Electronic Supplementary Material

Genetic evidence confirms severe extinction risk for critically endangered swift parrots: implications for conservation management Dejan Stojanovic, George Olah, Matthew Webb, Rod Peakall, Robert Heinsohn

Table S1. Hardy–Weinberg Equilibrium (HWE) tests in GenePop for the swift parrot population. W&C = Weir & Cockerham's (1984) estimate; R&H Robertson & Hill's (1984) estimate; Steps = the number of switches (change of genotypic matrices) performed. *Cfor3031 locus was excluded from the analyses.

	Hardy Weinberg Probability test				HWE test for heterozygote deficit				HWE test for heterozygote excess						
Locus	P	S F	F _{IS} esti	timates Steps	P	S.F.	F _{IS} esti	imates	Stens	D	F _{IS} estimates				
	-	0.2.	W&C	R&H	Juch	-	5.2.	W&C	R&H	Steps		S.E.	W&C	R&H	Steps
CI3	0.8467	0.0073	-0.0693	-0.023	11202	0.8249	0.0169	-0.0693	-0.023	11202	0.2337	0.019	-0.0693	-0.023	11202
C1415	0.7668	0.0107	0.0643	0.0557	24808	0.0783	0.0081	0.0643	0.0557	24808	0.9223	0.0081	0.0643	0.0557	24808
SCMA04	0.673	0.0312	0.0127	0.0083	6076	0.3659	0.0338	0.0127	0.0083	6076	0.6341	0.0338	0.0127	0.0083	6076
C2627	0.1882	0.0284	-0.0242	-0.0038	5319	0.4898	0.0344	-0.0242	-0.0038	5319	0.5102	0.0344	-0.0242	-0.0038	5319
SCMA01	0.5736	0.0349	0.0076	-0.0049	5944	0.4246	0.0354	0.0076	-0.0049	5944	0.5754	0.0354	0.0076	-0.0049	5944
SCMA07	0.2132	0.0154	0.0435	-0.0006	14837	0.4477	0.0227	0.0435	-0.0006	14837	0.5541	0.0225	0.0435	-0.0006	14837
SCMA29	0.8406	0.0033	0.0517	0.0222	28067	0.3325	0.0132	0.0517	0.0222	28067	0.7368	0.0121	0.0517	0.0222	28067
Cfor3031*	0	0	0.3447	0.3864	11159	0	0	0.3447	0.3864	11159	1	0	0.3447	0.3864	11159

Table S2. Pairwise population genetic differentiations of swift parrot samples originating from discrete breeding sites. F_{ST} values below diagonal. *P* values, based on 1,000 permutations, are shown above diagonal. Location of each population indicated in Fig. 1.

					P)			
		BN	BS	BU	ET	ME	RH	SF	WI
		(N = 32)	(N = 23)	(N = 13)	(N = 16)	(N = 8)	(N = 5)	(N = 6)	(N = 6)
	BN	-	0.161	0.444	0.122	0.430	0.176	0.229	0.093
F _{ST}	BS	0.005	-	0.450	0.195	0.321	0.244	0.390	0.241
	BU	0.000	0.000	-	0.443	0.315	0.448	0.443	0.462
	ET	0.007	0.006	0.000	-	0.191	0.457	0.104	0.216
	ME	0.000	0.004	0.006	0.010	-	0.085	0.452	0.187
	RH	0.013	0.010	0.000	0.000	0.030	-	0.457	0.493
	SF	0.008	0.003	0.000	0.020	0.000	0.000	-	0.394
	WI	0.019	0.009	0.000	0.010	0.020	0.000	0.004	-

Table S3. STRUCTURE output and implementing the Evanno method (Earl, D. and vonHoldt, B. 2012).

