Supplementary Information Along-shelf connectivity and circumpolar gene flow in Antarctic silverfish (Pleuragramma antarctica)

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Table S1 Genetic variability of *Pleuragramma antarctica* for each locus across all nineteen sampling locations. Sample size (*n*), number of alleles (N_A), % of total observed alleles per locus per sampling location, allelic richness based on a minimum sample size of 14 individuals (A_R), observed heterozygosity (H_O), unbiased heterozygosity (H_E) and probability of deviation from Hardy-Weinberg equilibrium (*pHWE*) are shown. Values in bold indicate significant HWE deviations after correction for multiple tests as implemented in SGoF+¹ (threshold for significance with 304 comparisons P = 0.008). Sampling location acronyms are as in Table 1.

				CI1	0						MB()1						MBO	2		
Locus	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	$N_{\rm A}$	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE
Ch126	60	3	42.86	2.21	0.45	0.51	0.300	28	3	42.86	2.38	0.46	0.51	0.711	49	4	57.14	3.00	0.59	0.55	0.763
Ch520	60	4	50.00	3.09	0.33	0.38	0.488	28	4	50.00	3.38	0.39	0.51	0.020	49	5	62.50	3.70	0.47	0.48	0.754
Ch623	60	23	69.70	14.35	0.92	0.94	<0.001	28	20	60.61	12.75	0.96	0.92	0.219	49	20	60.61	12.82	0.90	0.92	0.975
Ch10105	60	6	66.67	3.54	0.30	0.31	0.146	28	3	33.33	1.86	0.07	0.07	1.000	49	5	55.56	2.89	0.24	0.24	0.182
Ch10857	60	9	52.94	6.34	0.68	0.69	0.903	28	9	52.94	6.03	0.54	0.66	0.091	49	7	41.18	5.59	0.57	0.67	0.702
Ch13222	60	3	50.00	2.26	0.17	0.16	1.000	28	2	33.33	1.94	0.18	0.16	1.000	49	4	66.67	1.75	0.06	0.06	1.000
Ch18085	60	10	71.43	7.29	0.83	0.83	0.852	28	9	64.29	6.98	0.86	0.82	0.790	49	9	64.29	7.02	0.86	0.83	0.998
Ch2931	60	10	58.82	6.96	0.73	0.83	0.891	28	10	58.82	7.55	0.86	0.82	0.165	49	11	64.71	7.09	0.84	0.82	0.370
Ch4796	60	13	72.22	9.07	0.75	0.87	0.002	28	12	66.67	9.71	0.96	0.88	0.985	49	14	77.78	10.09	0.94	0.89	0.793
Ch10441	60	9	39.13	5.36	0.65	0.65	0.643	28	10	43.48	7.17	0.86	0.76	0.535	49	9	39.13	6.00	0.61	0.68	0.524
Ch11230	60	7	50.00	4.90	0.77	0.72	0.744	28	5	35.71	4.69	0.71	0.71	0.996	49	7	50.00	5.00	0.78	0.72	1.000
Ch11483	60	10	66.67	5.43	0.47	0.56	0.675	28	9	60.00	6.26	0.57	0.64	0.409	49	9	60.00	4.67	0.45	0.46	0.096
Ch17977	60	8	53.33	5.08	0.58	0.63	0.722	28	4	26.67	3.60	0.57	0.56	0.955	49	7	46.67	4.60	0.65	0.62	0.509
Ch19846	60	8	66.67	6.26	0.70	0.78	0.690	28	8	66.67	6.56	0.86	0.78	0.996	49	8	66.67	6.52	0.86	0.80	0.941
Ch24332	60	6	60.00	4.02	0.60	0.62	0.096	28	5	50.00	3.92	0.57	0.60	0.320	49	5	50.00	3.70	0.73	0.62	0.184
Ch25478	60	4	25.00	2.96	0.57	0.56	0.422	28	4	25.00	3.04	0.46	0.56	0.036	49	6	37.50	3.69	0.61	0.63	0.341

				MB1	.0						MB1	1						JI0'	7		
Locus	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\mathbf{R}}$	H_0	$H_{\rm E}$	pHWE
Ch126	60	4	57.14	2.40	0.43	0.52	0.205	83	3	42.86	2.77	0.58	0.55	0.661	34	5	71.43	3.22	0.56	0.54	1.000
Ch520	60	5	62.50	4.07	0.62	0.56	0.855	83	5	62.50	3.58	0.45	0.40	0.712	34	3	37.50	2.95	0.50	0.41	0.362
Ch623	60	24	72.73	13.71	0.93	0.93	0.038	83	25	75.76	13.58	0.89	0.93	0.012	34	22	66.67	13.01	0.97	0.92	0.106
Ch10105	60	4	44.44	2.88	0.22	0.21	0.594	83	5	55.56	3.16	0.25	0.23	0.680	34	4	44.44	2.99	0.24	0.22	1.000
Ch10857	60	11	64.71	6.36	0.75	0.73	0.121	83	10	58.82	6.32	0.73	0.71	0.937	34	7	41.18	5.29	0.76	0.74	0.716
Ch13222	60	4	66.67	2.40	0.25	0.22	1.000	83	3	50.00	1.94	0.10	0.10	<0.001	34	3	50.00	2.31	0.21	0.23	1.000
Ch18085	60	10	71.43	7.21	0.83	0.83	0.171	83	11	78.57	7.05	0.87	0.82	0.959	34	10	71.43	7.47	0.76	0.82	0.715
Ch2931	60	10	58.82	7.52	0.83	0.81	0.998	83	10	58.82	6.91	0.73	0.81	0.279	34	10	58.82	7.61	0.71	0.84	0.333
Ch4796	60	13	72.22	9.37	0.82	0.88	0.996	83	14	77.78	10.72	0.93	0.90	1.000	34	11	61.11	9.11	1.00	0.88	1.000
Ch10441	60	11	47.83	7.20	0.73	0.71	0.436	83	12	52.17	6.09	0.69	0.68	0.989	34	9	39.13	5.85	0.74	0.66	0.289
Ch11230	60	8	57.14	5.68	0.68	0.76	0.986	83	6	42.86	4.64	0.80	0.73	0.603	34	7	50.00	5.71	0.88	0.76	0.547
Ch11483	60	8	53.33	5.20	0.65	0.57	0.597	83	8	53.33	4.26	0.55	0.56	0.461	34	5	33.33	3.77	0.59	0.48	0.606
Ch17977	60	9	60.00	5.13	0.62	0.66	<0.001	83	10	66.67	4.85	0.58	0.58	0.013	34	7	46.67	5.73	0.68	0.69	0.828
Ch19846	60	8	66.67	6.45	0.87	0.80	0.216	83	8	66.67	6.04	0.71	0.78	0.618	34	7	58.33	5.72	0.76	0.77	0.385
Ch24332	60	6	60.00	4.64	0.60	0.65	0.934	83	7	70.00	4.55	0.69	0.63	0.914	34	4	40.00	3.79	0.62	0.64	0.800
Ch25478	60	6	37.50	4.43	0.68	0.64	0.018	83	8	50.00	4.45	0.64	0.63	0.686	34	7	43.75	4.12	0.47	0.61	0.019

