Vulnerability to Climate Change of Trichoptera species in Sierra Nevada and Pyrenees (Spain & Andorra)

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Máster de Biodiversidad 17/09/2020

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Journal: Hydrobiologia
Link to journal submission
guidelines:
https://www.springer.com/journal/10750/submission-guidelines#Instructions%20for%20Authors_General

Abstract

Global Climate Change (GCC) is expected to affect ecosystems worldwide. Freshwater ecosystems, such as lakes and streams, are among the ecosystems predicted to be greatly affected by GCC. Among them, the most endangered ones will be those located in high mountain areas and Mediterranean climate. Trichoptera is one of the most diverse freshwater insect orders and its ecosystemic role is of great importance. Furthermore, they are very useful to assess stream degradation. However, Trichoptera species are sensitive to changes in water temperature and as they have a high endemicity in high mountain areas and the Mediterranean basin in Europe, many species might be endangered by GCC. This study was focused in two high mountain ranges in the European Mediterranean Basin, Sierra Nevada and the Pyrenees. The aim of the study was to assess the vulnerability of Trichoptera species to GCC using biogeographic, ecological and biological traits. Overall, results showed a higher proportion of endangered species in high mountain areas than estimated in previous studies. Species with preference for spring zones and cold/very cold waters were the ones predicted to be more vulnerable to climate change. The most vulnerable species were located in the Pyrenees where the percentage of vulnerable taxa was much higher, especially in its central and eastern areas. Vulnerability studies using multiple traits could be useful to address species conservation measures in high mountain areas, and inform about what regions should be prioritized.

Keywords: Global Climate Change, Trichoptera, high mountains, freshwater ecosystems, species vulnerability, trait-based analysis

Introduction

Global Climate Change (GCC) is undoubtedly one of the main issues discussed in scientific papers in the last decades, particularly in those related to environmental sciences. This is because GCC is expected to become one of the major drivers of ecosystem change in the next decades (Bakun et al., 2015; García-Reyes et al., 2015; Hamann & Wang, 2006). Among the predicted and observed ecosystem effects of GCC, one of the most common and widely documented is biodiversity loss (Dawson et al., 2011; Maclean & Wilson, 2011; Sala et al., 2000; Urban, 2015). Species loss could pose a major threat to ecosystem functions and services, since these are strongly linked to biodiversity (Tilman et al., 2014). Besides extinction, species can also respond to GCC through local adaptation by microevolutionary processes and phenotypic plasticity, or through moving to more appropriated areas. For example, changes in environmental conditions due to GCC can modify the life cycle of species through advancing egg hatching or adult emergence, or increasing voltinism (Stange & Ayres, 2010; Stoks et al., 2014). In addition, GCC could also cause a shift in species distribution along the altitudinal gradient. As temperatures get warmer due to GCC, species outside of their thermal tolerance range are moving to higher altitudes searching for their thermal optimum (Comte & Grenouillet, 2013; Flousek et al., 2015; Inouye, 2020).

Freshwater ecosystems are thought to be greatly affected by GCC (Firth & Fisher, 2012). Stream water temperature has significantly increased during the last decades, and it is expected to continue increasing, while river runoff and flow will also be affected, with many river reaches shifting from perennial to intermittent (Isaak et al., 2012; Leith & Whitfield, 1998; Nelson & Palmer, 2007). Lakes will also suffer the effects of GCC in the form of higher water temperatures, longer stratification season, deeper thermocline, higher oxygen demand, and stronger upward flux of phosphorus from sediments (Komatsu et al., 2007).

High mountain streams and lakes will be specially affected by GCC. On the one hand, the hydrology of freshwater ecosystems in high mountain areas is influenced by precipitation, groundwater inputs, and water stored as snow and in glaciers (Milner et al., 2010). At the current rate of 0.025ºC increase per year, alpine glaciers, for example, are expected to lose between 30 and 70% of the volume they had in 1990 (Kaltenborn et al., 2010). This melting process would lead to an increase of the spring runoff and reduction of summer runoff in the short-term (Huss et al., 2008), and a long-term total discharge reduction (Jansson, Hock & Schneider, 2003). On the other hand, GCC is also expected to affect precipitation regimes worldwide, including high mountain regions (Bravo et al., 2008; Dore, 2005; Gobiet et al., 2014; Urrutia & Vuille, 2009). High mountain lakes, for example, are expected to suffer from stronger water level fluctuations, reduced nutrient absorption, changes in vegetal community composition and extended stratification periods due to GCC (De Vicente et al., 2010; Michelutti et al., 2016; Toro et al., 2006). All this could have a profound impact in the hydrology and ecology of high mountain lakes but also of streams, and therefore, their invertebrate communities, leading many species towards extinction (Giersch et al., 2017; Labaj et al., 2017; Muhlfeld et al., 2011). For example, De Figueroa et al. (2010) found that twenty-one taxa of Plecoptera in southern Europe are particularly threatened with extinction by GCC.

Although the biodiversity loss may be the most obvious effect of GCC on the macroinvertebrate communities in high mountain streams, there are also other equally important effects. Observed upstream movement of species (Sáinz-Bariáin et al., 2016) could lead to a shortening of the altitudinal range inhabited by a species, since some species that are driven out of their tolerance ranges in their lowest end might be unable to colonize further upstream (Giersch et al., 2015; Muhlfeld et al., 2011; Woodward et al., 2010). Furthermore, this altitudinal shift could have another side-effect, the change in community composition. This change could be driven by a turnover between species, as downstream species replace those that disappear or migrate upstream, or by just the addition of new, upstream-moving, species from lower reaches (Finn et al., 2010; Jourdan et al., 2018; Sáinz-Bariáin et al., 2016).

In freshwater ecosystems, as in many other ecosystems, insects are among the major regulators of ecosystem processes that sustain the delivery of services (Schowalter, 2013). They act as consumers at intermediate trophic levels (i.e. grazers, shredders, gatherers, filterers, and predators) and are essential to the food web and ecosystem functions (nutrient cycles, primary productivity, decomposition, and translocation of materials). They are also influenced by both bottom-up and top-down forces and serve as valuable indicators of stream degradation (Wallace & Webster, 1996). For this last purpose, Trichoptera are specially interesting, due to their sensitivity to environmental change, their high species richness and wide ecological and biological trait diversity.

Trichoptera, along with Diptera, is the most species-rich order among primary aquatic insects (Holzenthal et al., 2007). They probably have a high degree of local endemism in high-mountain environments (Holzenthal et al., 2015). In general, Trichoptera are tightly bound to freshwater ecosystems since, although adults are terrestrial, their larval stages usually develop in freshwater. Thus, their reproduction is influenced by the water conditions around them. They are opportunistic feeders that are able to use a wide range of resources: diatoms, filamentous algae, vascular plants, dead organic materials and living animals (Mackay & Wiggins, 1979). Because of their diversity and density in most clean, freshwater ecosystems, the significance of Trichoptera for processing nutrients and transferring energy is often significant (Morse, 2009). However, Trichoptera are sensitive to temperature along elevation gradients and GCC may affect their body size and other related parameters such as survival and fitness (Cogo et al., 2020). They might also be specially sensitive to GCC as many endemisms show great specialization to cold habitats (Holzenthal et al., 2015). Furthermore, the lateral dispersal ability (i.e. the capacity to migrate to another basin) of Trichoptera seems to be quite limited, as most species usually travel less than 200m (Petersen et al., 1999; Sode & Wiberg-Larsen, 1993). This makes them significantly vulnerable to the effects of GCC.

