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Supplementary Information for

Hybridization and population variation during an adaptive radiation

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Hybridization and population variation during an adaptive radiation

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Section 1 Hybridization and mate choice

Mates are chosen on the basis of morphological and song cues learned in early life by imprinting on parents (1-3). *G. fortis* and *G. scandens* on Daphne Major Island differ in beak proportions and in song. Why, then, do finches mate with members of another species? We identified two causes of interbreeding. First, it arises when the normal imprinting process is perturbed, for example through the death of the father during the period of parental dependence as nestlings or fledglings. The young birds may then learn the song of a territorial neighbor of another species (2, 4). Second, interbreeding occurs through extra-pair mating (EPM). Three of six hybrids that could be checked for paternity by DNA were the product of EPM (5). Although the sample size is small, the fraction (0.5) is significantly higher than the fraction expected (0.23) based on 93 EPMs of 488 matings (both species combined) with identified paternity (5). Extra-pair mating is an opportunistic behavior, and not known to be adaptive (6). The frequency is 0.171 in *G. fortis* (n = 1,248 chicks) and 0.103 in *G. scandens* (n = 368 chicks). The incidence of heterospecific EPM is relatively high because courtship, involving species-specific signals and responses, is likely to be minimal or absent.

Section 2 Hybridization of *G. fuliginosa* and *G. fortis*

G. fuliginosa numbers varied in the annual samples of measured birds from 0 to 22 (mean 5.8 ± 0.07) in the years 1978 to 2012. Not all were breeders, and the few that bred hybridized with *G. fortis* and not with *G. scandens* or *G. magnirostris*. Numbers of admixed individuals of *G. fortis* and *G. fuliginosa* are much larger over the same time period, from 0 to 62, but when combined with *G. fortis* individuals their proportions are only 0-0.19 (mean 0.08 ± 0.01 SE). Proportions fluctuated but with no net change (Fig. S1).

Section 3 Variances

Annual estimates of variances of beak lengths and beak depths are shown in Fig. S2. The *G. fortis* variances fluctuated synchronously without a net increase or decrease across 38 years. Beak depth variances of *G. scandens* remained stable throughout and consistently lower than beak length variances. In contrast to beak depth, beak length variance of *G. scandens* increased strongly in 1991 and remained stable at the elevated level until 1999, when it fell to the previous level and remained there. Beak length variance was consistently higher than beak depth variance in this species, but not in *G. fortis*.

Coefficients of variation scale the variances by the mean. These are reported in the main text. Annual variation in the *G. fortis* coefficients almost exactly parallels the annual variation in variances. Coefficients for *G. scandens* beak depth increase after 1998, and thereby differ from the temporarily stable variances.

Section 4 Skewness

Frequency distributions are predicted to be right-skewed, positively, in *G. fortis* towards the larger species, and left-skewed, negatively, in *G. scandens* (8). Most skew estimates match the predictions (Fig. S3). Skewness values in *G. fortis* remained stable in the range 0 to +0.4 until 1997 when an upward trend towards more positive values began in both of the strongly covarying beak traits. The late increase is a signature of possible introgression without a counterpart in CVs. The two beak traits of *G. scandens* differed in skewness. Beak depth values varied in the range -0.5 to +0.5 with an average of +0.09 ± 0.05 (SE) and no net change, whereas skewness in the beak length distribution became strongly negative in 1986, and only returned to the pre-1986 level in 2002. The temporal pattern of beak length skewness parallels the pattern of change in CVs.

Section 5 Museum specimens

David Lack (7) measured male and female specimens from Museums. Relying on information on the labels and plumage indications of juveniles, he calculated means and

standard deviations for single sex samples of adults. We have calculated coefficients of variation and associated confidence limits (Table S1 and Table S2). With few exceptions we have used his samples of only males because they outnumber females in all but seven of the 99 island population samples. The alternative to single-sex analysis is a weighted average of the coefficients of variation for males and females, calculated by multiplying each by their respective sample sizes, adding the products and dividing by the combined sample size. We chose not to do this because it is likely to increase the uncertainty of the estimate of the coefficients, even though this may be offset by the increased sample size. However, for samples of males under 10 we have combined male and female samples in the manner indicated above. None of these are significantly different from zero. Lack (7) was unable to reliably measure beak depth of *Certhidea* specimens.