				JI10							Л1	2						SSI9	96		
Locus	n	NA	%	$A_{\mathbf{R}}$	Ho	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\mathbf{R}}$	H_0	$H_{\rm E}$	pHWE
Ch126	148	6	85.71	3.08	0.63	0.55	0.384	54	5	71.43	3.00	0.50	0.54	1.000	14	2	28.57	2.00	0.57	0.49	0.620
Ch520	148	8	100.00	4.15	0.57	0.58	0.713	54	6	75.00	3.87	0.44	0.51	0.210	14	5	62.50	4.15	0.50	0.42	1.000
Ch623	148	28	84.85	13.77	0.91	0.94	0.574	54	20	60.61	12.56	0.80	0.92	0.092	14	15	45.45	11.59	0.86	0.91	<0.001
Ch10105	148	6	66.67	3.72	0.32	0.37	0.074	54	5	55.56	3.35	0.20	0.25	0.359	14	3	33.33	2.66	0.36	0.30	1.000
Ch10857	148	12	70.59	6.60	0.70	0.74	0.858	54	11	64.71	6.33	0.74	0.71	0.090	14	5	29.41	4.59	0.64	0.69	0.170
Ch13222	148	4	66.67	2.26	0.16	0.17	0.718	54	3	50.00	2.10	0.15	0.14	1.000	14	3	50.00	2.29	0.14	0.14	1.000
Ch18085	148	12	85.71	7.69	0.84	0.85	0.931	54	8	57.14	6.78	0.85	0.81	0.507	14	6	42.86	5.74	0.71	0.80	0.879
Ch2931	148	10	58.82	7.02	0.78	0.81	0.297	54	8	47.06	6.19	0.67	0.82	0.999	14	7	41.18	6.39	0.79	0.82	0.475
Ch4796	148	14	77.78	9.56	0.82	0.89	0.491	54	12	66.67	8.86	0.98	0.87	0.991	14	11	61.11	8.89	0.93	0.86	0.009
Ch10441	148	17	73.91	6.54	0.67	0.70	<0.001	54	7	30.43	5.13	0.67	0.66	0.910	14	6	26.09	5.48	0.64	0.72	0.136
Ch11230	148	9	64.29	5.23	0.74	0.74	0.860	54	7	50.00	5.18	0.69	0.70	0.222	14	5	35.71	4.59	0.57	0.73	0.842
Ch11483	148	12	80.00	5.06	0.55	0.58	<0.001	54	8	53.33	4.43	0.54	0.57	0.739	14	5	33.33	3.80	0.29	0.37	0.082
Ch17977	148	9	60.00	5.20	0.60	0.63	0.035	54	8	53.33	5.52	0.63	0.63	0.308	14	4	26.67	3.53	0.64	0.56	0.663
Ch19846	148	10	83.33	6.36	0.80	0.79	0.250	54	8	66.67	6.50	0.78	0.79	0.386	14	8	66.67	7.07	0.86	0.83	0.811
Ch24332	148	8	80.00	4.87	0.62	0.65	0.842	54	6	60.00	3.80	0.57	0.59	0.013	14	5	50.00	3.97	0.71	0.62	0.006
Ch25478	148	9	56.25	4.45	0.67	0.63	0.267	54	5	31.25	3.39	0.56	0.59	0.023	14	3	18.75	2.65	0.50	0.53	0.594

				SOI	1						LB0	7						LB1	1		
Locus	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE
Ch126	47	3	42.86	2.24	0.40	0.50	0.140	46	3	42.86	2.72	0.65	0.49	0.152	52	3	42.86	2.57	0.60	0.52	0.879
Ch520	47	6	75.00	4.54	0.51	0.60	0.108	46	5	62.50	3.96	0.50	0.60	0.328	52	7	87.50	4.52	0.58	0.57	0.948
Ch623	47	18	54.55	12.21	0.91	0.92	1.000	46	20	60.61	12.82	0.87	0.93	0.997	52	21	63.64	13.57	0.88	0.93	1.000
Ch10105	47	6	66.67	3.93	0.36	0.35	0.329	46	4	44.44	2.66	0.17	0.18	0.001	52	5	55.56	2.88	0.23	0.21	1.000
Ch10857	47	9	52.94	6.25	0.72	0.66	0.938	46	9	52.94	5.74	0.54	0.63	0.816	52	10	58.82	6.23	0.73	0.74	0.713
Ch13222	47	3	50.00	2.24	0.19	0.21	1.000	46	3	50.00	1.86	0.04	0.12	0.001	52	2	33.33	1.91	0.15	0.14	1.000
Ch18085	47	8	57.14	6.59	0.91	0.80	0.997	46	8	57.14	6.69	0.83	0.82	0.861	52	9	64.29	7.20	0.88	0.82	0.869
Ch2931	47	9	52.94	6.75	0.70	0.82	0.444	46	11	64.71	7.75	0.78	0.83	0.939	52	9	52.94	6.96	0.77	0.83	0.172
Ch4796	47	12	66.67	8.74	0.79	0.87	0.902	46	10	55.56	7.88	0.91	0.86	0.921	52	12	66.67	9.13	0.92	0.88	1.000
Ch10441	47	7	30.43	4.88	0.49	0.56	0.111	46	11	47.83	6.74	0.70	0.70	0.142	52	10	43.48	6.07	0.58	0.63	0.046
Ch11230	47	6	42.86	4.79	0.64	0.68	0.847	46	6	42.86	5.12	0.70	0.74	0.781	52	7	50.00	5.19	0.73	0.72	0.105
Ch11483	47	7	46.67	4.85	0.60	0.55	0.764	46	7	46.67	4.63	0.61	0.55	0.597	52	7	46.67	4.49	0.48	0.50	0.001
Ch17977	47	8	53.33	5.58	0.60	0.59	0.728	46	6	40.00	4.38	0.54	0.57	0.982	52	8	53.33	4.82	0.67	0.64	0.996
Ch19846	47	7	58.33	5.79	0.66	0.73	0.744	46	9	75.00	6.46	0.83	0.78	0.587	52	10	83.33	7.11	0.88	0.82	0.852
Ch24332	47	8	80.00	4.81	0.64	0.67	0.757	46	6	60.00	4.35	0.57	0.65	0.579	52	8	80.00	4.97	0.60	0.69	0.455
Ch25478	47	8	50.00	4.50	0.55	0.64	<0.001	46	5	31.25	3.79	0.67	0.59	0.784	52	7	43.75	3.82	0.50	0.59	0.005

