

Title

Genomic patterns of Iberian wild bees reveal levels of diversity, differentiation and population structure supporting the "refugia within refugia" hypothesis.

Authors

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Supplementary Material

Supplementary information

Tables

Table S1: Information of sampled bees. Specimens that have a longer ID were samples not used in the analysis and for that reason their field code was not changed.

Sample ID	Species	Sampling Site code	Sampling Site name	Latitude	Longitude
Bet_01	Andrena agilissima	Bet	Baetic mountains	36.9556777	-2.9593338
Bet_02	Andrena agilissima	Bet	Baetic mountains	36.9853001	-3.1198463
Bet_03	Andrena agilissima	Bet	Baetic mountains	36.9853001	-3.1198463
Bet_04	Andrena agilissima	Bet	Baetic mountains	36.9853001	-3.1198463
Cat_01	Andrena agilissima	Cat	Catalonia	40.8754721	0.5007094
And_Cat_001_Poll128	Andrena agilissima	Cat	Catalonia	40.8754721	0.5007094
Fzz_01	Andrena agilissima	Fzz	Ferreira do Zêzere	39.6350900	-8.2916300
Gal_01	Andrena agilissima	Gal	Galicia	42.4680732	-7.2613812
Gal_02	Andrena agilissima	Gal	Galicia	42.4680732	-7.2573163
Gal_03	Andrena agilissima	Gal	Galicia	42.4680732	-7.2613812
Gal_04	Andrena agilissima	Gal	Galicia	42.4747255	-7.2782602
And_Gua_001_Poll_121	Andrena agilissima	Gua	Guadalajara	40.7815084	-3.2310376
Jac_01	Andrena agilissima	Jac	Jaca	42.5653434	-0.5154760
Jac_02	Andrena agilissima	Jac	Jaca	42.5653434	-0.5154760

And_Jac_045_Poll_130	Andrena agilissima	Jac	Jaca	42.5653434	-0.5154760
And_TurA_016_Poll71	Andrena agilissima	Tur	Turcufal	39.046187	-9.271455
And_01	Andrena flavipes	And	Andalucia	37.4890027	-3.6120980
And_02	Andrena flavipes	And	Andalucia	37.4890027	-3.6120980
And_03	Andrena flavipes	And	Andalucia	37.4890027	-3.6120980
And_04	Andrena flavipes	And	Andalucia	37.4890027	-3.6120980
And_05	Andrena flavipes	And	Andalucia	37.4890027	-3.6120980
And_06	Andrena flavipes	And	Andalucia	37.4890027	-3.6120980
Bej_01	Andrena flavipes	Bej	Bejar	40.3727018	-5.8220921
Bej_02	Andrena flavipes	Bej	Bejar	40.3727018	-5.8220921
Bej_03	Andrena flavipes	Bej	Bejar	40.3727018	-5.8220921
Bej_04	Andrena flavipes	Bej	Bejar	40.3727018	-5.8220921
Bet_01	Andrena flavipes	Bet	Baetic mountains	36.9853001	-3.1198463
Bet_02	Andrena flavipes	Bet	Baetic mountains	36.9853001	-3.1198463
Bet_03	Andrena flavipes	Bet	Baetic mountains	36.9853001	-3.1198463
And_Bet_015_Poll123	Andrena flavipes	Bet	Baetic mountains	36.9853001	-3.1198463
Cat_01	Andrena flavipes	Cat	Catalonia	40.8632260	0.3866056
Cat_02	Andrena flavipes	Cat	Catalonia	40.8632260	0.3866056
Cat_03	Andrena flavipes	Cat	Catalonia	40.8632260	0.3866056
Cat_04	Andrena flavipes	Cat	Catalonia	40.8632260	0.3866056
Clm_01	Andrena flavipes	Clm	Castilla-La Mancha	39.0334127	-1.9019997
Clm_02	Andrena flavipes	Clm	Castilla-La Mancha	39.0334127	-1.9019997
Clm_03	Andrena flavipes	Clm	Castilla-La Mancha	39.0334127	-1.9019997
Clm_04	Andrena flavipes	Clm	Castilla-La Mancha	39.0334127	-1.9019997
Fvf_01	Andrena flavipes	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Fvf_02	Andrena flavipes	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Fvf_03	Andrena flavipes	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
AND_FVFN_009_poll015	Andrena flavipes	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Gal_01	Andrena flavipes	Gal	Galicia	42.4747255	-7.2782602
Gal_02	Andrena flavipes	Gal	Galicia	42.4747255	-7.2782602
Gal_03	Andrena flavipes	Gal	Galicia	42.4628360	-7.2573163
Gal_04	Andrena flavipes	Gal	Galicia	42.4628360	-7.2573163
Jac_01	Andrena flavipes	Jac	Jaca	42.5653434	-0.5154760
Jac_02	Andrena flavipes	Jac	Jaca	42.5653434	-0.5154760
Jac_03	Andrena flavipes	Jac	Jaca	42.5653434	-0.5154760
Jac_04	Andrena flavipes	Jac	Jaca	42.5653434	-0.5154760
Mal_01	Andrena flavipes	Mal	Malcata mountain	40.3043900	-6.9626670
Mal_02	Andrena flavipes	Mal	Malcata mountain	40.3043900	-6.9626670
Mal_03	Andrena flavipes	Mal	Malcata mountain	40.3043900	-6.9626670
Mal_04	Andrena flavipes	Mal	Malcata mountain	40.3043900	-6.9626670
Mal_05	Andrena flavipes	Mal	Malcata mountain	40.3043900	-6.9626670
Mal_06	Andrena flavipes	Mal	Malcata mountain	40.3043900	-6.9626670
Mgr_01	Andrena flavipes	Mgr	Marinha Grande	39.8628610	-8.9329570
Mgr_02	Andrena flavipes	Mgr	Marinha Grande	39.8584490	-8.8355940
Mgr_03	Andrena flavipes	Mgr	Marinha Grande	39.8584490	-8.8355940
Mgr_04	Andrena flavipes	Mgr	Marinha Grande	39.8582690	-8.8840710
Ode_01	Andrena flavipes	Ode	Odeceixe	37.8163900	-8.6834380
Ode_02	Andrena flavipes	Ode	Odeceixe	37.8163900	-8.6834380
Ode_03	Andrena flavipes	Ode	Odeceixe	37.4237790	-8.7659270

