

Natural history and conservation of the wolf spider  
*Vesubia jugorum* (Simon, 1881) (Araneae, Lycosidae),  
assessed as Endangered in the IUCN Red List

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A female of *Vesubia jugorum* (Simon, 1881) carrying the egg sac. Valdeblore, Parc national du Mercantour. Photo credit: Nicolas Henon.

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# Natural history and conservation of the wolf spider *Vesubia jugorum* (Simon, 1881) (Araneae, Lycosidae), assessed as Endangered in the IUCN Red List

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## ABSTRACT

*Vesubia jugorum* (Simon, 1881) (Lycosidae Sundevall, 1833) is a wolf spider inhabiting high altitude rocky areas of the Southwestern Alps. Due to its restricted geographic range, its sensitivity to global warming and its continuing decline, this species has been recently assessed as Endangered (EN) by the IUCN Red List. On the basis of the research carried out in the recent years, we here provide updated information about its distribution, habitat characterisation, life history and conservation. Field observations and laboratory rearing suggest a multi-annual life cycle for this species, with a growing season of five-six months and 10-12 instars to reach the adult stage. Adult males are found for a short period, and die after mating. During winter, adult females and immatures at different stages likely survive in the upper layers of the rocky debris under the snow, where the temperature remains stable around 0-2°C. Recent studies based on species distribution modelling have demonstrated a significant relationship between habitat suitability and functional traits related to species performance, which we briefly recall here. In light of this relation, a long-term monitoring programme was designed in collaboration with Parc national du Mercantour (France) and Parco Alpi Marittime (Italy), aiming at providing the conservation status of the species and possible future trends. Here, we present the results of the baseline phase of the monitoring programme, based on 17 sites across the French-Italian border, that confirm the positive relationship between functional traits and habitat suitability, and corroborate it as a practical, non-invasive approach to the assessment of species health.

## KEY WORDS

Endemic species,  
monitoring,  
phenology,  
life cycle,  
climate change,  
habitat suitability,  
biodiversity  
conservation.



## RÉSUMÉ

*Histoire naturelle et conservation de l'araignée-loup Vesubia jugorum (Araneae, Lycosidae), une espèce "En danger" sur la Liste rouge de l'UICN.*

*Vesubia jugorum* (Simon, 1881) (Lycosidae Sundevall, 1833) est une espèce d'araignée-loup habitant les zones rocheuses de haute altitude des Alpes du Sud-Ouest. En raison de son aire de répartition géographique restreinte, de sa sensibilité au réchauffement climatique et de son déclin continu, cette espèce a récemment été classée comme « espèce en danger d'extinction (EN) » sur la Liste rouge de l'UICN. Sur la base des recherches menées ces dernières années, nous fournissons ici des informations actualisées sur sa distribution, la caractérisation de son habitat, son cycle biologique et sa conservation. Les observations sur le terrain et l'élevage en laboratoire suggèrent un cycle de vie pluriannuel pour cette espèce, avec une saison de croissance de cinq à six mois et 10 à 12 stades pour atteindre le stade adulte. Les mâles adultes sont trouvés pendant une courte période et meurent après l'accouplement. Pendant l'hiver, les femelles adultes et les immatures à différents stades survivent probablement dans les couches supérieures des débris rocheux sous la neige, où la température reste stable autour de 0-2°C. Des études récentes ont démontré une relation significative entre la qualité de l'habitat et les traits fonctionnels liés à la performance de l'espèce, que nous rappelons ici brièvement. En fonction de cette relation, un programme de monitoring sur le long terme a été conçu en collaboration avec le Parc national du Mercantour (France) et le Parco Alpi Marittime (Italie), visant à décrire l'état de conservation de l'espèce et les tendances futures possibles. Nous présentons ici les résultats de la phase de référence du monitoring, qui confirme la relation positive entre traits fonctionnels et qualité de l'habitat, et qui s'est avérée être une approche pratique et non invasive pour évaluer la santé de l'espèce.

## MOTS CLÉS

Espèce endémique, surveillance, phénologie, cycle biologique, changement climatique, qualité de l'habitat, conservation de la biodiversité.

## INTRODUCTION

*Vesubia jugorum* (Simon, 1881) is an alpine endemic species of wolf spider inhabiting high altitude rocky areas at high elevations in the Southwestern Alps (Tongiorgi 1969; Mammola *et al.* 2016). The species was originally described by Simon (1881) based on a specimen collected in unspecified locality nearby Saint-Martin-Vésubie, in the département des Alpes-Maritimes, south-eastern France. The distribution was later investigated by Tongiorgi (1968; 1969), Maurer & Thaler (1988) and more recently by Isaia *et al.* (2007; 2015), Mammola *et al.* (2016; 2019), and Milano *et al.* (2019). The current number of verified occurrences for this species published in scientific literature is 101 (detailed in Mammola *et al.* 2019), mostly encompassing the Province of Cuneo in north-western Italy (65 localities), the Département des Alpes-Maritimes (19 localities) and the Département des Alpes-de-Haute-Provence (17 localities), in south-eastern France. The known distribution range covers an area of approximately 2500 km<sup>2</sup>. Most of the species occurrences fall within protected areas and sites of the Natura 2000 network, namely the Special Area of Conservation and Special Protection Area of the Maritime Alps (SAC/SPA IT1160056 Alpi Marittime), the Natural Park of Marguareis (EUAP0214 and SAC/SPA IT1160057 Alte Valli Pesio e Tanaro) and the Special Protection Area Alte Valli Stura e Maira (SPA IT1160062) in Italy, and the Special Area of Conservation and Special Protection Area of the Mercantour National Park (SAC FR9301559 and SPA FR9310035 Le Mercantour) in France.

