SUPPLEMENTARY PAGES

Rampant introgressive hybridization in tinkerbirds [Piciformes: Lybiidae] despite millions of years of divergence

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Contents

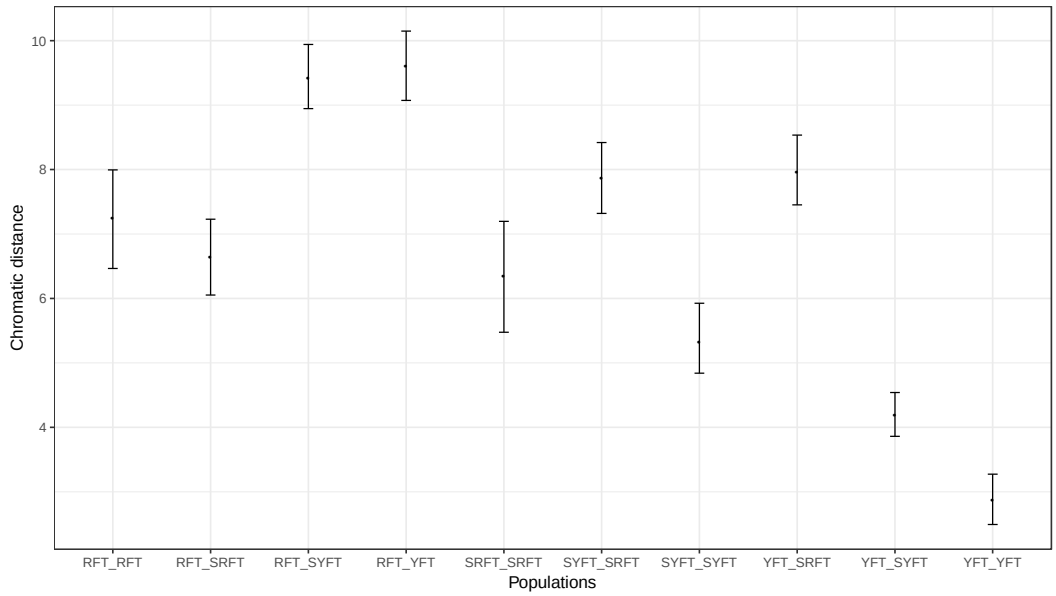

Figure S1. Chromatic distance within and among populations of each phenotype in sympatry and allopatry. Comparisons are between allopatric *P. p. pusillus* (RFT), sympatric *P. p. pusillus* (SRFT), allopatric *P. c. extoni* (YFT) and sympatric *P. c. extoni* (SYFT).

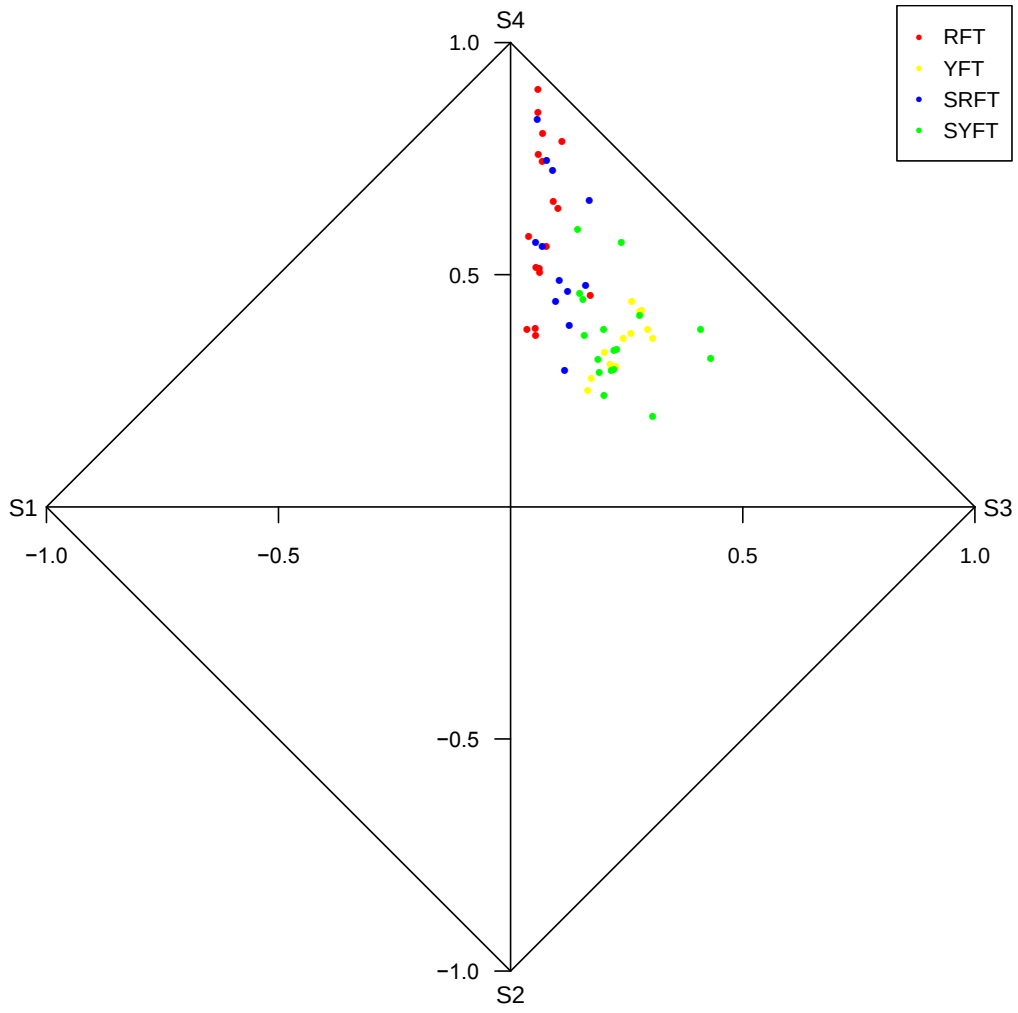

Figure S2. Segment classification plot for allopatric and sympatric populations. Allopatric *P. p. pusillus* (RFT) in red, sympatric *P. p. pusillus* (SRFT) in blue, allopatric *P. c. extoni* (YFT) in yellow and sympatric *P. c. extoni* (SYFT) in green.