The Mediterranean is one of the regions predicted to be strongly affected by GCC, with reduced rain and snow during winter and higher temperatures and more frequent droughts in summer (Giorgi & Lionello, 2008). High mountain streams in this area might be important in terms of Trichoptera endemisms, since this region is inhabited by almost a 40% of the caddisfly species present in the Palearctic region (De Figueroa et al., 2012). Changes in hydrology and climate in the Mediterranean could mean the extinction of species unable to adapt to the new conditions or lacking the ability to disperse and colonize new areas, following their thermal range as it shifts northwards (Hering et al., 2009). Moreover, Trichoptera species in the Iberic-Macaronesian region are thought to be especially vulnerable to GCC (Hering et al., 2009). This could pose a threat, not only to their local species diversity, but also to the genetic diversity of these species (Finn et al., 2013).

In terms of biodiversity conservation, vulnerability to GCC is defined by the IPCC (Intergovernmental Panel on Climate Change) as “the degree to which a system is susceptible to, or unable to cope with, adverse effects of climate change, including climate variability and extremes” (Fuessel, 2005). Vulnerability assessments are important to minimize biodiversity loss, since conservationists need to identify those species that are likely to be most vulnerable to the impacts of GCC. The Trait-based Vulnerability Assessment (TVA) is an approach that uses biological traits to assess species vulnerability. It is easier to use than other approaches and allows a relatively rapid assessment for multiple species that can be used to prioritize conservation planning and implementation of adaptive measures (Pacifici et al., 2015). TVA has already been used to assess the vulnerability to GCC of birds, plants, amphibians, reptiles, corals, and insects, among others (Anacker et al., 2013; Böhm et al., 2016; Foden et al., 2013; Sandin et al., 2014).

The aim of this study was to assess the level of vulnerability to GCC of high-mountain Trichoptera species of the Pyrenean and Sierra Nevada mountain ranges, and to identify areas of high vulnerability to GCC for future conservation purposes.

Materials and methods

Study area

The study area covered two high mountain ranges in the Iberian Peninsula, the Pyrenees in the north and Sierra Nevada in the south. The altitude ranged from 300 to 3404 m.a.s.l. in the Pyrenees and from 700 to 3478 m.a.s.l. in Sierra Nevada. In the Pyrenees there are still ten glaciers an around fifty glacial-masses, while in Sierra Nevada only a shrinking remnant of what once was a glacier remains (Gómez Ortiz et al., 2014). Both mountain ranges are important in terms of protected natural areas. In the Pyrenees there are 24 National Parks/Natural Parks in the Andorran and Spanish side only, while, Sierra Nevada is a National and Natural Park.

For defining the Pyrenees area, the boundaries drawn by the *Observatorio Pirenaico del Cambio Climático* (OPCC, Pyrenean Climate Change Observatory) (2018) were used. According to literature, Pyrenees could be divided in three subareas western (Western Pyrenees), central (Central Pyrenees) and eastern Pyrenees (Eastern Pyrenees) (Mattauer & Henry, 1974; OPCC, 2018). Only Spanish and Andorran Pyrenees were considered in this study (Fig. 1), which extends over more than 25000km^2. For defining the Sierra Nevada area, the boundaries of the Sierra Nevada Natural Park were used, which has an 862.08 km² extension. The area was divided in two subareas, north and south (North and South Sierra Nevada, respectively), regarding its two slopes (Sainz-Bariain et al., 2016) (Fig. 1).

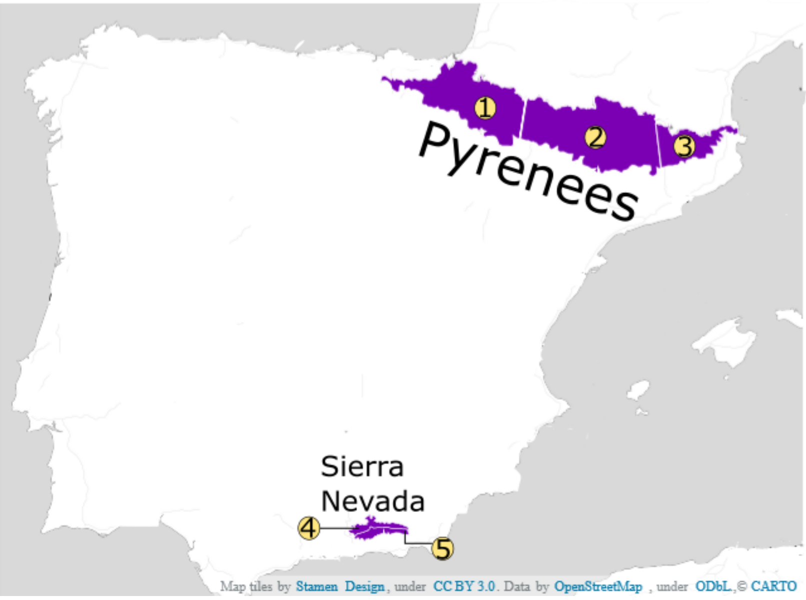


Fig. 1: Map of the two studied areas, highlighted in purple; the Pyrenees in the north and Sierra Nevada in the south. Each mountain range is divided in several subareas: Western Pyrenees (1), Central Pyrenees (2) and Eastern Pyrenees (3), and North Sierra Nevada (4) and South Sierra Nevada (5).

Data description

Records of Trichoptera species present in each high mountain range were obtained from the database stored in www.geobrink.uclm.es. This database contains all the knowledge on Trichoptera from the Iberian Peninsula gathered from 1905 to 2017. It includes a total of 13961 records, of which 1687 records belong for the study area: 1331 for the Pyrenees and 356 for Sierra Nevada.

Vulnerability traits description and vulnerability assessment

Vulnerability traits description

Twenty traits from previous works (Abellan et al., 2005; De Figueroa, 2010; Domisch et al., 2010; Graf et al., 2008; Hering et al., 2009; Hershkovitz et al., 2015; Sarremejane et al., 2020) were evaluated together for the Trait-based Vulnerability Assessment. Fourteen traits were finally selected by expert criteria due to their potential for assessing vulnerability to GCC. The traits not considered were: “pH preference”, “Current preference”, “Hydrologic preference”, “Respiration”, “Habitat rarity” and “Habitat loss”. These fourteen traits were grouped into biogeographic, ecological and biological traits. Each trait was divided in different categories, which were coded as presence/absence, single category assignment, or by a scoring system (10-point system) similar to the fuzzy-coding approach (Chevenet et al., 1994). The score reflected the affinity of the taxon with a particular category of each trait, where ‘0’ indicates no affinity and ‘10’ high affinity (Table 1). Information for each trait and Trichoptera species present in Sierra Nevada and the Pyrenees were extracted from www.freshwaterecology.info and www.geobrink.uclm.es databases (Table 1).