Recent studies based on species distribution modelling focusing on the sensitivity of the species to global warming, showed a significant reduction in its future bioclimatic range (Isaia *et al.* 2016; a forthcoming paper by Milano *et al.*), raising concerns to the long-term survival of this species. In view of this, *Vesubia jugorum* was assessed by the International Union for Conservation of Nature (IUCN), and classified as Endangered (EN) in the IUCN Red List of Threatened species, on the basis of its limited geographic range and the projected continuing decline of its natural habitat in the near future due to climate change (Isaia & Mammola 2018).

Mammola *et al.* (2019) demonstrated a significant relationship between habitat quality, predicted by species distribution models, and the individual performance of *Vesubia jugorum*, measured by means of functional traits (femur I length and egg-case size). This is a well-known and widely adopted ecological principle in the monitoring of many plants and vertebrates (Thuiller *et al.* 2004; Michel *et al.* 2017; Lunghi *et al.* 2018; Benito Garzón *et al.* 2019), but was rarely investigated among arthropods (Mammola *et al.* 2019). In *V. jugorum*, the length of femur I and egg-case (cocoon) size were found to be positively related with habitat suitability. The largest individuals (i.e., the individuals with longer femurs) and females with larger cocoons, occurred in the core of the species distribution, where the amount of predicted high-quality habitat was greatest and the related habitat suitability value was higher (> 0.7 in a range between 0 and 1). Conversely, in areas with lower habitat suitability (< 0.25), individuals had smaller femurs and smaller cocoons. On these bases, measuring variation in

morphological traits of *V. jugorum* has been suggested as a practical, non-invasive means of assessing population health through time (Mammola *et al.* 2019).

In the context of the species conservation, a long-term monitoring programme has been designed for evaluating the ongoing impact of climate change on the species survival and for detecting changes in populations, aiming at setting conservation actions and at informing stakeholders about the future management of the species. According to the IUCN, monitoring is one of the main sources of information on the population status, and a significant tool in the conservation strategy of the species. In 2019, a transnational monitoring programme involving Italy and France, coordinated by the University of Turin in collaboration with Parc national du Mercantour and Parco Naturale Alpi Marittime, has started.

Thanks to the work conducted during these years by our research team, we gathered new data on the ecology, the distribution and the life history of this species, that we sum up in this work along with the description of the monitoring programme and the presentation of the results obtained during the baseline phase of 2019.

## MATERIAL AND METHODS

### SAMPLINGS AND LABORATORY REARING

Field observations and collection of living specimens were conducted across the known species distribution range from mid-June to late September, in the years from 2016 to 2019. Additional samplings, aiming at extending the known distribution range of the species, have been performed in different localities of France and Italy in 2019 (see Acknowledgements), and during the “Explor’Nature Colmars-les-Alpes 2021” and “Explor’Nature Valdeblorre 2022” events organised by Parc national du Mercantour (1-4 July 2021 and 30 June-3 July 2022, respectively), within the territory of the municipalities of Colmars-les-Alpes (département des Alpes-de-Haute-Provence) and Valdeblorre (département des Alpes-Maritimes). Both areas were predicted as suitable by species distribution models (see Mammola *et al.* 2019), but never investigated before. The specimens are preserved in EtOH95% and the material is stored in the Marco Isaia collection (coll. MI) at the Department of Life Sciences and Systems Biology of the University of Torino and in the collection of the Muséum national d’Histoire naturelle, Paris (MNHN).

During our surveys in summer 2016 and summer 2018, 50 spiders were collected alive in the field for laboratory rearing. Spiders were collected by hand and placed in individual Falcon® Tubes of 50 mL. In laboratory, we housed the collected specimens individually in plastic terraria (18 cm × 12 cm × 7.5 cm), supplied with wet sponge or cotton wool as a source of water. The specimens were kept at room temperature during the day (19–26°C), and in an IPP 30 Peltier Memmert climatic chamber at night where the temperature was kept stable for approximately 12 hours at 4–6°C. The spiders were fed *ad libitum* with insects collected during the

TABLE 1. — Overview of the sites designed by the monitoring programme, with relative country, elevations (**Elev.**) in meters and coordinates in decimal degrees (Datum: WGS84).

Site	Country	Locality	Elev.	Latitude	Longitude
1	France	Col de la Bonette	2564	44.347	6.797
2	France	Col de la Cayolle	2420	44.265	6.736
3	France	Col de Mallemort	2570	44.474	6.854
4	France	Col de Vars	2289	44.534	6.692
5	France	Col du Trem	2472	44.050	7.430
6	France	Grande Séolane	2536	44.333	6.551
7	France	Lac de l’Agnel	2356	44.120	7.460
8	France	Rocca dell’Abisso	2611	44.140	7.510
9	France	Serrière de la Lombarde	2359	44.200	7.161
10	Italy	Colle del Chiapous	2540	44.181	7.319
11	Italy	Colle dell’Arcana	2260	44.461	6.941
12	Italy	Colle della Ciriegia	2534	44.140	7.280
13	Italy	Corborant	2764	44.265	7.000
14	Italy	Marguareis – Canale dei Genovesi	2000	44.180	7.687
15	Italy	Mongioie – Bocchin dell’Aseo	2298	44.175	7.793
16	Italy	Passo della Gardetta	2599	44.404	6.996
17	Italy	Passo Sant’Anna	2390	44.220	7.090

sampling and with laboratory-reared house crickets, *Acheta domesticus* (Linnaeus, 1758). Spiderlings that emerged from egg sacs, were reared in the laboratory, and fed with fruit flies, *Drosophila melanogaster* Meigen, 1830. The numbering of ontogenetic stadia was counted after the emergence from egg (not considering the first molt inside egg sac, see Dolejš *et al.* 2014).