Ian Abbott (in reference 9) measured the same *Geospiza* specimens from Museums. We have not used those measurements here for two reasons. First, he measured only *Geospiza* specimens. Second, he measured the upper mandible, and we consider this to be less replicable than total beak depth as measured by Lack (7). However, Lack's and Abbott's measurements of *Geospiza* mean beak length are perfectly correlated ($r = 1.0$).

Section 6 Ambiguous pattern of variation

G. magnirostris coexists on Darwin with only one resident species, the much smaller *G. septentrionalis*. The coefficient of beak depth variation is exceptionally large (13.13) and conspicuously larger than any other (main text, Table 2). This might be the result of interbreeding with *G. septentrionalis*, resulting in a small swarm of hybrids. Consistent with this explanation, the two species differ much less in beak length (32.7 percent), a less varying dimension, than in beak depth (83.3 percent). Alternatively, the sample might be a mixture of resident *G. magnirostris* and immigrant *G. conirostris* or *G. propinqua* (8, 10 – 12). The situation appears to have been ephemeral. On a visit in January 1997 we saw and tape-recorded singing *G. magnirostris* but saw no sign of unusual morphological variants. The population appears to be very rare.

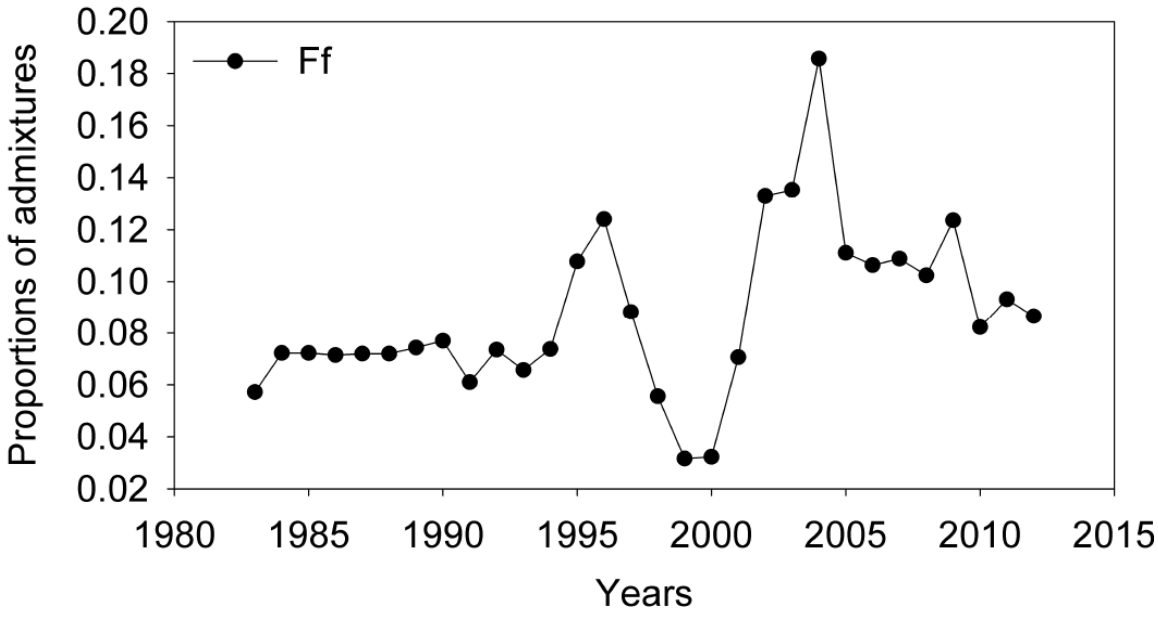

Fig. S1. The proportion of admixed individuals (*G. fuliginosa* and *G. fortis*) in the total sample of *G. fortis.*

