The tale of the black viper: distribution and bioclimatic niche modelling of melanistic *Vipera aspis* in Italy

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SUPPLEMENTARY MATERIAL

Fig. S1. Examples of pattern variability in non-melanistic individuals among the three *Vipera aspis* subspecies from Italy (males on the left and females on the right. **A.** *Vipera aspis aspis*, male and female from Piedmont; **B.** *Vipera aspis francisciredi*, male from Lombardy and female from Abruzzo; **C.** *Vipera aspis hugyi*, male from Sicily and female from Calabria. Photo credit: Matteo R. Di Nicola.

Fig. S2. Examples of “dark” *Vipera aspis* individuals deemed ineligible for the dataset. **A.** *V. aspis* cf. *francisciredi* from Valle Spluga, Lombardy, with dark grey ground colour and clearly distinguishable dark pattern; **B.** *V. aspis hugyi* from Palermo Province, Sicily, with dark grey ground colour and clearly distinguishable dark pattern; **C.** *V. aspis aspis* from Aosta Valley with extended dark pattern on a light ground colour that is still visible. Photo credit: Valter Rossotti (A); Giandomenico La Barbera (B); Matteo R. Di Nicola (C).

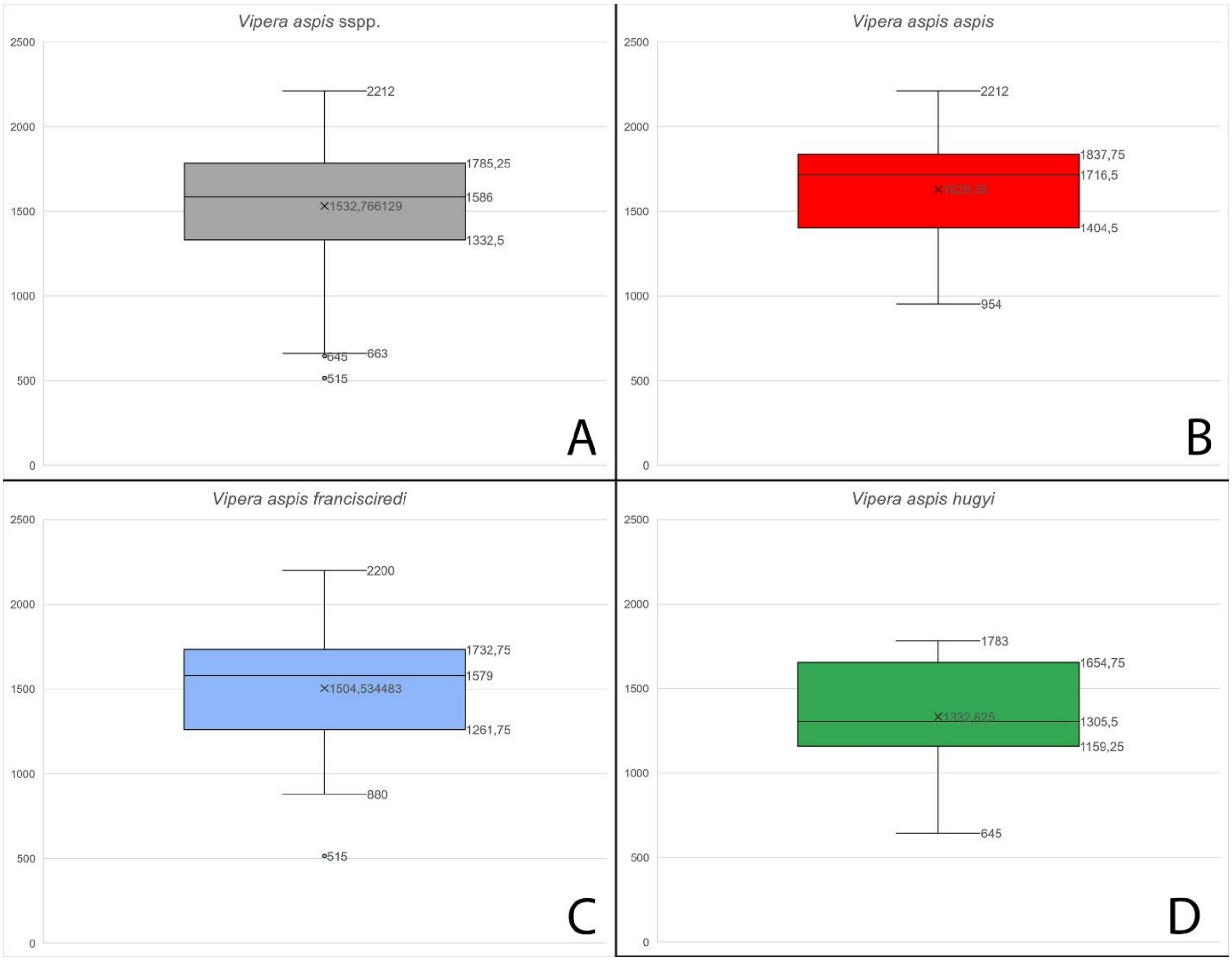


Fig. S3. Altitudinal plot of melanistic *Vipera aspis* records. **A.** total observations; **B.** *V. a. aspis* records; **C.** *V. a. francisciredi* records; **D.** *V. a. hugyi* records.