				FT1	4						HB	89						HB9	1		
Locus	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE
Ch126	50	4	57.14	3.07	0.50	0.56	1.000	19	2	28.57	2.00	0.53	0.49	1.000	41	3	42.86	2.49	0.51	0.51	1.000
Ch520	50	5	62.50	3.79	0.56	0.49	0.791	19	4	50.00	3.91	0.63	0.60	0.228	41	7	87.50	4.67	0.51	0.57	<0.001
Ch623	50	17	51.52	12.04	0.96	0.92	0.025	19	11	33.33	8.73	0.74	0.85	0.516	41	21	63.64	13.07	0.98	0.93	0.569
Ch10105	50	4	44.44	2.69	0.18	0.17	1.000	19	4	44.44	3.69	0.47	0.46	0.545	41	5	55.56	3.21	0.27	0.28	0.364
Ch10857	50	8	47.06	5.62	0.58	0.70	0.389	19	6	35.29	5.32	0.74	0.65	0.487	41	8	47.06	6.51	0.78	0.75	0.773
Ch13222	50	2	33.33	1.99	0.30	0.26	0.046	19	3	50.00	2.36	0.16	0.15	1.000	41	4	66.67	2.45	0.15	0.14	1.000
Ch18085	50	10	71.43	7.23	0.72	0.83	0.121	19	9	64.29	7.46	0.89	0.83	0.534	41	7	50.00	6.59	0.78	0.82	0.999
Ch2931	50	11	64.71	7.55	0.80	0.83	0.553	19	8	47.06	7.06	0.63	0.82	0.574	41	9	52.94	6.72	0.63	0.81	0.249
Ch4796	50	13	72.22	9.41	0.96	0.88	0.999	19	9	50.00	7.77	0.84	0.86	1.000	41	13	72.22	9.69	0.98	0.88	1.000
Ch10441	50	8	34.78	5.72	0.62	0.64	0.855	19	8	34.78	6.19	0.84	0.73	0.102	41	7	30.43	5.15	0.46	0.62	<0.001
Ch11230	50	6	42.86	4.86	0.64	0.70	0.811	19	6	42.86	5.11	0.63	0.71	0.884	41	7	50.00	5.55	0.71	0.73	0.698
Ch11483	50	8	53.33	5.34	0.52	0.54	0.772	19	6	40.00	4.42	0.47	0.58	<0.001	41	6	40.00	4.06	0.56	0.54	0.543
Ch17977	50	8	53.33	5.08	0.58	0.59	0.082	19	4	26.67	3.70	0.58	0.54	0.690	41	6	40.00	4.47	0.56	0.63	0.419
Ch19846	50	9	75.00	6.60	0.74	0.81	0.999	19	8	66.67	6.63	0.89	0.80	0.075	41	10	83.33	7.17	0.80	0.81	0.996
Ch24332	50	8	80.00	4.91	0.68	0.65	1.000	19	4	40.00	3.52	0.42	0.63	0.011	41	6	60.00	4.31	0.76	0.64	0.666
Ch25478	50	8	50.00	4.53	0.66	0.62	0.975	19	6	37.50	4.13	0.47	0.55	1.000	41	7	43.75	4.47	0.68	0.64	0.542

				HB1	4						AB1	4						RS96	6		
Locus	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE	n	NA	%	$A_{\rm R}$	H_0	$H_{\rm E}$	pHWE
Ch126	82	5	71.43	2.81	0.57	0.53	0.017	25	3	42.86	2.84	0.60	0.55	1.000	91	3	42.86	2.62	0.55	0.52	0.331
Ch520	82	7	87.50	4.23	0.57	0.54	0.391	25	5	62.50	3.76	0.48	0.47	<0.001	91	7	87.50	4.04	0.58	0.52	0.069
Ch623	82	21	63.64	12.94	0.87	0.93	0.946	25	15	45.45	11.64	0.96	0.91	1.000	91	23	69.70	13.70	0.88	0.93	0.988
Ch10105	82	4	44.44	3.18	0.27	0.27	0.159	25	4	44.44	2.33	0.12	0.12	1.000	91	5	55.56	3.27	0.25	0.26	0.600
Ch10857	82	8	47.06	5.38	0.57	0.69	0.931	25	8	47.06	6.06	0.76	0.73	0.480	91	11	64.71	6.70	0.71	0.76	0.625
Ch13222	82	4	66.67	2.50	0.20	0.20	1.000	25	2	33.33	1.96	0.16	0.21	0.335	91	3	50.00	2.20	0.16	0.18	0.203
Ch18085	82	9	64.29	7.30	0.83	0.83	0.996	25	7	50.00	6.24	0.84	0.81	0.997	91	11	78.57	7.69	0.89	0.84	1.000
Ch2931	82	12	70.59	6.92	0.73	0.81	0.795	25	9	52.94	7.06	0.80	0.83	0.008	91	12	70.59	6.85	0.68	0.79	0.630
Ch4796	82	15	83.33	9.70	0.89	0.88	0.166	25	10	55.56	7.67	0.88	0.85	<0.001	91	14	77.78	9.73	0.91	0.89	1.000
Ch10441	82	10	43.48	5.54	0.48	0.64	0.010	25	7	30.43	5.48	0.68	0.60	0.973	91	11	47.83	6.12	0.67	0.71	0.827
Ch11230	82	8	57.14	5.14	0.67	0.72	0.978	25	6	42.86	4.59	0.68	0.70	0.004	91	7	50.00	4.79	0.70	0.68	0.831
Ch11483	82	10	66.67	4.60	0.54	0.54	0.543	25	6	40.00	4.71	0.68	0.58	0.308	91	9	60.00	4.82	0.55	0.55	0.966
Ch17977	82	7	46.67	4.71	0.62	0.61	0.777	25	7	46.67	5.08	0.76	0.63	0.021	91	9	60.00	5.22	0.62	0.62	0.940
Ch19846	82	10	83.33	7.23	0.78	0.82	0.477	25	7	58.33	6.13	0.68	0.79	0.088	91	10	83.33	6.88	0.87	0.80	0.997
Ch24332	82	8	80.00	4.72	0.60	0.65	0.315	25	5	50.00	3.57	0.64	0.57	0.422	91	10	100.00	4.87	0.64	0.67	0.564
Ch25478	82	9	56.25	3.70	0.50	0.58	0.208	25	6	37.50	4.21	0.68	0.62	0.005	91	9	56.25	4.37	0.63	0.61	0.970