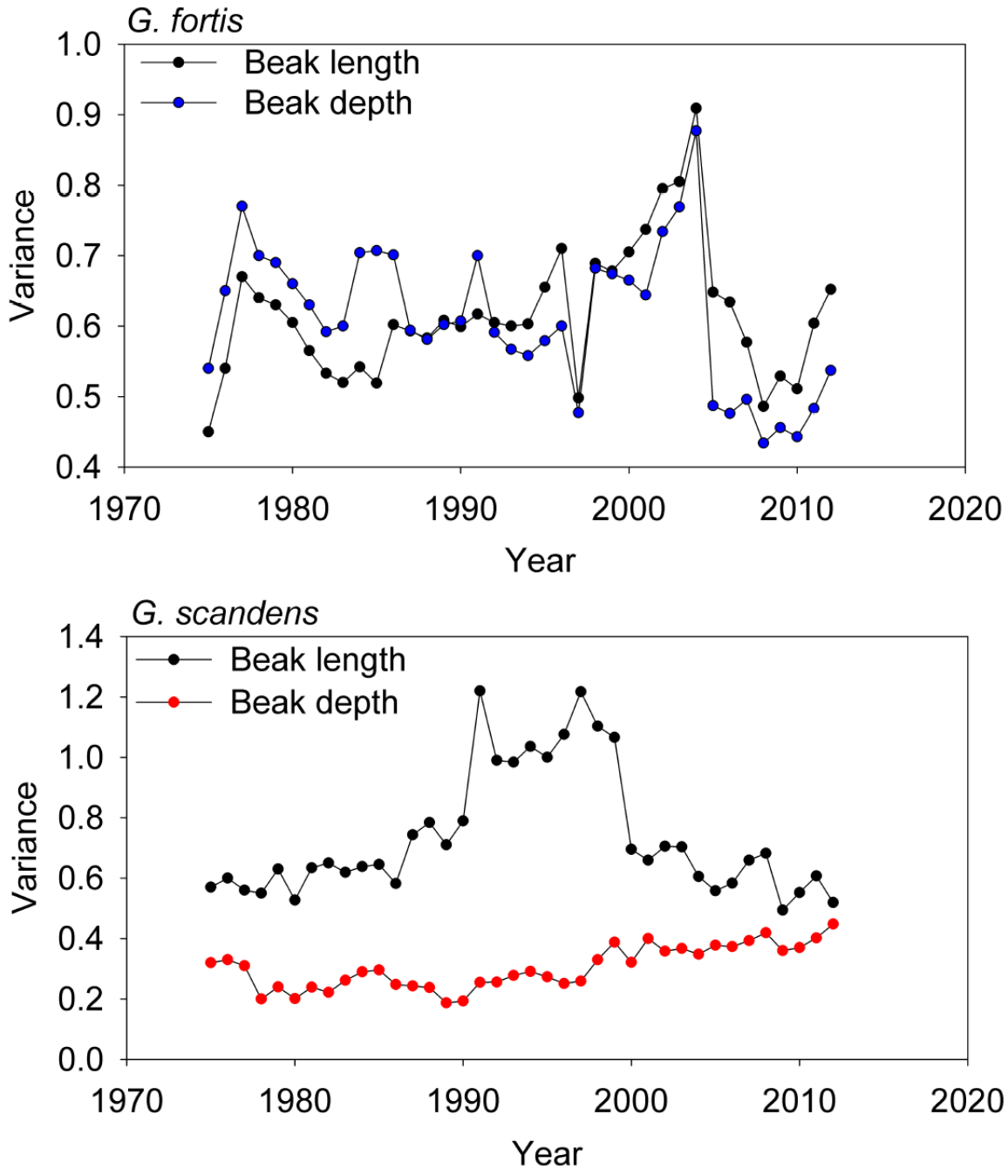

Fig. S2. Annual estimates of variances of beak lengths and beak depths of *G. fortis* and *G. scandens*.