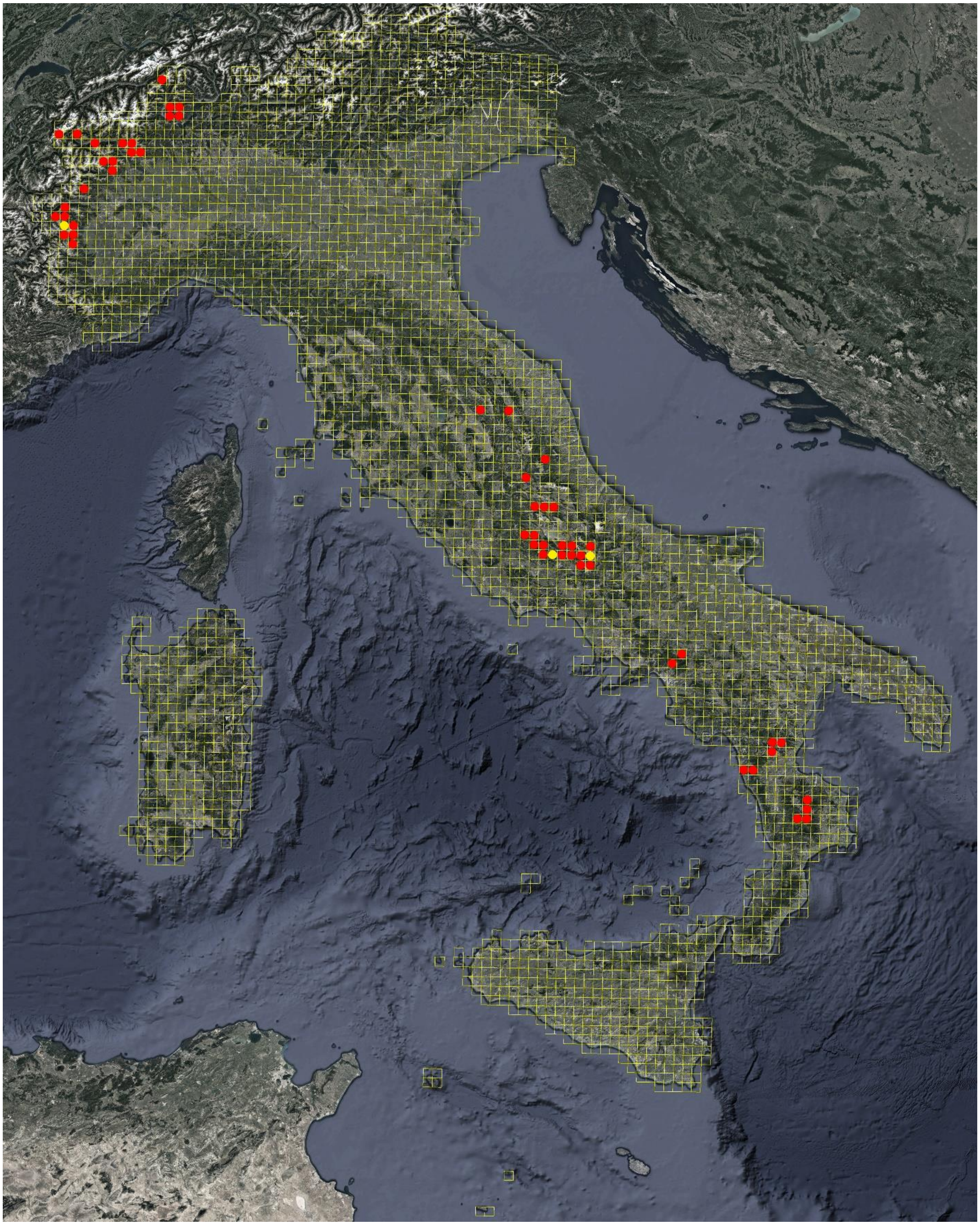


Fig. S4. 10x10 km ETRS89 / ETRS-LAEA grid with the Italian distribution of melanistic *Vipera aspis* (red circles: cells with records from 2010 onwards; yellow circles: cells with pre-2010 records). Map credits: Google Earth. Data SIO, NOAA, U.S. Navy, NGA, GEBCO. Image Landsat / Copernicus (modified).

