap-18	-	-	1
Hap-19	-	1	-
Hap-20	-	-	3
Hap-21	-	3	-
Hap-22	4	-	-
Hap-23	1	-	-
Hap-24	4	-	-
Hap-25	1	-	-
Hap-26	1	-	-
Hap-27	1	-	-
Hap-28	6	-	-
Hap-29	-	1	-
Hap-30	2	-	-

Hap-31	3	-	-
Hap-32	1	-	-
Hap-33	1	-	-
Hap-34	-	1	-
Hap-35	1	-	-
Hap-36	1	-	-
Hap-37	1	-	-
Hap-38	1	-	-
Hap-39	2	-	-
Hap-40	1	-	-
Hap-41	8	-	-
Hap-42	1	-	-
Hap-43	-	-	1
Hap-44	1	-	-
Hap-45	-	2	-
Hap-46	1	-	-
Hap-47	1	-	-
Hap-48	-	1	-
Hap-49	1	-	-
Hap-50	1	-	-
Hap-51	3	-	-
Hap-52	2	-	-
Hap-53	1	-	-
Hap-54	1	-	-
Hap-55	-	1	-
Hap-56	-	1	-
Hap-57	1	-	-
Hap-58	1	-	-
Hap-59	1	-	-
Hap-60	-	-	1
Hap-61	2	-	-
Hap-62	-	1	-
Hap-63	1	-	-
Hap-64	2	-	-

Hap-65	2	-	-
Hap-66	1	-	-
Hap-67	1	-	-
Hap-68	1	-	-
Hap-69	-	1	-
Hap-70	1	-	-
Hap-71	1	-	-
Hap-72	1	-	-
Hap-73	-	1	-
Hap-74	1	-	-
Hap-75	1	-	-
Hap-76	2	-	-
Hap-77	1	-	-
Hap-78	1	-	-
Hap-79	1	-	-
Hap-80	-	1	
Hap-81	1	-	-
Hap-82	1	-	-
Hap-83	1	-	-
Hap-84	1	-	-
Hap-85	1	-	-
Hap-86	1	-	-
Hap-87	1	-	-
Hap-88	1	-	-
Hap-89	1	-	-
Hap-90	1	-	-
Hap-91	1	-	-
Hap-92	-	1	-
Hap-93	1	-	-
Hap-94	1	-	-
Hap-95	1	-	-
Hap-96	1	-	-
Priv.	77	15	4

Table S5. Numbers of observations of mtDNA partial control region (D-loop) haplotypes from 147 western Atlantic Ocean *Tetrapturus pfluegeri* individuals in each sampling region. Priv.: number of private haplotypes.

Haplotype	Region		
	North Atlantic	Gulf of Mexico	Caribbean Sea
Hap-1			1
Hap-2			1
Hap-3			2
Hap-4			1
Hap-5		2	4
Hap-6			1
Hap-7			2
Hap-8		1	1
Hap-9			2
Hap-10		1	
Hap-11		1	
Hap-12		1	3
Hap-13			1
Hap-14			
Hap-15			2
Hap-16			1
Hap-17			
Hap-18			
Hap-19			
Hap-20			2
Hap-21			1
Hap-22			3
Hap-23			
Hap-24			3
Hap-25			
Hap-26			
Hap-27			
Hap-28			4
Hap-29			
Hap-30			1
Hap-31			2
Hap-32			

Hap-33		
Hap-34		
Hap-35		
Hap-36		
Hap-37		
Hap-38		
Hap-39	1	1
Hap-40		1
Hap-41		8
Hap-42		1
Hap-43		1
Hap-44		1
Hap-45		2
Hap-46		1
Hap-47		1
Hap-48		1
Hap-49		1
Hap-50		1
Hap-51		3
Hap-52		2
Hap-53		1
Hap-54		1
Hap-55		1
Hap-56		1
Hap-57		1
Hap-58		1
Hap-59		1
Hap-60		1
Hap-61		2
Hap-62		1
Hap-63		1
Hap-64		2
Hap-65		2
Hap-66		1
Hap-67		1
Hap-68		1
Hap-69		1

Hap-70			1
Hap-71			1
Hap-72			1
Hap-73			1
Hap-74			1
Hap-75			1
Hap-76			2
Hap-77			1
Hap-78			1
Hap-79			1
Hap-80			1
Hap-81			1
Hap-82			1
Hap-83			1
Hap-84			1
Hap-85			1
Hap-86			1
Hap-87			1
Hap-88			1
Hap-89			1
Hap-90	1		
Hap-91	1		
Hap-92			1
Hap-93			1
Hap-94			1
Hap-95			1
Hap-96			1
Priv.	2	2	62