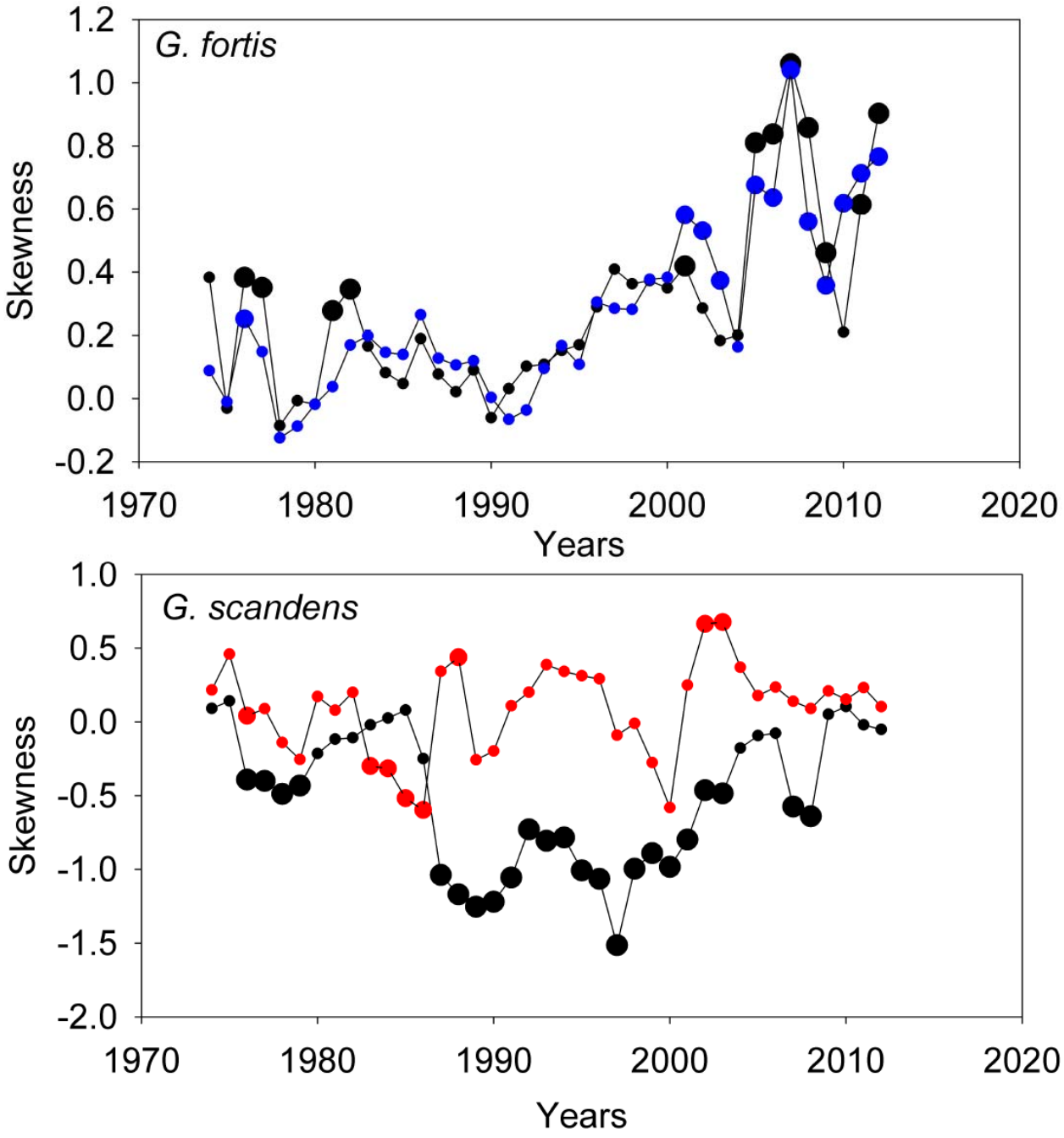

Fig. S3 Changes in skewness in frequency distributions of beak length (black) and beak depth (colored). Skewness estimates (g_1) that are significantly different from zero ($P < 0.05$) are indicated by large symbols.

