

Supplementary Information

Genetic diversity of Italian greater horseshoe bats (*Rhinolophus ferrumequinum*) and distinction of the Sardinian colonies

N. Mucci, A. Palladini, M. Mucedda, E. Pidinchedda, A. Martinoli, D. Russo, G. Bogliani

**Table S1:** Haplotype distribution across colonies: heatmap of the percent coverage of the haplotypes present in each colony. Growing intensity of green indicates a higher proportion of the bats sampled in the colony exhibited that haplotype. Percent values are calculated by dividing the number of individuals with a given haplotype in a colony by the total number of bats sampled in that colony.

	AO	CN_A	CN_B	SV	CO	GO	FC	MO_SC	MO_T	PC_A	PC_T	MC	PG	MT_A	MT_J	FG_C	FG_I	FG_F	S_OG	S_OR	S_CC	S_CA
IT1	100%	91%	36%	38%		50%	8%	52%		26%	21%	64%	63%			82%	9%	64%	29%			
IT2						25%	8%	30%	100%	21%	13%	9%										
IT3							23%															
IT4	9%	45%	36%																			
IT5	9%																					
IT6																9%	36%					
IT7				19%						21%	17%	9%										
IT8				6%			3%	4%				9%	13%									
IT9												9%	13%									
IT10			45%				8%	13%			25%											
IT11							18%						13%									
IT12		9%								5%												
IT13			9%	13%							13%											
IT14			9%	6%																		
IT15				13%																		
IT16						25%																
IT17										11%	8%											
IT18										5%												
IT19										5%												
IT20										5%												
IT21											4%											
IT22																			29%			20%
IT23																			29%	83%	90%	20%
IT24																			14%			
IT25																				17%		60%
IT26																					10%	
IT27							5%															
IT28							3%															
IT29					80%																	
IT30					20%																	
IT31														94%	100%							
IT32														6%								

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**Table S2:** Pairwise analysis for  $F_{st}$  (A), and  $G_{st}$  (B).  $F_{st}$  and  $G_{st}$  indicate the genetic differentiation between two colonies. Their values are below the diagonal, while their respective  $p$ -values are above the diagonal. Probability,  $p(\text{rand} \geq \text{data})$  is based on 999 permutations. Significance threshold is 0.0042 after applying the Bonferroni correction (0.05/12). Significant adjusted  $p$ -values are shown in **bold**.