Ode_04	Andrena flavipes	Ode	Odeceixe Aire e Candeeiros	37.6012850	-8.6945060
Sda_01	Andrena flavipes	Sda	mountain	39.4924514	-8.9299633
Sin_01	Andrena flavipes	Sin	Sintra	38.8961060	-9.4332770
Sin_02	Andrena flavipes	Sin	Sintra	38.8910270	-9.3962270
Sin_03	Andrena flavipes	Sin	Sintra	38.8910270	-9.3962270
Sin_04	Andrena flavipes	Sin	Sintra	38.8976390	-9.4191060
Sma_01	Andrena flavipes	Sma	São Mamede mountain	39.3751530	-7.3891110
Sma_02	Andrena flavipes	Sma	São Mamede mountain	39.3489450	-7.3493450
Sma_03	Andrena flavipes	Sma	São Mamede mountain	39.3751530	-7.3891110
Sma_04	Andrena flavipes	Sma	São Mamede mountain	39.3751530	-7.3891110
Sma_05	Andrena flavipes	Sma	São Mamede mountain	39.3751530	-7.3891110
Sma_06	Andrena flavipes	Sma	São Mamede mountain	39.3751530	-7.3891110
And_01	Lasioglossum malachurum	And	Andalucia	37.0556040	-4.5907680
And_02	Lasioglossum malachurum	And	Andalucia	36.6799310	-5.7001470
And_03	Lasioglossum malachurum	And	Andalucia	36.6799310	-5.7001470
And_04	Lasioglossum malachurum	And	Andalucia	36.6658950	-5.6745270
Bet_01	Lasioglossum malachurum	Bet	Baetic mountains	37.3201375	-3.3356380
Bet_02	Lasioglossum malachurum	Bet	Baetic mountains	37.3201375	-3.3356380
Bet_03	Lasioglossum malachurum	Bet	Baetic mountains	37.3200153	-3.3355448
Lgl_Bet_015_Poll40	Lasioglossum malachurum	Bet	Baetic mountains	37.3200153	-3.3355448
Cat_01	Lasioglossum malachurum	Cat	Catalonia	40.8632260	0.3866056
Cat_02	Lasioglossum malachurum	Cat	Catalonia	40.8632260	0.3866056
Cat_03	Lasioglossum malachurum	Cat	Catalonia	40.8746670	0.3968107
Cat_04	Lasioglossum malachurum	Cat	Catalonia	40.8632260	0.3866056
Clm_01	Lasioglossum malachurum	Clm	Castilla-La Mancha	38.9280097	-1.7369577
Fvf_01	Lasioglossum malachurum	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Fvf_02	Lasioglossum malachurum	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Fvf_03	Lasioglossum malachurum	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Fvf_04	Lasioglossum malachurum	Fvf	Foros de Vale Figueira	38.7184430	-8.3245650
Lgl_FvfN_009_poll016	Lasioglossum malachurum	Fvf	Foros de Vale Figueira	38.7108960	-8.3135350
Lgl_FvfN_012_poll016	Lasioglossum malachurum	Fvf	Foros de Vale Figueira	38.7108960	-8.3135350
Fzz_01	Lasioglossum malachurum	Fzz	Ferreira do Zezere	39.6894830	-8.3109010
Fzz_01	Lasioglossum malachurum	Fzz	Ferreira do Zezere	39.6875990	-8.3143900