				RS9	7		
Locus	n	NA	%	$A_{\rm R}$	Ho	$H_{\rm E}$	pHWE
Ch126	84	3	42.86	2.60	0.54	0.53	1.000
Ch520	84	6	75.00	3.88	0.42	0.48	0.393
Ch623	84	24	72.73	13.54	0.93	0.93	0.786
Ch10105	84	4	44.44	3.12	0.26	0.25	0.759
Ch10857	84	10	58.82	6.29	0.73	0.76	0.065
Ch13222	84	3	50.00	2.27	0.23	0.23	0.311
Ch18085	84	10	71.43	7.83	0.86	0.83	1.000
Ch2931	84	9	52.94	6.44	0.68	0.79	0.883
Ch4796	84	15	83.33	9.44	0.83	0.87	0.398
Ch10441	84	10	43.48	6.04	0.60	0.64	0.517
Ch11230	84	8	57.14	4.72	0.71	0.68	0.002
Ch11483	84	8	53.33	4.19	0.55	0.51	0.740
Ch17977	84	8	53.33	5.09	0.57	0.62	0.377
Ch19846	84	10	83.33	6.88	0.82	0.80	0.870
Ch24332	84	7	70.00	4.35	0.68	0.64	1.000
Ch25478	84	8	50.00	4.01	0.60	0.60	0.111

Table S2 Primer sequences and amplification conditions used to amplify 16 EST-linked microsatellite loci in *Pleuragramma antarctica*. Locus names, repeat motifs, primer pair sequences, fluorescent dye of forward primer, size range of fragments in base pairs, annealing temperature for single-locus amplification, and the respective multiplex reaction set are shown (adapted from Agostini *et al.*²).

Locus	Motif	Primer sequences (5' - 3')	Dye	Size range	T ^a (°C)	Locus set for multiplexing
Ch10105 ^b	(CTG)12	F: TGC CTT GGT TAG GAT TAA ACG T	6-FAM	96 - 131	57	1
		R: AGA AGT GCT CCA TCA AGT CCA				
Ch10441 ^b	(CA)10	F: GTC CTT ACC TGG CAG TGC AGA	PET	110 - 162	60	2
		R: GCA CAT CAG TCC AAA ATC GGT G				
Ch10857 ^b	(CA)9	F: GCT TAA ATC ACC ATG TGC CCA	6-FAM	161 - 195	57	1
		R: TGG ACA TGA ATG TAC CAA AAC G				
Ch11230 ^b	(GA) ₇	F: ATC ACA CAG CTG ACT GGG GCT	NED	90 - 116	60	2
		R: TGA GAC TCG TCC GCG AAT GGA				
Ch11483 ^b	(TAA) ₇	F: ATC AAA CTG AAC AGC CTG TGT	NED	124 - 168	57	2
		R: TGG AGA ACA GGA ATG GAC AGA				
Ch126 ^c	(AAT) ₇	F: CGG TTT TTA TGC ATG TTG CCA	NED	264 - 282	56	1
		R: ACT GCT CAT TCA CAC TGG TTC				
Ch13222 ^b	(TTG) ₆	F: GTC CCA TGG TGA CTG ATA GGT	PET	82 - 98	57	1
		R: ACG AGT CAA TGT AAC CGG AAG				
Ch17977 ^b	(GAT) ₈	F: TTG ACT GAC AGC TTG GGT GCA	6-FAM	162 - 234	57	2
		R: GAA TGA GCC ATG TTG AGC CC				
Ch18085 ^b	(TGA) ₆	F: TTT AGG GGT GGC ACA TTG GAC	VIC	98 - 137	57	1
		R: ACA AAC CTT CTC CTT GTC TTC T				
Ch19846 ^b	(GT)14	F: CGA CCT TTG AGA AAG GCG GCA	6-FAM	116 - 141	60	2
		R: CGA CGT GTA TCA CAA GGG TCA				
Ch24332 ^b	(GCA) ₈	F: ATC GGT TGT CGT CCT CCA CAC	6-FAM	67 - 96	60	2
		R: GGC TGT TTC TGG ATC AGC GGT				
Ch25478 ^b	(AC) ₈	F: ACA GTG TTG TCT TGG AGA GGT	PET	143 - 175	57	2
		R: GGA GAG AAG TAC ATG AGG AGG A				
Ch2931 ^b	(TCT)8t-(TTC) ₆	F: GGG CTT TCA GGA GCA TCG GGA	VIC	79 - 132	60	2
		R: ACT TGA ACC TGA CGT GGC AAC				
Ch4796 ^b	(GAT) ₈	F: ACA ATG GTT GGT GAG AGC TCC	VIC	156 - 208	57	2
		R: TGA GTT AAG CAG AAC AAG TGC				
Ch520 ^b	(TC)7	F: GGA ACA ACT TGA GCC AAG ACA	6-FAM	259 - 273	57	1
		R: CCA TAA AAG TGC ATC ATC GCT				
Ch623 ^c	(AAC)7	F: GCT GTT TGA TTC CCT CGT GAG G	PET	174 - 295	62	1
		R: AAA AGT GGT CCT CCG CTG CAG T				

^a Reported annealing temperature refers to single-locus PCR. Loci were amplified together in two multiplex reactions. Multiplex PCR reaction volume was 10 μ L, containing 1x QIAGEN Multiplex PCR Master mix (HotStartTaq DNA Polymerase, Multiplex PCR Buffer, dNTP Mix; QIAGEN, Hilden, Germany), 0.2 μ M primer mix and 100 ng of genomic DNA. The PCR amplification profile for all loci consisted of: (1) an initial activation step of 15 min at 95 °C; (2) 30 cycles of denaturation at 94 °C for 30 s, annealing at 57 °C for 90 s and extension at 72 °C for 60 s; and (3) a final extension of 30 min at 60 °C.

^b Molecular Ecology Resources Primer Development Consortium et al.³

^c Molecular Ecology Resources Primer Development Consortium et al.⁴

Table S3 Analysis of Molecular Variance (AMOVA) among 19 populations of *Pleuragramma antarctica*, among six groups based on geographic origin, and among individuals within populations. Global results for fixation indices F_{ST} , F_{SC} , and F_{CT} reported in upper table, results by locus reported in lower table.

