Ecography

ECOG-03074
Williams, P. H., Lobo, J. M. and Meseguer, A. S. 2017. Bumblebees take the high road: climatically integrative biogeography shows that escape from Tibet, not Tibetan uplift, is associated with divergences of present-day *Mendacibombus*. – Ecography doi: 10.1111/ecog.03074

Supplementary material

Appendix 1

Table A1. Primer sequences for the sequenced regions of the four genes.

Gene	Primer name	Primer sequence	Reference
COI	LepF	5’- ATTCAACCAATCATAAAGATATTGG -3’	Hebert et al. 2004
	LepR	5’- TAAACTTCTGGATGTCCAAAAAATCA -3’	Hebert et al. 2004
16S	16SWb	5'- CACCTGTTTATCACCTGTTTATCAAAAACAT -3'	Dowton and Austin 1994
	874-16SlR	5'- TATAGATAGAAACCAAYCTG -3'	Cameron et al. 1992
Opsin	LWRhF	5'- AATTGCTATTAYGARACNTGGGT -3'	Mardulyn and Cameron 1999
	LWRhR	5'- ATATGGAGTCCANGCCATRAACCA -3'	Mardulyn and Cameron 1999
PEPCK	FHv4	5'- TGTATRATAATTCGCAAYTTCAC -3'	Cameron et al. 2007
	RHv4	5'- CTGCTGGRGTYCTAGATCC -3'	Cameron et al. 2007

Table A2. GenBank accession numbers for gene sequences with the species' names (in the genus *Bombus*) listed according to Williams *et al*. (2016), together with COI barcode numbers from the BOLD database (boldsystems.org). Outgroup species are shown with *. Sequences are also available from PW.

Species	COI barcode	16S	Opsin	PEPCK
*confusus**	6876D11	DQ787992	DQ788324	EF051016
*soroeensis**	1550E10	DQ788107	AF493008	EF051013
*difficillimus**	6876C09	DQ787998	DQ788327	EF050990
*gerstaeckeri**	FBHAP779-09	DQ788017	DQ788335	EF051003
*balteatus**	NOAPI567	DQ787974	AY739455	EF050870
*bimaculatus**	4485F03	DQ787978	AY739456	EF050847
*nobilis**	HJNO00012	DQ788071	AY739485	EF050912
superbus	HJS1	KX452097	KX452101	KX452104
waltoni	1550D06	DQ788134	DQ788385	EF051022
convexus	6876B05	DQ787993	DQ788325	EF051021
makarjini	6875G01	–	–	–
marussinus	19992G06	–	–	–
avinoviellus	1552D05	AY268416	AY268394	EF051020
himalayanus	6876D08	–	–	–
turkestanicus	6875F08	KX452098	KX452102	KX452105
defector	6875A05	KX452099	KX452103	KX452106
margreiteri	9808D12	KX452100	AF493025	KX452107
handlirschianus	1549E10	DQ788022	DQ788337	EF051017
mendax	6876B08	DQ788057	AF493024	EF051019

Table A3. Principal events in the most likely biogeographic reconstruction for species of the subgenus *Mendacibombus* from the S-DIVA analysis shown in Fig. 5a using the estimate of phylogeny for all species from Fig. 4 and the short-distance dispersal model in Fig. 3 with other bumblebees as the outgroup. Letters represent the areas listed in Table 1; arrows, range-state transitions; caret, local duplicate daughter populations; vertical lines, daughter species' ranges. Where alternative explanations of events exist, only one is shown.

Event	Ancestral range	Processes	Dispersal	Vicariance	Extinction	Daughter ranges
1	GHP	GHP → GH|P	0	1	0	GH, P
2	G	GH → G|H	0	1	0	G, H
3	P	P → P^P → P|PT	1	0	0	P, PT
4	P	P → P^P → P|PCA	2	0	0	P, PCA
5	P	P → P^P → PH|HPT	3	0	0	PH, HPT
6	HPT	HPT → PT|H	0	1	0	PT, H
7	PH	PH → P|H	0	1	0	P, H
8	PCA	PCA → PCATM → PCA|TM	2	1	0	PCA, TM

Table A4**.** Principal events in the most likely biogeographic reconstruction for species of the subgenus *Mendacibombus* from the S-DEC analysis shown in Fig. 5b using the estimate of phylogeny for all species from Fig. 4 and the short-distance dispersal model in Fig. 3 with other bumblebees as the outgroup. Letters represent the areas listed in Table 1; arrows, range-state transitions; caret, local duplicate daughter populations; vertical lines, daughter species' ranges. Where alternative explanations of events exist, only one is shown.

