

# Diversity, distribution, habitat preferences and community assemblages of Amphibians and Reptiles in the “Cilento, Vallo di Diano e Alburni” National Park (Campania, Southern Italy)

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**Abstract.** Amphibians and reptiles are important ecological indicators of ecosystem status and play significant ecological roles. However, their populations are declining globally due to factors such as climate and land-use change. To effectively manage these species, it is crucial to study their distribution in protected areas. In this study, we provide new information on the distribution and ecology of amphibians and reptiles in the “Cilento, Vallo di Diano e Alburni” (CVDA) National Park in southern Italy. We used a comprehensive dataset consisting of 2465 records obtained from fieldwork, citizen science, and literature to evaluate species’ rarity, habitat preferences and community assemblage. We investigated whether differences in species richness across different habitat types and elevations resulted from species selection of different environmental conditions. We quantified species probability of observation in different habitat types and estimated the significance of species-habitat associations. Species rarity was assessed by considering geographic range, population abundance, and habitat breadth. Collected data substantially improved knowledge of the distribution of numerous species compared to published data. Our findings are generally consistent with the species’ ecological information available in Italy, but we also found some species’ peculiar ecological aspects that are little-known. The CVDA National Park emerges as an effective protected area that ensures good conservation status of the herpetofauna, also in the case of species showing population and range declines at the national scale.

**Keywords.** Herpetofauna, protected areas, community ecology, elevation range, species rarity.

## INTRODUCTION

Amphibians and reptiles play important roles in nutrient cycling and connecting aquatic and terrestrial environments. They are also considered ecological indicators of ecosystems’ status and stability (Pereira et al., 2012). Nonetheless, the global biodiversity crisis includes a decline in amphibian populations worldwide, with over 40% of amphibian species at risk of extinction (Stuart et al., 2004; IUCN, 2023). Significant population declines have also been found for reptile populations, with more

than 21% of the world’s reptiles threatened by extinction (Cox et al., 2022). Amphibians and reptiles face similar stressors, including agriculture intensification, overexploitation of natural resources, urbanization, introduction of alien species and climate change. In particular, panzootic chytridiomycosis is a major threat to amphibians and it is causing widespread population collapses and extinctions worldwide (Scheele et al., 2019). These factors mainly operate at small spatial scales, resulting in numerous cases of local extinctions. Local conservation action within protected areas can be the key to halting the decline of

amphibians and reptiles. To achieve this, it is crucial to have a comprehensive picture of species' ecology and distribution in these areas. Studying the distribution of herpetofauna in protected areas provides information to identify areas of high conservation value and prioritize conservation actions, such as habitat and connectivity restoration, to improve landscape-scale conservation (Beale et al., 2013). This information can also guide decisions about land-use planning to ensure sustainable development in complex landscapes where people and biodiversity coexist (Guillera-Arroita et al., 2015). This is the case of the Cilento (Campania Region), a highly diverse but yet under-investigated area of southern Italy protected by the "Cilento, Vallo di Diano e Alburni" (CVDA) National Park, which ensures a remarkable equilibrium between nature and society. The Park hosts many endemic species and subspecies of herpetofauna (Romano et al. 2010; Romano, 2014) whose distribution has been historically overlooked. Here we provide detailed information on terrestrial herpetofauna distribution and ecology in the CVDA National Park by presenting and using a dataset resulting from 14 years of fieldwork, citizen science and literature. Using the distribution dataset, we evaluated species rarity, the influence of elevation on species distribution and richness, the terrestrial and aquatic habitat preferences and, finally, the assemblage of species at terrestrial and water sites.

## MATERIAL AND METHODS

### *Study area*

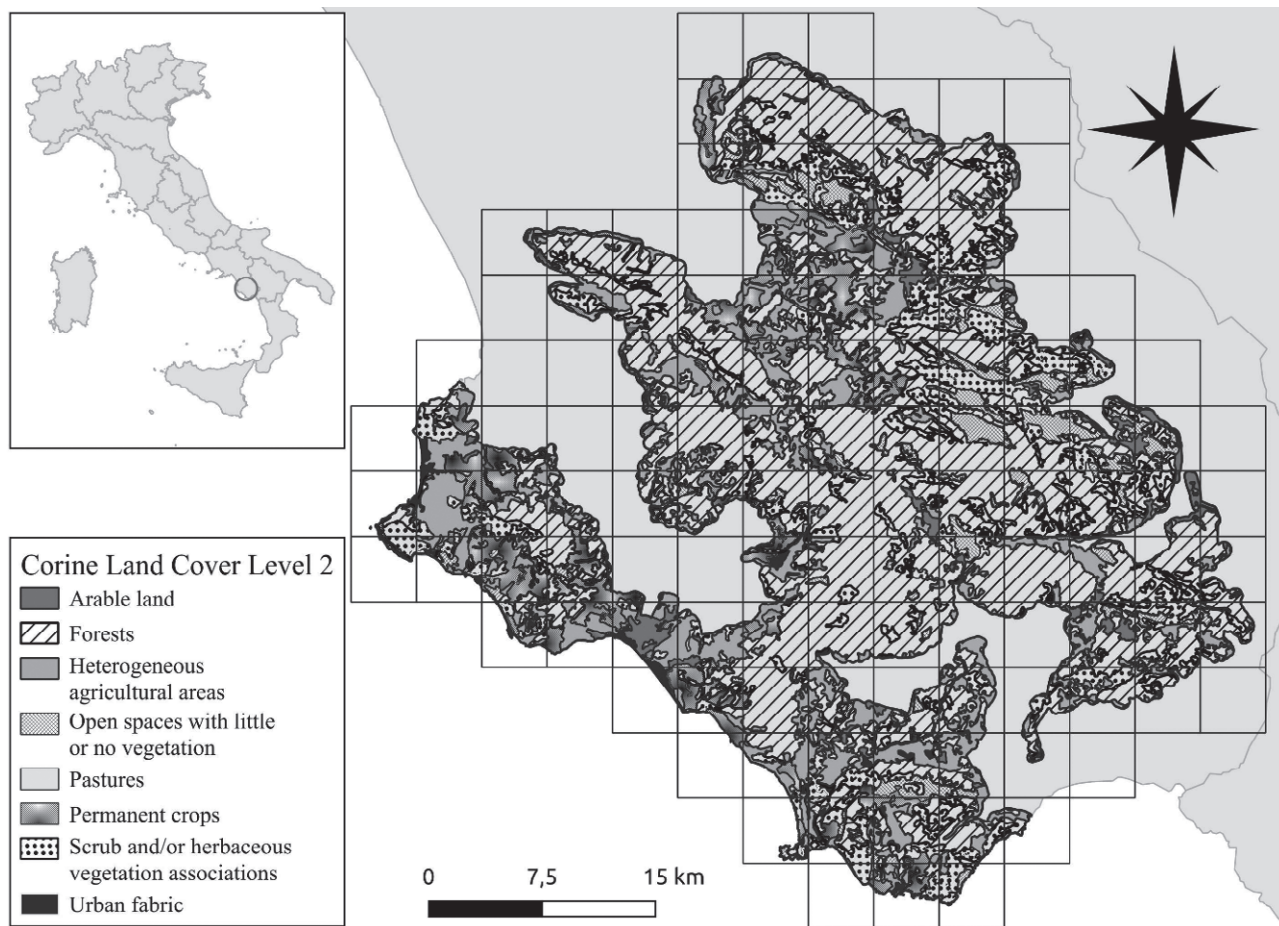
The "Cilento, Vallo di Diano e Alburni" National Park (CVDA) National Park is located in the Province of Salerno, southern Italy, and has a total area of 181,048 hectares, making it the largest national park in Italy. The park extends between 40°00' and 40°30'N and 14°50' and 15°40'E, stretching from the Tyrrhenian coast to the foot of the Apennines in Campania and Basilicata regions. It includes the peaks of Alburni Mountains, the Mt. Cervati (the highest mountain of the park, 1897 m asl), the Mt. Gelbison and the coastal buttresses of Mt. Bulgheria and Mt. Stella (Fig. 1). The park's topography is mainly hilly and mountainous except for the tectonic depression of the Vallo di Diano, which is the northern portion of the park. This complex topography translates into high environmental and climatic heterogeneity (Guarino et al., 2012; Guglietta et al., 2015). The lowland and coastal zone is characterized by a typically Mediterranean climate, with dry winters and warm summers, while the highland zone is more temperate, with dry winters and cold summers (Peel et al., 2007). According to the Corine