Source of variation	df	Proportion of variation	<i>P</i> -value	Fixa	tion Indices
Among groups	5	0.16	$\begin{array}{c} 0.00218 \pm \\ 0.00050 \end{array}$	$F_{\rm ST}$	0.00161
Among populations within groups	13	0.00	$\begin{array}{c} 0.63050 \pm \\ 0.00429 \end{array}$	$F_{\rm SC}$	0.00002
Among individuals within populations	2115	99.84	$\begin{array}{c} 0.00644 \pm \\ 0.00087 \end{array}$	F _{CT}	0.00159

Locus	F _{SC}	P -value	F _{ST}	P -value	F _{SC}	P -value
Ch126	0.00222	0.21436	0.00322	0.11287	0.00100	0.31168
Ch520	0.00272	0.14782	0.00583	0.01525	0.00311	0.16238
Ch623	-0.00118	0.93178	-0.00083	0.93248	0.00035	0.18139
Ch10105	0.00264	0.16455	0.00529	0.02604	0.00266	0.18634
Ch10857	0.00030	0.50772	0.00173	0.24465	0.00143	0.14436
Ch13222	0.00253	0.21772	0.00267	0.19158	0.00014	0.44020
Ch18085	-0.00079	0.67248	-0.00131	0.81139	-0.00051	0.79634
Ch2931	0.00256	0.18356	0.00231	0.16030	-0.00025	0.57743
Ch4796	-0.00143	0.84980	-0.00073	0.78752	0.00070	0.38485
Ch10441	0.00197	0.24109	0.00197	0.19099	0.00000	0.42762
Ch11483	0.00111	0.32614	-0.00120	0.65604	-0.00231	0.99822
Ch17977	-0.00251	0.87376	-0.00239	0.93010	0.00012	0.46446
Ch19846	-0.00047	0.61356	0.01231	0.00000	0.01277	0.00059
Ch24332	-0.00156	0.71347	-0.00190	0.84584	-0.00034	0.64149
Ch25478	-0.00297	0.87149	0.00042	0.50792	0.00339	0.01970

Table S4 Results of power analysis for the Fisher's and Chi-square tests by using POWSIM ver. 4.1^5 . To assess the statistical power of the 15 microsatellites to detect F_{ST} values ≤ 0.01 , observed sample sizes and allele frequencies were taken from the 19 sampling locations using 1000 replicates. Following the POWSIM manual, several combinations of N_e (1000 – 10000) and generations (t = 2 – 201) were tested to account for the variability in N_e estimation. Power is given in % for Fisher's and Chi-square tests.

N		$F_{\rm ST} =$	0.01		$F_{\rm ST} =$	0.005		$F_{\rm ST} =$	0.0025		$F_{\rm ST} =$	0.001
INe	t	Fisher	Chi-square	t	Fisher	Chi-square	t	Fisher	Chi-square	t	Fisher	Chi-square
1000	20	100	100	10	100	100	5	100	100	2	96.2	95.3
5000	100	100	100	50	100	100	25	100	100	10	96.4	94.1
10000	201	100	100	100	100	100	50	100	100	20	97.4	94.6

Table S5 Comparison of tests of genetic differentiation among *Pleuragramma antarctica* samples from the Antarctic Peninsula included in the analysis of Agostini *et al.*². Pairwise F_{ST} estimates (above the diagonal) and corresponding *P*-values (below the diagonal) are shown. Values in bold were significant after correction for multiple tests as implemented in SGoF+. Comparisons within the same geographic region are delineated with dotted lines. **a** Original pairwise F_{ST} analysis adapted from Table 3 in Agostini *et al.*² (threshold for significance with 36 comparisons P = 0.0180). **b** Pairwise F_{ST} analysis of samples from Agostini *et al.*² using 15 loci after the removal of locus Ch11230 (threshold for significance with 36 comparisons P = 0.0160). Values shaded in light gray indicate gains of significance compared to the original analysis². Values shaded in dark gray indicate losses of significance compared to the original analysis². Numbers in parentheses adjacent to group names indicate sample size (*n*). Sampling location acronyms are as in Table 1.

a									
Original		Wester	n Antarctic P	eninsula		Northe	rn Antarctic Pe	eninsula	Larsen Bay
Analysis	CI10 (60)	MB01 (28)	MB02 (49)	MB10 (60)	MB11 (83)	JI07 (34)	JI10 (148)	JI12 (54)	LB07 (46)
CI10	-	-0.0011	-0.0008	0.0007	-0.0001	-0.0009	0.0065	0.0102	0.0061
MB01	0.7027	-	-0.0015	-0.0026	-0.0015	0.0017	0.0057	0.0086	0.0063
MB02	0.6216	0.7207	-	0.0005	0.0009	0.0007	0.0037	0.0093	0.0035
MB10	0.3333	0.8018	0.4054	-	0.0004	-0.0028	0.0017	0.0062	0.0029
MB11	0.5496	0.6757	0.2342	0.2793	-	-0.0004	0.0041	0.0060	0.0089
JI07	0.7838	0.2973	0.3874	0.8559	0.5315	-	-0.0015	0.0008	0.0001
JI10	< 0.0001	0.0180	< 0.0001	0.0541	< 0.0001	0.8108	-	0.0020	0.0020
JI12	< 0.0001	0.0090	< 0.0001	0.0270	< 0.0001	0.3874	0.0991	-	0.0057
LB07	0.0180	0.0991	0.0811	0.0451	< 0.0001	0.4955	0.0991	0.0090	-

b

15 loci w/o		Wester	n Antarctic Pe	eninsula		Norther	Larsen Bay		
Ch11230	0 CI10 (60) MB01 (28) MB02 (49) MB10 (60) M		MB11 (83)	JI07 (34)	JI10 (148)	JI12 (54)	LB07 (46)		
CI10	-	0.0021	0.0003	0.0003	-0.0010	-0.0006	0.0068	0.0111	0.0057
MB01	0.2793	-	0.0019	0.0002	0.0004	-0.0001	0.0041	0.0059	0.0042
MB02	0.4454	0.2315	-	0.0007	0.0002	0.0012	0.0035	0.0084	0.0041
MB10	0.4566	0.4512	0.3341	-	0.0009	-0.0020	0.0027	0.0085	0.0025
MB11	0.7577	0.3860	0.3902	0.2479	-	0.0002	0.0051	0.0082	0.0083
JI07	0.6186	0.4475	0.2680	0.8035	0.3916	-	-0.0010	0.0019	0.0008
Л10	< 0.0001	0.0556	0.0160	0.0234	< 0.0001	0.7230	-	0.0025	0.0019
Л12	< 0.0001	0.0444	0.0006	0.0003	0.0001	0.2279	0.0514	-	0.0060
LB07	0.0160	0.1147	0.0441	0.1259	0.0002	0.3588	0.1176	0.0120	-

Table S6 Standard length (SL) analysis. **a** SL-derived groupings independent of sampling location. **b** – **g** Pairwise F_{ST} estimates (above the diagonal) and corresponding *P*-values (below the diagonal) are shown. Values in bold were significant after correction for multiple tests as implemented in SGoF+¹. Group numbers correspond to respective groupings in **a**. Group SL range (cm) in parentheses to the left of each matrix. Sample size (*n*) in parentheses above each matrix.