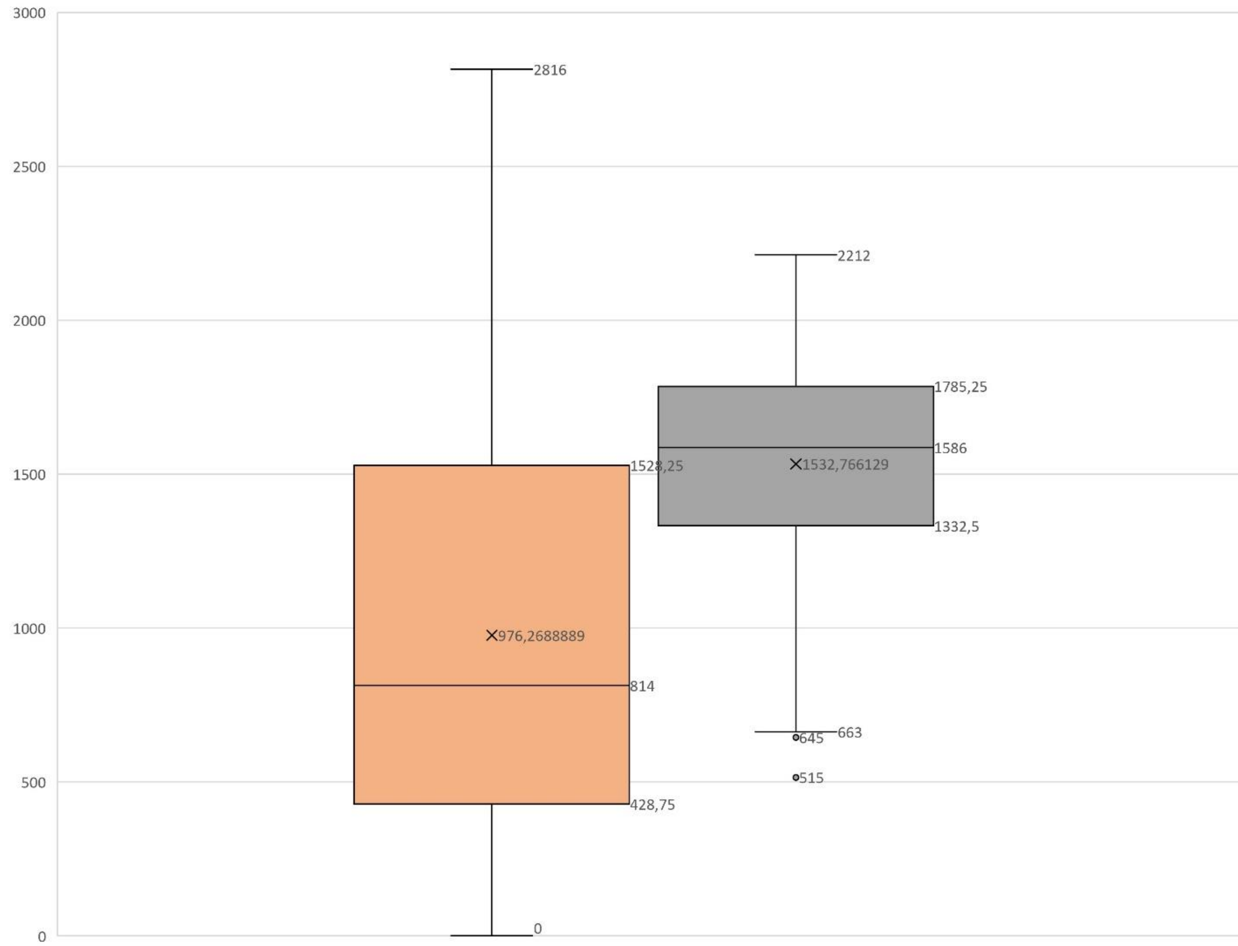


Fig. S5. Altitudinal plot of *Vipera aspis* in general (on the left) and melanistic individuals (on the right) in comparison.