Land Cover Level 2 (Copernicus, 2018), the study area is mainly covered by Forests (52.13%) and Heterogeneous agricultural areas (hereafter Heterogeneous agrosystems, 15.02%) and Shrub and/or herbaceous vegetation associations (hereafter Scrub/herb vegetation, 13.49%), followed by Permanent crops (8.78%), Open spaces with little or no vegetation (6.29%, hereafter Scarce Vegetation), Arable land (2.43%), Urban fabric (1.37%) and Pastures (0.47%) (Fig. 1). Coastal areas are mainly characterized by Mediterranean scrubland while inner zones present extensive forests with interspersed natural grasslands. Several human settlements existed since ancient times in the Cilento area, however, human population density in the Park (85 municipalities, total population about 200.000) is rather low, with a median of 60 inhabitants per km<sup>2</sup> (ISTAT, 2023). In general, human settlements coexist in harmony with the natural environment and reflect local people's traditional lifestyles (Cerreta & Girard 2021). Traditional agro-pastoral practices have created complex patterns of agrosystems, mainly based on low-intensity annual crops, and olive groves as permanent crops; these agrosystems host significant patches of natural vegetation throughout the study area.

### *Data collection*

We obtained data on species occurrences from three main sources: literature, citizen science, and fieldwork. To ensure complete coverage of the study area, we divided it into a grids of cells of 5×5 km, which are submultiples of the 10×10 km UTM grid, resulting in 117 cells overlapping with the Park area. Grid-based sampling provides a good trade-off between sampling bias and costs at both the single- and multi-species levels (Marta et al., 2019). During springs and summers of the last 14 years (from 2008 to 2022), we carried out at least one field survey in each cell (mean surveys per cell: 3.692) targeting historical sites known from the literature and new sites potentially suitable for herpetofauna. We identified potentially suitable sites using the Military Geographical Institute Map (IGM) (MATTM, 2017), which accurately maps the presence of water, and aerial photographs from Google Maps (<https://www.google.it/maps>) using the software QGIS 3.22 (<https://qgis.org/>).

To search for amphibians and reptiles on the field, we followed standard methodologies (McDiarmid et al., 2012; Dodd, 2009; 2016). In particular, amphibians' potential aquatic habitat types have been identified and sampled following the methods reported by Romano et al. (2010; 2012). For both, we gathered additional information about species presence from local people, primarily shepherds and farmers. We obtained bibliographic



**Fig. 1.** Study area, the “Cilento, Vallo di Diano e Alburni” National Park, with Corine Land Cover level 2 categories and the used UTM grid of 5×5 km cells.

data from Caputo et al. (1985), Caputo and Guarino (1992), and Ruffo and Stoch (2005). We georeferenced species occurrences following the “Guide to Best Practices for Georeferencing” (Chapman and Wieczorek 2006) using QGIS and the IGM (MATTM 2017), which provides toponyms used in the above-mentioned sources. In 2021 we created a citizen science project on iNaturalist entitled “Herpetofauna of Cilento” (<https://www.inaturalist.org/projects/herpetofauna-del-cilento>).

#### *Species richness and elevation range*

Using the chosen grid of 5×5 km, we obtained maps of species  $\alpha$  diversity (species number per grid cell) for amphibians, reptiles and the two combined using the QGIS function “count points inside polygons” with species as value of the “class” parameter. We used the Copernicus Digital Elevation Model (DEM) at 20 m resolu-

tion (Copernicus, 2021) to extract the elevation (m asl) of each occurrence point in QGIS. For observations that could present the species’ higher elevation limits, we used a GPS Garmin device to record the elevation in the field. We reclassified the DEM into nine elevation classes (Fig. S1) of 200 meters (except for the highest class that ranged from 1600 to 1879 m asl) using QGIS. To investigate whether there are significant differences among species’ elevation preferences, we used Kruskal-Wallis one-way analysis of variance (KW), followed by Mann-Whitney U test and Bonferroni correction. We tested for correlation between elevation classes and a diversity by applying the Spearman rank correlation test for herpetofauna, amphibians and reptiles, using the function `cor.test` in R software version 4.2.3 (<https://www.r-project.org/>). Furthermore, we used the Chi-square test to test the hypothesis that changes in species richness reflect only the available land area. This involved comparing the percentage of land area and the percentage of species occurrence in each eleva-

tion class. KW, Mann-Whitney U and Chi-square tests were performed as in the statistical package PAST ver.4 (Hammer et al., 2001).

### *Species richness and rarity*

To describe species diffusion and rarity, we considered only those 5×5 km cells containing at least one record of amphibians (N=85) or reptiles (N=102) of the 117 5x5 km cells. We estimated species rarity using two methods, the Index of Species Rarity (ISR) and Rabinowitz's forms of rarity. The ISR was originally proposed by Gheu & Gheu (1980) and was calculated as follows:  $ISR = (1 - n/N) \times 100$ , where  $n$  is the number of grid cells occupied by a species and  $N$  is the total number of grids of its class. This index ranges from 0 (very common species) to 100 (very rare species, i.e. only one record). The second method is based on Rabinowitz's approach (Rabinowitz et al., 1984) as implemented in the "Rare7" R package (Maciel and Arlé, 2020), which employs a combination of three parameters to evaluate species' rarity: (1) geographic range, (2) population abundance, and (3) habitat breadth. The combination of these three parameters results in the classification of a species as either common or as one of seven types of increasing rarity. We estimated the size of the geographic range for each species by counting the number of occupied 5x5 km cells. We used the mean number of records per cell as a proxy for local abundance, and we calculated habitat specificity by examining the number of Corine Land Cover level 3 types (CLC3) in which the species were found. To account for the effect of possible local extinctions, we calculated ISR and Rabinowitz's only using post-2000 data. We chose three thresholds to classify species in a wide or restricted range, various or uniform habitat and scarce or abundant populations: the 25<sup>th</sup> percentile of occupied cells/total number of cells, the median number of mean occurrences per cell and the median number of frequented habitat types (CLC3).

### *Habitat preferences*

To determine the terrestrial habitat type in which the species was recorded, we used the Corine Land Cover (CLC), a European program that collects and disseminates data on land cover and land use throughout Europe (Copernicus, 2018). The CLC classifies land cover into different levels of detail, namely CLC Level 2 (CLC2) and Level 3 (CLC3). The first comprises 15 broad land cover types that distinguish between macro environmental conditions, e.g. Urban Fabric, Arable Land, For-

ests etc. The second provides more detailed information within each of the Level 2 land cover types, resulting in 44 subcategories. For instance, the Level 2 category "Forests" is further subdivided into three Level 3 categories: broad-leaved forest, coniferous forest and mixed forest. We assigned a CLC2 category to each occurrence point in QGIS 3.2 with the "join attributes by location" function. CLC2 categories were considered relevant for species' broad distribution in the study area as they identify general differences among environments. Eight CLC2 categories are present in the study area (Fig. 1). For aquatic habitat types, we classified amphibians' breeding sites into nine categories based on their characteristics (Romano et al., 2010; Romano, 2014): ditch (small running waters occurring in agricultural or urban areas), drinking trough for livestock grazing (rectangular and elongated, singular or multiple container, made of concrete or stone, filled by running water), pond (small still waters), river (perennial running waters greater than streams), spring (pools generating from a water spring), stream (running waters with a torrential regime), tank (generally a single quadrangular stone or cement cistern designed to collect water for irrigation), vernal pool (small seasonal still waters), and well (holes drilled to access groundwater) (for detailed differences among artificial water sites see Romano, 2012).