Table S1. Coefficients of variation (CV) of beak length, and upper and lower 95-percent confidence limits (CL). Adult male samples only, except for a few indicated by an asterisk, where a small sample of males (<10) has been combined with females, with appropriate weighting by sample size. All species with the first letter *G* are members of the genus *Geospiza*. Other genera are *Platyspiza (crassirostris), Pinaroloxias (inornata), Certhidea (fusca* and *olivacea)* and *Camarhynchus* (all others). Original data in reference 7.

Species	**Island**	**Sample (*n*)**	**Mean**	**Variation (CV)**	**Lower CL**	**Upper CL**
G. magnirostris	Darwin	17	14.99	5.831	3.734	8.099
G. magnirostris	Wolf	10	15.65	4.064	2.006	6.325
G. magnirostris	Genovesa	29	16.52	3.626	2.665	4.649
G. magnirostris	Pinta	50	16.04	4.096	3.285	4.948
G. magnirostris	Marchena	41	15.19	5.069	3.955	6.245
G. magnirostris	Santiago	46	15.91	4.412	3.499	5.373
G. magnirostris	Rábida	35	15.29	4.709	3.593	5.892
G. magnirostris	Santa Cruz	13	15.28	5.229	3.008	7.661
G. fortis	Pinta	48	11.25	4.542	3.622	5.510
G. fortis	Marchena	36	11.66	4.031	3.081	5.037
G. fortis	Santiago	46	11.49	4.839	3.838	5.893
G. fortis	Rábida	10	11.52	3.273	1.165	5.093
G. fortis	Seymour	26	11.69	6.672	4.794	8.679
G. fortis	Santa Cruz	105	11.95	6.770	5.852	7.720
G. fortis	Pinzón	30	11.26	6.465	4.784	8.255
G. fortis	Fernandina	15	11.55	4.165	2.548	5.920
G. fortis	N. Isabela	65	11.49	6.319	5.226	7.460
G. fortis	S. Isabela	70	12.45	6.675	5.562	7.835
G. fortis	San Cristóbal	109	12.20	5.475	4.747	6.229
G. fortis	Floreana	181	11.65	7.579	6.799	8.381
G. fortis	Daphne	31	10.51	5.157	3.840	6.558
G. fuliginosa	Pinta	48	8.22	4.720	3.765	5.725
G. fuliginosa	Marchena	24	8.08	4.889	3.449	6.430
G. fuliginosa	Santiago	23	8.43	4.555	3.181	6.028
G. fuliginosa	Rábida	12	8.48	5.660	3.128	8.428
G. fuliginosa	Seymour	45	8.60	4.151	3.281	5.068
G. fuliginosa	Santa Cruz	80	8.42	4.466	3.769	5.190
G. fuliginosa	Pinzón	84	8.59	4.750	4.027	5.501
G. fuliginosa	Fernandina	19	8.07	3.234	2.145	4.408
G. fuliginosa	N. Isabela	87	8.31	4.356	3.704	5.033
G. fuliginosa	S. Isabela	63	8.43	4.496	3.706	5.321
G. fuliginosa	Santa Fé	41	8.76	4.224	3.295	5.204
G. fuliginosa	San Cristóbal	125	8.79	7.702	6.744	8.691
G. fuliginosa	Española	32	8.78	4.112	3.078	5.209
G. fuliginosa	Gardner*	14	8.49	5.823	3.464	8.390