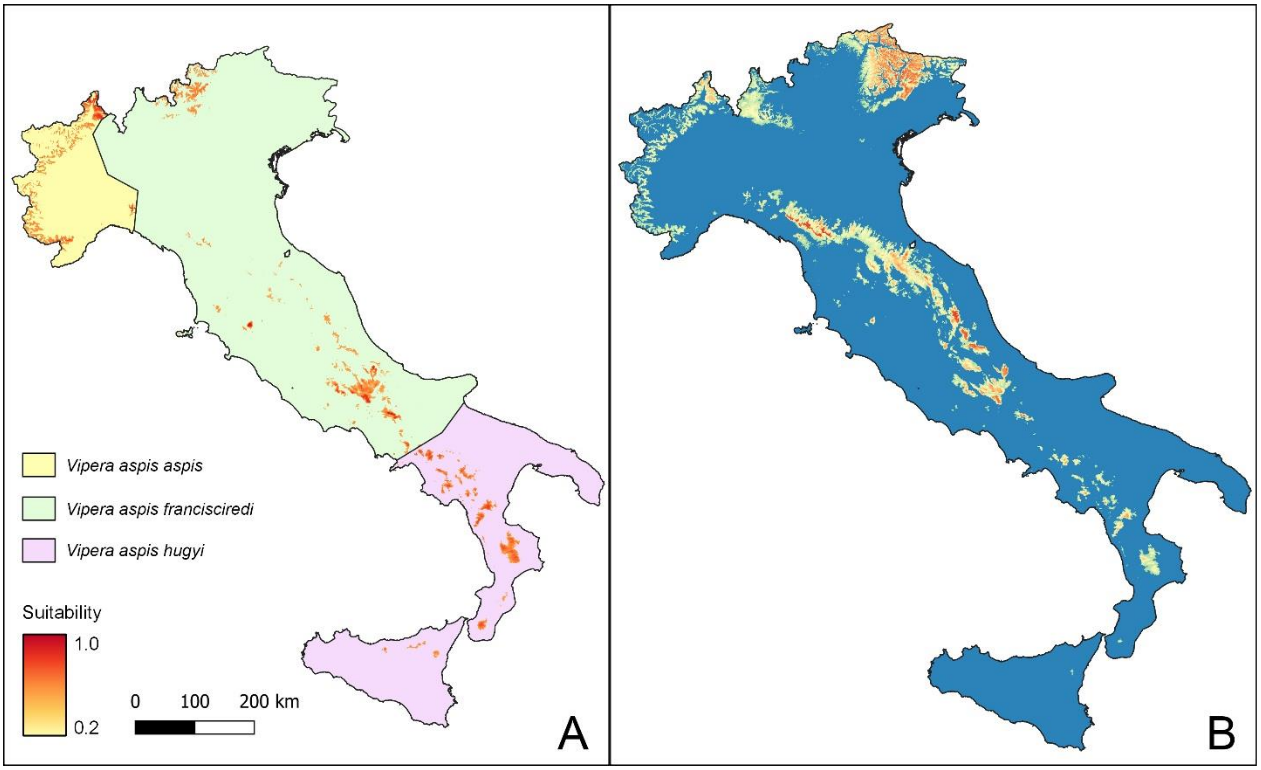
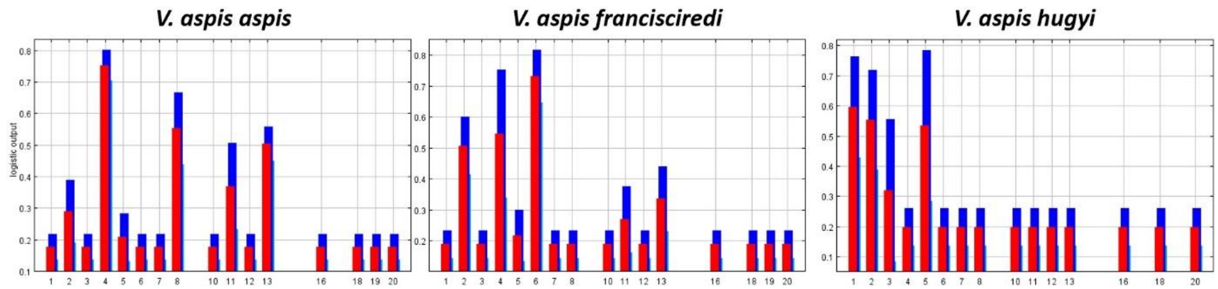


Fig. S6. Predicted bioclimatic suitability for melanistic individuals of *Vipera aspis* in Italy. **A.** Suitability models computed independently for subspecies *V. a. aspis, V. a. francisciredi* and *V. a. hugyi* using a minimum bounding polygon background. **B.** Ensemble model computed using all occurrence records together and the collection of the three minimum bounding polygons generated around each subspecies records. The outputs shown represent the average of four model combinations, each replicated 10 times (see materials and methods for details). Warmer colours indicate higher suitability, the background falls below the minimum suitability threshold (OR10).



#	Class Name	#	Class Name
1	**Broadleaf evergreen forest**	**11**	**Cropland**
2	**Broadleaf deciduous forest**	12	Paddy field
3	Needleleaf evergreen forest	**13**	**Cropland or other vegetation mosaic**
4	**Needleleaf deciduous forest**	14	Mangroves
5	**Mixed forest**	15	Wetland
6	**Tree open**	16	Bare area, consolidated (gravel, rock)
7	Shrubland	17	Bare area, unconsolidated (sand)
8	Herbaceous vegetation	18	Urban
9	Sparse trees and shrubs	19	Snow / Ice
10	Sparse vegetation	20	Water bodies

Fig. S7. Response curves showing the change in suitability (y axis) in response to variation in the predictor variable land cover (x axis) for the three subspecies of *V. aspis* modelled in the present study. Each bar represents the mean response (red) – and standard deviation (blue) – of 10 replicate MaxEnt runs. Land cover classes are explained in the table. Categories connected with high suitability (> 0.5) for one or more of the three subspecies are indicated in bold.