To investigate species-habitat associations, we calculated the probability of observing the species in each habitat type (aquatic for amphibians and terrestrial for both) and the significance of each possible association. In order to examine the relationship between species and habitat type, we made a 2×2 contingency table (Table 1) with the following division of species occurrences: *i*) the number of occurrences of the focal species in the focal habitat type (1,1: row 1 and column 1), *ii*) the number of occurrences of all species recorded in the habitat type (1,2), *iii*) the number of occurrences of the focal species in the focal habitat type (2,1), and *iv*) the number of occurrences of all species recorded in all habitat types (2,2). We used this table to estimate species' probability of being

**Table 1.** The 2x2 contingency table used to calculate odds ratios for species-habitat relationships with species' occurrences on rows and habitat types on columns.

	Habitat type	All habitat types
Species	Number of occurrences of the focal species in the focal habitat type.	Number of all occurrences of the species except those in the focal habitat type.
All species	Number of all occurrences recorded in the habitat type except those of the focal species.	Number of all occurrences recorded in all habitat types except those recorded in the focal habitat type.

observed in each habitat type as by calculating the odds ratio by dividing the product of the counts in cells 1,1 and 2,2 by the product of the counts in cells 1,2 and 2,1 and then by transforming the odds ratio in probability of observing in each habitat type using the equivalence:  $\text{Probability} = \text{Odds} / (\text{Odds} + 1)$  (McHugh, 2009; Grant, 2014), obtaining values ranging from 0 (low probability of observing the species in the habitat type) to 1 (high probability). To determine if the found habitat-species associations were statistically significant, we performed the Fisher’s Exact Test for count data using the ‘fisher.test’ R function. Finally, we used the Chi-square test to test the hypothesis that changes in species richness between habitat types reflect the available habitat types’ area. This involved comparing the percentage of land area and the percentage of species occurrence in each habitat type in the software package PAST ver.4 (Hammer et al., 2001).

### Community assemblages

To investigate species association, we first defined species assemblages using distances between occurrence points and habitat types. To create amphibians sampling units, we used a 100 meter buffer from each occurrence and dissolved the resulting circles according to the occupied aquatic habitat types, merging multiple buffer polygons of the same habitat type into one single polygon. This method ensured that if two species occurred within a distance of 100 m but in two different habitat types (e.g. a stream and a stony well), they would be considered from two different species assemblages. Similarly, we created sampling units for both amphibians and reptiles using a buffer of 250 m from each occurrence and dissolving the resulting circles according to the occupied terrestrial habitat types. We used classes from the CLC3 (Copernicus, 2018), which we deem suited to describe the environmental filters behind the community assemblage process, and QGIS for spatial operations.

We created a matrix of species presence/absence per sampling unit using the function “ddply” from the “plyr” R package and “dcast” functions from the “madr” R package. We used it to perform community dissimilarity analyses for both amphibians and reptiles using the Raup-Crick Dissimilarity Index with Unequal Sampling Densities of Species (Raup and Crick 1979) using the ‘raupcrick’ function (Chase et al., 2011) in the R package ‘vegan’ version 2.6-4 (Oksanen et al., 2022). The Raup-Crick Index is a measure of  $\beta$  diversity used to identify differences in species assemblages occurring in different environments. This index describes the probability that compared sampling units have non-identical species composition. This probability is calculated through

“oecosimu” simulation (R package ‘vegan’) (Oksanen et al., 2022), where the observed amount of shared species between the tested sampling units is used as the test statistic and compared to a community null model where the probability of selecting a species is directly proportional to its frequency (Oksanen et al., 2022). The Raup-Crick Index ranges from 0 (identical assemblages) to 1 (distinct assemblages). Finally, we performed a cluster analysis based on Raup-Crick index values using the R function ‘hclust’ with Ward’s minimum variance clustering method (method=“ward.D”) to investigate how species relate to each other based on the similarity of their assemblages. We excluded two species found in only one location (*Emys orbicularis* and *Testudo hermanni*).

## RESULTS

### Species distribution

Following the taxonomy of Speybroeck et al. (2020), we recorded 16 species of reptiles and 11 of amphibians (Table 2). The collected dataset amounted to 2465 records (835 for amphibians and 1630 for reptiles). We georeferenced 330 records from bibliographic sources (194 and 143, for amphibians and reptiles respectively). From the iNaturalist project (at the 5<sup>th</sup> February 2023) we obtained 401 observations (130 and 271) by 180 users (excluding fieldwork personnel); finally, we collected 1734 occurrence points during fieldwork (498 and 1217). Bibliographic sources mostly covered the decades from 1980 to 2000 while most of the data were collected during the last 2 decades and recent years (mean = 2014, median = 2020). Maps showing the distribution of each species are available on Fig. S2.

Species a diversity per grid cell ranged from 1 species (3.6% of total species present in the area) to 18 (66.7%), with a median diversity value of 8 (Fig. 2). When considering amphibians and reptiles separately, cells with amphibians ranged from 0 to 9 per cell (81.8% of total), while those with reptiles ranged from 1 (6.25%) to 11 (68.75%) per cell. Interestingly, highly diverse cells were distributed throughout the territory but did not exhibit a uniform or random pattern. Instead, they tended to cluster together, particularly towards the inner regions of the territory.

Elevation distributions of amphibian and reptile species are shown in Fig. 3. The KW test showed highly significant differences among species in each of these two classes ( $H = 175.4$ ,  $H = 375.4$  for amphibians and reptiles respectively;  $P < 0.001$  for both classes). Among amphibians, *S. salamandra* and *T. carnifex* were present at the highest elevations, with the second species also charac-

**Table 2.** Amphibians and reptiles that occur in the “Cilento, Vallo di Diano e Alburni” National Park with short names used in figures, number of sites of occurrence and occupied 5x5 km cells. \*Species endemic to Italy.

Species	Short name	Records pre-2005	Records Post-2005	Total Records	Number of Cells
<b>AMPHIBIA</b>					
<i>Salamandra salamandra</i>	<i>Salsal</i>	10	19	29	11
<i>Salamandrina terdigitata</i> *	<i>Salter</i>	6	30	36	25
<i>Lissotriton italicus</i> *	<i>Lisita</i>	38	170	208	66
<i>Triturus carnifex</i>	<i>Tricar</i>	18	32	50	14
<i>Bombina variegata</i>	<i>Bomvar</i>	22	24	46	23
<i>Bufo bufo</i>	<i>Bufbuf</i>	30	69	99	47
<i>Bufo viridis</i>	<i>Bufvir</i>	0	4	4	4
<i>Hyla intermedia</i> *	<i>Hylint</i>	4	57	61	34
<i>Pelophylax lessonae</i> complex	<i>Pelles</i>	16	52	68	37
<i>Rana dalmatina</i>	<i>Randal</i>	7	11	18	12
<i>Rana italica</i> *	<i>Ranita</i>	34	182	216	66
<b>REPTILIA</b>					
<i>Emys orbicularis</i>	<i>Emyorb</i>	0	1	1	1
<i>Testudo hermanni</i>	<i>Tesher</i>	0	1	1	1
<i>Hemidactylus turcicus</i>	<i>Hemtur</i>	3	14	17	12
<i>Tarentola mauritanica</i>	<i>Tarmaur</i>	3	111	114	47
<i>Anguis veronensis</i> *	<i>Angver</i>	3	33	36	30
<i>Lacerta bilineata</i>	<i>Lacbil</i>	20	152	172	56
<i>Podarcis muralis</i>	<i>Podmur</i>	14	57	71	20
<i>Podarcis siculus</i>	<i>Podsic</i>	22	678	700	103
<i>Chalcides chalcides</i>	<i>Chacha</i>	12	19	31	24
<i>Coronella austriaca</i>	<i>Coraus</i>	6	11	17	14
<i>Elaphe quatuorlineata</i>	<i>Elaqua</i>	11	72	83	56
<i>Hierophis viridiflavus</i>	<i>Hievir</i>	17	201	218	82
<i>Natrix helvetica</i>	<i>Nathel</i>	11	53	64	37
<i>Natrix tessellata</i>	<i>Nettes</i>	3	10	13	12
<i>Vipera aspis</i>	<i>Vipasp</i>	10	45	55	40
<i>Zamenis lineatus</i> *	<i>Zamlin</i>	8	29	37	27
<b>TOTAL</b>		<b>328</b>	<b>2137</b>	<b>2465</b>	