When trait information was not available for a species, it was completed using expert criteria, with the information from the taxonomically closest species, or from all species belonging to the same genus.

Biogeographic traits

1. Distribution: The distribution area is a key factor of extinction vulnerability. Species with a smaller distribution area are more sensitive to extinction since they usually are less dispersive, more specialised and inhabit in rarer climates (Korkeamäki & Suhonen, 2002; Malcolm et al., 2006; Ohlemüller et al., 2008). Moreover, species with small ranges are typically rare within their ranges in comparison to more widely distributed species, making them even more vulnerable (Pimm, & Raven, 2000). The categories used were the next five:

- Transiberic: Species present in Europe north of the Pyrenees, the Iberian Peninsula and North Africa.
- Iberoeuropean: Species present in Europe north of the Pyrenees and some regions of the Iberian Peninsula but not in North Africa.
- Iberoafrican: Species present in North Africa and some regions of the Iberian Peninsula but not in Europe north of the Pyrenees.
- Discrete: Species present in some regions of the Iberian Peninsula and central and eastern Mediterranean region, but not in Europe north of the Pyrenees.
- Endemic: Species endemic to the Iberian Peninsula, including the northern mountainside of the Pyrenees, and the Balearic Islands in some cases.

2. Endemicity: Endemicity is a vulnerability factor closely related to distribution. The more restricted the geographic range of a species, the more vulnerable to changes. In this case, only the species that are endemic to the Iberian Peninsula were considered, the rest were given the minimum scoring. The categories used were the next four:

- Not strict: Species that inhabit both mountain ranges, Sierra Nevada and the Pyrenees.
- General: Species endemic to one mountain range, Sierra Nevada or the Pyrenees, but are widely distributed through all its area.
- Restricted: Species endemic to one mountain range but that even if widely distributed are not present in all subareas. Only applied to the Pyrenees, since Sierra Nevada was divided in two subareas only.
- Exclusive: Species endemic one mountain range, Sierra Nevada or the Pyrenees, and are restricted to one subarea.

Ecological traits

3. Stream zonation preference: Species inhabiting different stream zones have different ecological requirements. Species inhabiting spring zones are adapted to cold and fast waters, while species that live in the lower reaches are adapted to slower and warmer water. However, species inhabiting the upper reaches of streams will be more vulnerable to GCC since they will be unable to move to higher altitudes, as it has been observed in crayfish species (Bland, 2017).

4. Temperature preference: Species adapted to cold temperatures (cold stenothermic) are more vulnerable to GCC since the temperature increase will force them to migrate upstream or reduce their distribution range. That could pose a notable threat for species that have "nowhere to go" (Domisch et al., 2011).

5. Persistence: The persistence level refers to the continuity of populations in the study area. The species that have not been addressed in a while would be more vulnerable than the ones that are still regularly captured.

6. Microhabitat specialist: Most Trichoptera species have a preference for one or more substrates during their larval stage (Graf et al., 2008). In some species certain substrate types are critical to their larval or pupal development. Thus, species that depend heavily on a small number of substrate types, microhabitat specialists, could be more vulnerable than more generalist ones due to their narrow microhabitat requirements. Thirteen substrate categories were considered after Graf et al. (2008), for which every species had an affinity-score based in a 10-point system. For a species to be considered in one feeding category it had to score a minimum of 10% affinity to that substrate (i.e. 1 out of 10). The substrates considered were the following:

- Pelal: Mud. Grain size < 0.063 mm.
- Argyllal: Silt, loam, clay. Grain size < 0.063 mm.
- Psammal: Sand. Grain size 0.063-2 mm.
- Akal: Fine to medium-sized gravel. Grain size 0.2-2 cm
- Micro-/mesolithal: Coarse gravel to hand-sized cobbles. Grain size 2-20 cm
- Macro-/megalithal: Stones, boulders, bedrock. Grain size > 20 cm
- Hygropertric habitats: Thin layers of water over bedrocks, waterfalls.
- Algae: Micro- and macroalgae.
- Macrophytes: Macrophytes, mosses, Characeae, living parts of terrestrial plants
- POM: Coarse and fine particulate organic matter.
- Woody debris (xylal): Woody debris, twigs, roots, logs. Size > 10 cm
- Madicol habitats: Edge of water bodies, moist substrates.

- Other habitats: e.g. host of a parasite.

Altitude preference: High-altitude areas are expected to suffer the most the effects of GCC (Ohmura, 2012). Furthermore, high-altitude endemic species could face a disproportionate extinction risk due to GCC-induced habitat loss (Dirnböck et al., 2011).

Biological traits

7. Reproductive cycle: The reproductive-rate of a species could be key for the adaptability of a species to GCC since the more generations, the faster the natural selection can act. Thus, species with a higher voltinism could be more adaptable than species with lower reproductive-rate since the last ones are dependent on more stable environments. This has already been observed in butterflies (Altermatt, 2010; Soga & Koike, 2012).

8. Species lifespan: Species with long life-cycles are potentially less adaptable and less resilient to changes since they usually reproduce with less frequency and have less offspring (Laurence et al., 2010; Poff et al., 2010).

9. Female maximum wing length: Species that have a greater ability to disperse could keep up with the different GCC scenarios by migrating to more suitable areas. Thus, less dispersive species would be more vulnerable to extinction (Nadeau & Fuller, 2016). Females are the main actors of dispersal through oviposition and dispersal capacity of Trichoptera species has been shown to be positively correlated with their wing length (Hoffsten, 2004).

10. Emergence season: The timing of emergence reflects the requirements of both, the aquatic and terrestrial life stages. GCC may affect the timing of insect emergence (Harper & Peckarsky, 2006). This could make especially vulnerable those species with summer emergence, since it is the season on which GCC is predicted to have a greater impact in the Mediterranean (Giorgi & Lionello, 2008). Spring, the main wet season, could also be considerably harmed as well as the poorly surveyed autumn (Gallinat et al., 2015).

11. Emergence period length: Species with a long emergence period are less prone to environmental perturbations because risks of droughts or spates are minimised. A short emergence period enhances the likelihood of finding mating partners but synchronised development of aquatic stages makes species more vulnerable (Graf et al., 2008; Kotiaho et al., 2005).

12. Drought resistance: The ability of a species to cope with droughts is important to evaluate its vulnerability since it is an adaptation to survive environmental changes. For our study it is especially relevant since more frequent droughts are expected in the Mediterranean area due to GCC (Giorgi & Lionello, 2008).