Natural microclimatic conditions experienced by *Vesubia jugorum* throughout the year, were derived from data-loggers positioned for one year at the ground level under stones, in a suitable area at 2589 m in the nearby of the meteorological station of Rocca dell’Abisso (2753 m a.s.l., Valdieri, Province of Cuneo). Meteorological data from the station of Rocca dell’Abisso were provided by Arpa Piemonte ([https://www.arpa.piemonte.it/rischinaturali/accesso-ai-dati/annali\\_meteorologici/annali-meteo-idro/banca-dati-meteorologica.html](https://www.arpa.piemonte.it/rischinaturali/accesso-ai-dati/annali_meteorologici/annali-meteo-idro/banca-dati-meteorologica.html)).

### THE MONITORING PROGRAMME

The monitoring programme was designed on the basis of the currently known distribution range of *Vesubia jugorum*, and on the basis of the significant relationship between habitat quality and the performance of individuals recovered in Mammola *et al.* (2019). Accordingly, the programme focussed on morphological and reproductive traits that might constitute reliable proxies for the health of the populations of *V. jugorum*: the size of the femur of the fourth leg (femur IV), as a representative measure of the overall body size, and the size of the cocoons, as trait related to the reproductive success. The monitoring programme was designed to be regularly repeated at five-year intervals, considering the life cycle of the species.

The surveys of the baseline phase were carried out from 11 July to 20 September 2019 (72 days), during the peak of the growing season of the species, in 17 sites selected by

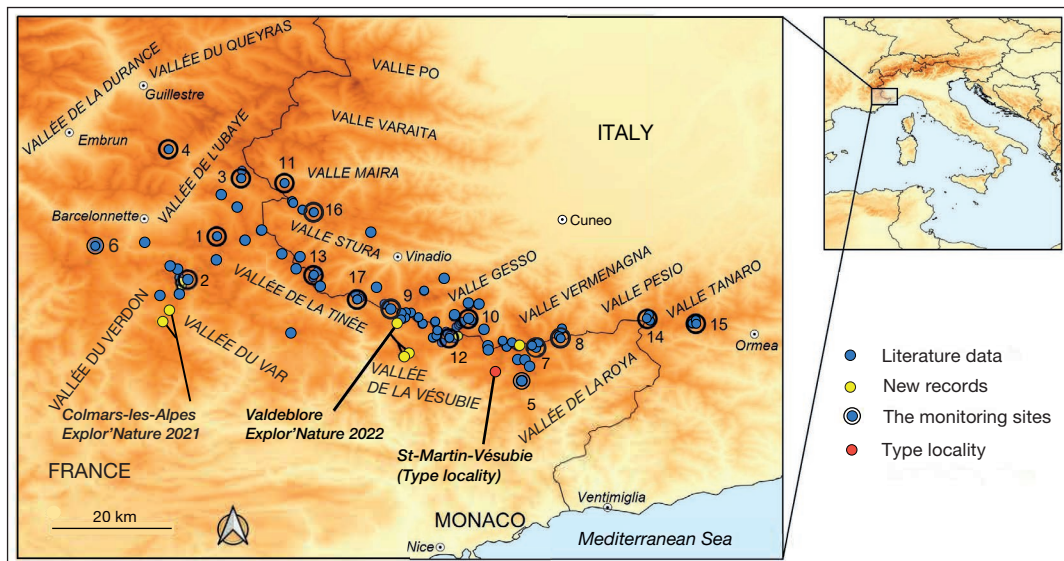


FIG. 1. — The updated distribution of *Vesubia jugorum* (Simon, 1881). Symbols: ●, type locality; ●, literature data; ●, new records published in this work; ●, the 17 sites chosen for the long-term monitoring programme.

TABLE 2. — New occurrences of *Vesubia jugorum* (Simon, 1881) collected in 2019, 2021 and 2022. For each locality, coordinates are in decimal degrees (Datum: WGS84), municipality, elevation in meters, numbers of specimens collected (n) and date of discovery are specified.

Country	Locality	Municipality	n	Date	Elevation (m)	Lat.	Long.
Italy	Lago Bianco dell’Agnello	Entracque	1	8.VIII.2019	2200	44.124	7.424
France	Col du Mercantour	Saint-Martin-Vésubie	1	18.VIII.2019	2532	44.145	7.299
France	Lac des Garrets	Entraunes	1	22.VIII.2019	2299	44.248	6.728
France	Col de la Petite Cayolle	Uvernet-Fours	1	5.IX.2019	2460	44.256	6.726
France	Tête de l’Encombrette	Colmars-les-Alpes	2	3.VII.2021	2200	44.198	6.701
France	Dent de Lièvre	Colmars-les-Alpes	5	3.VII.2021	2138	44.174	6.686
France	Vallon des Millefontes	Valdeblore	1	30.VI.2022	2150	44.103	7.185
France	Mont Pepoiri	Valdeblore	5	1.VII.2022	2360	44.110	7.196
France	Tête de la Roubine	Valdeblore	1	3.VII.2022	2274	44.171	7.173

our own expert opinion as representatives of the overall distribution range of the species (Table 1 and Figs 1; 2). For each sampling site, we set the collection of a minimum of five adult females with cocoons over a limit of searching time of three hours.