terized by a narrow elevation range. The two species are thus restricted to mountain areas in the Park. *Bombina variegata* also preferred high elevations although some sites were recorded in hilly and lowland areas. Conversely, *B. viridis* and *P. lessonae* complex occurred mainly in lowland, although the latter exhibits a significantly wider elevation range than the former. The other amphibian species generally had a wide elevation range, with *B. bufo* reaching the highest quote (1850 m asl) in a vernal pool on the Cervati mountain peak. Another species showing a wide elevation range is *R. dalmatina*, which occurs from 75 to 1600 m asl. Among reptiles, *P. muralis* and *C. austriaca* mainly occur at high elevations. Similarly, *C. chalcides* and *L. bilineata* also occur mainly at high elevation, however both are present at lower elevations as well. Lowland species are *H. turcicus*, *T. mauritanica* and

*N. tessellata*, which only occasionally reached the mid-range elevations; while *E. quatuorlineata* and *Z. lineatus* showed a wider elevation range.

The nine elevation classes occupy an area percentage ranging from 0.6% and 19.7% (Fig. 4a). Species  $\alpha$  diversity generally decreases with increasing elevation classes (Spearman correlation test,  $r_s = -0.853$ ;  $P = 0.005$ ), with the middle elevation classes (200-400, 400-600 and 600-800 m) holding the highest number of species (Fig. 4a). However, when considering the two classes separately, amphibians  $\alpha$  diversity was not significantly correlated to elevation classes while reptiles  $\alpha$  diversity was ( $r_s = -0.579$ ,  $P = 0.108$ ;  $r_s = -0.900$ ,  $P = 0.001$ ). Amphibians and reptiles  $\alpha$  diversity showed a different pattern along elevation classes, with the first showing high richness at all classes except for the last two, and the latter a clear

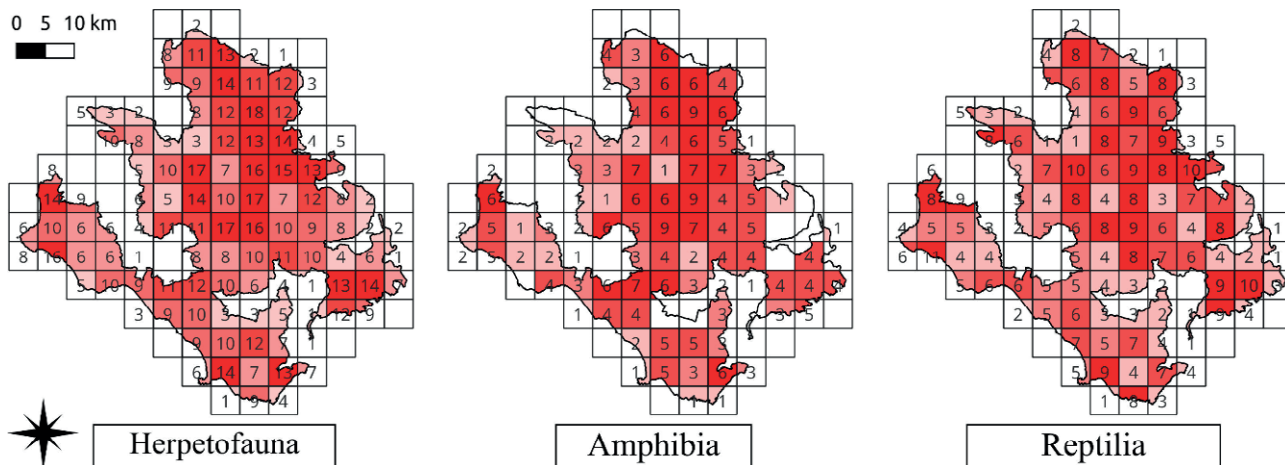


Fig. 2. Maps of species richness for the whole herpetofauna in the study area (a), and for amphibians (b) and reptiles (c) separately.

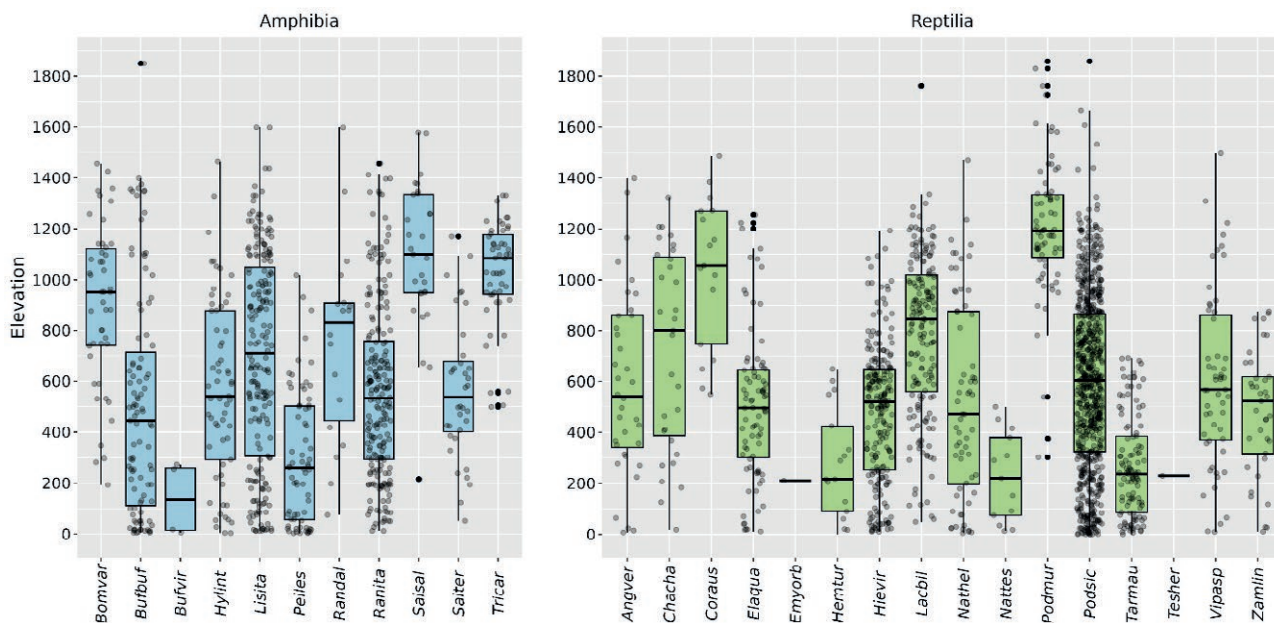
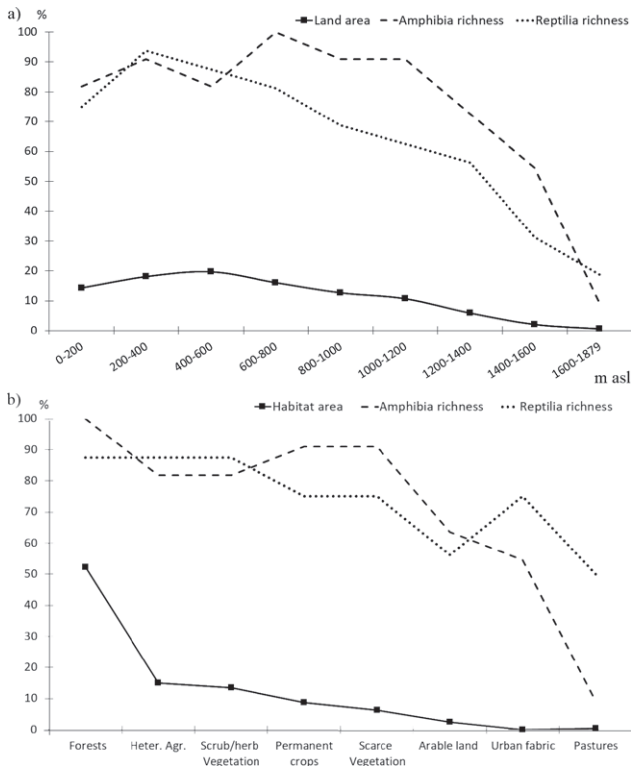