Fzz_03	Lasioglossum malachurum	Fzz	Ferreira do Zezere	39.6350900	-8.2916300
Fzz_04	Lasioglossum malachurum	Fzz	Ferreira do Zezere	39.6875990	-8.3143900
Gua_01	Lasioglossum malachurum	Gua	Guadalajara	40.7562230	-3.1701773
Gua_02	Lasioglossum malachurum	Gua	Guadalajara	40.7815084	-3.2310376
Gua_03	Lasioglossum malachurum	Gua	Guadalajara	40.7562230	-3.1701773
Gua_04	Lasioglossum malachurum	Gua	Guadalajara	40.7562230	-3.1701773
Mgr_01	Lasioglossum malachurum	Mgr	Marinha Grande	39.8582690	-8.8840710
Mgr_02	Lasioglossum malachurum	Mgr	Marinha Grande	39.8582690	-8.8840710
Mgr_03	Lasioglossum malachurum	Mgr	Marinha Grande	39.8582690	-8.8840710
Lgl_MgrN_004_Poll097	Lasioglossum malachurum	Mgr	Marinha Grande	39.8633300	-8.9293000
Lgl_MgrN_004_Poll098	Lasioglossum malachurum	Mgr	Marinha Grande	39.8680390	-8.9236690
Ode_01	Lasioglossum malachurum	Ode	Odeceixe	37.4377909	-8.7987614
Ode_02	Lasioglossum malachurum	Ode	Odeceixe	37.8163900	-8.6834380
Ode_03	Lasioglossum malachurum	Ode	Odeceixe	37.4377909	-8.7987614
Sda_01	Lasioglossum malachurum	Sda	Aire e Candeeiros mountain	39.4924514	-8.9299633
Sin_01	Lasioglossum malachurum	Sin	Sintra	38.8956340	-9.4297740
Sin_02	Lasioglossum malachurum	Sin	Sintra	38.8016958	-9.4844644
Sin_03	Lasioglossum malachurum	Sin	Sintra	38.7972590	-9.4796230
Lgl_SinN_016_Poll102	Lasioglossum malachurum	Sin	Sintra	38.7827830	-9.4702320
Lgl_SinN_002_Poll105	Lasioglossum malachurum	Sin	Sintra	38.8851760	-9.3969830
Sma_01	Lasioglossum malachurum	Sma	São Mamede mountain	39.3489450	-7.3493450

Table S2: Distribution of samples across sampling area. For each species the sampling sites, the number of samples per sampling site and the total number of individuals retained after filtering (Total) is given.