13. Feeding specialist: Species that are feeding specialists are more vulnerable to GCC than more generalist species, since the first ones are limited to a narrower food resource (Nadeau & Fuller, 2016). Nine feeding strategies were defined after Graf et al. (2008), for which every species had an affinity-score based in a 10-point system. The parasite category was excluded since no parasite species of Trichoptera is known in Europe. For a species to be considered in one feeding category it had to score a minimum of 20% affinity to that strategy (i.e. 2 out of 10). The feeding strategies considered were the following:

- Grazer/scraper
- Miner
- Xylophagous
- Shredder
- Gatherer/collector
- Active filter feeder
- Passive filter feeder
- Predators
- Other feeding type

Vulnerability assessment

Depending on the category of a trait to which a species belonged, a score for each species was given (Table1). The different categories in each trait were given a whole numeric value from ‘0’ to ‘3’ depending on their relevance for the vulnerability to GCC. ‘0’ was given for low vulnerability, ‘1’ for medium vulnerability, ‘2’ for high vulnerability and ‘3’ for very high vulnerability. In the case of traits coded with a 10-point system the category with the highest affinity was selected (i.e. the category with a 5< score). In cases where there was not a clear option in a category, the most conservative (the lowest score) option was chosen. The sum of the scores obtained by a species in all the different categories was defined as the total vulnerability score of that species, and had a potential maximum of 39 points. The traits for which data was not available and could not be obtained were given a value of ‘0’ for the sum of scores.

Habitat loss was also considered as a vulnerability factor, since species suffering strong habitat destruction are more vulnerable to extinction (Abellan et al., 2015). However, since the study area is focused in protected natural areas it was excluded from the study.

Table 1: Description of the categories and scoring of each trait assessed in the study. (pts = points)

TRAITS	CATEGORIES & SCORING	FROM OR MODIFIED FROM
DISTRIBUTION	▪ Transiberic -- 0pts ▪ Iberoafrican -- 1pts ▪ Discrete -- 2pts ▪ Endemic -- 3pts	Abellán et al. (2005); Sánchez-Fernández et al. (2004)
ENDEMICITY	▪ Not strict -- 0pts ▪ General -- 1pts ▪ Restricted -- 2pts ▪ Exclusive -- 3pts	Abellán et al. (2005); Sánchez-Fernández et al. (2004)
STREAM ZONATION PREFERENCE	▪ Hyperliterally (grayling region) -- 0pts ▪ Epicortical (barbel region) -- 0pts ▪ Metapotamal (bream region) -- 0pts ▪ Hypopotamal (brackish water) -- 0pts ▪ Littoral (lake and stream, shorelines, ponds, etc.) -- 0pts ▪ Profundal (bottom of stratified lakes) -- 0pts ▪ Metarhithral (lower trout region) -- 1pt ▪ Epirhithral (upper trout region) -- 2pts ▪ Eucrenal (spring region) -- 3pts ▪ Hypocrenal (spring brook) -- 3pts	Hering et al. (2009); Hershkovitz et al. (2015); Tierno de Figueroa et al. (2010)
TEMPERATURE PREFERENCE	▪ Warm (>18Cº) or eurytherm -- 0pt ▪ Moderate 10 -18Cº -- 1pt ▪ Cold 6 - 10Cº -- 2pts ▪ Very cold < 6Cº -- 3pts	Domisch et al. (2011); Graf et al. (2008); Tierno de Figueroa et al. (2010); Hering et al. (2009)
PERSISTENCE	▪ Last capture between 3 and 8 years ago -- 0pts ▪ Last capture between 9 and 14 years ago -- 1pt ▪ Last capture between 15 and 20 years ago -- 2pts ▪ Last capture between 21 years ago or before – 3pts	Abellán et al. (2005); Sánchez-Fernández et al. (2004)
MICROHABITAT SPECIALIST	▪ > 7 Microhabitats -- 0pts ▪ 5-6 Microhabitats -- 1pt ▪ 3-4 Microhabitats -- 2pts ▪ 1-2 Microhabitats -- 3pts	
ALTITUDE PREFERENCE	▪ Montane or lower (< 1900m) -- 0pts ▪ Subalpine (1900 - 2400m) -- 1pt ▪ Alpine (2400 - 2900m) -- 2pts ▪ Nival-subnival (> 2900m) -- 3pts	Graf et al. (2008)
REPRODUCTIVE CYCLE	▪ Multivoltine -- 0pt ▪ Bivoltine -- 1pt ▪ Univoltine -- 2pts ▪ Semivoltine -- 3pts	Graf et al. (2008); Hershkovitz et al. (2015)
SPECIES LIFESPAN	▪ < 1year -- 0pts ▪ ≥ 1year -- 3pts	Hering et al. (2009)

MAX. FEMALE WING LENGTH	▪ ≥15mm -- 0pts ▪ ≥10-15mm -- 1pt ▪ ≥5-10mm -- 2pts ▪ <5mm -- 3pts	Sarremejane et al. (2020)
EMERGENCE SEASON	▪ Emergence all year -- 0pts ▪ Mainly winter -- 1pt ▪ Mainly spring/autumn -- 2pts ▪ Mainly summer -- 3 pts	Graf et al. (2008)
EMERGENCE PERIOD LENGTH	▪ Long: ≥ 2 months -- 0pts ▪ Short: < 2 months -- 3pts	Graf et al. (2008)
DROUGHT RESISTANCE	▪ Adult diapause – 0pts ▪ Aquatic diapause – 2pts ▪ No drought resistance -- 3pts	Graf et al. (2008)
FEEDING SPECIALIST	▪ ≥ 4 Feeding strategies -- 0pts ▪ 3 Feeding strategies -- 1pt ▪ 2 Feeding strategies -- 2pts ▪ 1 Feeding strategy -- 3pts	Hering et al. (2009)

Data analysis

Biodiversity databases are usually built with records provided by many different authors. This information is useful to characterize the geographic distribution of species but three types of biases can occur: the lack of reliable absence information on the real distribution of species, the unknown survey effort, and the unknown recurrence of species in sampling sites (Lobo et al., 2018). Completeness refers to the percentage of species found in a surveyed area in relation to the total number of species estimated by a species accumulation curve for the same area. It has a direct and positive relation with the sampling effort. In order to know how reliable our data was to accomplish our objective; completeness was calculated using the KnowBR software and following the approach proposed by Lobo et al. (2018). A square cell resolution of two minutes (10.84 km^2) was used to carry out the calculations, since Trichoptera species usually do not disperse through an area larger than that.

To graphically compare the vulnerability of species of both mountain ranges, a species vulnerability curve was made. Species were sorted from most to least vulnerable in the x-axis and their total vulnerability (i.e. the sum of the scores of all traits) was drawn on the y-axis. As the sample of the two mountain ranges was very different in size (147 in the Pyrenees and 37 in Sierra Nevada), samples were randomized. For that purpose, 37-species long samples were randomly taken 1000 times from the Pyrenees species pool. Then, samples were sorted from most to least vulnerable and the mean value for each of the 37 positions was calculated. This new 37-species sample was used to compare with the Sierra Nevada sample.