a		
<u>SL G</u>	rouping 1	
Groups	cm	n
1	3 - 4	3
2	4 - 5	46
3	5 - 6	14
4	6 - 7	16
5	7 - 8	5
6	8 - 9	14
7	9 - 10	56
8	10 - 11	83
9	11 - 12	81
10	12 - 13	58
11	13 - 14	30
12	14 - 15	45
13	15 - 16	78
14	16 - 17	72
15	17 - 18	32
16	18 - 19	23
17	19 - 20	7
18	20 - 21	4
19	21 - 22	4

SL Grouping 2										
Groups	cm	n								
1	3 - 5	49								
2	5 - 7	30								
3	7 - 9	19								
4	9 - 11	139								
5	11 - 13	139								
6	13 - 15	75								
7	15 - 17	150								
8	17 - 19	55								
9	19 - 21	11								
10	21 - 23	4								
SL Grouping 3										

SL Grouping 3										
Groups	cm	n								
1	3 - 6	63								
2	6 - 9	35								
3	9 -12	220								
4	12 - 15	133								
5	15 - 18	182								
6	18 - 21	34								
7	21 - 24	4								

SL Grouping 4											
Groups	cm	n									
1	3 - 7	79									
2	7 - 11	158									
3	11 - 15	214									
4	15 - 19	205									
5	19 - 23	15									

SL Grouping 5											
Groups	cm	n									
1	3 - 8	84									
2	8 - 13	292									
3	13 - 18	257									
4	18 - 23	38									

SL Grouping 6											
Groups	cm	n									
1	3 - 9	98									
2	9 - 15	353									
3	15 - 22	220									

h	CI	Groun	ina 1	
ν	SL	Uloup	III III III III III III III III III II	

(cm) (n)	1 (3)	2 (46)	3 (14)	4 (16)	5 (5)	6 (14)	7 (56)	8 (83)	9 (81)	10 (58)	11 (30)	12 (45)	13 (78)	14 (72)	15 (32)	16 (23)	17 (7)	18 (4)	19 (4)
1 (3 - 4)	-	0.0486	0.0163	0.0515	0.0097	0.0454	0.0509	0.0575	0.0457	0.0571	0.0407	0.0563	0.0395	0.0472	0.0485	0.0541	0.0559	-0.0081	0.0330
2 (4 - 5)	0.0180	-	0.0039	0.0047	-0.0016	-0.0016	0.0031	0.0018	0.0012	0.0042	0.0003	0.0004	0.0020	0.0012	0.0019	0.0025	0.0016	0.0083	-0.0059
3 (5 - 6)	0.3153	0.2613	-	0.0070	-0.0037	0.0081	0.0120	0.0047	0.0023	0.0070	-0.0028	0.0092	0.0042	0.0035	-0.0007	-0.0015	0.0196	-0.0057	-0.0101
4 (6 - 7)	0.0631	0.1351	0.1892	-	-0.0240	0.0012	0.0033	-0.0012	-0.0031	-0.0013	-0.0020	0.0025	-0.0008	0.0031	-0.0031	0.0053	0.0270	0.0107	-0.0103
5 (7 - 8)	0.6306	0.5405	0.5405	0.9550	-	-0.0185	-0.0173	-0.0115	-0.0134	-0.0200	-0.0094	-0.0095	-0.0104	-0.0129	-0.0113	-0.0040	0.0068	-0.0357	-0.0241
6 (8 - 9)	0.1261	0.7387	0.1351	0.3694	0.9730	-	0.0024	-0.0055	-0.0029	-0.0003	-0.0011	-0.0034	-0.0046	-0.0006	-0.0036	-0.0007	-0.0113	-0.0153	-0.0196
7 (9 - 10)	0.0541	0.1081	0.0180	0.2793	0.9640	0.3514	-	0.0030	0.0016	-0.0001	0.0053	0.0024	0.0024	0.0020	-0.0002	0.0064	0.0048	0.0028	-0.0008
8 (10 - 11)	0.0180	0.1532	0.1351	0.6667	0.8919	0.9369	0.0541	-	-0.0019	-0.0003	-0.0008	-0.0013	-0.0013	0.0005	-0.0032	-0.0012	0.0058	0.0042	-0.0093
9 (11 - 12)	0.0180	0.2072	0.2613	0.8378	0.9279	0.7568	0.1261	0.9369	-	-0.0002	-0.0010	-0.0014	-0.0021	-0.0004	-0.0013	-0.0009	0.0033	0.0020	-0.0120
10 (12 - 13)	< 0.0001	0.0090	0.0901	0.6667	0.9820	0.5045	0.5676	0.5766	0.6126	-	0.0022	0.0021	0.0023	0.0004	0.0003	0.0028	0.0050	0.0061	-0.0112
11 (13 - 14)	0.0631	0.5045	0.7658	0.6487	0.8649	0.6487	0.0541	0.5946	0.6306	0.2072	-	0.0015	0.0001	0.0037	0.0004	-0.0048	0.0119	0.0063	-0.0133
12 (14 - 15)	0.0360	0.4955	0.0451	0.3243	0.8018	0.8108	0.2072	0.7568	0.7928	0.1622	0.3153	-	-0.0013	0.0026	-0.0012	0.0004	0.0027	0.0104	-0.0132
13 (15 - 16)	0.0541	0.1261	0.1351	0.5676	0.8559	0.8829	0.0541	0.8288	0.9550	0.1081	0.4414	0.7748	-	0.0004	-0.0010	0.0003	0.0023	0.0019	-0.0068
14 (16 - 17)	0.0180	0.3063	0.2793	0.2162	0.9460	0.5946	0.1441	0.2342	0.6126	0.3964	0.0901	0.1081	0.3964	-	0.0018	0.0016	0.0047	0.0077	0.0012
15 (17 - 18)	0.0270	0.3243	0.5946	0.7478	0.8919	0.8378	0.5496	0.9279	0.7658	0.4505	0.4505	0.6577	0.7748	0.3514	-	0.0027	0.0097	0.0050	-0.0152
16 (18 - 19)	0.0270	0.2252	0.6036	0.1441	0.7297	0.5676	0.0631	0.5766	0.5676	0.1982	0.9369	0.3874	0.4505	0.2613	0.2523	-	0.0106	0.0174	-0.0139
17 (19 - 20)	0.1441	0.5315	0.0901	< 0.0001	0.4324	0.9460	0.3063	0.2703	0.3694	0.3153	0.1712	0.4505	0.3964	0.3423	0.1802	0.1351	-	0.0040	-0.0040
18 (20 - 21)	0.7568	0.3333	0.6577	0.2162	0.9550	0.8739	0.4865	0.4505	0.6216	0.3604	0.3243	0.2252	0.5586	0.3964	0.3694	0.1261	0.5586	-	-0.0083
19 (21 - 22)	0.4324	0.7478	0.7838	0.7928	0.9279	0.9640	0.6396	0.7387	0.7658	0.7838	0.8829	0.9099	0.6847	0.4865	0.8829	0.8108	0.7478	0.6126	-