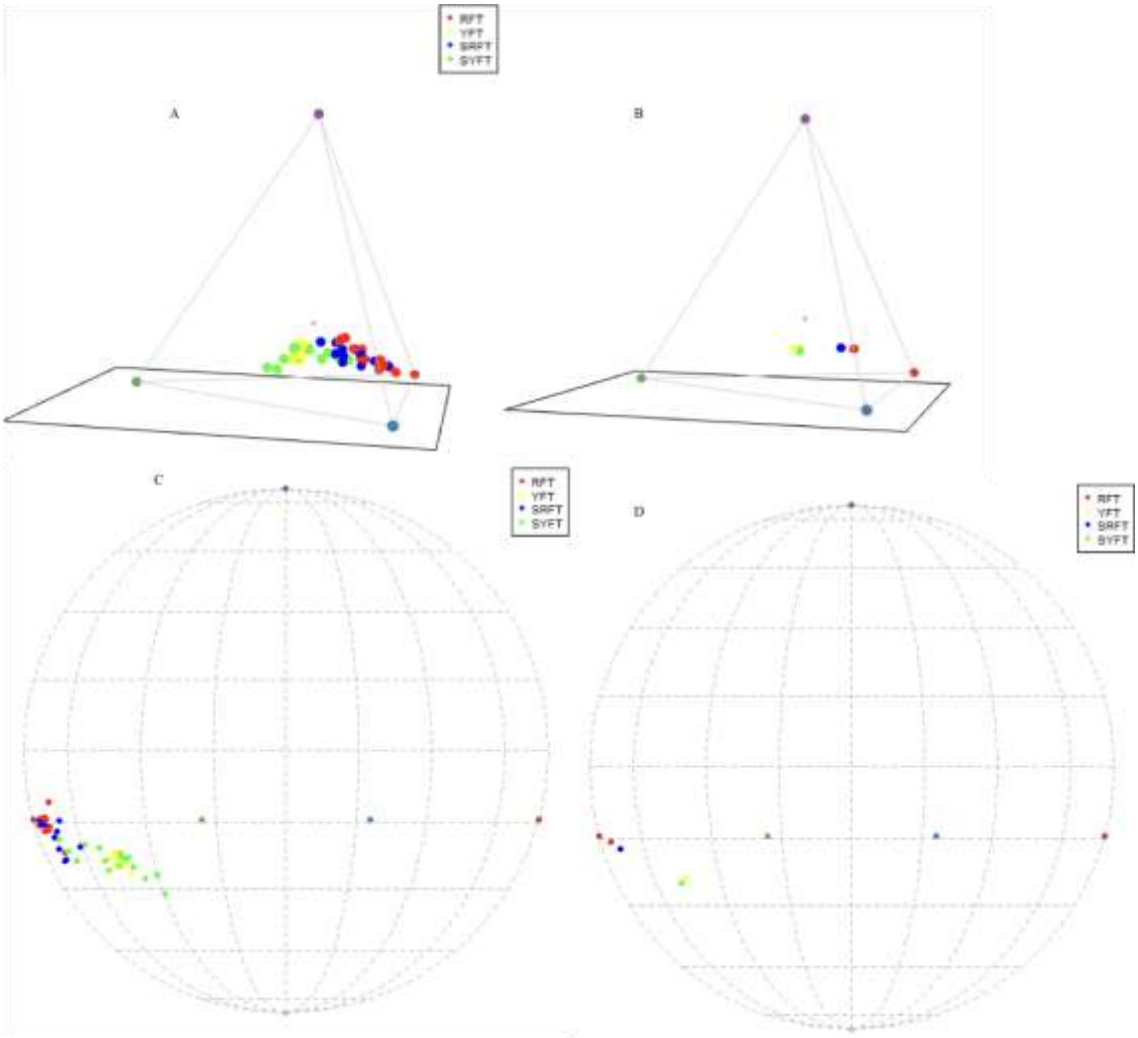

Figure S3. Tetracolourspace plot at A) individual level and B) population mean. Hue projection plot encapsulated within a sphere at C) individual level and D) population mean.