G. fuliginosa	Floreana	86	8.59	4.622	3.926	5.344
G. fuliginosa	Los Hermanos	12	9.32	9.367	5.177	13.948
G. septentrionalis	Darwin	32	11.32	3.763	2.817	4.768
G. septentrionalis	Wolf	67	10.67	3.899	3.235	4.592
G. acutirostris	Genovesa	52	9.42	4.002	3.225	4.818
G. difficilis	Pinta	18	9.67	3.154	2.058	4.338
G. difficilis	Santiago	44	10.25	3.776	2.976	4.618
G. difficilis	Santa Cruz	33	9.60	3.333	2.509	4.208
G. scandens	Pinta	17	14.55	4.137	2.649	5.747
G. scandens	Marchena*	22	14.80	4.045	2.794	5.389
G. scandens	Santiago	23	12.92	3.839	2.681	5.080
G. scandens	Rábida	15	13.63	4.644	2.842	6.601
G. scandens	Seymour	40	14.74	4.864	3.782	6.008
G. scandens	Santa Cruz	87	14.95	5.130	4.363	5.928
G. scandens	Pinzón*	18	14.46	4.756	3.103	6.542
G. scandens	S. Isabela	16	14.64	4.816	3.020	6.762
G. scandens	Santa Fe	61	14.40	4.889	4.016	5.801
G. scandens	San Cristóbal*	15	12.86	6.112	3.740	8.688
G. scandens	Floreana	102	13.88	4.452	3.840	5.087
G. propinqua	Genovesa	43	14.35	5.310	4.172	6.510
G. conirostris	Española	87	15.43	6.079	5.170	7.024
G. conirostris	Gardner	72	14.64	6.742	5.634	7.897
P. crassirostris	Pinta	16	10.43	3.816	2.393	5.358
P. crassirostris	Marchena*	19	10.36	3.075	2.040	4.192
P. crassirostris	Santiago	26	10.55	3.242	2.329	4.217
P. crassirostris	Santa Cruz	28	10.43	2.704	1.973	3.483
P. crassirostris	S. Isabela	56	10.26	2.768	2.253	3.308
P. crassirostris	San Cristóbal	37	10.47	3.047	2.339	3.795
P. crassirostris	Floreana	24	10.69	3.302	2.330	4.343
P. inornata	Cocos	124	10.41	3.660	3.203	4.132
C. pauper	Floreana	80	9.02	4.479	3.780	5.206
C. heliobates	Isabela	32	10.49	4.480	3.353	5.678
C. psittacula	Pinta	18	10.13	3.277	2.138	4.508
C. psittacula	Marchena	24	10.49	3.603	2.542	4.740
C. psittacula	Santiago	17	9.84	3.394	2.174	4.715
C. psittacula	Santa Cruz*	18	9.41	4.381	2.858	6.025
C. psittacula	N. Isabela*	10	8.44	4.007	1.978	6.237
C. psittacula	S. Isabela	17	8.53	4.865	3.115	6.758
C. psittacula	Floreana*	17	9.57	2.298	1.472	3.192
C. parvulus	Santiago	28	6.95	2.863	2.090	3.688
C. parvulus	Santa Cruz	19	7.49	5.915	3.923	8.062
C. parvulus	N. Isabela	14	6.97	3.099	1.844	4.465
C. parvulus	S. Isabela	24	7.28	4.451	3.140	5.854
C. parvulus	Floreana	86	7.33	4.052	3.442	4.685
C. parvulus	San Cristóbal	91	7.96	3.894	3.325	4.486
C. pallidus	Santiago	13	12.56	4.084	2.350	5.976
C. pallidus	Santa Cruz*	21	12.04	2.888	1.970	3.874
C. pallidus	S. Isabela	53	11.24	3.843	3.081	4.642
C. fusca	Darwin*	19	8.36	1.860	1.234	2.535

C. fusca	Wolf*	14	7.94	5.924	3.524	8.535
C. fusca	Genovesa	41	8.18	2.518	1.965	3.103
C. fusca	Pinta	17	8.10	2.827	1.810	3.927
C. fusca	Marchena	18	8.20	3.780	2.466	5.200
C. fusca	Pinzón	21	7.56	6.151	4.197	8.251
C. fusca	Santa Fé	40	7.92	3.788	2.945	4.679
C. fusca	San Cristóbal	46	7.89	4.347	3.448	5.294
C. fusca	Española	44	8.02	3.429	2.703	4.194
C. fusca	Gardner	43	8.00	3.625	2.844	4.448
C. fusca	Floreana	25	7.74	3.747	2.669	4.900
C. olivacea	Santiago	31	7.30	3.836	2.856	4.877
C. olivacea	Santa Cruz	44	7.60	4.474	3.526	5.472
C. olivacea	Fernandina	10	7.30	2.877	1.420	4.477
C. olivacea	N. Isabela	20	7.30	3.973	2.675	5.370
C. olivacea	S. Isabela	28	7.50	4.533	3.308	5.839