After the sampling, the leg IV of each specimen collected was removed and stored in EtOH95%. As demonstrated in literature (Wrinn & Uetz 2008), the removal of a leg IV in a spider does not represent harm to the individual. The measurement of the femur IV was carried out in laboratory, through Leica M80 stereoscopic microscope (up to 60 × magnification). To standardize data acquisition, we derived measurements from digital pictures taken with a Leica EC3 digital camera, and we calculated them with Leica LAS EZ 3.0 software (Leica Microsystems, Switzerland). After the removal of the leg IV in the field, the specimens were released in their natural habitat. Whenever present, the cocoon diameter was measured with a digital calliper (Fig. 2). After the measurement, the cocoon was returned to the individuals.

#### REGRESSION ANALYSES

We conducted all analyses using R software (R Core Team 2021). We tested the relationship between the length of femur IV collected in the monitoring and the habitat suitability index of the sampling sites as in Mammola *et al.* (2019) by means of linear mixed models (LMMs) with the ‘lmer’ R package (Bates *et al.* 2015). Mixed models allowed us to address the violation of models assumption of spatial independence, caused by multiple measurements of the same population.

In R notation, the structure of the linear mixed models was:

$$Y \sim H + 1|S$$

where S are the sampling sites (excluding Col de Vars, where no specimens were collected) used as random factors, Y represents one of morphological variables (i.e. femur IV length and cocoon size), and H represents values of habitat suitability, expressed in a range between 0 and 1. Values of habitat suitability were calculated by averaging the values of each pixel of the species distribution model from Mammola *et al.* (2019) with a 500-m buffer around each sampling site, using QGIS software (version 3.14) (QGIS Development Team 2022).





FIG. 2. — Long-term monitoring programme of *Vesubia jugorum* (Simon, 1881): **A**, a typical high-altitude rocky area colonized by *Vesubia jugorum*; **B**, a female with its cocoon; **C**, measurement of the cocoon diameter with a digital calliper; **D**, removal of leg IV from a female; **E**, measurement of leg IV through Leica M80 stereoscopic microscope; **F**, a female with cocoon found in its retreat.

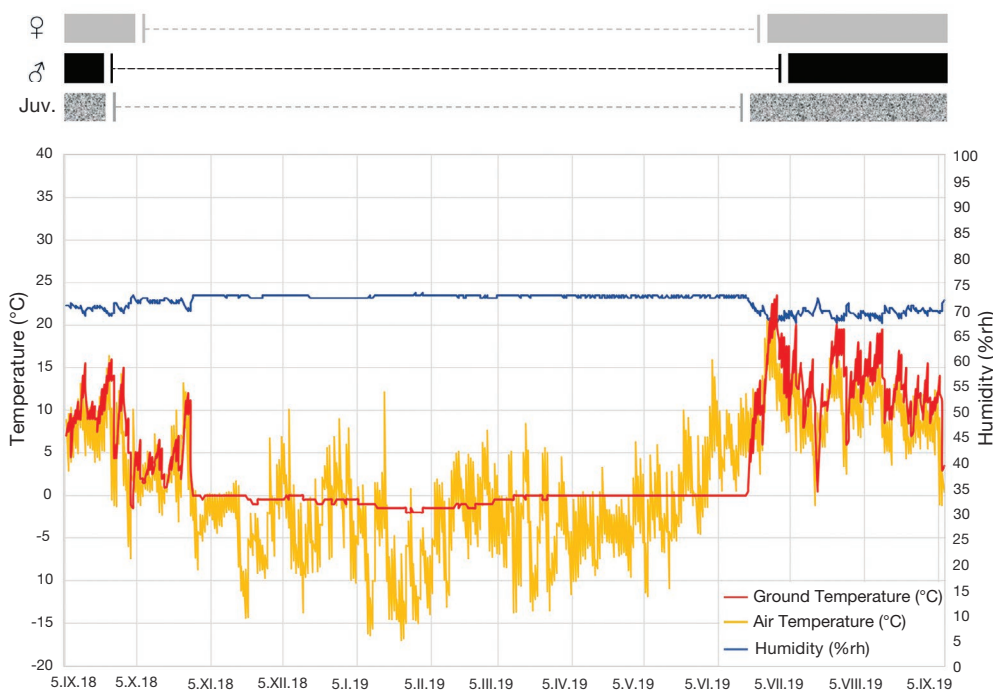


FIG. 3. — Life cycle of *Vesubia jugorum* (Simon, 1881) derived from field observations conducted across the species distribution range during the 2016–2019 sampling seasons. Daily fluctuations in temperature (red line) and humidity (blue line) derived from data-logger positioned across 2018 and 2019 at the ground level under stones, in the nearby of Rocca dell’Abisso (Valdieri, 2589 m a.s.l.). Daily fluctuations in air temperature (orange line) derived from the meteorological station of Rocca dell’Abisso (2753 m a.s.l.). Growing season is indicated by light grey (females), black (males) and dotted dark grey (juveniles) bars. Dash line refers to the overwintering. See text for further details.

RESULTS

Order ARANEAE Clerck, 1757  
 Family LYCOSIDAE Sundevall, 1833

*Vesubia jugorum* (Simon, 1881)

*Trabea jugorum* Simon, 1881: 83.

*Vesubia jugorum* – Simon 1909: 402.

MATERIAL. — **France** • 2 ♀; département des Alpes-Maritimes, Valdeblorre, Mont Pepoiri, western slope, in rocky debris; 2360 m a.s.l.; 1.VII.2022; Isaia and Tolve leg., coll. MI • 1 ♀; same data as for preceding; MNHN • 2 ♀; Colmars-les-Alpes, Tête de l’Encombrette, southern slope; steep alpine scree; 2200 m a.s.l.; 3.VII.2021; Isaia and Tolve leg., coll. MI • 5 ♀; Colmars-les-Alpes, Dent de Lièvre, northern slope; flat rocky debris area; 2138 m a.s.l.; 3.VII.2021; Isaia and Tolve leg., coll. MI.