A linear regression was performed to test if there was any relation between family species number in the surveyed area and mean total vulnerability score of species in a family.

To analyse the most relevant vulnerability traits for Trichoptera species, a Principal Component Analysis (PCA) was performed using the R software (version 3.5.2) (R Core Team, 2018). The species that appeared in less than 5 samplings were excluded from the analysis. “Drought resistance” trait was also excluded from data analysis due to excessive lack of information about species. Data matrix for the PCA consisted of 13 vulnerability traits and 162 Trichoptera species. The scores for each vulnerability factor of species were used in this matrix. To deal with the missing values for some vulnerability traits in many species in the matrix, the Ipca method was used to consider missing values of cells, since it is the method that outperforms the rest in most situations (Dray & Josse, 2015). To know the most important traits for vulnerability, the total vulnerability score of each species was correlated with its projection in the X and Y axes of the PCA.

To calculate the area and subareas with higher vulnerability, the percentage of highly (total vulnerability score 20-29pts) or very highly (total vulnerability score 30-39pts) endangered species of each area and subarea was calculated.

Results

General results

In total, out of the 469 caddisfly species that inhabit the Iberian Peninsula, 162 species were found in the study area, of which 147 inhabited the Pyrenees and 37 Sierra Nevada. Both mountain ranges had 22 Trichoptera species in common. The number of species found in each subarea was the next: 23 species in South Sierra Nevada, 37 in North Sierra Nevada, 66 in Western Pyrenees, 93 in Central Pyrenees and 79 in Eastern Pyrenees. Out of the 14 vulnerability traits studied, four contained information for all 162 species in both mountain ranges (Table 2). The percentage of encoded taxa by mountain range was slightly higher in Sierra Nevada (90.59%) than in the Pyrenees (89.86%), but differences were small. The trait with the lowest percentage of the total taxa encoded was “Drought resistance” (28%).

Table 2: Percentages of taxa encoded for each trait in total and by mountain range.

Trait	% Total taxa encoded	% Sierra Nevada taxa encoded	% Pyrenean taxa encoded
Distribution	100	100	100
Endemicity	100	100	100
Stream zonation preference	100	100	100
Temperature preference	89	84	88
Persistence	100	100	100
Microhabitat specialist	98	100	97
Altitude preference	100	100	100
Reproductive cycle	84	84	84
Species lifespan	69	59	70
Max. female wing length	100	100	100
Emergence season	99	100	99
Emergence period length	99	100	99
Drought resistance	28	41	25
Feeding specialist	96	100	96

Completeness

In terms of data completeness, the two mountain ranges were very different. On the one hand, in the Pyrenees, sampling effort was very biased. Samplings and sampling sites were quite abundant in Eastern and Central Pyrenees, but very scarce in Western Pyrenees (Fig. 2A). However, 53% of the surveyed areas fell in the moderate-well surveyed category (i.e. >50 completeness; (Fig. 2C). On the other hand, in Sierra Nevada, sampled points were quite homogeneously distributed across the whole mountain range and blank spaces were scarce (Fig. 2B). Most of the surveyed areas (61%) in the mountain range had above 41% of data completeness and 33% of the total fell in the moderate-well surveyed category (Fig. 2D).

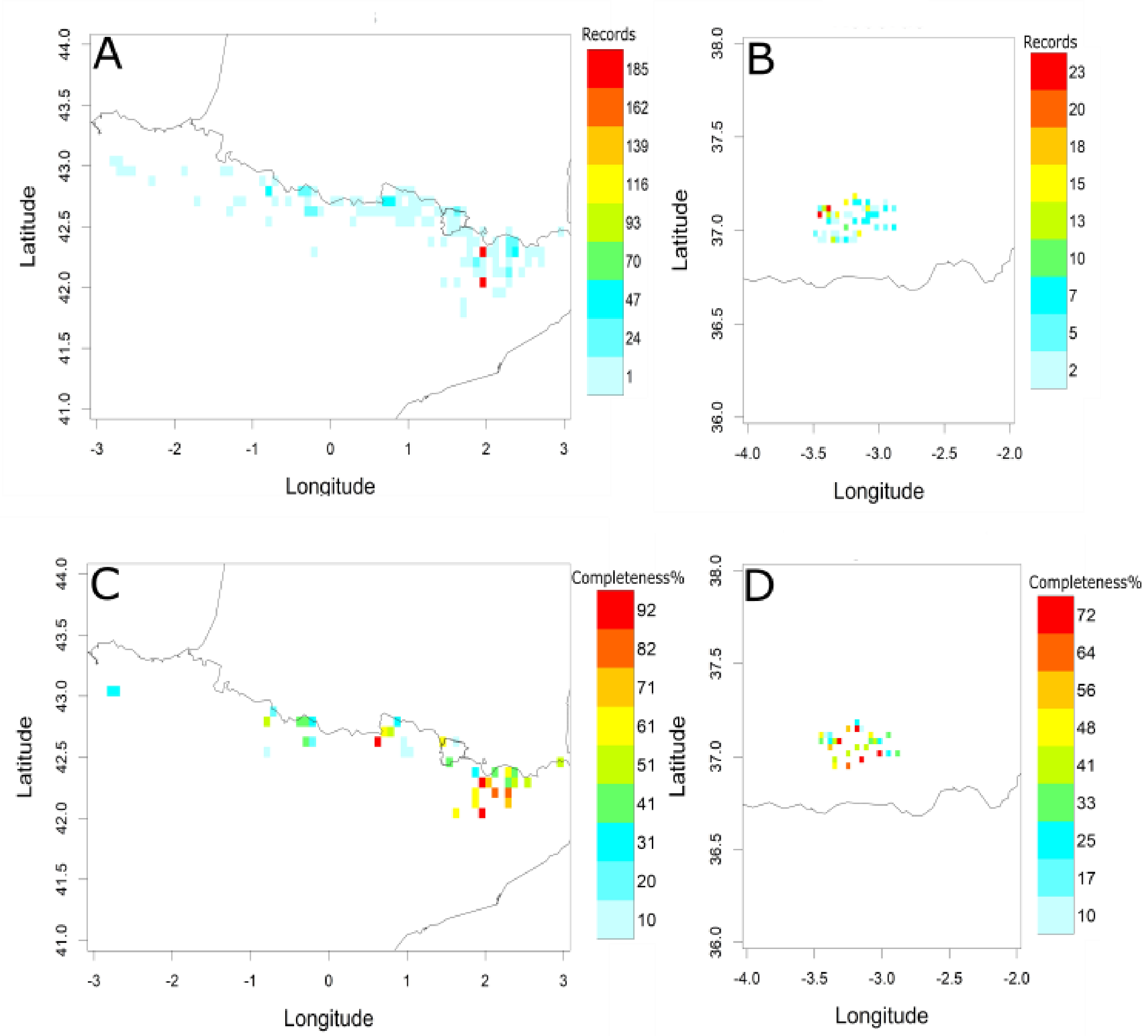


Fig. 2: Maps of the records (A and B) and completeness (C and D) of the sampling areas in the Pyrenees (A and C) and Sierra Nevada (B and D).