Species	And	Bej	Bet	Cat	Clm	Fvf	Fzz	Gal	Gua
<i>A. agilisissima</i>	0	0	4	1	0	0	1	4	0
<i>A. flavipes</i>	6	4	3	4	4	3	0	4	0
<i>L. malachurum</i>	4	0	3	4	1	4	4	0	4

Species	Jac	Mal	Mgr	Ode	Sda	Sin	Sma	Total
<i>A. agilisissima</i>	2	0	0	0	0	0	0	12
<i>A. flavipes</i>	4	6	4	4	1	4	6	57
<i>L. malachurum</i>	0	0	3	3	1	3	1	35

Table S3: Filtering SNPs. From left to right for each species: number of single nucleotide polymorphic sites (SNPs) before filtering with *vcftools*; number of SNPs after filtering with *vcftools* with only one random SNP per locus and with all SNPs per locus; number of sampled individuals and number of analyzed individuals.

Species	SNPs before filtering	SNPs after filtering		Sampled Individuals	Analyzed Individuals
		One random SNP per locus	All SNPs per locus		
<i>A. agilissima</i>	43451	14230	26321	16	12
<i>A. flavipes</i>	62620	27822	543914	59	57
<i>L. malachurum</i>	72772	33810	256976	43	35

Table S4: Summary statistics by populations: Number of individuals per population (**Npop**); Observed heterozygosity (**Ho**); Expected heterozygosity (**He**); Inbreeding coefficient (**F_{IS}**); and Nucleotide diversity (**π**). For **Ho**, **F_{IS}** and **π** it is provided the average value and respective standard deviation.

Species/Populations	Npop	Ho	He	F _{IS}	π
<i>A. agilissima</i>					
West	5	0.2277 ± 0.273	0.2819	0.165 ± 0.563	0.2819 ± 0.22
Ebro	3	0.2301 ± 0.299	0.2735	0.0968 ± 0.593	0.2735 ± 0.253
Baetic	4	0.1736 ± 0.248	0.2985	0.3767 ± 0.581	0.2985 ± 0.229
<i>A. flavipes</i>					
Iberia	53	0.0809 ± 0.122	0.1027	0.2791 ± 0.405	0.1027 ± 0.123
Pyrenean	4	0.0781 ± 0.190	0.0847	0.0415 ± 0.477	0.0847 ± 0.176
<i>L. malachurum</i>					
Ebro	4	0.1799 ± 0.239	0.1921	0.0303 ± 0.417	0.1921 ± 0.217
Iberia	31	0.1813 ± 0.155	0.1951	0.081 ± 0.279	0.1951 ± 0.148

Table S5: Global differentiation levels by species: Weir and Cockrham's (1984) **F_{ST}**; Hedrick (2005) **G'_{ST}**; and **Jost's D** (2008).

Species	F _{ST}	G' _{ST}	Jost's D
<i>A. agilissima</i>	0.0957	0.1973	0.0654
<i>A. flavipes</i>	0.5611	0.4352	0.1579
<i>L. malachurum</i>	0.0663	0.0896	0.0187

Table S6: Pairwise differentiation statistics. Hedrick (2005) G'_{ST} values below the diagonal and Jost's D (2008) above diagonal.

<i>A. agilisima</i>			
	West	Ebro	Baetic
West	-	0.0646	0.0650
Ebro	0.199	-	0.0668
Baetic	0.1933	0.1999	-
<i>A. flavipes</i>			
	Iberia	Pyrennes	
Iberia	-	0.1579	
Pyrenean	0.6686	-	
<i>L. malachurum</i>			
	Ebro	Iberia	
Ebro	-	0.0187	
Iberia	0.0896	-	