OBSERVATIONS. — **Italy** • 1 specimen; Province of Cuneo, Entracque, Lago Bianco dell’Agnello; 2200 m a.s.l.; 8.VIII.2019; Giordana vid. **France** • 1 ♀; département des Alpes-Maritimes, Entraunes, Lac des Garrets; 2299 m a.s.l.; 22.VIII.2019; Lucas vid. • 1 ♀; Saint-Martin-Vésubie, Col du Mercantour; 2532 m a.s.l.; 18.VIII.2019; Assmann vid. • 1 ♀; Valdeblorre, Vallon des Millefontes, western slope, in rocky debris; 2150 m a.s.l.; 30.VI.2022; Breton vid. • 2 ♀; Mont Pepoiri, western slope, in rocky debris; 2360 m a.s.l.; 1.VII.2022; Isaia and Tolve vid. • 1 ♀; Valdeblorre Tête de la Roubine, southern slope, in rocky debris; 2274 m a.s.l.; 3.VII.2022; Isaia and Tolve vid. • 1 ♀; département des Alpes-de-Haute-Provence, Uvernet-Fours, Col de la Petite Cayolle; 2460 m a.s.l.; 5.IX.2019; Lucas vid.

REMARK

These new findings extend the current known distribution range of the species from the previous 2456 km<sup>2</sup> to the current 2628 km<sup>2</sup>. To date, the known occurrences of *Vesubia jugorum* are 110, i.e., 66 in Italy (Province of Cuneo) and 44 in France (24 in département des Alpes-Maritimes and 20 in département des Alpes-de-Haute-Provence). The new updated distribution of *V. jugorum* is presented in Fig. 1, and the new records are detailed in Table 2.

MICROCLIMATIC DATA

Temperature data derived from data-loggers positioned under stones showed daily fluctuations in temperature and relative humidity during the warm season until late October, when the temperature quickly drops to 0°C and remains almost constant until the following mid-June. Data from the meteorological station of Rocca dell’Abisso showed strong fluctuations of the air temperatures during the year, ranging from –17° in winter to 22°C in summer (Fig. 3).

OBSERVATIONAL DATA ON LIFE CYCLE, PHENOLOGY AND DIET

Observational data likely confirm for *Vesubia jugorum* a stenochronous life cycle. Females and juveniles were generally found throughout the summer season, from mid-June to late September (only one record from Mongioie at the beginning of October), whereas adult males were found for a shorter period, from July to mid-September (Fig. 3). No observations



were ever carried out in winter due to the high snow coverage in suitable areas. During the summer season, different cohorts were found simultaneously. The highest abundance of adult specimens occurred in July and August, likely corresponding with the mating period. Males were encountered less frequently and their density was generally low, an observation that may be possibly biased by the higher mobility of males and the greatest difficulty in catching them.

Females with cocoons have been found – always in their retreats – from the end of June to the early days of September. Females build circular and silk-lined retreat under stones, with a small opening in the silken walls, occasionally digging an additional small recess into the soil. Females with cocoons did not seem to be territorial, as up to three females have been found in adjoining retreats under the same stone. The cocoons are globular, white and contain on average 200 eggs (89–343,  $n = 10$ ). According to our observations, females produce more egg sacs in the same season. The production of a second egg sac has been observed in laboratory-reared specimens.

Females of *Vesubia jugorum* exhibit maternal cares of both cocoons and spiderlings. They carry their cocoon underneath their abdomens attached to the spinnerets. If they lose their egg sac, or if the egg sac is removed, they look for it in the surrounded area until they found it or a surrogate (e.g. a rounded piece of cotton). Under laboratory conditions, females fed while carrying egg sacs or pulli. Female looks after the cocoon for around one month after the laying, until the offspring hatch. The first moult occurs inside the egg sac while the second-instar juveniles emerge from the cocoon through a cleft in the seam, and climb onto their mother opisthosoma, and, occasionally, carapace. Maternal care for spiderlings lasts one week to ten days, after which the spiderlings disperse by falling off from the female body. Shortly after, they moult to the third instar. On average, the duration of the instars is 40 days, with earlier stages moulting every two weeks and later stages being longer up to two months. Given the remarkable size reached by adult specimens, and considering the duration of the growing season, it seems likely that the specimens require an average of 10–12 instars to complete the development, reaching the adult stage over multiple years.

As proved by the specimen collected in earliest time of the growing season, overwintering individuals are generally adult females or immatures at different stages in their development. Unfortunately, we have no data on overwintering behaviour. However, we observed a tendency of the individuals to aggregate during the growing season in areas of high suitability (see, e.g., the high population density observed in July 2019 at Colle della Ciriégia, a highly suitable site where many specimens have been found in a few minutes), and to disaggregate when the temperature drops (no specimens found in samplings carried out in the same locality in mid-October). It seems likely that specimens are very mobile within the rocky areas, showing a higher tendency to aggregate in the snow-free period and to disaggregate as the cold season approaches.

*Vesubia jugorum* is a cursorial hunter which preys actively. The spider approaches the prey and pounces on it from a close distance, grabbing and surrounding it using the strong, spiny

TABLE 3. — Measurements of the functional traits for each monitoring site, with relative values of habitat suitability. For each locality, the mean length of femur IV (in mm), mean cocoon size (in mm), and sample size ( $n$ ) measured for each site was reported. Habitat suitability is derived from Mammola *et al.* (2019), setting an average value calculated on a 500-m circle buffer around each site.