c SL Grouping 2

e de orot	<u>apm8</u>	-								
(cm) (n)	1 (49)	2 (30)	3 (19)	4 (139)	5 (139)	6 (75)	7 (150)	8 (55)	9 (11)	10 (4)
1 (3 - 5)	-	0.0016	0.0016	0.0019	0.0025	0.0002	0.0012	0.0016	0.0024	-0.0061
2 (5 - 7)	0.2523	-	0.0014	0.0013	-0.0011	0.0003	0.0005	-0.0028	0.0134	-0.0118
3 (7 - 9)	0.3874	0.3694	-	-0.0025	-0.0020	-0.0010	-0.0014	-0.0011	-0.0089	-0.0160
4 (9 - 11)	0.1982	0.2793	0.8649	-	-0.0011	-0.0003	-0.0002	-0.0018	0.0029	-0.0065
5 (11 - 13)	0.0541	0.7117	0.7297	0.9460	-	-0.0002	-0.0002	-0.0007	0.0029	-0.0115
6 (13 - 15)	0.4505	0.4505	0.6487	0.6306	0.5856	-	0.0006	-0.0020	0.0062	-0.0135
7 (15 - 17)	0.2252	0.3964	0.6667	0.6216	0.6306	0.3243	-	-0.0001	0.0031	-0.0030
8 (17 - 19)	0.2252	0.8919	0.5676	0.9369	0.6396	0.9189	0.6487	-	0.0083	-0.0152
9 (19 - 21)	0.4595	0.0721	0.9189	0.2973	0.3243	0.1171	0.3063	0.0991	-	-0.0051
10 (21 - 23)	0.7838	0.8198	0.9820	0.6757	0.8018	0.9009	0.5496	0.9099	0.6036	-

d SL Grouping 3

(cm) (n)	1 (63)	2 (35)	3 (220)	4 (133)	5 (182)	6 (34)	7 (4)
1 (3 - 6)	-	0.0044	0.0022	0.0020	0.0012	-0.0002	-0.0072
2 (6 - 9)	0.0451	-	-0.0010	-0.0008	-0.0001	0.0015	-0.0124
3 (9 - 12)	0.0270	0.8108	-	-0.0009	-0.0007	-0.0012	-0.0082
4 (12 - 15)	0.0901	0.6847	0.9369	-	0.0002	-0.0013	-0.0129
5 (15 - 18)	0.1892	0.6036	0.9460	0.2973	-	-0.0005	-0.0051
6 (18 - 21)	0.5856	0.3243	0.7838	0.6757	0.5946	-	-0.0135
7 (21 - 24)	0.6757	0.8739	0.7207	0.9279	0.6216	0.8649	-

e SL Grouping 4						f SL Grouping 5					g SL Grouping 6			
(cm) (n)	1 (79)	2 (158)	3 (214)	4 (205)	5 (15)	(cm) (n)	1 (84)	2 (292)	3 (257)	4 (38)	(cm) (n)	1 (98)	2 (353)	3 (220)
1 (3 - 7)	-	0.0015	0.0004	0.0003	0.0033	1 (3 - 8)	-	0.0007	-0.0002	0.0008	1 (3 - 9)	-	0.0002	-0.0003
2 (7 - 11)	0.1171	-	-0.0006	-0.0004	-0.0001	2 (8 - 13)	0.1802	-	-0.0002	-0.0007	2 (9 - 15)	0.3964	-	-0.0003
3 (11 - 15)	0.3423	0.8469	-	-0.0002	0.0005	3 (13 - 18)	0.5225	0.7658	-	-0.0004	3 (15 - 22)	0.7838	0.7748	-
4 (15 - 19)	0.4595	0.7478	0.7748	-	0.0025	4 (18 - 23)	0.4144	0.6487	0.5496	-				
5 (19 - 23)	0.2613	0.5315	0.3784	0.2883	-									

Table S7 Genetic differentiation among *Pleuragramma antarctica* samples including small and large size clusters from Joinville Island 2010. Pairwise F_{ST} estimates (above the diagonal) and corresponding *P*-values (below the diagonal) are shown. Values in bold were significant after correction for multiple tests as implemented in SGoF+¹ (threshold for significance with 190 comparisons P = 0.0453). Comparisons within the same geographic region are delineated with dotted lines. Values shaded in light gray indicate gains of significance compared to the original analysis². Values shaded in dark gray indicate losses of significance compared to the original analysis². Numbers in parentheses adjacent to group names indicate sample size (*n*). Sampling location acronyms are as in Table 1. J110S, Joinville Island 2010 small cluster; J110L, Joinville Island 2010 large cluster; SOI, South Orkney Islands.