Figures

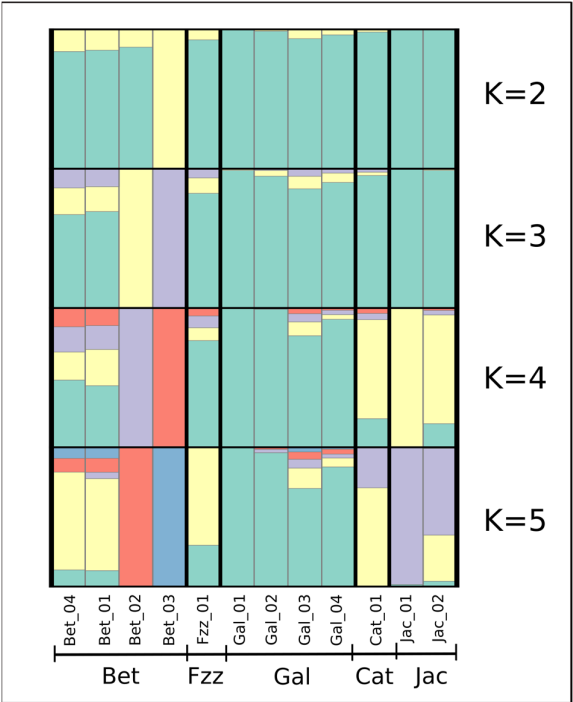


Figure S1: Structure analysis for *Andrena agilissima* using ALStructure for $k = [2:5]$.

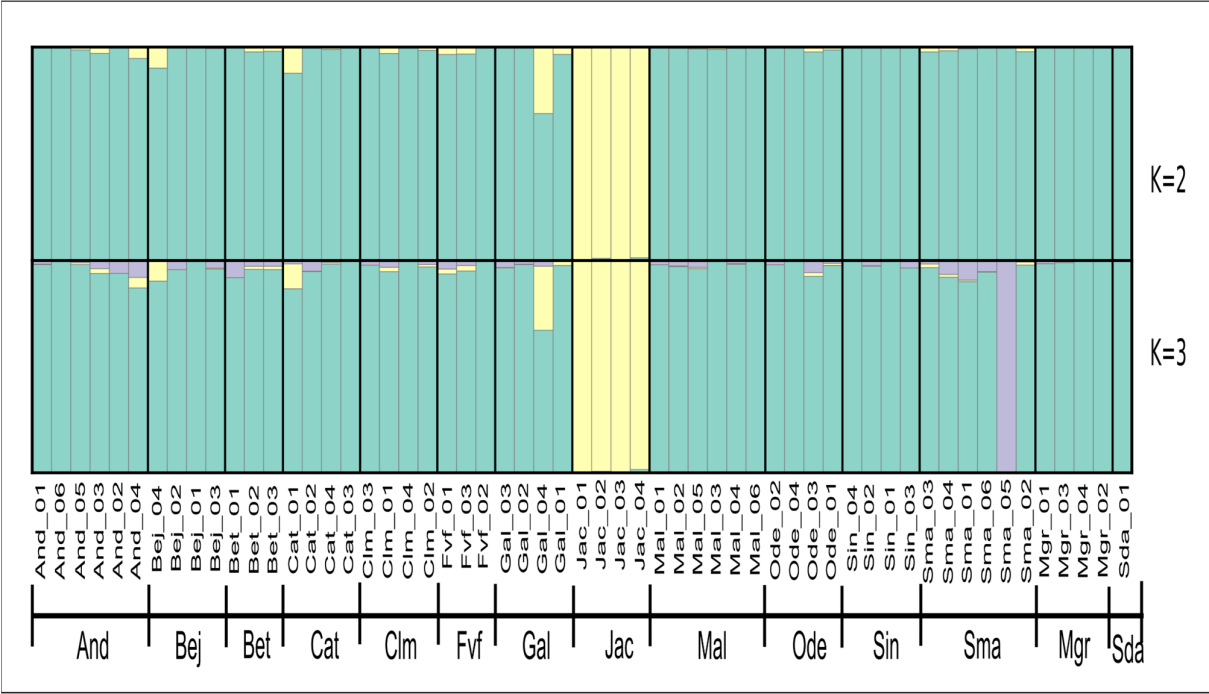


Figure S2: Structure analysis for *Andrena flavipes* using ALStructure for $k = [2:3]$.