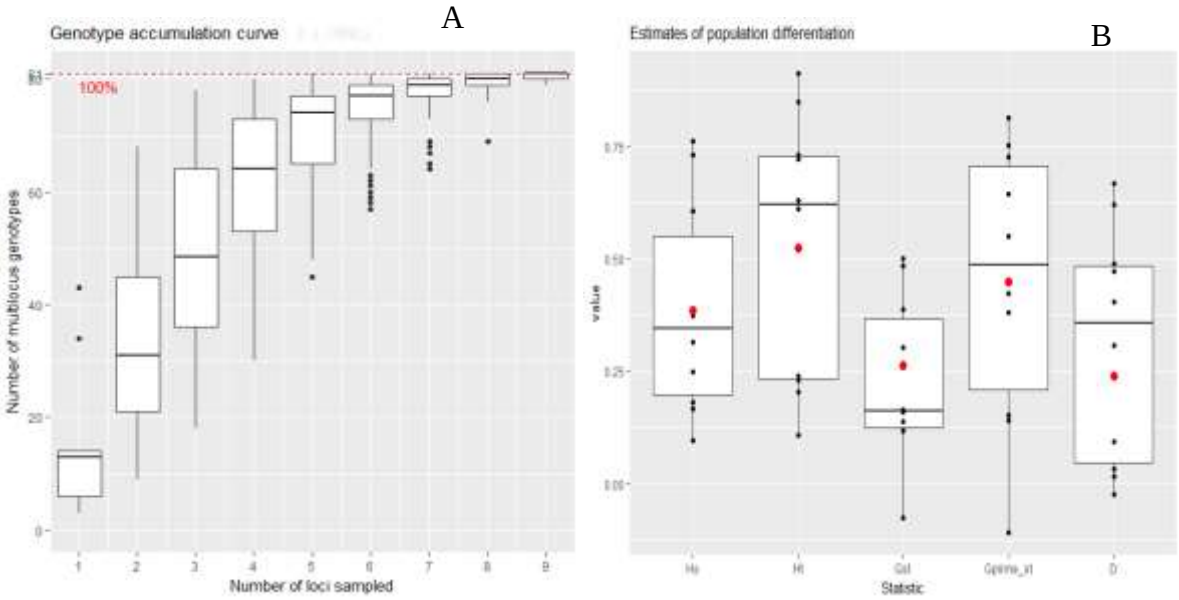

Figure S4. A) Genotype accumulation curve based on a Permutational test on the optimum number of markers required to sufficiently distinguish between individuals in the populations plateaued at six loci. B) Estimates of global indices for population differentiation based on microsatellite data. Heterozygosity with population structure (H_{ST}, G_{ST}) was lower than heterozygosity without population structure (H_T, G_{prime_ST}). The extent of population differentiation (D) ranging from 0 for a panmictic population to 1 for complete differentiation revealed high differentiation at two loci (CAM13 and TG13009, Table A7).

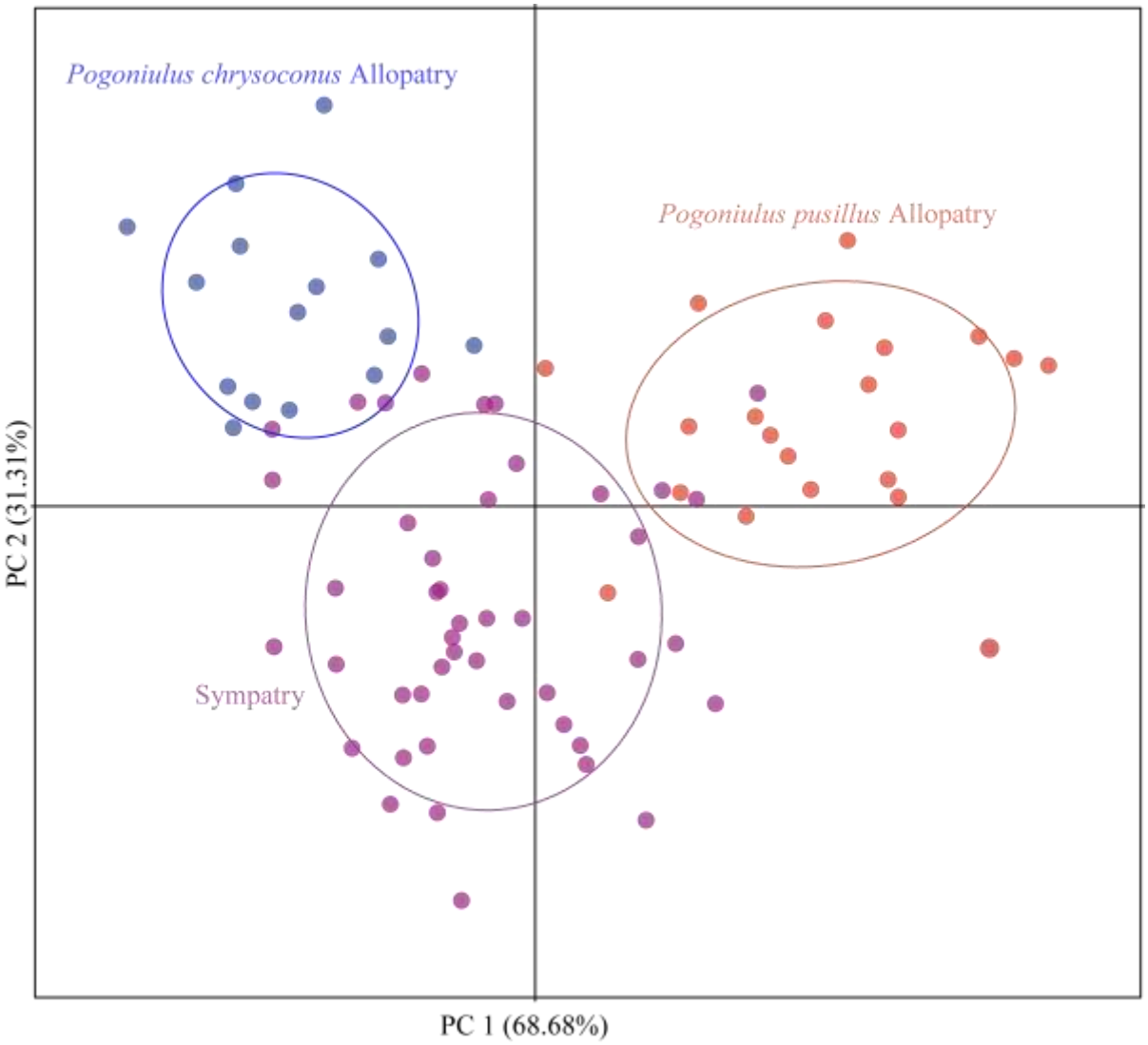

Figure S5. Discriminant Principal component analysis illustrating genetic differentiation among samples from *P. pusillus pusillus* allopatric, *P. chrysoconus extoni* allopatric, and sympatric samples based on microsatellite data

