

# Spatial and temporal boundaries to gene flow between *Chaenocephalus aceratus* populations at South Orkney and South Shetlands

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**Appendix 1.** Differentiation tests between population samples were performed using CHIFISH ver. 1.3 (Ryman 2006). The software calculates chi-squared tests at each locus, sums the test statistics and their associated degrees of freedom, directly from the genotypic data file, and provides p-values associated with these sums (Ryman 2006). The p-values from Fisher's exact test, estimated using a Markov chain algorithm (Raymond & Rousset 1995), are combined by means of Fisher's method. p-values were obtained by 500 000 iterations (100 batches with 5000 iterations per batch). Ryman & Jorde (2001) noted that the Fisher's approach might in some settings (small contingency tables with few populations and few alleles) result in low statistical power, potentially generating a high type II error rate. The chi-squared test can be a better alternative to Fisher's method, being very powerful even with weak population differentiation (Ryman & Jorde 2001, Waples & Gaggiotti 2006).

Table A1. p-values of population differentiation for and across all loci. Fisher test (p(Fisher)), relative standard errors (SEp), chi-squared ( $\chi^2$ ) and the degrees of freedom (df) values are reported for (a) all populations, (b) EI02 against SS02, (c) EI02 to EI06, (d) EI02 to SO06, (e) EI06 to SS02, (f) EI06 to SO06, (g) SS02 to SO06, (h) EI02+EI06-SO06– and (i) EI02+EI06-SS02. Fisher's and chi-squared tests for population differentiation were performed including (Total) and excluding (–noHWE) loci that deviated from Hardy-Weinberg equilibrium

Locus	p(Fisher)	SE(p)	$\chi^2$	df	p( $\chi^2$ )
(a)					
<b>All populations</b>					
Ca21	<0.00001	0.00000	773.508	276	<0.00001
Ca26	0.04239	0.00588	96.1770	81	0.11964
Ca40	0.02076	0.00388	185.616	144	0.01109
Ca48	0.21630	0.00800	27.3520	18	0.07263
Ca55	0.13661	0.00844	99.3550	81	0.08119
Ca86	0.00239	0.00087	176.633	129	0.00344
Ca88	0.00024	0.00010	97.7930	54	0.00025
Cr15	0.26436	0.01071	16.1340	15	0.37319
Cr127	0.40404	0.00975	26.2630	24	0.33993
Cr171	<0.00001	0.00000	377.612	174	<0.00001
Cr259	0.03069	0.00437	88.7890	63	0.01786
Total	<0.00001		1965.232	1059	<0.00001
(–noHWE)	0.00007		539.7070	402	0.00001

## Appendix 1 (continued)

Locus	p(Fisher)	SE(p)	$\chi^2$	df	p( $\chi^2$ )
<b>(b)</b>					
<b>EI02 to SS02</b>					
Ca21	0.10999	0.00657	77.766	59	0.05131
Ca26	0.08361	0.00507	33.013	26	0.16172
Ca40	0.79944	0.00657	32.864	43	0.86854
Ca48	0.33076	0.00393	6.4700	5	0.26313
Ca55	0.12524	0.00485	32.871	26	0.16588
Ca86	0.47545	0.01021	41.111	41	0.46574
Ca88	0.61771	0.00586	15.136	15	0.44169
Cr15	0.18210	0.00369	5.5680	4	0.23379
Cr127	0.89407	0.00253	3.3240	8	0.91245
Cr171	0.03470	0.00343	59.722	44	0.05717
Cr259	0.40691	0.00733	20.795	20	0.40931
Total	0.10040		328.640	291	0.06363
(-noHWE)	0.23417		137.493	125	0.20974
<b>(c)</b>					
<b>EI02 to EI06</b>					
Ca21	0.39860	0.00578	5.90300	5	0.31581
Ca26	<0.00001	0.00000	370.525	80	<0.00001
Ca40	0.78370	0.00584	19.3850	25	0.77832
Ca48	0.07068	0.00381	64.1070	45	0.03202
Ca55	0.09413	0.00232	11.2740	5	0.04621
Ca86	0.28723	0.00667	31.7690	26	0.20086
Ca88	0.16170	0.00710	51.5170	41	0.12570
Cr15	0.00500	0.00075	35.8710	16	0.00302
Cr127	0.43018	0.00618	8.01300	8	0.43218
Cr171	<0.00001	0.00000	136.633	43	<0.00001
Cr259	0.54703	0.00759	19.5200	20	0.48826
Total	<0.00001		754.518	314	<0.00001
(-noHWE)	0.01968		163.731	126	0.01339
<b>(d)</b>					
<b>EI02 to SO06</b>					
Ca21	0.72826	0.00376	2.05700	4	0.72523
Ca26	<0.00001	0.00000	341.439	77	<0.00001
Ca40	0.03456	0.00300	39.4050	25	0.03354
Ca48	0.00506	0.00091	74.8240	46	0.00459
Ca55	0.20495	0.00293	5.14900	4	0.27230
Ca86	0.82304	0.00484	20.2210	25	0.73514
Ca88	0.00406	0.00071	68.7970	43	0.00749
Cr15	0.00229	0.00041	39.4300	16	0.00094
Cr127	0.14401	0.00402	12.3020	8	0.13823
Cr171	<0.00001	0.00000	148.182	53	<0.00001
Cr259	0.00426	0.00115	45.6080	21	0.00144
Total	<0.00001		797.415	322	<0.00001
(-noHWE)	0.00050		187.361	125	0.00026
<b>(e)</b>					
<b>EI06 to SS02</b>					
Ca21	0.01197	0.00066	7.12200	2	0.02841
Ca26	<0.00001	0.00000	149.132	61	<0.00001
Ca40	0.33343	0.00645	22.5390	21	0.36906
Ca48	0.43606	0.00758	37.5370	36	0.39857
Ca55	0.78988	0.00193	3.34800	5	0.64658
Ca86	0.02168	0.00162	35.9760	25	0.07196
Ca88	0.00873	0.00101	41.6850	25	0.01942
Cr15	0.00335	0.00047	31.2530	15	0.00812
Cr127	0.27201	0.00472	8.53500	7	0.28777
Cr171	<0.00001	0.00000	76.8720	38	0.00019
Cr259	0.62015	0.00589	16.9300	19	0.59462
Total	<0.00001		430.929	254	<0.00001
(-noHWE)	0.00007		150.458	100	0.00083