(A) The Evaluate output for $K = 1$ to with repeats of 1
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# K	Reps	mean est.	stdev est.
		LnP(Data)	LnP(Data)
1	10	-2462.14	0.70899
2	10	-2497.5	31.690482
3	10	-2501.99	31.660787
4	10	-2485.58	46.346756
5	10	-2510.52	62.770425
6	10	-2529.36	93.628061
7	10	-2493.27	52.014828
8	10	-2501.3	48.428527
9	10	-2523.52	96.138624
10	10	-2502.56	61.415293

(B) The raw STRUCTURE output

		Fst. In	Mean	Variance
Run #	к	nroh of	value of	ofIn
	ĸ	data	Ln	likelihood
		uata	likelihood	incentiood
10	1	-2461.9	-2445.3	33.4
9	1	-2462	-2445.9	32.2
6	1	-2462.6	-2446	33.1
5	1	-2462.1	-2445.8	32.5
1	1	-2461.8	-2446	31.5
3	1	-2461.2	-2445.6	31.3
7	1	-2463.1	-2445.9	34.5
8	1	-2463.4	-2446.2	34.3
4	1	-2461.3	-2446.3	30
2	1	-2462	-2445.9	32.2
13	2	-2519.2	-2433.1	172.2
12	2	-2471.9	-2436	71.8
11	2	-2478.2	-2439.1	78.2
20	2	-2465.4	-2443.5	43.8
17	2	-2464.7	-2445.5	38.5
16	2	-2558.3	-2432.1	252.4
14	2	-2476.1	-2442.5	67.2
19	2	-2516.8	-2439.7	154.2
15	2	-2496.8	-2438	117.6
18	2	-2527.6	-2430.3	194.6
25	3	-2534.9	-2425.6	218.5
27	3	-2475.8	-2436.1	79.5
22	3	-2517.4	-2439.5	155.8
30	3	-2465	-2444.6	40.8
26	3	-2481.3	-2435.9	90.7
21	3	-2475.5	-2443.1	64.8
24	3	-2541.2	-2429.4	223.6
23	3	-2468.1	-2438.8	58.6
28	3	-2540.9	-2432.5	216.7
29	3	-2519.8	-2428.1	183.4
32	4	-2476.9	-2438.3	77.1
39	4	-2473.8	-2438.7	70.1
40	4	-2470.2	-2443.3	53.8
31	4	-2465.6	-2440.6	50
35	4	-2470.1	-2438.7	62.8

34	4	-2465.4	-2439.6	51.7
36	4	-2478.2	-2439.6	77.1
38	4	-2616.9	-2420.6	392.7
33	4	-2471.5	-2441.7	59.6
37	4	-2467.2	-2441.5	51.4
47	5	-2490.5	-2437.6	105.7
49	5	-2505.5	-2432	146.8
50	5	-2464	-2443.8	40.4
44	5	-2469.3	-2440.4	57.8
46	5	-2463.4	-2442.8	41.2
45	5	-2536.5	-2436.9	199.1
42	5	-2669.6	-2425.3	488.6
48	5	-2538.2	-2431.7	212.8
41	5	-2464	-2444.4	39.3
43	5	-2504.2	-2435.1	138.2
60	6	-2465.9	-2442.1	47.7
55	6	-2465.6	-2441.4	48.5
54	6	-2551.3	-2431.2	240.2
59	6	-2473	-2441.9	62.2
58	6	-2644.9	-2418.1	453.7
51	6	-2469	-2439.8	58.3
53	6	-2480.2	-2437.5	85.4
57	6	-2465.7	-2441.3	48.8
52	6	-2738.1	-2425.7	624.8
56	6	-2539.9	-2429.5	220.8
62	7	-2470.5	-2439.5	62.1
70	7	-2476	-2435	81.9
67	7	-2607	-2423.5	367.1
63	7	-2465.6	-2441.5	48.3
61	7	-2464.3	-2443.5	41.6
68	7	-2464.3	-2441.9	44.9
65	7	-2574	-2432.1	283.8
64	7	-2464.1	-2442.5	43.1
69	7	-2472.9	-2443.2	59.3
66	7	-2474	-2438.6	70.8
77	8	-2469.5	-2441.9	55
75	8	-2467.4	-2440.9	53
80	8	-2540.4	-2437.7	205.4
74	8	-2462.4	-2442.8	39.3
78	8	-2550.9	-2432.1	237.5
71	8	-2462.4	-2442.4	40