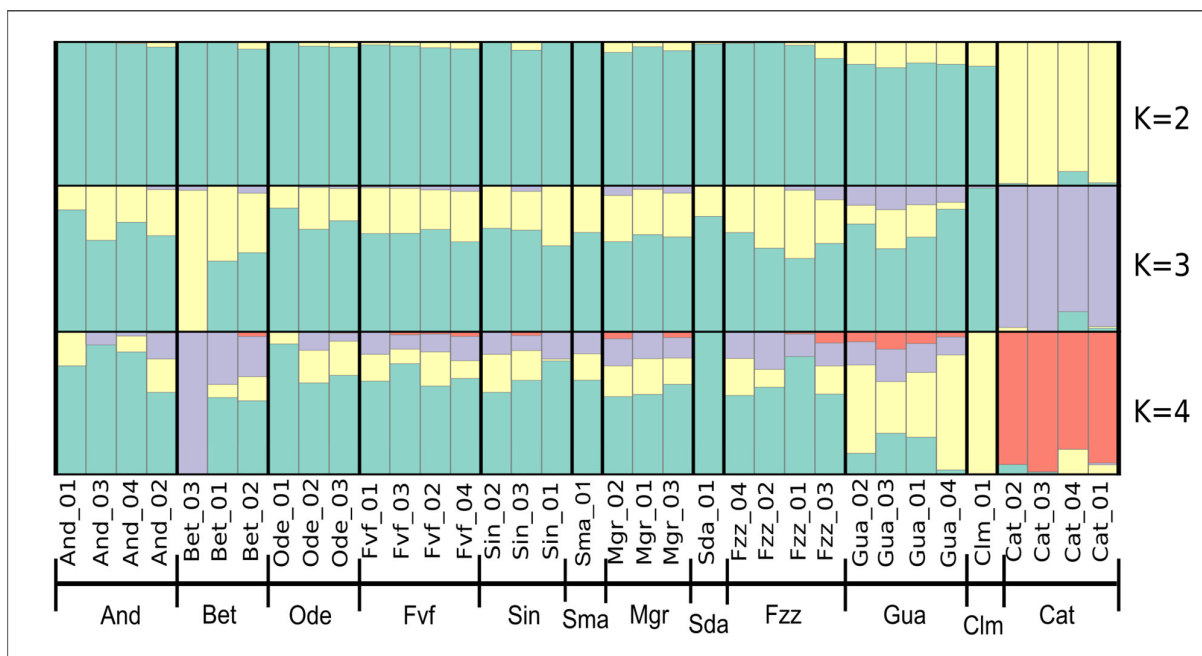


Figure S3: Structure analysis for *Lasioglossum malachurum* using ALStructure for $k= [2:4]$.

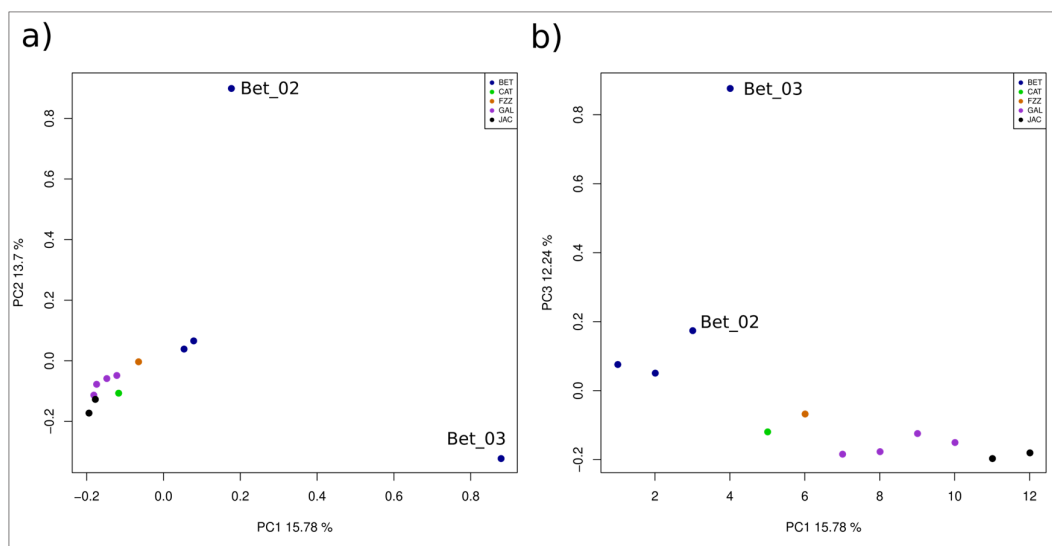


Figure S4: Principal components analysis of *Andrena agilissima*. a) PC1 and PC2 and b) PC1 and PC3.