Locus	p(Fisher)	SE(p)	$\chi^2$	df	p( $\chi^2$ )
(f)					
<b>EI06 to SO06</b>					
Ca21	0.13349	0.00228	3.29400	2	0.19263
Ca26	0.00074	0.00026	74.2240	53	0.02874
Ca40	0.09837	0.00370	24.7260	19	0.16976
Ca48	0.01091	0.00106	58.9560	40	0.02701
Ca55	0.73757	0.00205	3.51800	5	0.62066
Ca86	0.02406	0.00170	35.8120	23	0.04311
Ca88	0.00196	0.00031	42.5340	24	0.01124
Cr15	0.34117	0.00628	18.4140	17	0.36315
Cr127	0.09268	0.00278	11.9170	7	0.10333
Cr171	0.00028	0.00016	78.8260	51	0.00746
Cr259	0.00611	0.00068	33.7580	20	0.02781
Total	<0.00001		385.980	261	<0.00001
(-noHWE)	0.00131		140.215	97	0.00272
(g)					
<b>SS02 to SO06</b>					
Ca21	0.31714	0.00187	1.17900	1	0.27758
Ca26	<0.00001	0.00000	138.425	55	<0.00001
Ca40	0.00054	0.00022	39.9830	21	0.00747
Ca48	0.14905	0.00532	44.3310	37	0.18997
Ca55	0.90860	0.00128	2.55600	5	0.76804
Ca86	0.35067	0.00598	24.0680	22	0.34370
Ca88	0.05656	0.00271	35.9080	26	0.09337
Cr15	0.00765	0.00068	30.1760	16	0.01711
Cr127	0.51895	0.00399	5.46200	6	0.48607
Cr171	<0.00001	0.00000	81.0060	46	0.00110
Cr259	0.03119	0.00173	28.4260	19	0.07557
Total	<0.00001		431.518	254	<0.00001
(-noHWE)	0.00106		139.331	97	0.00316
(h)					
<b>EI02+EI06-SO06</b>					
Ca21	0.06047	0.00127	10.098	5	0.07251
Ca26	0.21570	0.00735	53.884	46	0.19826
Ca40	<0.00001	0.00000	64.855	33	0.00077
Ca48	0.00012	0.00009	42.804	19	0.00138
Ca55	0.00368	0.00048	36.098	18	0.00685
Ca86	0.00001	0.00001	86.363	48	0.00057
Ca88	0.66927	0.00160	1.2410	2	0.53773
Cr15	0.36421	0.00112	1.0900	1	0.29636
Cr127	1.00000	0.00000	0.2100	4	0.99484
Cr171	0.00658	0.00074	41.665	24	0.01406
Cr259	<0.00001	0.00000	59.937	19	<0.00001
Total	<0.00001		398.246	219	<0.00001
(-noHWE)	<0.00001		229.457	82	<0.00001
(i)					
<b>EI02+EI06-SS02</b>					
Ca21	0.14625	0.00558	13.182	10	0.21368
Ca26	<0.00001	0.00000	528.569	168	<0.00001
Ca40	0.30591	0.01239	56.649	54	0.37645
Ca48	0.28172	0.01301	104.323	90	0.14349
Ca55	0.18510	0.00571	20.252	12	0.06247
Ca86	0.06375	0.00479	72.041	54	0.05093
Ca88	0.12781	0.00830	99.818	82	0.08805
Cr15	0.01345	0.00154	52.507	32	0.01260
Cr127	0.68351	0.00681	12.575	16	0.70356
Cr171	<0.00001	0.00000	212.770	94	<0.00001
Cr259	0.53224	0.00987	40.269	40	0.45835
Total	<0.00001		1212.954	652	<0.00001
(-noHW)	0.11041		153.299	130	0.07961

## LITERATURE CITED

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