Locality	Femur length mean (n)	Cocoon size mean (n)	Habitat suitability
Colle del Chiapous	7.206 (5)	NA	0.784
Corborant	6.794 (5)	NA	0.784
Serrière de la Lombarde	7.288 (5)	10.828 (4)	0.737
Colle della Ciriégia	7.074 (5)	11.262 (5)	0.716
Passo della Gardetta	6.218 (5)	8.407 (4)	0.638
Col de la Bonette	6.494 (5)	NA	0.470
Passo Sant'Anna	6.938 (5)	9.535 (5)	0.452
Marguarais – Canale dei Genovesi	6.165 (2)	NA	0.436
Colle dell'Arcana	6.526 (5)	9.776 (4)	0.388
Col de la Cayolle	6.202 (5)	NA	0.341
Col du Trem	6.996 (5)	11.468 (2)	0.315
Rocca dell'Abisso	6.820 (2)	NA	0.300
Lac de l'Agnel	7.053 (4)	NA	0.294
Grande Séolane	7.050 (3)	NA	0.211
Mongioie – Bocchin dell'Aseo	5.778 (5)	9.093(4)	0.170
Col de Mallemort	5.882 (5)	NA	0.109

legs. After grabbing it, the prey is bitten with the chelicerae and released with the legs. *Vesubia jugorum* was observed masticating the prey organisms with the chelicerae, maximizing food assimilation and thus increasing the total energy extracted from each prey item. Laboratory observations showed a generalist predatory habit for *V. jugorum*. Accepted preys included Orthoptera, Diptera, Lepidoptera, Coleoptera, Isopoda Oniscidea and Araneae. Cannibalism was common, often involving juveniles or smaller individuals, and in general occurring between pairs of specimens with great differences in both mass and size.

*Vesubia jugorum* shares its habitat with other Alpine spider species such as *Alopecosa alpicola* (Simon, 1876), *Drassodes thalери* Hervé, 2009, *Drassodes simoni* Hervé, Roberts & Murphy, 2009, *Attulus longipes* (Canestrini, 1873), *Xysticus desidiosus* Simon, 1875 and *Pardosa nigra* (C. L. Koch, 1834) (see Isaia *et al.* 2015). Tentatively, areas where *V. jugorum* is locally abundant are generally avoided by *Pardosa nigra*, which is slightly smaller in comparison, but possibly shares similar ecological requirements.

#### RESULTS OF THE MONITORING BASELINE PHASE

During the monitoring surveys, 71 females and 29 cocoons were collected. In 12 out of 17 sampling sites, the target of five adult females was achieved, whereas in the remaining five sites the sampling stopped after three searching hours, without reaching the target of five adult females collected. Col de Vars was the only site where no individual was captured. According to the models performed in Mammola *et al.* (2019), this site has a very low value of habitat suitability. For this reason, Col de Vars was excluded from the monitoring programme.

The results of the measurements of the functional traits collected in each locality with their relative values of habitat suitability (extracted from Mammola *et al.* 2019) are reported

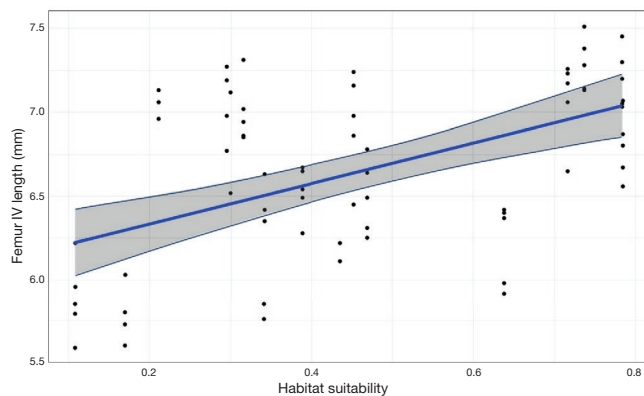


Fig. 4. — Predicted relationship (blue line) and 95% confidence intervals (gray stripe) between length of femur IV and habitat suitability of each monitoring site derived from the Ecological Niche Model presented in Mammola *et al.* (2019) (see Material and Methods for further details).

in Table 3. The femur IV length ranged from 5.590 mm (Col de Mallemort) to 7.510 mm (Serrière de la Lombarde). In the locality with smallest femurs, Mongioie – Bocchin dell’Aseo where the mean femur IV length was 5.778 mm ( $n = 5$ ), the habitat suitability index was 0.170. The site with the longest femurs IV was Serrière de la Lombarde, with a mean length of 7.288 mm ( $n = 5$ ). This site had a high habitat suitability index, corresponding to 0.737.

Only in seven sampling sites we were able to collect cocoons, and their finding was generally rare. The cocoon size ranged from 6.915 mm (Passo della Gardetta) to 12.545 mm (Serrière de la Lombarde). The site with the largest average cocoon size was Col du Trem (11.467 mm;  $n = 2$ ), whereas the locality with the smaller average cocoon size was Passo della Gardetta (8.407 mm;  $n = 4$ ).

Results of the regression analyses, highlighted relationship very close to statistical significance ( $P$ -value = 0.0516) between the length of the femurs IV and the habitat suitability predicted by the model (Fig. 4). No significant relationship was observed between habitat suitability and cocoon size, possibly due to the low sample size.

Specimens collected at Grande Séolane showed an unexpected great length of the femur IV. Accordingly, when excluding this observation, the significance of the regression improves, reaching a  $P$ -value of 0.0202.