Event	Ancestral range	Processes	Dispersal	Vicariance	Extinction	Daughter ranges
1	GHP	GHP → GP → G|P	0	1	1	G, P
2	G	G → G^G → G|GH	1	0	0	G, GH
3	P	P → P^P → P|PT	1	0	0	P, PT
4	P	P → P^P → P|PTC	2	0	0	P, PTC
5	P	P → P^P → PH|HPT	3	0	0	PH, HPT
6	HPT	HPT → PT|H	0	1	0	PT, H
7	PH	HP → P|H	0	1	0	P, H
8	PTC	PTC → TC → TMCA → CA|TM	2	1	1	CA, TM

Table A5. Ancestral state reconstruction results for the selected variables. Node numbers correspond with node numbers in Fig. A1.

Node number		1	2	3	4	5	6	7	8	9	10	11
Annual variation in precipitation	Ancestral state	81.29	85.33	69.93	70.33	70.84	114.31	66.05	65.44	79.24	69.23	68.94
	Variance	200.92	192.63	129.21	85.63	75.67	137.67	74.43	77.04	88.88	78.34	71.43
	Lower 95% CI	53.51	58.13	47.65	52.19	53.79	91.31	49.14	48.24	60.76	51.88	52.37
	Upper 95% CI	109.07	112.53	92.20	88.46	87.89	137.31	82.96	82.64	97.71	86.58	85.50
Aridity	Ancestral state	64.67	66.58	59.32	60.15	57.60	59.19	52.43	51.37	60.03	70.30	72.39
	Variance	250.31	239.98	160.96	106.68	94.27	171.51	92.72	95.98	110.73	97.60	88.98
	Lower 95% CI	33.66	36.22	34.45	39.91	38.57	33.52	33.56	32.17	39.41	50.93	53.90
	Upper 95% CI	95.68	96.94	84.18	80.40	76.63	84.86	71.30	70.57	80.66	89.66	90.87
Max month temperature	Ancestral state	10.08	9.57	11.51	12.19	12.36	10.01	12.75	12.94	12.13	12.74	12.93
	Variance	2.77	2.65	1.78	1.18	1.04	1.90	1.02	1.06	1.22	1.08	0.98
	Lower 95% CI	6.82	6.38	8.90	10.07	10.36	7.31	10.77	10.92	9.97	10.70	10.99
	Upper 95% CI	13.34	12.76	14.13	14.32	14.36	12.71	14.73	14.96	14.30	14.77	14.88
Min month precipitation	Ancestral state	9.04	7.58	13.16	16.47	14.89	4.53	13.06	12.43	14.29	27.38	34.62
	Variance	207.13	198.58	133.20	88.27	78.00	141.93	76.73	79.42	91.63	80.77	73.63
	Lower 95% CI	-19.17	-20.04	-9.46	-1.94	-2.42	-18.82	-4.11	-5.04	-4.47	9.76	17.80
	Upper 95% CI	37.25	35.20	35.78	34.89	32.20	27.88	30.23	29.90	33.05	44.99	51.44

Table A6. Model fit comparisons for the studied variables based on the Akaike information criterion with a correction for the small sample-size (AICc). AICc differences (delta) equal to zero indicate the best model and are highlighted in bold. Comparisons were performed assuming no measurement error, and with estimation of this error. The models compared were: Brownian motion (BM, neutral drift); Ornstein-Uhlenbeck (OU, adaptive selection); early burst (EB, initial rapid evolution); and white noise (WN, random signal).

		BM	OU	EB	WN
Annual variance in precipitation	AICc	114.24	117.91	117.12	118.75
	AICc_SE	117.91	122.62	121.83	122.41
	dAICc	0.00	3.67	2.88	4.51
	dAICc_SE	0.00	4.71	3.92	4.51
Aridity	AICc	116.88	116.26	120.54	112.59
	AICc_SE	120.54	120.97	125.25	116.26
	dAICc	4.29	3.67	7.95	0.00
	dAICc_SE	4.28	4.71	9.00	0.00
Maximum monthly temperature	AICc	62.81	66.43	66.48	64.34
	AICc_SE	66.37	71.15	71.09	68.01
	dAICc	0.00	3.62	3.67	1.53
	dAICc_SE	0.00	4.77	4.71	1.64
Minimum monthly precipitation	AICc	114.60	113.51	118.27	109.90
	AICc_SE	118.27	118.23	122.98	113.57
	dAICc	4.70	3.61	8.37	0.00
	dAICc_SE	4.70	4.66	9.42	0.00