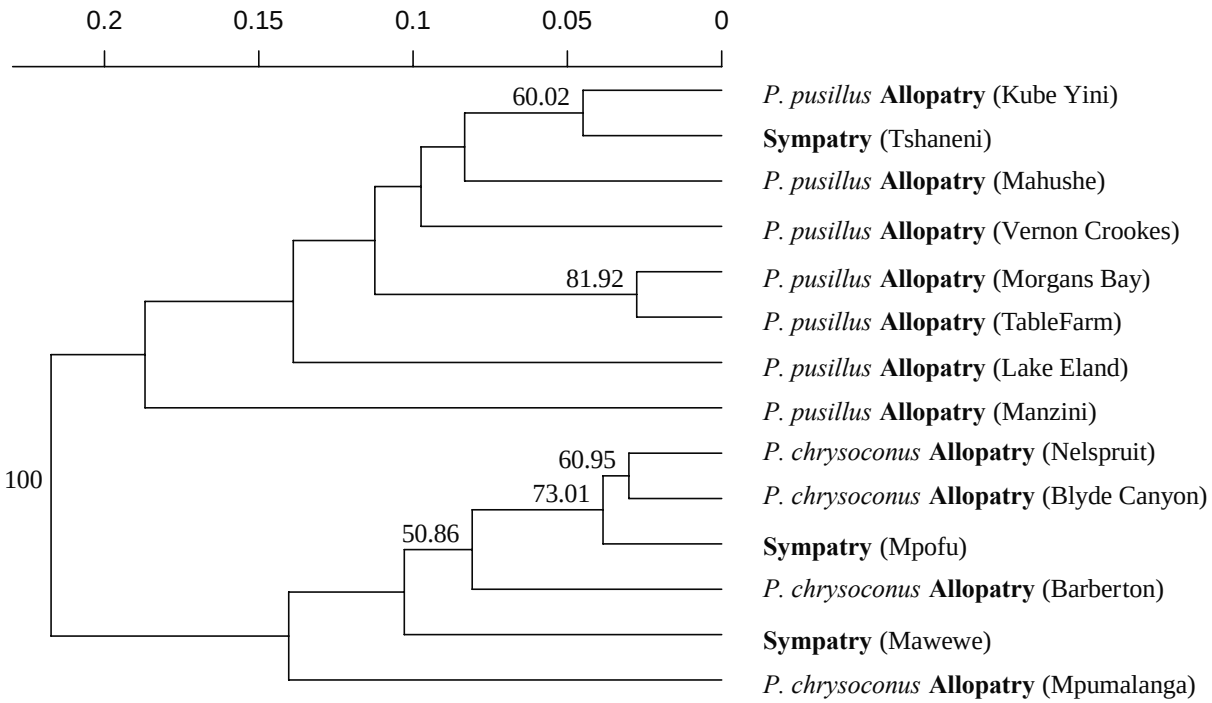

Figure S6. Dendrogram on the genetic distance of the populations using the microsatellite data. Allopatric *P. pusillus pusillus* are more closely related to the sympatric *P. chrysoconus extoni /P. p. pusillus* population from Tshaneni and an allopatric *P. c. extoni* population at Mahushe Shongwe. Allopatric *P. chrysoconus extoni* are more closely related to the sympatric *P. chrysoconus extoni/P. p. pusillus* populations from Mawewe and Mpofu.