## DISCUSSION

In this work, we provide an updated frame of the knowledge on the biology of *Vesubia jugorum*, with original information on its distribution, habitat characterisation, life history and conservation.

*Vesubia jugorum* occurs almost exclusively in rocky areas at high elevations, such as rocky debris, boulder fields and scree, mostly from 2000 m upwards (Tongiorgi 1969; Mammola *et al.* 2016, 2019). More specifically, when considering the current dataset of occurrences, the known altitudinal

range spans from 1800 (Pian della Casa, Rifugio Regina Elena, Valdieri) to 3010 m a.s.l. (top of Mount Corborant, Vinadio). Specimens are generally observed wandering on the rocks or sheltering under stones (Mammola *et al.* 2016) and occasionally in prairies at the edge of alpine screes. According to Tongiorgi (1969), individuals are most active at night, while they shelter under stones during the day. Our observations generally confirm these data; moreover, we also observed individuals basking in full sun at high elevations.

Considering the harsh condition of the habitat, the growing season seems likely limited to 5-6 months per year, corresponding to the snow-free periods of maximum intake of solar energy, which is fundamental for increasing metabolic rate in poikilothermic invertebrates. It seems plausible that the spider overwinters under the snow for 6-7 months (Fig. 3), when conditions at the interface snow-soil remain stable around 0°C and the metabolic rate of the spider possibly decreases (Danks 2006; Knapp & Řeřicha 2020).

In light of the number of instars required to complete the development and to reach adulthood, as seen in large entelegyne spiders (Eason & Whitcomb 1965; Eason 1969; Dolejš *et al.* 2014), and the duration of the growing season (approximately three instars per growing season), it is expected that maturity is reached at least after four seasons. Multi-annual life cycles are common in spiders having a long overwintering period (Pickavance 2001; Hammel 2005; Høye & Forchhammer 2008). It seems likely that, similarly to other Lycosids, males reach adulthood at the 10th instar, and females at 11th or 12th. After mating, males die (generally at the end of summer). Females overwinter once more, and die the following year, presumably at the age of five years. The presence of females with cocoons at the beginning of the growing season, before the appearance of males, suggests that females store sperm in their receptacula during overwintering, and then lay eggs when the temperature increases, as seen on other Lycosids (Eason & Whitcomb 1965; Dolejš *et al.* 2010). The cocoon construction is similar to that reported for other Lycosids (Eason 1964; Engelhardt 1964; Eason & Whitcomb 1965). The ability to produce more than one egg sac seen in laboratory-reared females is widespread in wolf spiders, not only in the temperate zone but also at higher altitudes and latitudes (see for example Buddle 2000; Pickavance 2001; Høye *et al.* 2020; Viel *et al.* 2022). As seen in Mammola *et al.* (2019), average clutch size is positively correlated with the average female size.

The generalist predatory habit of *Vesubia jugorum* is coherent with the general opportunistic habit found in most high alpine spiders, dwelling in habitat with limited resources. The hunting behaviour, with the adoption of the “full leg basket” technique, is common in lycosoid spiders, which hunt without web (see Eggs *et al.* 2015). Feeding behaviour is similar to that of other wolf spiders (Nyffeler & Benz 1988; Nyffeler & Breene 1990): *V. jugorum* was observed masticating the prey organisms with the chelicerae, maximizing food assimilation and thus increasing the total energy extracted from each prey item. The cannibalism as well has been observed in other wolf spiders (Samu *et al.* 1999; Anthony 2003; Buddle *et al.* 2003).

Under laboratory conditions, adult females fed while carrying egg sacs or pulli, differently from other burrowing species of lycosids (e.g. in the genus *Trochosa* C. L. Koch, 1847; see Engelhardt 1964), generally fasting after laying eggs. Together with the presence of a small opening in the silken walls of the retreat (Tongiorgi 1969), this may suggest that females leave their retreats during the egg sac-guarding and pulli-carrying period.

As the result of the samplings carried out during the “Explor’Nature Colmars-les-Alpes 2021” and “Explor’Nature Valdeblore 2022”, five new localities have been discovered. The new occurrences in Colmars-les-Alpes extended the known limits of the species geographic distribution towards south-west, in an area predicted as suitable by the species distribution models but never investigated before. Occurrences in Valdeblore were highly expected, being located close to the center of the distribution range, in a highly suitable area, according to the models.

All the new findings here reported validate the distribution limits in projecting the potential current geographic distribution of *Vesubia jugorum*, confirming the importance of these tools to overcome gaps in spatial data in threatened species.

Ecological Niche Modelling presented in Mammola *et al.* (2019) points out a specific relationship between some climatic feature and the probability of presence of *Vesubia jugorum*. In particular, timing, duration and thickness of seasonal snow coverage seems to play a major role in determining the distribution of the species by possibly influencing the duration of its growing season. Accordingly, observational data seems to support that the development of *V. jugorum* occurs during a relatively short snow-free period in summer and early autumn (Mammola *et al.* 2016). During the rest of the year, the individuals most likely survive under stones in the upper layers of the rocky debris, which are insulated by a deep blanket of snow (see, e.g. Zhang 2005). According to the interpretation of the statistical models, habitat quality for *V. jugorum* declines in areas where the mean annual number of days of snow cover during the year is < 40, which is typical for lower elevations. In addition, habitat quality declines where the mean annual number of days of snow coverage is > 100, a condition occurring either above 2800-3000 m within the core of the species distribution or at northern latitudes within the species range.