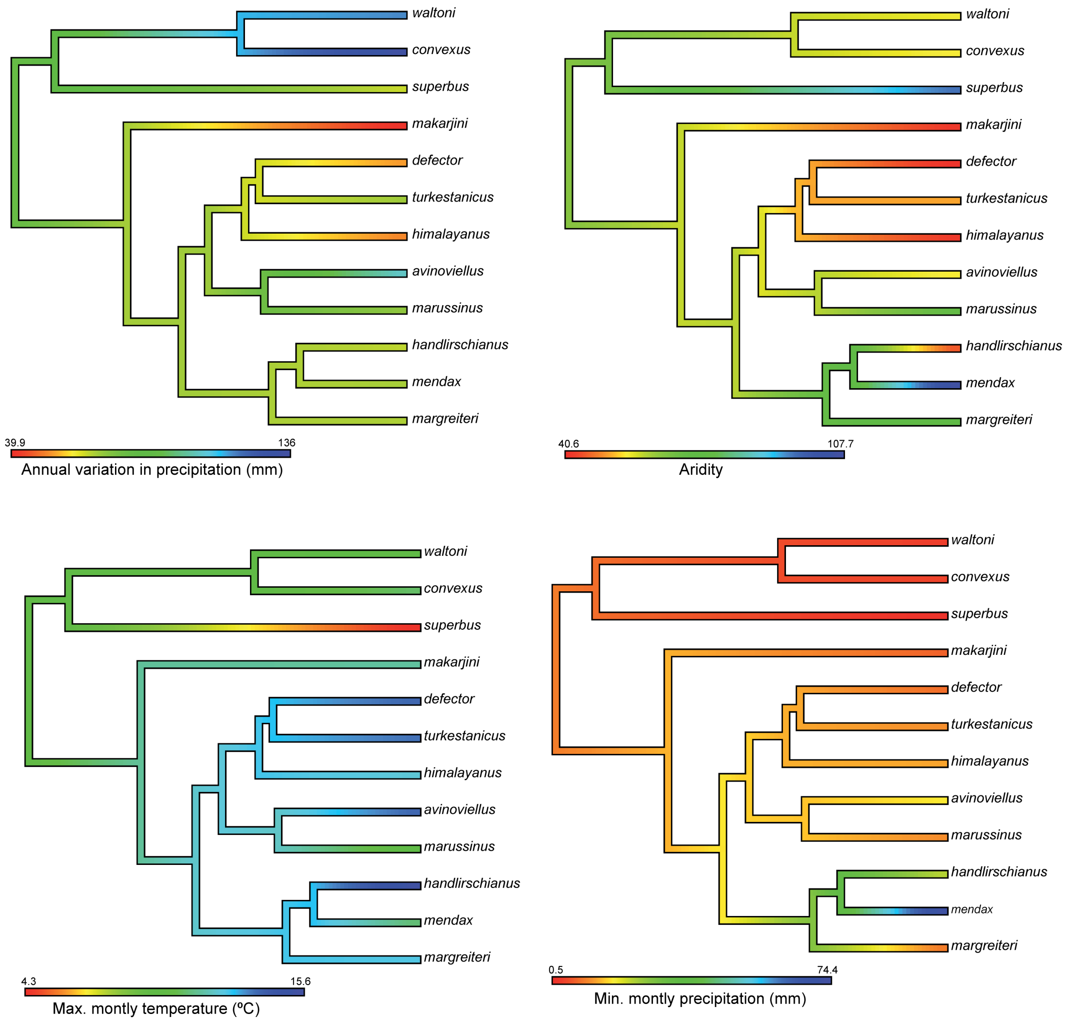

Figure A1. Trees showing the evolution of the ancestral climate niche within each of the selected climate variables along the dated tree of *Mendacibombus* species, estimated with *Phytools* from species' current climate niches from their current distributions (Fig. 1a) and the chronogram in Fig. 4. For each tree, colours on branches correspond with the estimated value of the trait on the scale bars shown below each tree.