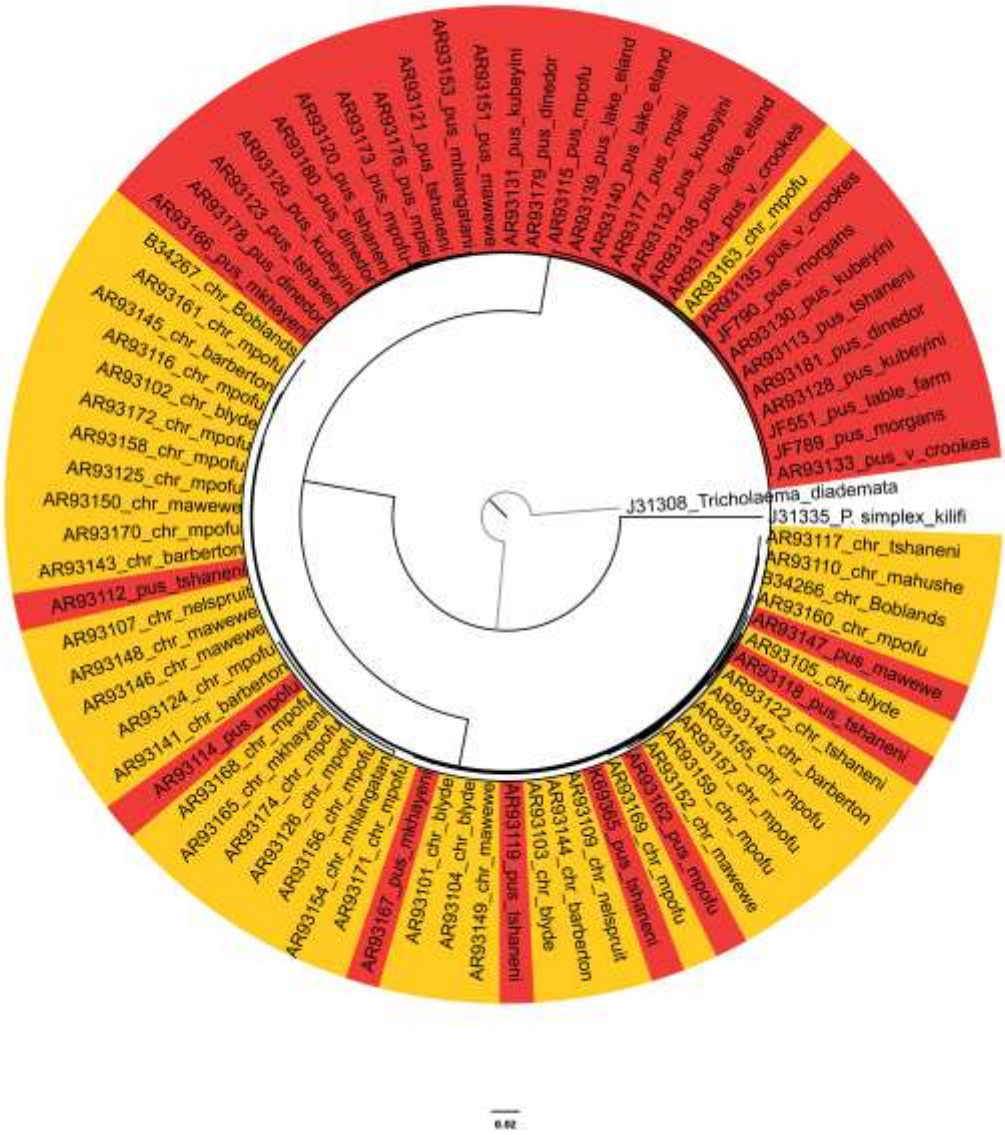

Figure S7. Maximum likelihood phylogenetic tree (RAxML best tree) based on 1033 bp of Cytochrome *b*. Individuals are labelled based on their phenotype such that those highlighted in red are phenotypically red-fronted tinkerbirds and those highlighted in yellow are phenotypically yellow-fronted tinkerbirds. There are eight individuals with red forecrowns and *P. c. extoni* haplotypes, but only one individual with a yellow forecrown and a *P. p. pusillus* haplotype.

Table S1. Factor Loadings on body size variables with varimax rotated PC1 and PC2.

Variables	PC1	PC2
Mass	0.5458	0.1837
Wing	-0.0591	0.8837
Tarsus	0.7025	-0.2795
Tail	0.4529	0.3274

	PC1	PC2
Variance	1.7860	1.2144
Difference	0.5716	.
Proportion	0.4465	0.3036
Cumulative	0.4465	0.7501

Table S2. Factor Loadings on bill size variables with varimax rotated PC1 and PC2.

Variables	PC1	PC2
Culmen	0.5723	0.0573
Bill length	0.6268	-0.0529
Bill depth	0.5287	0.0040
Bill width	-0.0017	0.9969

	PC1	PC2
Variance	2.0366	1.0052
Difference	1.0314	.
Proportion	0.5091	0.2513
Cumulative	0.5091	0.7604

Table S3. GLMM results for Body size (PC1). Values for each variable are from last model in which they were included based on AICc score. Best supported model included only significant fixed effects.

	Estimate	**St. Err.**	**t**	**Pr(>F)**
Intercept	-0.8840	0.1547	-5.714	**<0.00001**
Latitude	0.0967	0.1244	0.778	0.4368
Distance to contact zone	0.6915	0.0759	9.105	**<0.00001**
VCF	-0.0045	0.0092	-0.488	0.6254
Species	-0.1072	0.3395	-0.316	0.7522
Elevation	0.00007	0.0011	0.068	0.9460

Table S4. GLMM results for Body size (PC2). Values for each variable are from last model in which they were included based on AICc score. Best supported model included only significant fixed effects.

	Estimate	St. Err.	t	Pr(>F)
Intercept	0.6008	0.2691	2.232	**0.0266**
Latitude	0.0354	0.2894	0.122	0.9027
Distance to contact zone	0.0857	0.1281	0.669	0.5038
VCF	-0.0063	0.0110	-0.568	0.5697
Species	-0.9422	0.3390	-2.779	**0.0054**
Elevation	0.0020	0.0011	1.791	0.0733

Table S5. GLMM results for Bill size (PC1). Values for each variable are from last model in which they were included based on AICc score. Best supported model included only significant fixed effects.

	Estimate	St. Err.	t	Pr(>F)
Intercept	-0.5674	0.4524	-1.254	0.2097
Latitude	0.2745	0.3781	0.726	0.4678
Distance to contact zone	0.2762	0.2304	1.199	0.2306
VCF	-0.0125	0.0161	-0.774	0.4389
Species	1.1090	0.5163	2.148	**0.0317**
Elevation	-0.0005	0.0027	-0.184	0.8538

Table S6. GLMM results for Bill size (PC2). Values for each variable are from last model in which they were included based on AICc score. Best supported model included only significant fixed effects.