Subspecies	Region	Province	Coordinates (hidden)	Altitude (meters a.s.l.)	Period
aspis	Aosta Valley	Aosta	45.*** 6.***	1959	September 2019
aspis	Aosta Valley	Aosta	45.*** 7.***	2028	July 2012
aspis	Aosta Valley	Aosta	45.*** 7.***	1663	> 2010
aspis	Aosta Valley	Aosta	45.*** 7.***	1880	> 2010
aspis	Aosta Valley	Aosta	45.*** 7.***	1680	> 2010
aspis	Piedmont	Biella	45.*** 7.***	1756	July 2019
aspis	Piedmont	Biella	45.*** 7.***	1013	August 2020
aspis	Piedmont	Biella	45.*** 7.***	1215	August 2022
aspis	Piedmont	Biella	45.*** 7.***	1762	June 2017
aspis	Piedmont	Turin	44.*** 7.***	1720	July 2022
aspis	Piedmont	Turin	44.*** 7.***	1837	September 2018
aspis	Piedmont	Turin	45.*** 7.***	1797	May 2020
aspis	Piedmont	Turin	44.*** 7.***	1514	March 2008
aspis	Piedmont	Turin	44.*** 7.***	1527	March 2021
aspis	Piedmont	Turin	44.*** 7.***	1367	2005
aspis	Piedmont	Turin	45.*** 7.***	2212	> 2010
aspis	Piedmont	Turin	45.*** 7.***	1437	June 2017
aspis	Piedmont	Turin	45.*** 7.***	1811	July 2018
aspis	Piedmont	Turin	44.*** 7.***	1225	June 2021
aspis	Piedmont	Turin	44.*** 6.***	1673	August 2021
aspis	Piedmont	Turin	44.*** 7.***	1837	September 2018
aspis	Piedmont	Turin	44.*** 7.***	1837	September 2018
aspis	Piedmont	Turin	45.*** 7.***	1406	August 2021
aspis	Piedmont	Turin	45.*** 7.***	1400	July 2020
aspis	Piedmont	Turin	45.*** 7.***	1501	> 2010
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1873	July 2013
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1844	July 2011
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1824	August 2018
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1735	May 2022
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1126	July 2017
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1840	June 2015
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1840	June 2015
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1902	June 2012
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1800	June 2014
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1428	August 2020
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1436	July 2021
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1387	May 2014
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1394	June 2014
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1383	June 2015
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1379	June 2015
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1170	May 2013
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1713	September 2021
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1755	August 2015
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	954	June 20013
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1595	2004
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1890	July 2016
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1867	July 2016
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1894	June 2016
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1606	July 2015
aspis	Piedmont	Verbano-Cusio-Ossola	46.*** 8.***	1786	September 2015
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1794	January 2018
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1722	July 2019
francisciredi	Abruzzo	L'Aquila	42.*** 13.***	908	September 2016
francisciredi	Abruzzo	L'Aquila	42.***13.***	997	May 2020
francisciredi	Abruzzo	L'Aquila	42.*** 13.***	1225	June 2018
francisciredi	Abruzzo	L'Aquila	42.*** 13.***	1401	April 2019
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1357	2019
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1892	2019
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1888	May 2017

francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1084	July 2017
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1949	August 2007
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1895	August 2011
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1504	2015
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1540	July 2018
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1080	April 2015
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1575	April 2017
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1604	2018
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1072	March 1984
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1224	May 1997
francisciredi	Abruzzo	L'Aquila	41.*** 14.***	1686	September 1997
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1095	November 2016
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1735	April 2019
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1680	July 2017
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1457	May 1985
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	2200	< 2010
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1850	May 2023
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1724	> 2010
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1910	> 2010
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1037	> 2010
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1864	October 2022
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1178	March 2019
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1510	September 2017
francisciredi	Abruzzo	L'Aquila	41.*** 13.***	1602	July 2021
francisciredi	Abruzzo	L'Aquila	42.*** 13.***	1589	2010
francisciredi	Abruzzo	Teramo	42.*** 13.***	1349	> 2010
francisciredi	Lazio	Frosinone	41.*** 13.***	1681	July 2019
francisciredi	Lazio	Frosinone	41.*** 13.***	1905	April 2016
francisciredi	Lazio	Frosinone	41.*** 13.***	1208	May 2008
francisciredi	Lazio	Frosinone	41.*** 13.***	1575	August 2021
francisciredi	Lazio	Frosinone	41.*** 13.***	1583	August 2022
francisciredi	Lazio	Frosinone	41.*** 13.***	1583	August 2022
francisciredi	Lazio	Frosinone	41.***13.***	1573	August 2022
francisciredi	Lazio	Frosinone	41.*** 13.***	1583	August 2022
francisciredi	Lazio	Frosinone	41.*** 13.***	1452	May 1985
francisciredi	Lazio	Frosinone	41.*** 13.***	1594	September 1985
francisciredi	Lazio	Frosinone	41.*** 13.***	1745	July 2015
francisciredi	Lazio	Frosinone	41.*** 13.***	1732	July 2018
francisciredi	Lazio	Frosinone	41.*** 13.***	1711	September 2018
francisciredi	Lazio	Frosinone	41.*** 13.***	1511	September 2019
francisciredi	Lazio	Frosinone	41.*** 13.***	1590	September 2016
francisciredi	Lazio	Frosinone	41.*** 13.***	1327	> 2010
francisciredi	Lazio	Rieti	42.*** 13.***	1020	June 2022
francisciredi	Lazio	Rome	41.*** 13.***	1481	July 2018
francisciredi	Lazio	Rome	41.*** 13.***	1274	April 2020
francisciredi	Marche	Macerata	43.*** 12.***	880	> 2010
francisciredi	Molise	Isernia	41.*** 13.***	1786	July 2018
francisciredi	Molise	Isernia	41.*** 13.***	1777	June 2020
francisciredi	Umbria	Perugia	43.*** 12.***	515	June 2017
hugyi	Basilicata	Potenza	39.*** 16.***	1654	August 2022
hugyi	Basilicata	Potenza	39,*** 16,***	1655	June 2022
hugyi	Basilicata	Potenza	39.*** 16.***	1535	August 2022
hugyi	Basilicata	Potenza	39.*** 16.***	1783	May 2019
hugyi	Calabria	Cosenza	39.*** 16.***	1734	June 2022
hugyi	Calabria	Cosenza	39.*** 16.***	1497	September 2015
hugyi	Calabria	Cosenza	39.*** 16.***	1715	October 2015
hugyi	Calabria	Cosenza	39.*** 16.***	1138	June 2021
hugyi	Calabria	Cosenza	39.*** 16.***	1223	September 2021
hugyi	Calabria	Cosenza	39.*** 16.***	1241	August 2022
hugyi	Calabria	Cosenza	39.*** 16.***	1318	May 2012
hugyi	Calabria	Cosenza	39.*** 15.***	645	April 2017

hugyi	Calabria	Cosenza	39.*** 16.***	1293	April 2016
hugyi	Calabria	Cosenza	39.*** 15.***	663	May 2023
*hugyi**	Campania	Avellino	40.*** 15.***	1278	August 2018
*hugyi**	Campania	Salerno	40.*** 14.***	950	September 2011

Table S1. List of verified observations of melanistic *Vipera aspis* considered in the study. *Individuals coming from an area of possible intergradation between the subspecies *hugyi* and *franciscíredi*.