Table S2. Coefficients of variation (CV) of beak depth, and upper and lower 95-percent confidence limits (CL). Adult male samples only, except for a few indicated by an asterisk, where a small sample of males (<10) has been combined with females, with appropriate weighting by sample size. All species with the first letter *G* are members of the genus *Geospiza*. Other genera are *Platyspiza (crassirostris), Pinaroloxias (inornata), Certhidea (fusca* and *olivacea)* and *Camarhynchus* (all others). Original data in reference 7.

Species	Island	Sample (n)	Mean	Variation (CV)	Lower CL	Upper CL
G. magnirostris	Darwin	16	16.53	13.134	8.235	18.442
G. magnirostris	Wolf	9	20.44	4.344	1.971	6.959
G. magnirostris	Genovesa	24	21.21	5.314	3.749	6.989
G. magnirostris	Pinta	47	20.01	4.703	3.741	5.714
G. magnirostris	Marchena	40	19.10	4.995	3.884	6.168
G. magnirostris	Santiago	43	20.50	5.000	3.928	6.130
G. magnirostris	Rábida	33	18.56	5.749	4.328	7.257
G. magnirostris	Santa Cruz	14	19.11	9.173	5.458	13.216
G. fortis	Pinta	42	11.82	5.592	4.378	6.873
G. fortis	Marchena	35	12.11	3.386	2.582	4.237
G. fortis	Santiago	43	12.50	5.712	4.488	7.003
G. fortis	Rábida	8	12.56	4.522	1.820	7.507
G. fortis	Seymour	24	12.55	7.625	5.380	10.030
G. fortis	Santa Cruz	100	12.82	10.304	8.873	11.787
G. fortis	Pinzón	25	11.46	7.522	5.357	9.837
G. fortis	Fernandina	15	12.30	4.195	2.567	5.963
G. fortis	N. Isabela	64	12.41	8.501	7.020	10.049
G. fortis	S. Isabela	65	13.86	8.434	6.976	9.958
G. fortis	San Cristóbal	101	13.21	8.562	7.378	9.788
G. fortis	Floreana	173	12.50	9.776	8.747	10.834
G. fortis	Daphne	26	10.67	6.823	4.902	8.875
G. fuliginosa	Pinta	43	7.70	5.727	4.500	7.021