	Estimate	St. Err.	t	Pr(>F)
Intercept	-1.1362	0.5516	-2.060	**0.0394**
Latitude	-0.0936	0.1322	-0.708	0.4787
Distance to contact zone	-0.3402	0.3673	-0.926	0.3543
VCF	-0.0073	0.0114	-0.643	0.5201
Species	0.5830	0.3737	1.560	0.1187
Elevation	0.0028	0.0012	2.374	**0.0176**

Table S7. Differentiation indices per locus

	H_{ST}	H_T	G_{ST}	G_{prime_ST}	D
Bb111TG	0.3147	0.6302	0.5005	0.7526	0.48923
CAM13	0.7608	0.9112	0.1650	0.7258	0.6683
CAM17	0.7307	0.8478	0.1381	0.5495	0.4712
CAM18	0.3739	0.6114	0.3883	0.6435	0.4030
HvoB1TTG	0.0947	0.1076	0.1198	0.1396	0.0151
TG06009	0.2482	0.2306	-0.0760	-0.1081	-0.0248
TG03031	0.1795	0.2034	0.1175	0.1511	0.0309
TG13009	0.3764	0.7303	0.4846	0.8120	0.6192
Tgu06	0.1663	0.2389	0.3037	0.3799	0.0925
TG02088	0.6071	0.7211	0.1580	0.4232	0.3082

Table S8 Genotypic richness and abundance indices

Populations	**N**	**MLG**	**eMLG**	**H**	**G**	**E.5**	**Hexp**
***P. c. extoni* Allopatry**	15	15	15	2.71	15	1	0.367
Sympatry	45	45	15	3.81	45	1	0.543
***P. p. pusillus* Allopatry**	21	21	15	3.04	21	1	0.436
Total	81	81	15	4.39	81	1	0.53

Table S9. Summary of samples used in this study and analyses conducted with the samples

Taxon	Locality	mtDNA	Biometrics/ Reflectance	Catalogue/ Ring number	Source
P. c. extoni	Malawi	MH364232(Cytb), MH364260(ATP6/8)	-	440453	FMNH
P. c. chrysoconus	Jos	MH364226(Cytb), MH364267(ATP6/8)	-	AP88977	Field
P. c. chrysoconus	Jos	MH364223(Cytb), MH364269(ATP6/8)	-	AP88983	Field
P. c. extoni	Blyde River Canyon, South Africa	MH364117(Cytb)	Biometrics+ Reflectance	AR93101	Field
P. c. extoni	Blyde River Canyon, South Africa	MH364118(Cytb)	Biometrics+ Reflectance	AR93102	Field
P. c. extoni	Blyde River Canyon, South Africa	MH364119(Cytb)	Biometrics+ Reflectance	AR93103	Field
P. c. extoni	Blyde River Canyon, South Africa	MH364120(Cytb)	Biometrics+ Reflectance	AR93104	Field
P. c. extoni	Blyde River Canyon, South Africa	MH364121(Cytb)	Biometrics+ Reflectance	AR93105	Field
P. c. extoni	Nelspruit, South Africa	MH364122(Cytb)	Biometrics+ Reflectance	AR93107	Field
P. c. extoni	Nelspruit, South Africa	MH364123(Cytb)	Biometrics+ Reflectance	AR93109	Field
P. c. extoni	Mahushe Shongwe, South Africa	MH364124(Cytb)	Biometrics+ Reflectance	AR93110	Field
P. p. pusillus	Tshaneni, Swaziland	MH364125(Cytb)	Biometrics+ Reflectance	AR93112	Field
P. p. pusillus	Tshaneni, Swaziland	MH364126(Cytb)	Biometrics+ Reflectance	AR93113	Field
P. p. pusillus	Mpofu, Swaziland	MH364127(Cytb)	Biometrics+ Reflectance	AR93114	Field
P. p. pusillus	Mpofu, Swaziland	MH364128(Cytb)	Biometrics+ Reflectance	AR93115	Field
P. c. extoni	Mpofu, Swaziland	MH364129(Cytb)	Biometrics+ Reflectance	AR93116	Field
P. c. extoni	Tshaneni, Swaziland	MH364130(Cytb)	Biometrics+ Reflectance	AR93117	Field