Fig. 3. Box plots of the altitudinal distribution of amphibians and reptiles in the “Cilento, Vallo di Diano e Alburni” National Park. The box represents the interquartile range (H-Spread), the median is the horizontal line inside the box; the lines extend from the lowest to the highest value within 1.5 H-Spread from the end of the box. Black circles at the end of the lines represent outliers. Species abbreviations as in Table 2.

almost monotonic decline in species richness after the 400-600 m class (Fig. 4a) with an important inflection after 1400-1600 m class. The results of the Chi-square test indicate that the species richness in the various elevation classes was proportional to the elevation class extent (Chi-square values were 13.34, 7.44 and 9.35 for amphibians, reptiles, and both, respectively;  $df = 9$  and  $P > 0.05$  for all tests).

Species rarity

The Index of Specific Rarity (Fig. 5), based on distribution and frequency, revealed a wide range of values among reptiles, with *P. siculus* as the most common species and others, i.e., *T. hermanni* and *E. orbicularis*, occurring in only in a single location. Snakes displayed varying degrees of rarity, with *H. viridiflavus* being relatively common and *C. austriaca* being the least common



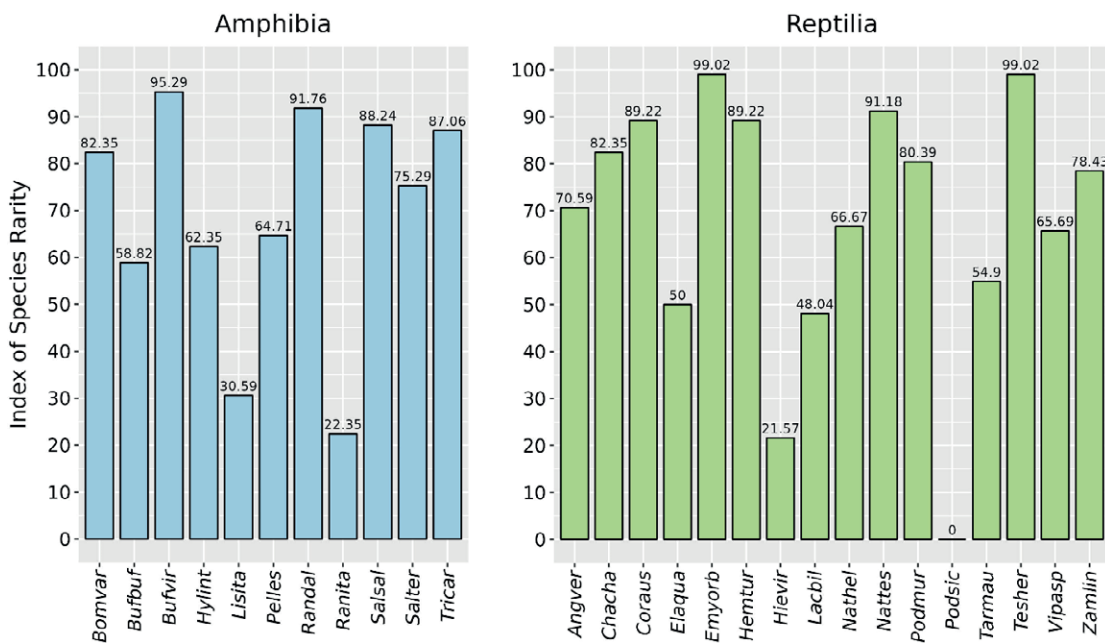
**Fig. 4.** Percentage comparisons between herpetological species richness and (a) the area availability of altitudinal ranges, and (b) the area availability of Habitat types in the “Cilento, Vallo di Diano e Alburni” National Park.

snake. Among amphibians, *B. viridis* is localized in only 4 sites, *R. dalmatina*, and *S. salamandra* exhibited relatively restricted distribution, whereas the two Italian endemic *R. italica* and *L. italicus* were widely distributed with a high number of sites.

Rabinowitz’s classification provided additional insights (Fig. 6). Common species were a minority for both amphibians and reptiles, with 3 (27%) and 6 (37%) species respectively classified in this category. On the opposite end of the spectrum, the seventh form of rarity had 2 (18%) species of amphibians and 7 (31%) of reptiles.

### Habitat preferences

The most frequented habitat type was Forests (35.50% of occurrences) followed by Scrub/herb vegetation (20.73%), Heterogeneous agrosystems (17.20%), Scarce Vegetation (10.71%), Permanent crops (8.84%), Urban fabric (4.06%), Arable land (2.47%) and Pastures (0.49%). (Fig. 4b). The habitat type that hosts the highest number of amphibian species is Forests, while Forests, Heterogeneous agrosystems, and Scrub/herb vegetation have the highest number of reptile species occurrences (Fig. 4b). The median number of frequented terrestrial habitat types (i.e., where the species was observed at least once) was 6.5 for reptiles and for 6 amphibians, out of 8 available terrestrial habitat types (Fig. S3). In addition, the median num-



**Fig. 5.** Bar plots of the Index of Specific Rarity (ISR) of amphibians and reptiles in the “Cilento, Vallo di Diano e Alburni” National Park. The higher the ISR value, the rarer the species. Species abbreviations as in Table 2.



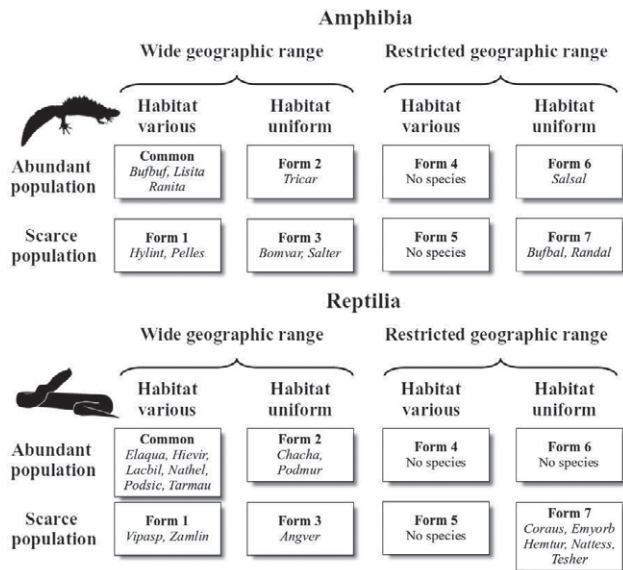


Fig. 6. Rabinowitz’s index of rarity of amphibians and reptiles in the “Cilento, Vallo di Diano e Alburni” National Park, as implemented in the “Rare7” R package (Maciel and Arlé, 2020). The index provides a classification of a species as either common or as one of seven types of increasing rarity. Species abbreviations as in Table 2.

ber of aquatic habitat types frequented by amphibians was 6 out of 9. However, the probability of observing a species in each habitat type was not the same, furthermore, not every species-habitat association was statistically significant according to the Fisher exact test (Figs. 7, 8).