Species and family vulnerability

Species vulnerability

Overall, the most vulnerable species was *Rhyacophila vandeli*, with 30 points, and the least vulnerable one *Mesophylax aspersus*, with 9 points (Table 3). In the Pyrenees, the most and least vulnerable species were the same two, respectively. However, in Sierra Nevada, though the least vulnerable species was still *Mesophylax aspersus*, the most vulnerable species was *Wormaldia granada* (25 points). The most vulnerable species were restricted to the Pyrenees. However, among the ten least vulnerable, five were restricted to the Pyrenees, two to Sierra Nevada and three appeared in both mountain ranges. The species vulnerability curve showed that overall, the vulnerability of Pyrenean species was slightly higher than the vulnerability of the species of Sierra Nevada. However, the profile of the vulnerability curve was very similar in both mountain ranges (Fig. 3).

Table 3: Table showing the ten most and least vulnerable species in the study and their total vulnerability scores.

10 most vulnerable species	Total vulnerability score	10 least vulnerable species	Total vulnerability score
Rhyacophila vandeli	30	*Polycentropus flavomaculatus*	13
Apatania stylata	28	*Potamophylax latipennis*	13
Rhyacophila rupta	28	*Cheumatopsyche lepida*	12
Rhyacophila praemorsa	27	*Hydropsyche incognita*	12
Rhyacophila sicorensis	27	*Hydropsyche siltalai*	12
Adicella filicornis	26	*Stenophylax crossotus*	12
Drusus marinettae	26	*Hydroptila Vectis*	11
Rhyacophila eatoni	26	*Schizopelex furcifera*	11
Rhyacophila laevis	26	*Hydropsyche dinarica*	10
Rhyacophila philopotamoides	26	*Mesophylax aspersus*	9

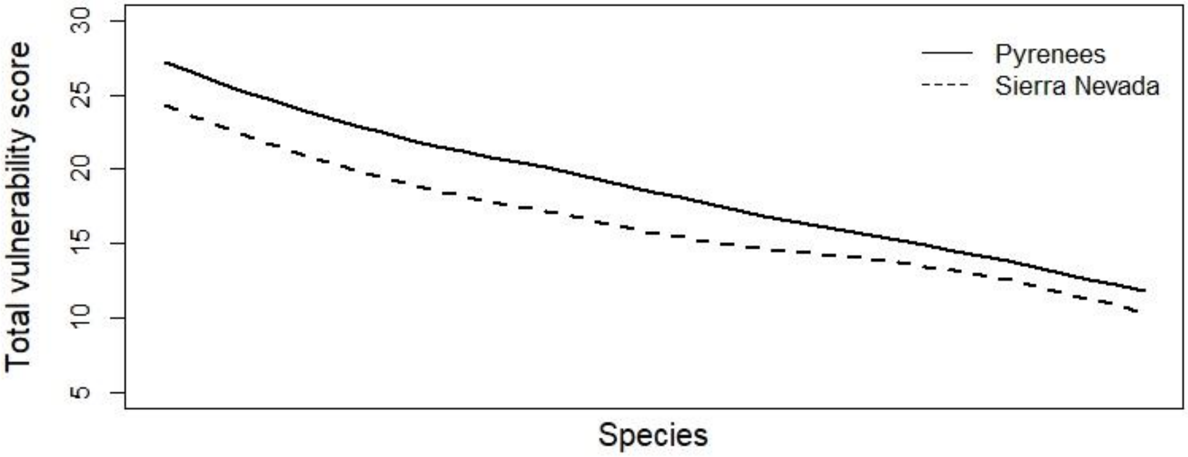


Fig. 3: Vulnerability curve of each mountain range. Species are sorted from most to least vulnerable in the x axis. See methods for a detailed description of the randomization performed.

Family vulnerability

The mean vulnerability of families was 19.03. The most vulnerable family was Apataniidae, with a score of 28, and the least vulnerable one Ecnomidae, with a score of 14.5 (Fig. 4A). When considering only the families with 3 or more species in our study area, the mean vulnerability of families was 18.51. The most vulnerable family was Glossosomatidae, with a score of 23.51, and the least vulnerable one Lepidostomatidae, with a score of 15.67. The linear regression showed no evidence of significant correlation between family species number and mean total vulnerability of species in a family ($p = 0.371$). When analysing each mountain range, the most vulnerable family in the Pyrenees was Apataniidae, with a score of 28, and the least vulnerable ones Phryganidae and Odontoceridae, with a score of 15 each (Fig. 4B). However, the most vulnerable family in Sierra Nevada was Philopotamidae, with a score of 18.67, and the least vulnerable one Polycentropodidae, with a score of 13 (Fig. 4C).