G. fuliginosa	Marchena	19	7.40	5.338	3.541	7.276
G. fuliginosa	Santiago	18	8.02	2.606	1.700	3.584
G. fuliginosa	Rábida	10	8.19	5.189	2.561	8.077
G. fuliginosa	Seymour	41	8.22	4.367	3.407	5.381
G. fuliginosa	Santa Cruz	76	8.17	4.651	3.907	5.426
G. fuliginosa	Pinzón	73	8.13	3.506	2.921	4.115
G. fuliginosa	Fernandina	15	7.91	3.692	2.259	5.247
G. fuliginosa	N. Isabela	70	8.13	3.506	2.921	4.115
G. fuliginosa	S. Isabela	59	8.31	5.078	4.157	6.043
G. fuliginosa	Santa Fé	36	8.09	4.994	3.817	6.240
G. fuliginosa	San Cristóbal	112	8.08	5.446	4.730	6.185
G. fuliginosa	Española	30	8.28	4.034	2.985	5.150
G. fuliginosa	Gardner*	13	8.05	5.215	3.000	7.360
G. fuliginosa	Floreana	71	8.14	4.939	4.121	5.791
G. fuliginosa	Los Hermanos	11	9.55	8.398	4.416	12.761
G. septentrionalis	Darwin	30	8.97	3.445	2.549	4.398
G. septentrionalis	Wolf	60	8.28	3.961	3.249	4.707
G. acutirostris	Genovesa	48	7.91	4.147	3.307	5.029
G. difficilis	Pinta	17	8.51	3.408	2.182	4.734
G. difficilis	Santiago	39	9.35	3.401	2.633	4.212
G. difficilis	Santa Cruz	29	8.67	3.818	2.806	4.895
G. scandens	Pinta	16	9.68	4.742	2.973	6.658
G. scandens	Marchena*	22	10.49	4.988	3.445	6.644
G. scandens	Santiago	21	8.84	4.140	2.825	5.554
G. scandens	Rábida	12	9.22	3.200	1.768	4.764
G. scandens	Seymour	34	9.74	4.220	3.193	5.308
G. scandens	Santa Cruz	80	9.75	4.441	3.748	5.161
G. scandens	Pinzón*	14	9.52	5.089	3.028	7.333
G. scandens	S. Isabela	16	9.59	3.806	2.387	5.344
G. scandens	Santa Fe	50	10.11	4.768	3.823	5.760
G. scandens	San Cristóbal*	12	9.54	2.943	1.626	4.382
G. scandens	Floreana	90	9.43	4.157	3.545	4.792
G. propinqua	Genovesa	40	12.98	7.381	5.737	9.116
G. conirostris	Española	80	15.97	6.913	5.835	8.034
G. conirostris	Gardner	67	15.05	7.395	6.136	8.710
P. crassirostris	Pinta	13	12.23	3.900	2.244	5.707
P. crassirostris	Marchena*	13	12.51	2.734	1.573	4.000
P. crassirostris	Santiago	13	12.79	3.839	2.209	5.617
P. crassirostris	Santa Cruz	23	12.31	2.746	1.918	3.634
P. crassirostris	S. Isabela	42	12.00	4.450	3.395	5.558
P. crassirostris	San Cristóbal	30	12.64	3.513	2.599	4.485
P. crassirostris	Floreana	15	12.60	3.786	2.316	5.381
P. inornata	Cocos	107	6.17	4.052	3.508	4.615
C. pauper	Floreana	66	8.83	4.858	4.025	5.729
C. heliobates	Isabela	29	8.15	3.423	2.516	4.389
C. psittacula	Pinta	16	10.40	4.019	2.520	5.644
C. psittacula	Marchena	20	10.54	3.055	2.057	4.130
C. psittacula	Santiago	12	11.15	6.018	3.326	8.961
C. psittacula	Santa Cruz*	17	10.11	5.883	3.767	8.172

C. psittacula	N. Isabela*	10	9.30	3.795	1.873	5.906
C. psittacula	S. Isabela	15	9.28	5.140	3.145	7.306
C. psittacula	Floreana*	13	10.45	3.239	1.863	4.739
C. parvulus	Santiago	23	7.36	4.701	3.283	6.221
C. parvulus	Santa Cruz	12	7.32	4.426	2.446	6.591
C. parvulus	N. Isabela	10	7.42	5.418	2.674	8.432
C. parvulus	S. Isabela	16	7.56	4.312	2.704	6.055
C. parvulus	Floreana	54	7.51	3.782	3.062	4.536
C. parvulus	San Cristóbal	71	7.91	4.185	3.492	4.907
C. pallidus	Santiago	9	9.29	3.068	1.392	4.914
C. pallidus	Santa Cruz*	18	8.80	3.042	1.984	4.183
C. pallidus	S. Isabela	48	8.12	4.926	3.929	5.974

Captions for databases S1 to S3

Database S1. Dataset for Fig 1 and Fig S1. Annual variation in the proportions of admixed individuals in the populations of *G. fortis* and *G. scandens* on Daphne Major Island.

Database S2. Dataset for statistical analysis. Proportions of admixed individuals in the populations of *G. fortis* and *G. scandens* produced in five years of extensive breeding on Daphne Major Island.

Database S3. Dataset for Figs 2, 3 and S3. Means, variances, coefficients of variation and skewness for annual samples of *G. fortis* and *G. scandens* on Daphne Major Island.

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