P. p. pusillus	Tshaneni, Swaziland	MH364131(Cytb)	Biometrics+ Reflectance	AR93118	Field
P. p. pusillus	Tshaneni, Swaziland	MH364132(Cytb)	Reflectance	AR93119	Field
P. p. pusillus	Tshaneni, Swaziland	MH364133(Cytb)	Biometrics+ Reflectance	AR93120	Field
P. p. pusillus	Tshaneni, Swaziland	MH364134(Cytb)	Biometrics+ Reflectance	AR93121	Field
P. c. extoni	Tshaneni, Swaziland	MH364135(Cytb)	Biometrics+ Reflectance	AR93122	Field
P. p. pusillus	Tshaneni, Swaziland	MH364136(Cytb)	Biometrics+ Reflectance	AR93123	Field
P. c. extoni	Mpofu, Swaziland	MH364137(Cytb)	Biometrics+ Reflectance	AR93124	Field
P. c. extoni	Mpofu, Swaziland	MH364138(Cytb)	Biometrics+ Reflectance	AR93125	Field
P. c. extoni	Mpofu, Swaziland	MH364139(Cytb)	Biometrics+ Reflectance	AR93126	Field
P. p. pusillus	Kube Yini, South Africa	MH364140(Cytb)	Biometrics+ Reflectance	AR93128	Field
P. p. pusillus	Kube Yini, South Africa	MH364141(Cytb)	Biometrics+ Reflectance	AR93129	Field
P. p. pusillus	Kube Yini, South Africa	MH364142(Cytb)	Biometrics+ Reflectance	AR93130	Field
P. p. pusillus	Kube Yini, South Africa	MH364143(Cytb) MK492415(ATP6/8)	Biometrics+ Reflectance	AR93131	Field
P. p. pusillus	Kube Yini, South Africa	MH364219(Cytb)	Biometrics+ Reflectance	AR93132	Field
P. p. pusillus	Vernon Crookes, South Africa	MH364144(Cytb)	Biometrics+ Reflectance	AR93133	Field
P. p. pusillus	Vernon Crookes, South Africa	MH364218(Cytb) MK492416(ATP6/8)	Biometrics+ Reflectance	AR93134	Field
P. p. pusillus	Vernon Crookes, South Africa	MH364145(Cytb)	Biometrics+ Reflectance	AR93135	Field
P. p. pusillus	Lake Eland, South Africa	MH364217(Cytb)	Biometrics+ Reflectance	AR93138	Field
P. p. pusillus	Lake Eland, South Africa	MH364146(Cytb) MK492417(ATP6/8)	Biometrics+ Reflectance	AR93139	Field
P. p. pusillus	Lake Eland, South Africa	MH364147(Cytb)	Biometrics+ Reflectance	AR93140	Field
P. c. extoni	Nelspruit, South Africa	MH364148(Cytb)	Biometrics+ Reflectance	AR93141	Field
P. c. extoni	Nelspruit, South Africa	MH364149(Cytb)	Biometrics+ Reflectance	AR93142	Field
P. c. extoni	Nelspruit, South Africa	MH364150(Cytb)	Biometrics+ Reflectance	AR93143	Field
P. c. extoni	Nelspruit, South Africa	MH364151(Cytb)	Biometrics+ Reflectance	AR93144	Field

P. c. extoni	Nelspruit, South Africa	MH364152(Cytb)	Biometrics+ Reflectance	AR93145	Field
P. c. extoni	Mawewe, South Africa	MH364153(Cytb)	Reflectance	AR93146	Field
P. p. pusillus	Mawewe, South Africa	MH364154(Cytb)	-	AR93147	Field
P. c. extoni	Mawewe, South Africa	MH364155(Cytb)	Reflectance	AR93148	Field
P. c. extoni	Mawewe, South Africa	MH364156(Cytb)	Reflectance	AR93149	Field
P. c. extoni	Mawewe, South Africa	MH364157(Cytb)	-	AR93150	Field
P. p. pusillus	Mawewe, South Africa	MH364158(Cytb)	Reflectance	AR93151	Field
P. c. extoni	Mawewe, South Africa	MH364159(Cytb)	Reflectance	AR93152	Field
P. p. pusillus	Mhlangatani (Mpofu), Swaziland	MH364160(Cytb)	-	AR93153	Field
P. c. extoni	Mhlangatani (Mpofu), Swaziland	MH364161(Cytb)	Reflectance	AR93154	Field
P. c. extoni	Mpofu, Swaziland	MH364162(Cytb)	Reflectance	AR93155	Field
P. c. extoni	Mpofu, Swaziland	MH364163(Cytb)	-	AR93156	Field
P. c. extoni	Mpofu, Swaziland	MH364164(Cytb)	Reflectance	AR93157	Field
P. c. extoni	Mpofu, Swaziland	MH364165(Cytb)	-	AR93158	Field
P. c. extoni	Mpofu, Swaziland	MH364166(Cytb)	Reflectance	AR93159	Field
P. c. extoni	Mpofu, Swaziland	MH364167(Cytb)	-	AR93160	Field
P. c. extoni	Mpofu, Swaziland	MH364168(Cytb)	-	AR93161	Field
P. p. pusillus	Mpofu, Swaziland	MH364169(Cytb)	Reflectance	AR93162	Field
P. c. extoni	Mpofu, Swaziland	MH364170(Cytb)	Reflectance	AR93163	Field
P. c. extoni	Mkhayeni (Tshaneni), Swaziland	MH364171(Cytb)	-	AR93165	Field
P. p. pusillus	Mkhayeni (Tshaneni), Swaziland	MH364172(Cytb)	Reflectance	AR93166	Field
P. p. pusillus	Mkhayeni (Tshaneni), Swaziland	MH364173(Cytb)	-	AR93167	Field
P. c. extoni	Mpofu, Swaziland	MH364174(Cytb)	Reflectance	AR93168	Field
P. c. extoni	Mpofu, Swaziland	MH364175(Cytb)	-	AR93169	Field