A) pairwise  $F_{st}$

	AO	CN_A	CN_B	SV	CO	GO	FC	MO_SC	MO_T	PC_A	PC_T	MC	PG	MT_A	MT_J	FG_C	FG_F	FG_I	CA	OG	OR	SS	
AO		0.391	0.019	0.012	0.005	<b>0.002</b>	0.005	0.049	0.186	0.009	0.028	0.075	0.067	0.076	0.259	0.001	0.015	0.005	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	AO
CN_A	0.012		0.655	0.847	0.014	0.055	0.876	0.853	0.562	0.198	0.905	0.51	0.83	0.819	0.847	0.075	0.216	0.112	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	CN_A
CN_B	0.021	0.014		0.705	0.086	0.334	0.187	0.122	0.12	0.252	0.51	0.344	0.109	0.08	0.226	0.024	0.201	0.009	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	CN_B
SV	0.02	0.012	0.015		0.103	0.046	0.541	0.528	0.372	0.335	0.96	0.585	0.586	0.26	0.535	0.33	0.245	0.047	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	SV
CO	0.055	0.044	0.039	0.039		0.008	0.045	<b>0.003</b>	0.019	0.013	0.015	0.005	0.005	<b>0.004</b>	0.007	<b>0.002</b>	<b>0.002</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.002</b>	<b>0.001</b>	CO
GO	0.03	0.021	0.019	0.023	0.055		0.041	<b>0.004</b>	0.046	0.06	0.022	0.053	0.145	<b>0.004</b>	0.068	<b>0.003</b>	0.008	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	GO
FC	0.019	0.011	0.018	0.014	0.04	0.022		0.417	0.255	0.041	0.327	0.068	0.556	0.537	0.186	0.273	0.22	0.018	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FC
MO_SC	0.015	0.01	0.017	0.013	0.048	0.023	0.012		0.458	0.408	0.622	0.318	0.653	0.093	0.316	0.049	0.232	0.172	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MO_SC
MO_T	0.028	0.024	0.033	0.029	0.06	0.04	0.029	0.024		0.389	0.13	0.108	0.443	0.178	0.522	0.117	0.043	0.314	<b>0.001</b>	<b>0.002</b>	<b>0.001</b>	<b>0.001</b>	MO_T
PC_A	0.017	0.015	0.017	0.015	0.044	0.02	0.017	0.012	0.027		0.551	0.392	0.412	0.048	0.481	0.004	0.014	0.011	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	PC_A
PC_T	0.018	0.01	0.015	0.01	0.042	0.022	0.014	0.011	0.031	0.012		0.579	0.389	0.059	0.362	0.028	0.285	0.018	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	PC_T
MC	0.023	0.018	0.021	0.019	0.055	0.028	0.024	0.019	0.037	0.019	0.017		0.49	0.134	0.629	0.093	0.127	0.01	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MC
PG	0.03	0.019	0.031	0.024	0.062	0.032	0.022	0.02	0.036	0.024	0.024	0.028		0.626	0.886	0.368	0.159	0.2	<b>0.001</b>	<b>0.002</b>	<b>0.001</b>	<b>0.001</b>	PG
MT_A	0.017	0.012	0.021	0.017	0.053	0.027	0.013	0.016	0.03	0.018	0.019	0.023	0.022		0.943	0.093	0.08	0.449	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MT_A
MT_J	0.021	0.017	0.026	0.021	0.061	0.031	0.024	0.021	0.033	0.019	0.022	0.024	0.024	0.016		0.347	0.339	0.326	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MT_J
FG_C	0.031	0.022	0.027	0.02	0.062	0.035	0.018	0.021	0.035	0.029	0.024	0.027	0.028	0.022	0.026		0.683	0.194	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FG_C
FG_F	0.028	0.021	0.024	0.023	0.057	0.032	0.021	0.02	0.04	0.027	0.021	0.028	0.033	0.025	0.028	0.02		0.208	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FG_F
FG_I	0.027	0.019	0.027	0.023	0.06	0.038	0.021	0.017	0.029	0.022	0.022	0.031	0.028	0.016	0.024	0.021	0.023		<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FG_I
CA	0.058	0.042	0.059	0.052	0.098	0.063	0.044	0.048	0.057	0.059	0.053	0.061	0.055	0.046	0.061	0.063	0.071	0.05		0.327	0.788	0.417	CA
OG	0.076	0.06	0.066	0.062	0.116	0.076	0.061	0.06	0.077	0.069	0.066	0.07	0.076	0.06	0.077	0.076	0.082	0.056	0.032		0.82	0.152	OG
OR	0.00075	0.054	0.061	0.059	0.1	0.068	0.053	0.057	0.073	0.069	0.062	0.067	0.069	0.059	0.077	0.074	0.081	0.061	0.026	0.028		0.812	OR
SS	0.058	0.042	0.051	0.047	0.088	0.062	0.04	0.044	0.063	0.059	0.049	0.052	0.055	0.046	0.055	0.059	0.066	0.047	0.024	0.033	0.024		SS

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**Table S2:** (continued) Pairwise analysis for  $F_{st}$  (A), and  $G_{st}$  (B).  $F_{st}$  and  $G_{st}$  indicate the genetic differentiation between two colonies. Their values are below the diagonal, while their respective  $p$ -values are above the diagonal. Probability,  $p(\text{rand} \geq \text{data})$  is based on 999 permutations. Significance threshold is 0.0042 after applying the Bonferroni correction (0.05/12). Significant adjusted  $p$ -values are shown in **bold**.