76	8	-2514.2	-2436.6	155.4
73	8	-2467.1	-2441	52.2
72	8	-2477.3	-2439.1	76.4
79	8	-2601.4	-2433.5	335.8
86	9	-2511.7	-2436.1	151.2
87	9	-2464.5	-2442.6	43.9
90	9	-2462.6	-2443.4	38.4
82	9	-2464.2	-2443.2	42.1
81	9	-2754.6	-2416.8	675.6
84	9	-2476.9	-2440.6	72.7
88	9	-2463.1	-2443.1	40
89	9	-2562	-2434.7	254.7
83	9	-2462.9	-2444.1	37.7
85	9	-2612.7	-2424.4	376.6
93	10	-2463.7	-2443.4	40.6
100	10	-2554.1	-2436.4	235.4
98	10	-2462.3	-2443.6	37.4
92	10	-2469.9	-2440.9	58
96	10	-2469.1	-2441.4	55.3
91	10	-2631.7	-2426.6	410.3
94	10	-2461.2	-2444.2	34
99	10	-2575.3	-2431.4	287.8
97	10	-2463.6	-2441.5	44.2
95	10	-2474.7	-2441.7	65.9

Earl, D., and vonHoldt, B. (2012) STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. *Conservation Genetics Resources* **4**(2), 359-361.

Figure S1. Performances of four non-likelihood relatedness estimators (L&L – Li et al. 1993, L&R – Lynch and Ritland 1999, Q&G – Queller and Goodnight 1989, and W – Wang 2002) on simulated data sets with the same locus characteristics as our own data. The 'related' package in R statistics (Pew et al., 2015) was used to perform the simulations with 100 simulated pairs of individuals for each type of relationship.



Pew J, Muir PH, Wang J, Frasier TR (2015) related: an R package for analyzing pairwise relatedness from codominant molecular markers. *Molecular Ecology Resources* **15**: 557-561.

Figure S2. Results from STRUCTURE Harvester (Earl, D. and vonHoldt, B. 2012) indicating that a single genetic cluster best explains population genetic structure of swift parrots (*Lathamus discolor*) across their breeding range in Tasmania.



(A) L(K) (mean \pm SD)

(B) DeltaK = mean(|L''(K)|) / sd(L(K))



Figure S3. Histograms of the expected relatedness values within each population, using the 'related' package of R (Pew et al., 2015). The red arrows indicate the observed values. The p-values indicate the percentage of randomized iterations where the expected values were greater than or equal to the observed value. The populations are: North (BN) and South Bruny Island (BS), Buckland (BU), Eastern Tiers (ET), Meehan Range (ME), Rheban (RH), Southern Forests (SF), and Wielangta (WI).



Pew J, Muir PH, Wang J, Frasier TR (2015) related: an R package for analyzing pairwise relatedness from codominant molecular markers. *Molecular Ecology Resources* **15**: 557-561.

Figure S4. Pairwise relatedness among different nesting sites of swift parrots across their breeding range (blue) with 95% confidence intervals (red) around the null hypothesis of zero relatedness using GenAlEx (Peakall and Smouse, 2012). Error bars represent bootstrapped confidence intervals around the mean. The column labelled '*Family' represents known siblings for comparison.



Peakall, R., and Smouse, P.E. (2012) GenAlEx 6.5: genetic analysis in Excel. Population genetic software for teaching and research - an update. *Bioinformatics* **28**(19), 2537-9.

Figure S5. Simulated estimates (10,000 runs) of (A) type I error and (B) power when scoring seven microsatellite markers and sampling N individuals from two populations of swift parrots (*Lathamus discolor*). (B) The expected divergences were set to $F_{ST} = 0.01$ ($N_e = 1,000$, t = 20; circles) and $F_{ST} = 0.005$ ($N_e = 1,000$, t = 10; squares). The program POWSIM v4.1 (Ryman and Palm 2006) was used to conduct the simulations.



Ryman, N., and Palm, S. (2006) POWSIM: a computer program for assessing statistical power when testing for genetic differentiation. *Molecular Ecology Notes* **6**(3), 600-602.

Figure S6. EasyPop simulations for two swift parrot (*Lathamus discolor*) populations (each with 250 males and 250 females) with different migration rates (m). All results represent the average F_{ST} value after 100 replicated simulations.

(A) The initial 10 generations were run with complete panmixia (m = 0.75), followed by 100 generations of restricted gene flow at different migration rates between the two populations. The insert shows a detailed view of the first 10 generations after the isolation event.



(B) The initial 40 populations developed $F_{ST} = 0.02$ (m = 0), followed by 70 generations with changed migration rates. The insert shows a detailed view of the first 10 generations after the migration rates have changed.