The results of the Chi-square test indicate that the species richness was not proportional to the amount of habitat type area (Fig. 4b; Chi-square values were 31.8, 43.6 and 38.3 for amphibians, reptiles, and both, respectively;  $df = 8$  and  $P < 0.01$  for all tests). We identified clear signals of habitat type preference (high probability of observing the species) or avoidance (low probability for all species). Reptiles showing significant habitat associations were 11 out of 16 while amphibians were 9 out of 11. The median number of significant associations was 2 for reptiles and 3 for amphibians. The species with the highest number of significant associations was, among reptiles, *P. siculus* (5) and, among amphibians, the *P. lessonae* complex (5). Regarding aquatic habitat types, all amphibians showed significant associations (median 2) with *Lissotriton italicus* and *P. lessonae* complex showing the highest number among species (6).

Species association

The Raup-Crick Index highlighted high community similarity between pairs of species. Among amphibians,

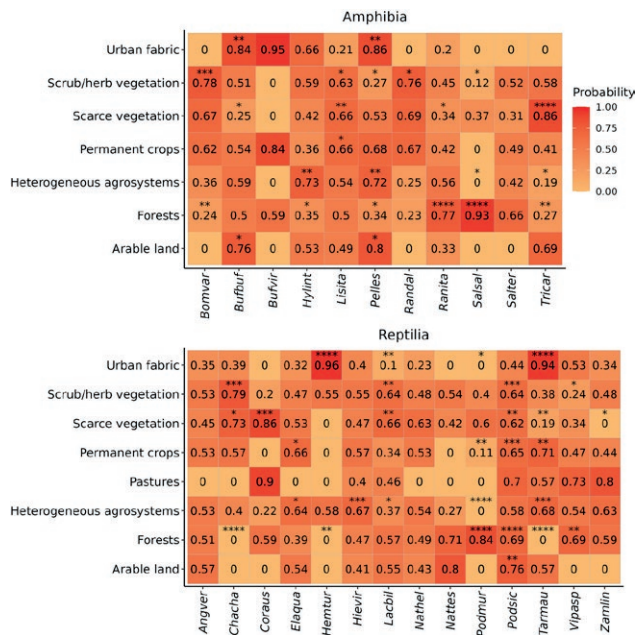


Fig. 7. Probability (Odds ratio / (Odds ratio + 1; see text) of amphibian and reptile preference for terrestrial habitat types (i.e. Corine Land Cover level 2 classes) in the study area. Statistically significant preferences are shown with asterisks: \*  $P \leq 0.05$ ; \*\*  $P \leq 0.01$ ; \*\*\*  $P \leq 0.001$ ; \*\*\*\*  $P \leq 0.0001$ . Species abbreviations as in Table 2.

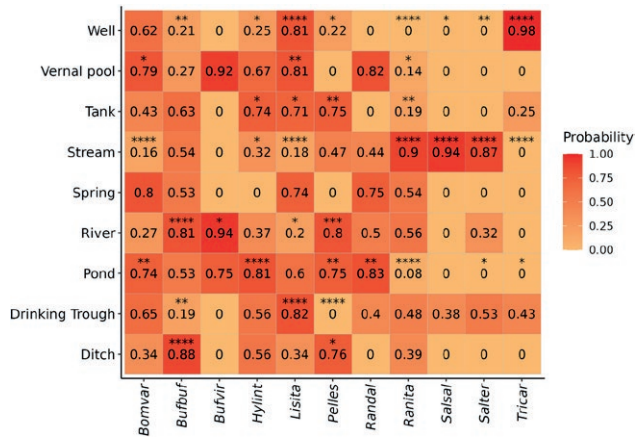


Fig. 8. Probability (Odds ratio / (Odds ratio + 1; see text) of amphibian preference for aquatic habitat types. Statistically significant preferences are shown by asterisks: \*  $P \leq 0.05$ ; \*\*  $P \leq 0.01$ ; \*\*\*  $P \leq 0.001$ ; \*\*\*\*  $P \leq 0.0001$ . Species abbreviations as in Table 2.

the pairs with the lowest Raup-Crick Index were *Triturus carnifex* and *Lissotriton italicus* (0.002), *Salamandrina terdigitata* and *Rana italica* (0.01), *Triturus carnifex* and *Bombina variegata* (0.12), while among reptiles, *Vipera aspis* and *Elaphe quatuorlineata* (0.001), *Tarentola mauritanica* and *Hemidactylus turcus* (0.001) and *Natrix tessellata* and *Natrix helvetica* (0.003) (Fig. S3).

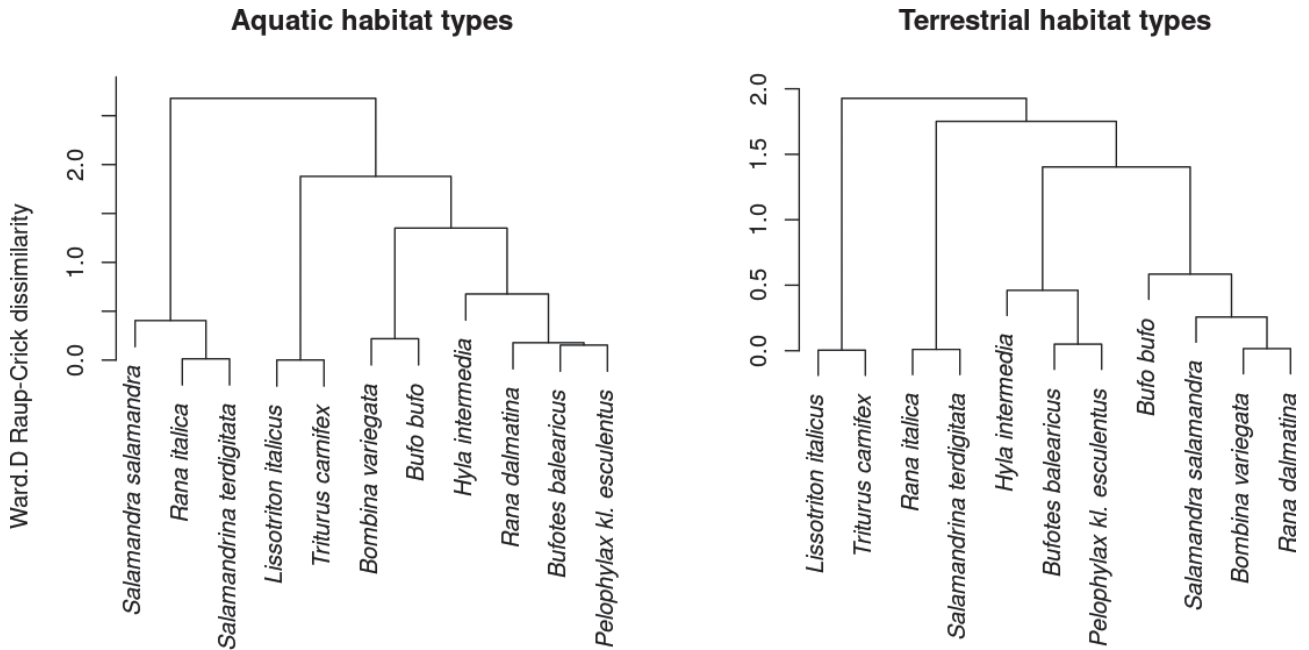


Fig. 9. Cluster analysis of amphibian species similarity assemblage based on Raup-Crick index values using the Ward's minimum variance.