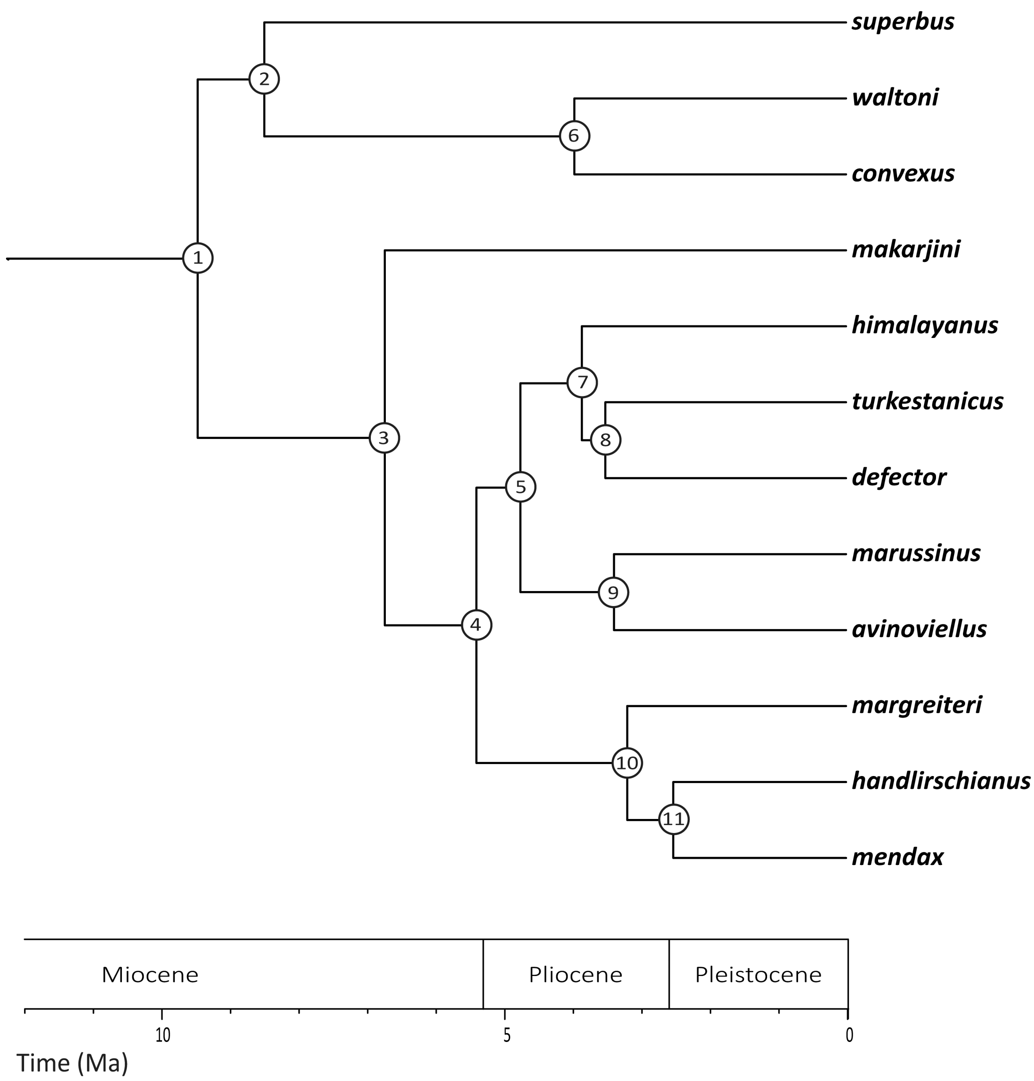

Figure A2. Dated estimate of phylogeny for all extant species of the subgenus *Mendacibombus* by linked Bayesian (BEAST) analysis of mitochondrial (COI, 16S) and nuclear (PEPCK, opsin) genes from two billion generations with a 1% burn-in based on the *Mendacibombus* root-date estimate of 34 Ma (Fig. 4). Nodes are numbered as in Table A3.

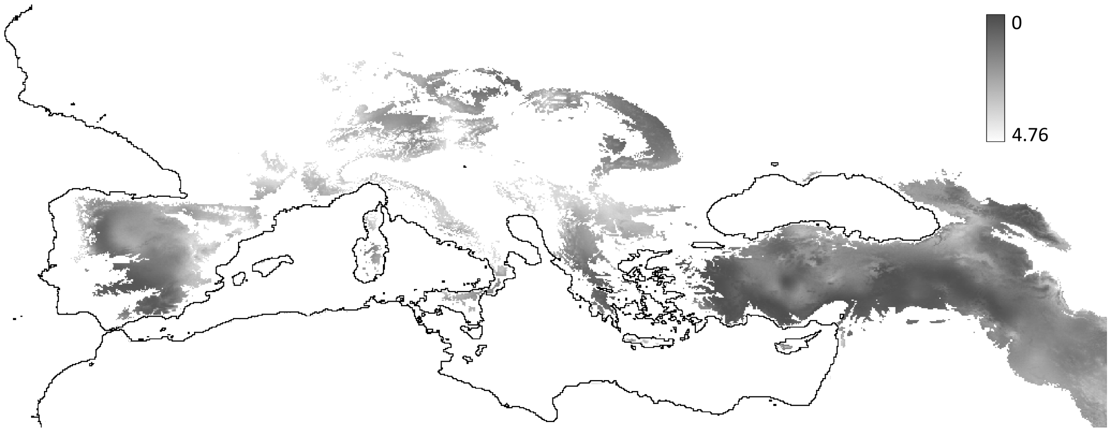

Figure A3. Reconstruction of climate suitability in the western Palaearctic for the Last Glacial Maximum (22 ka) based on the climate conditions of current observations for *Mendacibombus* species and following the same methodological protocol as in Fig. 7. Data from the GCM simulation available at WorldClim (www.worldclim.org).

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