Sp.	Model	# Samples (Train/Test)	Test AUC ± SD	OR10	OR10 Thresh.	Bio1	Bio2	Bio12	Bio19	Srad 04	Srad 05-10	LC	Exp	Elev
V. aspis sspp.	Climatic	59/24	0.974±0.008	0.15	0.270	**75.7**	4.2	2.9	6.6	**9.5**	1.2	-	-	-
	Climatic + lc		0.977±0.006	0.12	0.266	**84.1**	3.9	2.7	1.7	**5.9**	1	0.6	-	-
	Climatic + lc + exp		0.971±0.011	0.13	0.251	**82.1**	2.2	1.6	3.6	**7.6**	1.4	1.3	0.3	-
	Elevation ⇔ Bio1		0.975±0.006	0.14	0.269	-	0.9	3.6	3.9	3.5	1.6	1.1	0.3	**85.1**
V. aspis aspis	Climatic	26/10	0.953±0.015	0.20	0.228	**63.5**	3.8	5.7	8.9	**16.8**	1.3	-	-	-
	Climatic + lc		0.966±0.012	0.14	0.251	**72.8**	1.6	2.6	4.2	**12.7**	2.6	3.4	-	-
	Climatic + lc + exp		0.962±0.016	0.14	0.205	**64.0**	0.8	4	13.3	**12.2**	0.7	4.1	0.9	-
	Elevation ⇔ Bio1		0.953±0.018	0.21	0.260	-	0.3	4.8	8	**13.7**	1	3.3	1.3	**67.6**
V. aspis francisciredi	Climatic	26/10	0.983±0.008	0.10	0.251	**58.1**	0.3	11.6	2.8	1.3	**25.8**	-	-	-
	Climatic + lc		0.984±0.008	0.05	0.143	**66.9**	0.1	4.3	2.7	1.3	**24.2**	0.5	-	-
	Climatic + lc + exp		0.981±0.010	0.09	0.187	**59.0**	0.3	6.2	0.5	0.5	**32.3**	1.1	0.2	-
	Elevation ⇔ Bio1		0.984±0.007	0.10	0.205	-	1.4	5.3	3.3	1.3	**22.9**	1.3	0.6	**63.9**
V. aspis hugyi	Climatic	8/3	0.980±0.007	0.17	0.386	**42.9**	**31.3**	0	**19.3**	4	2.6	-	-	-
	Climatic + lc		0.981±0.005	0.20	0.405	**48.3**	**17.2**	0.1	**27.1**	0	0.4	6.9	-	-
	Climatic + lc + exp		0.980±0.006	0.27	0.357	**61.1**	**20.4**	0.1	4.9	0	2.3	**7.2**	4.1	-
	Elevation ⇔ Bio1		0.976±0.007	0.10	0.328	-	**56.7**	0	**10.7**	0	0.9	3.2	3.7	**24.8**

Table S2. Statistical results of the bioclimatic suitability models computed in MaxEnt. Four model combinations were computed for each species/subspecies (see section 2.2. for details). Mean (±SD) area under curve (AUC) values higher than 0.75 indicate good model quality, with values over 0.9 indicating excellent predictive performance (Fielding & Bell, 1997). Mean 10th percentile training omission rate (OR10) close to the predicted 0.10 denotes no overfitting (Boria et al., 2014). The OR10 threshold was used to set the minimum suitability values for the relative model. The contribution of each predictor variable to each model is expressed by its permutation importance (in %). The three most influential variables for each model are indicated in bold. Predictor variables used for modelling are as follows: bio1 = mean annual temperature; bio2 = mean diurnal temperature range; bio12 = annual precipitation; bio19 = precipitation of the coldest quarter; srad04 = average soral radiation of the month of April; srad05-10 = average solar radiation of the period May – October; lc = land cover; exp = slope exposition; elev = elevation / altitude.