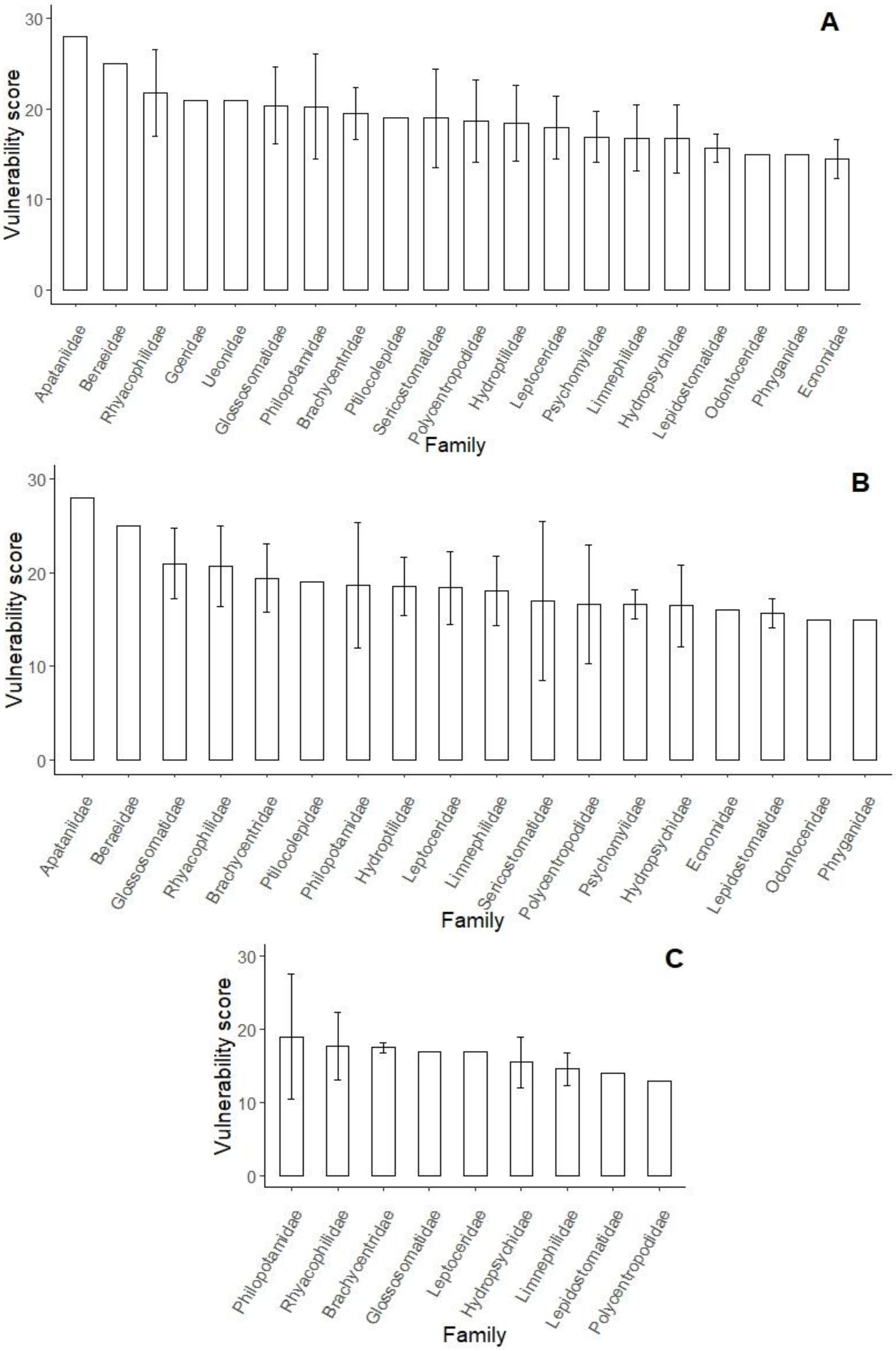


Fig. 4: Mean vulnerability-score and standard deviation for each family in total (A), the Pyrennes (B) and Sierra Nevada (C).

Vulnerability traits

When analysing the vulnerability scores by trait, the trait with higher percentage of species with very high vulnerability (i.e. a vulnerability score of '3') was "Emergence season" (66%), followed by "Microhabitat specialist" (54%) and "Endemicity" (51%), and the one with the lowest percentage the "Altitude preference" (0%). In contrast, the trait with higher percentage of species with low vulnerability (i.e. a vulnerability score of '0') was by far "Altitude preference" (96%) and the one with the lowest percentage the "Microhabitat specialist" (0%), followed by "Persistence" (1%) and "Reproductive cycle" (3%) (Appendix 1).

When analysing the vulnerability scores by trait and mountain region, the trait with higher percentage of species with very high vulnerability was "Emergence season" (70% in the Pyrenees and 54% in Sierra Nevada), followed by "Microhabitat specialist" (55% in the Pyrenees and 51% in Sierra Nevada), in both mountain regions. However, while in the Pyrenees "Endemicity" had a relevant percentage of species with very high vulnerability too (50%), in Sierra Nevada its relevance was much lower (24%). In contrast, the trait with higher percentage of species with low vulnerability was by far "Altitude preference" (97% in the Pyrenees and 97% in Sierra Nevada), followed at some distance by "Species lifespan" (66% in the Pyrenees and 59% in Sierra Nevada) and "Emergence period length" (53% in the Pyrenees and 51% in Sierra Nevada), in both mountain ranges. However, in Sierra Nevada taxa with low "Endemicity" vulnerability was abundant too (59%). The trait with the lowest percentage of low-vulnerability species was "Microhabitat specialist" (0% in the Pyrenees and 0% in Sierra Nevada) in both mountain ranges.

Regarding the PCA, the sum of the two first axes explained a 33.6% of the variance. The first axis was strongly related to "Stream zonation preference" (0.79) and "Temperature preference" (0.80), thus, to the altitudinal gradient. The second axis was strongly related to "Endemicity" (0.68) and "Persistence" (0.6) (Table 4). "Stream zonation preference" and "Temperature preference" ecological traits provided very similar information. "Emergence season", "Species lifespan" and "Reproductive cycle" ecological traits provided very different information. Biogeographical traits were mixed with the other trait variables. Species were quite homogeneously scattered along the vulnerability-traits axes, with no apparent pattern or grouping (Fig. 5). Total vulnerability score showed a significant correlation with the first ($p < 0.001$) and second ($p = 0.015$) dimensional axes, however the correlation value was notably higher for the first (0.75) than for the second axis (0.23).