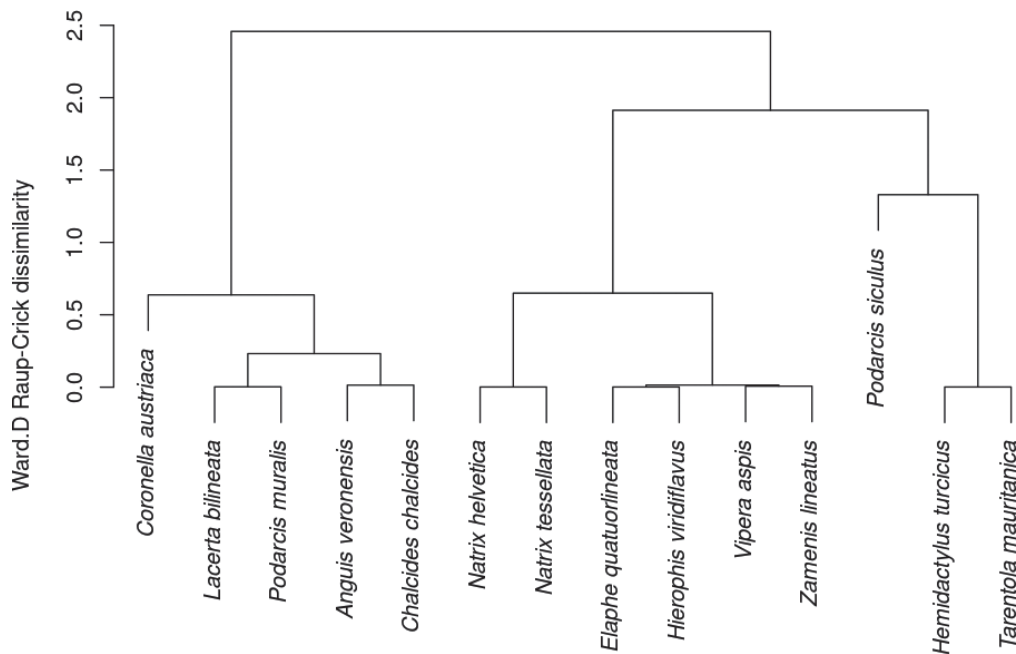


Fig. 10. Cluster analysis of reptile species similarity assemblage based on Raup-Crick index values using the Ward's minimum variance.

The cluster analyses based on The Raup-Crick Index produced a clear indication of species associations (Fig. 9 a,b; Fig. 10). Considering aquatic habitat types, among amphibians are recognizable two main clusters (Fig. 9a):

one including species associated with running waters and one including all the remaining species that preferred lentic or very slow-running waters. Considering the terrestrial habitat types, the association among species is

similar but shows some differences (Fig. 9b). Reptile species relationships are more complex (Fig. 10) with almost all snakes clustered together, with *N. helvetica* and *N. tessellata* grouped in a subcluster because both were found mainly in aquatic environments. The last cluster includes *P. siculus*, which may be considered almost ubiquitous, and the two geckos that are associated with urban and open areas and that avoid forests.

## DISCUSSION

### *Species distribution*

Protected areas are designated areas that are managed to conserve natural ecosystems and biodiversity (Rodríguez-Rodríguez and Martínez-Vega, 2022) and can provide critical refuge for species sensitive to habitat loss and fragmentation. However, the effectiveness of protected areas in conserving biodiversity depends on their ability to represent the diversity of species that occur within their boundaries. Using an extensive dataset based on decades-long fieldwork, citizen science and bibliographic sources, we investigated the distribution, ecology and community assembling of amphibians and reptiles in the “Cilento, Vallo di Diano e Alburni” National Park. Our work painted a comprehensive picture of the herpetofauna diversity inside the park, which has been overlooked in the past, and highlighted how the Park represents a hotspot area for amphibians and reptiles in Southern Italy. We found all the species that are potentially present in the area according to their known regional distribution (Guarino et al., 2012). They greatly differed in distribution and rarity. *Emys orbicularis* (classified by the IUCN as Endangered) and *Testudo hermanni* (Critically Endangered, Rondinini et al., 2022) are localized in only one site; in particular, the presence of a wild population of *T. hermanni* has not been confirmed, as only an individual in the wild was found, and the possibility of an escape from captivity cannot be ruled out. With the exception of these two species, the other ones have been classified in some category of threat by the IUCN (Rondinini et al., 2022), are quite extensively distributed inside the park. The conservation value of the study area is thus significant not just locally but also at a national scale, as several rare endemic species and subspecies (Table 2) were recorded (Fig. S2 and Fig. 5 and 6).

The highest values of  $\alpha$  diversity for both amphibians and reptiles are found in inland areas, particularly hilly and mountainous regions. However, some coastal or sub-coastal zones also exhibit high values of species richness (Fig. 2). Defining elevation ranges at a local scale can help determine the potential presence of certain species, making it easier to detect and monitor them within pro-

TECTED areas. Higher species richness is observed between 200 and 800 m for both amphibians and reptiles, while richness decreased with increasing elevation. Numerous studies have shown that animal and plant richness is highest at mid-elevations, and three hypotheses have been proposed to explain this pattern (reviewed in Rahbek, 1995). The “ends are bad” hypothesis suggests that there are limiting factors at high and low elevations, while the “middle is good” hypothesis cites optimal productivity at mid-elevations. The third hypothesis suggests that lower elevations experience more disturbance, leading to reduced diversity. Studies on herpetofauna suggest either a consistent decrease in richness with increasing elevation or a hump-shaped relationship with a peak at a certain elevation (Fauth et al., 1989; Nathan and Werner, 1999; Fu et al., 2006; Fischer and Lindenmayer, 2005; Wiens et al., 2007). Reptiles generally exhibit decreasing patterns (McCain, 2010), while preliminary studies on amphibians suggest mid-elevation peaks for salamanders and varied patterns for frogs. Amphibians and reptiles in the CVDA National Park follow a similar trend but reptiles begin to decrease at lower elevations than amphibians and more linearly (Fig. 4a). Indeed, the first mountain elevation zone is a hot spot for amphibian diversity, while almost all reptile species straddle between lowland and hilly elevation zones (Fig. 4a). Besides species richness, the conservation value of an elevation range is also determined by which species are present within it (Fig. 3). Species’ elevation preferences suggest that lowland areas are important for the conservation of *B. viridis*; hilly and low mountain areas (500-1000 m asl) are focal for *R. dalmatina*, and mountain ranges (above 900 m asl) are crucial for the conservation of *S. salamandra*. Regarding reptiles, *H. turcicus* and *N. tessellata* will benefit from additional sampling efforts toward lower elevation areas. Conversely, a targeted conservation effort for *C. austriaca* will mainly need to take into account mountain areas. Finally, it is noteworthy to mention that our data provide new upper elevation limits for seven species in the Campania Region in comparison to the limits reported by Guarino et al. (2012). Specifically, our findings indicate that *B. bufo* can be found at elevations up to 1850 m (compared to 1450 m), *H. intermedia* up to 1465 m (compared to 1180 m), *R. dalmatina* up to 1600 m (compared to 1350 m), *L. italicus* up to 1600 m (compared to 1450 m), *P. muralis* up to 1860 m (compared to 1597 m), *P. siculus* up to 1860 m (compared to 1600 m), and *C. austriaca* up to 1485 m (compared to 1375 m).