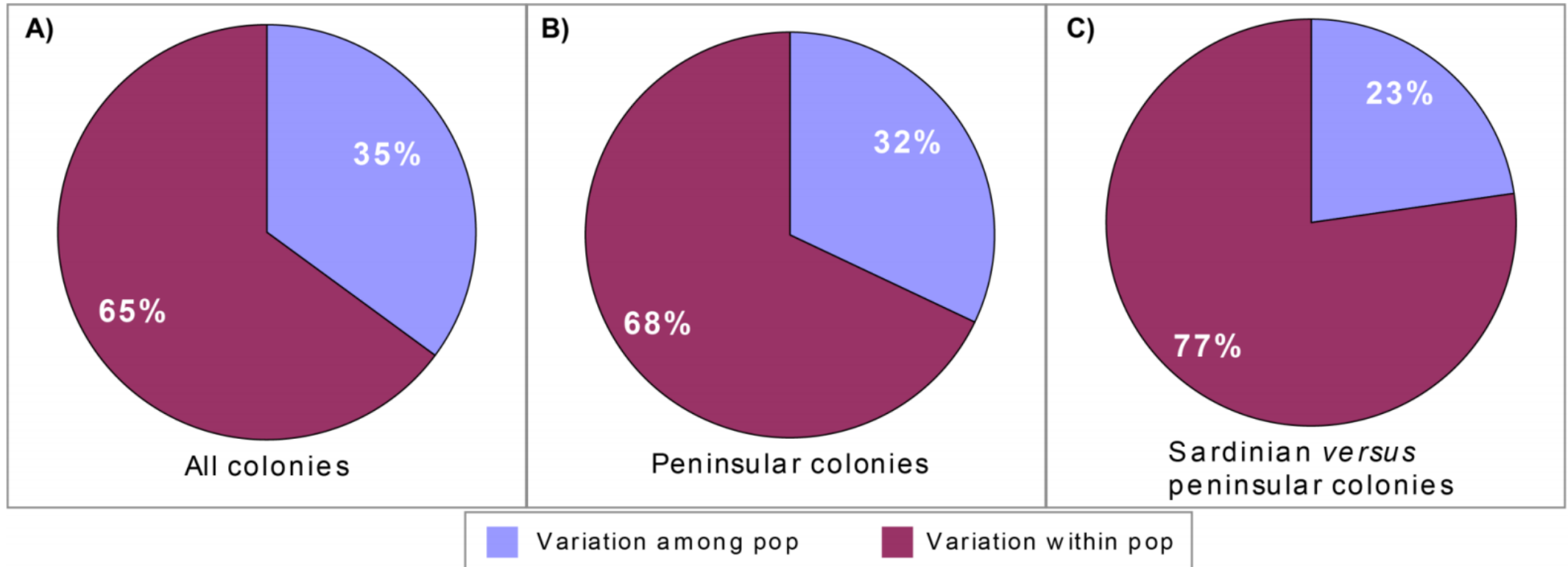
**B) pairwise  $G_{st}$**

	AO	CN_A	CN_B	SV	CO	GO	FC	MO_SC	MO_T	PC_A	PC_T	MC	PG	MT_A	MT_J	FG_C	FG_F	FG_I	CA	OG	OR	SS	
AO		0.391	0.019	0.012	0.005	<b>0.002</b>	0.005	0.049	0.186	0.009	0.028	0.075	0.067	0.076	0.259	<b>0.001</b>	0.015	0.005	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	AO
CN_A	0.012		0.655	0.847	0.014	0.055	0.876	0.853	0.562	0.198	0.905	0.51	0.83	0.819	0.847	0.075		0.112	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	CN_A
CN_B	0.021	0.014		0.705	0.086	0.334	0.187	0.122	0.12	0.252	0.51	0.344	0.109	0.08	0.226	0.024	0.201	0.009	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	CN_B
SV	0.02	0.012	0.015		0.103	0.046	0.541	0.528	0.372	0.335	0.96	0.585	0.586	0.26	0.535	0.33	0.245	0.047	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	SV
CO	0.055	0.044	0.039	0.039		0.008	0.045	<b>0.003</b>	0.019	0.013	0.015	0.005	0.005	<b>0.004</b>	0.007	<b>0.002</b>	<b>0.002</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.002</b>	<b>0.001</b>	CO
GO	0.03	0.021	0.019	0.023	0.055		0.041	<b>0.004</b>	0.046	0.06	0.022	0.053	0.145	<b>0.004</b>	0.068	<b>0.003</b>	0.008	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	GO
FC	0.019	0.011	0.018	0.014	0.04	0.022		0.417	0.255	0.041	0.327	0.068	0.556	0.537	0.186	0.273	0.22	0.018	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FC
MO_SC	0.015	0.01	0.017	0.013	0.048	0.023	0.012		0.458	0.408	0.622	0.318	0.653	0.093	0.316	0.049	0.232	0.172	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MO_SC
MO_T	0.028	0.024	0.033	0.029	0.06	0.04	0.029	0.024		0.389	0.13	0.108	0.443	0.178	0.522	0.117	0.043	0.314	<b>0.001</b>	<b>0.002</b>	<b>0.001</b>	<b>0.001</b>	MO_T
PC_A	0.017	0.015	0.017	0.015	0.044	0.02	0.017	0.012	0.027		0.551	0.392	0.412	0.048	0.481	0.004	0.014	0.011	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	PC_A
PC_T	0.018	0.01	0.015	0.01	0.042	0.022	0.014	0.011	0.031	0.012		0.579	0.389	0.059	0.362	0.028	0.285	0.018	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	PC_T
MC	0.023	0.018	0.021	0.019	0.055	0.028	0.024	0.019	0.037	0.019	0.017		0.49	0.134	0.629	0.093	0.127	0.01	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MC