Climatic evidence inferred from statistical models finds support in our data derived from temperature data-loggers positioned under stones in suitable areas for one year. The fast and high accumulation of snow at the beginning of the cold season, affects the ground climatic regime by insulating it from cold temperatures and variations occurring above ground, generating ideal conditions for the survival of ground arthropods at the snow-ground interface during winter. More specifically, the insulation effect of thick snow cover, has a significant influence on the ground thermal regime, and increase with increasing snow depth due to the thermal resistance of the snowpack (Zhang 2005; Luetschg *et al.* 2008). If, on one hand, during the cold season from November to June, the stable climatic conditions at the snow-ground interface allow

protection from the extreme winter temperatures and their variations, on the other hand, the early snowmelt occurring in the area and the consequent rapid temperature increase of the ground temperature lengthen the duration of the snow-free period, thus allowing a longer growing season for the species (Mammola *et al.* 2019).

Best available knowledge and updated information on the ecology and the distribution of this species are fundamental to provide an updated insight on the conservation status of this species. In the conservation context of the endangered species, monitoring is a fundamental tool to detect trends in populations and distribution, and to measure the impacts of threatening processes (Legge *et al.* 2018).

The results of the monitoring baseline phase confirmed the relationship between climatic suitability and femur size suggested by Mammola *et al.* (2019). Specimens with largest femur IV occurred in localities where the predicted habitat quality was highest (Fig. 4), confirming the use of this trait for monitoring purposes. Only at Grande Séolane specimens showed an unexpected great length of femur IV despite the low value of the predicted habitat quality. Interestingly, ongoing molecular analysis have highlighted issues and raised questions concerning the population of Grande Séolane, revealing a lack of shared haplotypes with the neighbouring populations and a genetic pattern which is hardly explainable in the framework of the current and historical population dynamics proposed for *V. jugorum* (a forthcoming paper by Milano *et al.*).

The use of the femur IV in our work rather than the femur I (as done in Mammola *et al.* 2019) does not seem to bias the significance of the relation. In predatory arthropods, specimens with greater body size have greater predatory efficiency and a higher benefit in terms of reproductive success and performance, compared to smaller ones. Therefore, populations having smaller specimens can be considered of higher concern in a conservative perspective. Body size in predatory arthropods determines their ability to thrive and the proportion of resources that they can allocate for reproduction (Jakob *et al.* 1996; Sokolovska *et al.* 2000). A positive relationship between body size and reproductive success has been reported in several spider species (Marshall & Gittleman 1994), including wolf spiders (Anderson 1990; Uetz *et al.* 2002; Ameline *et al.* 2018).

The size of the cocoon is a universal proxy of fitness, as a larger cocoon corresponds generally to a greater number of eggs (Marshall & Gittleman 1994; Bowden *et al.* 2013). However, in our case, we could not recover a significant relationship between cocoon size and habitat suitability, possibly due to low sample size (a similar issue was encountered in Mammola *et al.* [2019]).

Our approach to the monitoring turned out to be a practical and non-invasive tool for the assessment of the species health, and the morphological traits measured during the monitoring programme proved to be reliable proxies for the overall body size and for the reproductive success of the species. The results obtained during this first step of the monitoring (the “zero status”), provided baseline informa-



tion on the status of the populations, and will be compared with the results of the future monitoring campaign, planned for 2024, to detect potential population decline over time and to inform suitable response measures and coordinate conservation policies.

#### CONCLUDING REMARKS AND PERSPECTIVES

Despite the research carried out in these years and the above-mentioned advances in the knowledge of this species, several aspects still remain unknown, especially the ones inherent in the life cycle of this species, from post-embryo stages to adults. As a consequence, rather than based on direct measures, the life-span and the number of instars of the specimens have been estimated on the basis of the observed duration of the instars and of the information available in literature for other large entelegyne spiders. Laboratory rearing aiming to observe the growing pattern in captivity would be welcome to confirm our – inferred, but reliable – hypothesis.

In addition, there is uncertainty on the overwintering behaviour and on the seasonal movements of the specimens. According to literature data and field observations, no specimens have been found from the beginning of October (see Maurer & Thaler 1988) onwards, both in rocky areas and in other adjacent habitats.

During our laboratory experiments, we were not able to trigger any winter dormancy in laboratory conditions: specimens collected in late summer and placed in climatic chamber at low constant temperatures (0-1°C) did not enter dormancy, but remained active for 40-60 days and died afterwards, leaving open the question about overwintering behaviour in this species.

Another key aspect that needs further investigations is the courtship behaviour. Lycosids are known for their complex pre-copulatory behaviour, in which visual, vibratory and chemical signals are involved in sexual communication. Despite numerous attempts, the laboratory observations did not provide significant results. Males have been observed approaching the females and performing drumming against the substrate using the palps, but with no significant reactions elicited in the females. It seems likely that laboratory conditions where the mating was attempted, were not able to reproduce the basic conditions to ensure the mating.

Future studies are thus required to better clarify several aspects of the ecology and life history of this species, and additional samplings are needed to gain a further better understanding of the species distribution. In this regard, given that part of this species distribution falls within protected areas, training initiatives designed to provide to the staff of the parks and to volunteers a basic knowledge to detect and identify the species, could represent a valid contribution in collecting species occurrence information, and could help to attract more conservation attention to this species – one of the biggest wolf spiders in Europe – as seen for other charismatic species occurring in the Alps.

In light of its ecology, *Vesubia jugorum* has been recognized as a suitable model for the study of the effect of the climatic changes on the alpine biome (Isaia *et al.* 2016; Mammola

*et al.* 2019). Accordingly, enhancing awareness on the threats affecting this very emblematic species, may contribute to its conservation and may promote the general perception of the effects of climate change on the alpine ecosystems.

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