### *Habitat preferences, species rarity and association*

This study encompassed all types of habitats within the study area to investigate the distribution and ecology

of the herpetofauna. In terrestrial environments, amphibians and reptiles exhibit a richness that is proportional to the availability of habitat type area (Fig. 4b). Amphibian richness is proportional to the availability of Forests, Pastures, and Urban fabric but it is higher than expected in Scarce vegetation and Permanent crops. These habitat types, as a rule, are not suitable to ecological requirements of amphibians, however, it should be noted that in the Cilento area, where non-intensive agriculture is prevalent, they host numerous artificial water sites used for irrigation and watering of cattle and by amphibians as breeding sites. Reptiles appear to positively select Urban fabric and counter-select Forests, shaded environments which offer reduced basking opportunities only on their margins and ecotonal zones (Jaggi & Baur, 1999). The majority of reptile species in the CVDA National Park are active thermoregulators, while only a few species do not actively thermoregulate and prefer shaded habitats, such as the cover specialist *Anguis veronensis*. The preference for Urban fabric can also be attributed to the characteristics of the Cilento's landscape, in which urban settlements are sparsely distributed inside natural environments, with numerous ruined buildings. Even the continuous Urban fabric of main towns includes numerous dry stone walls, small vegetable gardens, and family chicken coops which offer high availability of shelters and trophic resources.

Considering species' habitat preferences, among amphibians (Fig. 7a) *B. bufo* and *P. lessonae* complex were the only two species that showed a positive association with Urban fabrics, consistent with their ecology on the Italian territory (Sindaco et al., 2006). *Lissotriton italicus* exhibited a positive association with Permanent crops (mainly olive groves), Scrub and Scarce vegetation. This endemic species spawns frequently in drinking troughs, stony wells and irrigation tanks (Romano et al., 2010; 2012), all man-made elements found in rural landscapes. Consequently, as expected for some species of amphibians, the importance of a particular terrestrial habitat type is determined by its association with certain aquatic habitat type preferences. Other species, such as *S. salamandra*, showed a negative correlation with open environments, while others, like *B. variegata* and *T. carnifex*, are negatively associated with Forests, in agreement with their general ecology (Sindaco et al., 2006). The habitat type with the most positive associations is Scarce vegetation, but this habitat type is also negatively correlated with the occurrence of many other species, along with Urban fabrics and Forests (Fig. 7b).

The relationship between amphibians and aquatic habitat types displays a considerable degree of interspecific variation (Fig. 8). *Triturus carnifex* and *S. salamandra* are the most stenoecious species because they are strictly

associated with only one habitat type (wells and streams, respectively) avoiding all other aquatic habitat types. While this is consistent with the knowledge of the Fire salamander on the national territory, it does not hold true for the Italian crested newt (Sindaco et al., 2006). Indeed this species appears to be relatively adaptable to several aquatic environments, preferring permanent or semi-temporary water bodies, in plains or moderately elevated areas (Sindaco et al., 2006; Vanni et al., 2007). Conversely, in the CVDA National Park, *T. carnifex* emerges as a stenoecious species, considering both aquatic (Fig. 8) and terrestrial habitat types (Fig. 7a), and for the elevation range (Fig. 3a).

On the other hand, *Lissotriton italicus* and *P. lessonae* complex showed a significant positive association with many aquatic environments. On the whole, ponds are the aquatic habitat type with the highest level of positive species association while wells are characterized by the highest rate of negative associations (Fig. 8).

Species association revealed interesting patterns. Two primary groups can be distinguished among amphibians based on the types of aquatic habitats they occupy (Fig. 9a): the first includes *Salamandra salamandra*, *Salamandrina terdigitata* and *Rana italica* that share running waters. The latter two are in a subcluster because they breed typically in syntopy as already known in the Apennines (Romano et al., 2007; 2012). The second cluster includes all the other species although is a much more branched group, where *T. carnifex* and *L. italicus* cluster together, *Bufo bufo* and *Bombina variegata* seems to share similar aquatic habitat types; the other anuran species are associated because they share still or very slow-running waters. Clearly, aquatic habitat preferences dictate amphibian association, furthermore they partially reflect phylogeny, as close species clustered together (e.g. geckos, newts, and water snakes).

Considering the terrestrial habitat types for amphibians (Fig. 9b), the two newts are associated, as for aquatic habitat types, because they often breed in stony wells and concrete tanks used in agriculture or pasture areas. The second cluster includes those species (*S. terdigitata* and *R. italica*) that preferred forest habitat types (from beech to maquis forests). The third species group is associated mainly with urban or suburban areas but also arable lands. The last cluster includes species occurring in a variety of habitat types and in particular in Forests, Scrub/herb vegetation but also Heterogeneous agrosystems (low-intensity), although *S. salamandra* significantly avoids agricultural areas (see Fig. 7a).

The complexity of relationships among reptile species is greater (Fig. 10). *Coronella austriaca* is the only colubrid markedly separated by the other snakes (that clustered together) and clusters with saurians experiencing a

great variety of habitat types but in particular with those associated with Scarce vegetation (see also Fig. 7b).

ISR identified species rarity considering their distribution only and was consistent with the Rabinowitz's forms of rarity that identified species potentially more vulnerable to habitat loss despite being relatively widespread, due to their restricted habitat breadth (Fig. 6). Species characterized by at least two of the three parameters used to assess rarity should be considered priority species for increased sampling effort and, when needed, conservation actions. Example of these species are, among amphibians, *B. variegata*, *S. salamandra*, *S. terdigitata*, *B. viridis* and *R. dalmatina*, and, among reptiles, *E. orbicularis*, *T. hermanni*, *C. austriaca*, *N. tessellata*, *H. turcicus*, and *A. veronensis*. The assessment of species rarity can however be influenced by low detection rates, a common issue for herpetofauna that can greatly vary among species, and uneven sampling effort across grid cells, which could artificially increase rarity estimations (Dodd, 2009, 2016; Gomez-Rodriguez et al., 2012; McDiarmid et al., 2012). However, our conclusions on species distribution and rarity can be considered robust, as our dataset derives from numerous, repeated surveys carried out by different observers in different seasons. In this perspective, opportunistic and ancillary data (i.e. those from citizen science) can provide a significant contribution, especially to improve the knowledge on distribution and ecology of rare and cryptic species (Hauser and Heise-Pavlov, 2017; Marshall et al., 2023).

#### Conclusions and conservation notes

Based on the analyzed data presented here, the “Cilento, Vallo di Diano e Alburni” National Park emerges as a protected area with high herpetological value, ensuring high levels of diversity. Our findings agree with the ecological information available in Italy, while for some species, we identified some peculiar characteristics in their local ecology, divergent from the rest of their range, that confirm their plasticity and adaptation to local conditions. Species showing population and range declines at national scale exhibit a good conservation status inside park boundaries. However, during fieldwork, we noticed many road kills of all reptile species excluding geckos. This threat should be addressed with specific surveys and assessments inside the CVDA National Park.

Surveys repeated after a period of about 15 years on the same sites revealed dramatic habitat alterations for many amphibians. In historical sites, *B. variegata*, experienced local extinctions while *S. terdigitata* and *T. carnifex* are close to disappearing. Habitat degradation was

caused by two opposite factors: the abandonment of traditional agro-pastoral activities resulting in the lack of maintenance of artificial aquatic sites that lose permeability or are filled by vegetation (Fig. S4), and the collection of water from spring through pipelines, that drastically reduced water availability in synergy with climate change. In the CVDA National Park, the management of artificial aquatic sites is crucial for amphibian conservation (Romano, 2012), similarly to other parts of the Apennines (Temple and Cox, 2009; Romano et al., 2012; Buono et al., 2019). Finally, our study confirms the importance of incorporating the opportunistic data from citizen science into research and monitoring plans.

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#### SUPPLEMENTARY MATERIAL

Supplementary material associated with this article can be found at < <http://www.unipv.it/webshi/appendix> > Manuscript number 14562.

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