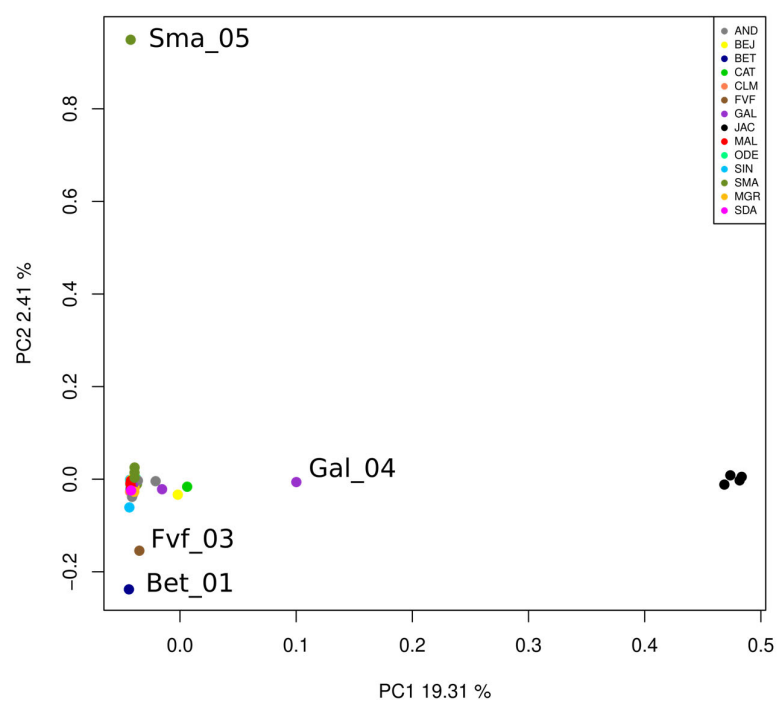


Figure S5: Principal components analysis of *Andrena flavipes*.

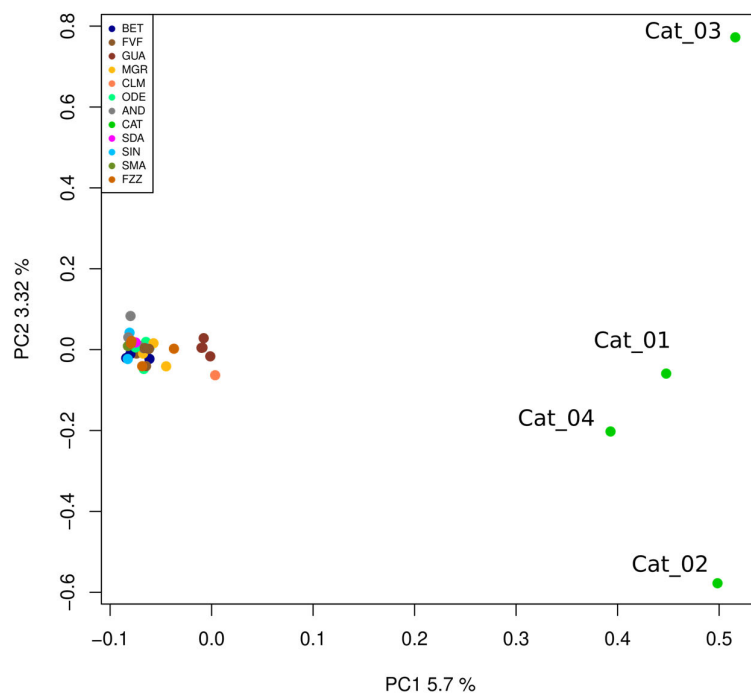


Figure S6: Principal components analysis of *Lasioglossum malachurum*.