PG	0.03	0.019	0.031	0.024	0.062	0.032	0.022	0.02	0.036	0.024	0.024	0.028		0.626	0.886	0.368	0.159	0.2	<b>0.001</b>	<b>0.002</b>	<b>0.001</b>	<b>0.001</b>	PG
MT_A	0.017	0.012	0.021	0.017	0.053	0.027	0.013	0.016	0.03	0.018	0.019	0.023	0.022		0.943	0.093	0.08	0.449	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MT_A
MT_J	0.021	0.017	0.026	0.021	0.061	0.031	0.024	0.021	0.033	0.019	0.022	0.024	0.024	0.016		0.347	0.339	0.326	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	MT_J
FG_C	0.031	0.022	0.027	0.02	0.062	0.035	0.018	0.021	0.035	0.029	0.024	0.027	0.028	0.022	0.026		0.683	0.194	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FG_C
FG_F	0.028	0.021	0.024	0.023	0.057	0.032	0.021	0.02	0.04	0.027	0.052	0.028	0.033	0.025	0.028	0.02		0.208	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FG_F
FG_I	0.027	0.019	0.027	0.023	0.06	0.038	0.021	0.017	0.029	0.022	0.022	0.031	0.028	0.016	0.024	0.021	0.023		<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	<b>0.001</b>	FG_I
CA	0.058	0.042	0.059	0.052	0.098	0.063	0.044	0.048	0.057	0.059	0.053	0.061	0.055	0.046	0.061	0.063	0.071	0.05		0.327	0.788	0.417	CA
OG	0.076	0.06	0.066	0.062	0.116	0.076	0.061	0.06	0.077	0.069	0.066	0.07	0.076	0.06	0.077	0.076	0.082	0.056	0.032		0.82	0.152	OG
OR	0.075	0.054	0.061	0.059	0.1	0.068	0.053	0.057	0.073	0.069	0.062	0.067	0.069	0.059	0.077	0.074	0.081	0.061	0.026	0.028		0.812	OR
SS	0.058	0.042	0.051	0.047	0.088	0.062	0.04	0.044	0.063	0.059	0.049	0.052	0.055	0.046	0.055	0.059	0.066	0.047	0.024	0.033	0.024		SS

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**Figure S3:** Analysis of the molecular variance (AMOVA) performed on the haplotypes of A) all colonies, B) peninsular colonies, C) Sardinian against peninsular colonies.

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**Genetic diversity of Italian greater horseshoe bats (*Rhinolophus ferrumequinum*) and distinction of the Sardinian colonies**

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**Figure S4:** Analysis of the molecular variance (AMOVA) performed on the microsatellite loci of A) all colonies, B) peninsular colonies, C) Sardinian against peninsular colonies.