P. c. extoni	Mpofu, Swaziland	MH364176(Cytb)	-	AR93170	Field
P. c. extoni	Mpofu, Swaziland	MH364177(Cytb)	-	AR93171	Field
P. c. extoni	Mpofu, Swaziland	MH364178(Cytb)	Reflectance	AR93172	Field
P. p. pusillus	Mpofu, Swaziland	MH364179(Cytb)	-	AR93173	Field
P. c. extoni	Mpofu, Swaziland	MH364180(Cytb)	-	AR93174	Field
P. c. extoni	Mpofu, Swaziland	MH364181(Cytb)	-	AR93175	Field
P. p. pusillus	Manzini, Swaziland	MH364182(Cytb)	Reflectance	AR93176	Field
P. p. pusillus	Manzini, Swaziland	MH364183(Cytb)	Reflectance	AR93177	Field
P. p. pusillus	Manzini, Swaziland	MH364184(Cytb)	Reflectance	AR93178	Field
P. p. pusillus	Manzini, Swaziland	MH364185(Cytb)	Reflectance	AR93179	Field
P. p. pusillus	Manzini, Swaziland	MH364186(Cytb)	Reflectance	AR93180	Field
P. p. pusillus	Manzini, Swaziland	MH364187(Cytb)	Reflectance	AR93181	Field
P. c. extoni	Mpumalanga, Boblands Farm, South Africa	MH364216(Cytb), MH364271(ATP6/8)	-	B34266, 165339	LSUMZ
P. c. extoni	Mpumalanga, Boblands Farm, South Africa	MH364215(Cytb), MH364272(ATP6/8)	-	B34267, 165340	LSUMZ
P. c. chrysoconus	Ghana	MH364214(Cytb), MH364273(ATP6/8)	-	B39241	LSUMZ
P. c. chrysoconus	Ghana	MH364213(Cytb), MH364274(ATP6/8)	-	B39310	LSUMZ
P. c. chrysoconus	Kongelai, Kenya	MH364209(Cytb), MH364276(ATP6/8)	-	J31301	Field
P. p. affinis	Kongelai, Kenya	MH364208(Cytb), MH364277(ATP6/8)	-	J31305	Field
Tricholaema diademata	Awasi, Kenya	MG697232(Cytb), MG230178(ATP6/8)	-	J31308	Field
P. c. chrysoconus	Awasi, Kenya	MH364207(Cytb), MH364278(ATP6/8)	-	J31310	Field

P. p. affinis	Awasi, Kenya	MH364206(Cytb), MH364279(ATP6/8)	-	J31312	Field
P. p. affinis	Watamu, Kenya	MH364205(Cytb), MH364280(ATP6/8)	-	J31313	Field
P. p. affinis	Diani, Kenya	MH364204(Cytb), MH364281(ATP6/8)	-	J31315	Field
P. simplex	Kilifi, Kenya	MH364282(ATP6/8)	-	J31335	Field
P. c. extoni	Migeregere, Tanzania	MH364203(Cytb), MH364286(ATP6/8)	-	J31360	Field
P. c. extoni	Migeregere, Tanzania	MH364202(Cytb), MH364287(ATP6/8)	-	J31361	Field
P. p. pusillus	Table Farm, South Africa	MH364200(Cytb), MH364289(ATP6/8)	-	JF551	MVZ
P. p. pusillus	Morgan's Bay, Eastern Cape, South Africa	MK492414(Cytb) MK492418(ATP6/8)	-	JF789	MVZ
P. p. pusillus	Morgan's Bay, Eastern Cape, South Africa	MH364292(ATP6/8)	-	JF790	MVZ
P. c. extoni	Katavi NP, Tanzania	MG211668(Cytb), MK492419(ATP6/8)	-	K69302	Field
P. c. extoni	Uvinza, Tanzania	MH364199(Cytb), MH364294(ATP6/8)	-	K69304	Field
P. c. extoni	Uvinza, Tanzania	MG211669(Cytb) MK492420(ATP6/8)	-	K69305	Field
P. simplex	Zanzibar, Bwejuu	MG437418(Cytb), MH364296(ATP6/8)	-	K69323	Field
P. c. extoni	Uvinza, Tanzania	MH364194(Cytb), MH364297(ATP6/8)	-	K69340	Field
P. p. affinis	Watamu, Kenya	MG437478(Cytb), MH364298(ATP6/8)	-	K69359	Field
P. p. affinis	Lamu, Kenya	MH364192(Cytb), MH364299(ATP6/8)	-	K69360	Field
P. leucomystax	Mdandu Forest	MH364243(Cytb)	-	134161	ZMUC

P. p. pusillus	Tshaneni, Swaziland	MK492413(Cytb)	Biometrics	K69365	Field
Lybius melanopterus	GenBank	AY279292(Cytb)	-	AY279292	GenBank
P. p. affinis	Wenge East, Kenya	MH364189(Cytb), MH364303(ATP6/8)	-	T37337	FMNH
Hemignathus flavus	GenBank	NC025608 (Mitogenome)	-	NC025608	GenBank
Hemignathus virens wilsoni	GenBank	KM078802 (Mitogenome)	-	KM078802	GenBank
Hemignathus virens virens	GenBank	KM078788 (Mitogenome)	-	KM078788	GenBank
Sasia ochracea	GenBank	NC028019 (Mitogenome)	-	NC028019	GenBank
Oreomystis bairdi	GenBank	KM078807 (Mitogenome)	-	KM078807	GenBank
Paroreomyza montana	GenBank	KM078771 (Mitogenome)	-	KM078771	GenBank

Table S10. Cline centres and widths and their two log-likelihood support limits for phenotypic traits hue, chroma and brightness of forecrown patch, wing length, as well as STRUCTURE Q scores from microsatellite markers, and the Cytochrome b haplotype.

Trait	Centre	Width	Centre LL(low)	Centre LL(high)	Width LL(low)	Width LL(high)
Hue	-1.0531	17.1062	-4.1545	2.3657	9.8200	29.8049
Chroma	0.0664	1.1003	-4.6985	9.2239	0.0016	29.3527
Brightness	-25.0871	69.3224	-57.799	-7.6021	0.3644	190.577
Wing	2.3918	2.5601	-3.1395	9.1250	2.0035	20.1003
Microsat Q score	15.5867	60.7615	4.3649	70.1248	28.3860	258.8522
Cytb haplotype	6.4703	27.0842	1.2917	15.6109	15.9355	54.9054