	Western Antarctic Peninsula (WAP)				Northern Antarctic Peninsula (NAP)				SOI	Larsen I	Bay (LB)	Weddell Sea (WS)			Ross Sea (RS)					
	CI10 (60)	MB01 (28)	MB02 (49)	MB10 (60)	MB11 (83)	JI07 (34)	J110S (68)	JI10L (72)	JI12 (54)	SSI96 (14)	SOI11 (47)	LB07 (46)	LB11 (52)	FT14 (50)	HB89 (19)	HB91 (41)	HB14 (82)	AB14 (25)	RS96 (91)	RS97 (84)
CI10	-	-0.0008	-0.0002	0.0007	-0.0008	-0.0033	0.0061	0.0048	0.0043	-0.0044	0.0100	0.0054	0.0021	0.0002	0.0067	0.0016	0.0001	-0.0012	0.0049	0.0024
MB01	0.6810		-0.0004	-0.0022	-0.0016	0.0006	0.0070	0.0043	0.0047	0.0016	0.0125	0.0057	0.0057	0.0007	0.0051	0.0037	0.0015	0.0008	0.0043	0.0042
MB02	0.5787	0.5126	-	0.0015	0.0010	-0.0013	0.0041	0.0016	0.0039	-0.0041	0.0103	0.0021	0.0006	0.0010	0.0062	0.0004	-0.0003	-0.0036	0.0023	0.0043
MB10	0.3758	0.7993	0.2069	-	0.0009	-0.0037	0.0032	0.0006	0.0037	-0.0012	0.0091	0.0027	-0.0005	-0.0000	0.0047	0.0008	-0.0011	-0.0021	0.0002	0.0010
MB11	0.7213	0.7356	0.2443	0.2427	-	-0.0008	0.0061	0.0047	0.0043	0.0012	0.0096	0.0094	0.0046	0.0008	0.0060	0.0030	0.0018	-0.0001	0.0041	0.0025
JI07	0.9567	0.4147	0.6655	0.9617	0.6194	-	0.0031	-0.0032	0.0010	-0.0084	0.0059	0.0009	-0.0034	-0.0034	0.0054	0.0002	-0.0027	-0.0053	-0.0018	-0.0026
JI10S	0.0041	0.0156	0.0284	0.0399	0.0005	0.1056		0.0040	0.0034	0.0013	0.0045	0.0055	0.0037	0.0019	-0.0004	0.0005	0.0021	0.0038	0.0001	0.0044
JI10L	0.0096	0.0691	0.1819	0.3252	0.0020	0.9572	0.0098	-	0.0024	-0.0042	0.0042	0.0003	-0.0018	0.0002	0.0049	0.0032	-0.0008	-0.0018	-0.0000	0.0019
JI12	0.0358	0.0902	0.0527	0.0388	0.0115	0.3543	0.0514	0.1143	-	0.0020	0.0013	0.0039	0.0027	-0.0006	0.0053	-0.0015	0.0015	0.0014	0.0020	0.0020
SSI96	0.8949	0.3972	0.8144	0.5788	0.3643	0.9734	0.4086	0.8656	0.3729	-	0.0010	0.0012	-0.0057	-0.0062	0.0034	-0.0018	-0.0050	-0.0072	-0.0016	-0.0016
SOI11	0.0006	0.0016	0.0009	0.0006	0.0001	0.0383	0.0265	0.0312	0.3132	0.4789	-	0.0058	0.0039	0.0003	0.0066	-0.0001	0.0037	0.0049	0.0066	0.0031
LB07	0.0188	0.0518	0.1708	0.0958	0.0001	0.3593	0.0095	0.4401	0.0598	0.4017	0.0212	-	0.0020	0.0020	0.0047	0.0056	0.0004	0.0004	0.0033	0.0033
LB11	0.1806	0.0453	0.3619	0.5866	0.0088	0.9430	0.0361	0.8908	0.1145	0.9241	0.0650	0.1872	-	-0.0012	0.0098	-0.0012	-0.0008	-0.0035	0.0016	0.0013
FT14	0.5172	0.4127	0.3118	0.4773	0.2877	0.9348	0.1716	0.4705	0.6511	0.9403	0.4792	0.1909	0.7452	-	0.0044	-0.0012	-0.0015	-0.0037	-0.0009	-0.0013
HB89	0.0826	0.1752	0.0647	0.1036	0.0501	0.1201	0.5773	0.1046	0.1347	0.3473	0.0970	0.1393	0.0140	0.1607	-	0.0047	0.0024	0.0066	0.0017	0.0057
HB91	0.2849	0.1499	0.4115	0.3282	0.0648	0.4695	0.4343	0.0736	0.7977	0.6767	0.5613	0.0245	0.7272	0.7308	0.1513	-	-0.0006	-0.0031	0.0015	0.0007
HB14	0.5501	0.3427	0.6011	0.8075	0.1110	0.9388	0.1104	0.7651	0.2477	0.9273	0.0570	0.4541	0.7321	0.8674	0.3111	0.6730	-	-0.0051	0.0011	-0.0009
AB14	0.6987	0.3851	0.8802	0.7514	0.4561	0.9474	0.1031	0.7432	0.3292	0.9054	0.0926	0.4136	0.9021	0.9088	0.0950	0.8464	0.9948	-	0.0011	-0.0004
RS96	0.0038	0.0557	0.0871	0.4265	0.0021	0.8266	0.4733	0.5089	0.1134	0.6513	0.0019	0.0387	0.1593	0.7272	0.3082	0.2056	0.2117	0.3128	-	0.0024
RS97	0.0946	0.0675	0.0183	0.2533	0.0392	0.9226	0.0061	0.1035	0.1422	0.6717	0.0702	0.0508	0.2248	0.8198	0.0698	0.3866	0.8077	0.5514	0.0391	-

Table S8 Genetic differentiation among *Pleuragramma antarctica* sampling locations from the eastern Weddell Sea. Pairwise F_{ST} estimates (above the diagonal) and corresponding *P*-values (below the diagonal) are shown. Numbers in parentheses adjacent to group names indicate sample size (*n*). FT, Filchner Trough; CL, Coats Land; HB, Halley Bay; AB, Atka Bay.

	west FT (23)	east FT (27)	CL (29)	HB (52)	AB (25)
west FT	-	0.0027	-0.0053	-0.0019	-0.0019
east FT	0.2798	-	-0.0015	0.0007	-0.0039
CL	0.9602	0.7372	-	-0.0028	-0.0060
HB	0.7556	0.4143	0.9051	-	-0.0062
AB	0.6489	0.8309	0.9830	0.9954	-



Figure S1 FLOCK clustering results for *Pleuragramma antarctica* populations. a Mean LLOD scores are ordered by geographic area. b Normalized likelihoods, individuals are ordered by geographic area. ref. reference k group cluster (user-define k = 6). Initial partition mode by random choice of samples. Number of iterations (re-allocations) per run = 20. Number of runs per parameter k = 50. Log likelihood difference (LLOD) threshold = 0 (allocation occurs as soon as there is a highest log-likelihood difference between populations)⁶. Group acronyms are as in Table 1.



Figure S2 Discriminant analysis of principal components (DAPC) analysis criteria. **a** Bayesian Information Criterion (BIC) plotted for increasing numbers of clusters, indicating the optimal number of clusters at k = 9. **b** Plot of a-score optimization based on the *adegenet* R package function *optim.a.* score, used to quantify the trade-off between discrimination power and over-fitting by calculating the difference between the proportion of observed and random discrimination, indicating the optimal number of principal components (PCs) to retain is nine⁷.



Figure S3 Discriminant analysis of principal components (DAPC) clustering results for *Pleuragramma antarctica* based on location-independent SL groupings (as in Table S6a).



Figure S4 East Antarctica. Arrows indicate suggested positions according to Fig.1 in Ryan *et al.*⁸ and Whitworth *et al.*⁹ for flow from the ACC and ASC, respectively. **a** Map of the Antarctic. Areas of interest in subsequent maps are indicated in squares and ordered by movement along the ASC. **b** George V and Adélie Coasts. **c** Prydz Bay north to Iceberg Alley. **d** Lützow Bay. Maps created using the Norwegian Polar Institute's Quantarctica 2.0 package¹⁰ in the software QGIS version 2.18.9 <u>http://qgis.osgeo.org</u>¹¹.

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