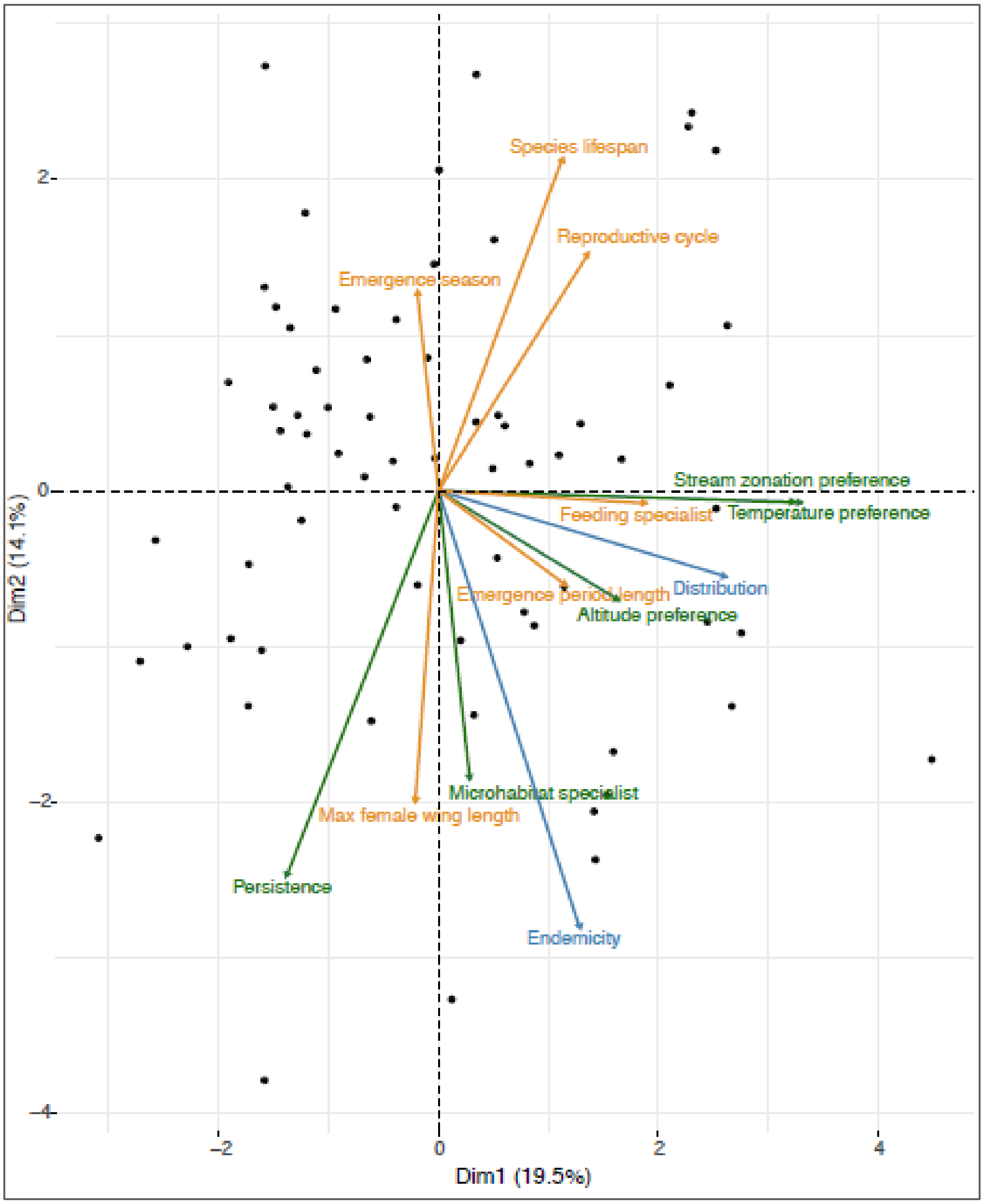


Fig. 5: PCA biplot showing the position of species (black points) in relation to the variables (arrows). The variables are divided in three categories: biogeographical traits (blue), ecological traits (green) and biological traits (orange).

Table 4: Correlation of the different trait variables with each dimension of the PCA

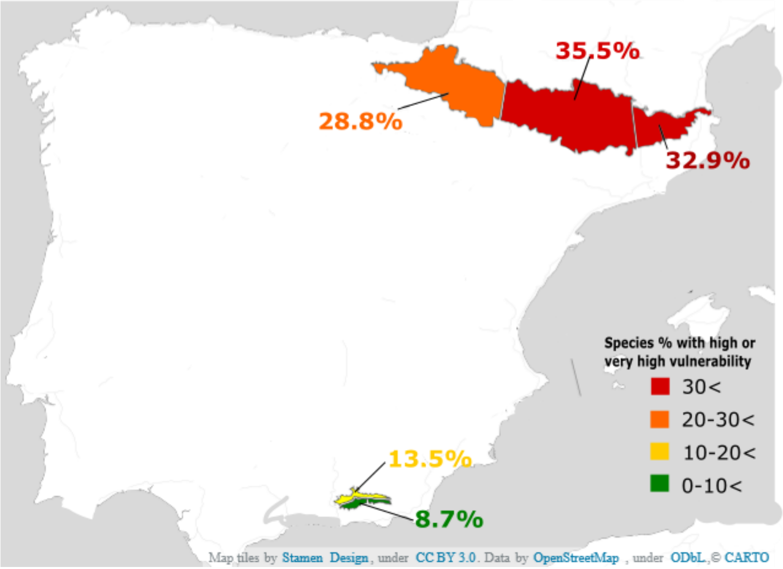
Trait	Dim.1	Dim.2
Distribution	0.633	-0.133
Endemicity	0.311	-0.681
Stream zonation preference	0.791	-0.017
Temperature preference	0.803	-0.018
Persistence	-0.335	-0.600
Microhabitat specialist	0.069	-0.451
Altitude preference	0.399	-0.170
Reproductive cycle	0.330	0.369
Species lifespan	0.273	0.517
Max. female wing length	-0.051	-0.486
Emergence season	-0.046	0.310
Emergence period length	0.283	-0.147
Feeding specialist	0.460	-0.019

Region vulnerability

Overall, the percentage of highly and very highly vulnerable species (i.e. species with a total vulnerability score between 20 and 39pts) was of 38.8% in the Pyrenees and of 13.5% in Sierra Nevada, and ranged from 8.7% to 35.5% each of the five subareas. In general, the Pyrenean subareas were more vulnerable than those in Sierra Nevada. The most vulnerable subarea was Central Pyrenees an the least vulnerable Southern Sierra Nevada (Fig. 6). In South Sierra Nevada the most vulnerable species was *Rhyacophila nevada* (23pts.) and in North Sierra Nevada *Wormaldia granada* (25pts.). The most vulnerable species in Western Pyrenees was *Rhyacophila eatoni*, *Apatania stylata* and *Rhyacophila rupta* in Central Pyrenees, and *Rhyacophila vandeli* in Eastern Pyrenees (Table 5).

Table 5: The five most vulnerable species of each subarea with their total vulnerability score between brackets. (pts = points)

South Sierra Nevada	North Sierra Nevada	Western Pyrenees	Central Pyrenees	Eeastern Pyrenees
Rhyacophila nevada (23pts)	*Wormaldia granada* (25pts)	*Rhyacophila eatoni* (26pts)	*Apatania stylata* (28pts)	*Rhyacophila vandeli* (30pts)
Sericostoma vittatum (21pts)	*Wormaldia occipitalis* (24pts)	*Wormaldia triangulifera* (24pts)	*Rhyacophila rupta* (28pts)	*Adicella filicornis* (26pts)
Annitella iglesiasi (20pts)	*Rhyacophila nevada* (23pts)	*Drusus bolivari* (23pts)	*Rhyacophila praemorsa* (27pts)	*Drusus marinettae* (26pts)
Hydropsyche tibialis (19pts)	*Sericostoma vittatum* (21pts)	*Polycentropus telifer* (23pts)	*Rhyacophila sicorensis* (27pts)	*Rhyacophila laevis* (26pts)
Hydropsyche infernalis (10pts)	*Annitella iglesiasi* (20pts)	*Psychomyia ctenophora* (23pts)	*Rhyacophila laevis* (26pts)	*Beraea maurus* (25pts)



Map tiles by Stamen Design, under CC BY 3.0. Data by OpenStreetMap, under ODbL, © CARTO

Fig. 6: Map showing the vulnerability (% of species with high or very high total vulnerability score) of each mountain range and subareas included in the study.

Discussion

In this study, the biogeographic, ecological and biological traits of Trichoptera species of Pyrenees and Sierra Nevada have been analysed in order to assess their vulnerability to GCC. Previous studies, such as Hering et al. (2009) or Hershkovitz et al. (2015), focused more in the ecological preferences of species. This study added a wider range of biogeographic, ecologic and biological variables, much more similar to the approach of Conti et al. (2014) but focused on caddisflies and with several differences in the traits addressed. For example, in this study traits such as "Maximum female wing length" and "Persistence" of species were incorporated, as a measure of dispersal and continuity of a species in an area, while traits with low potential for assessing vulnerability to GCC, such as "pH preference" and "Locomotion type" were excluded. Furthermore, this study targeted a much smaller area than the previous ones, which were focused at the continental scale, aiming to be more conservation-oriented.

The most vulnerable species, such as *Rhyacophila vandeli*, are mainly rare and endemic species of the Pyrenees that have not been observed since the twentieth century (Universidad de Castilla-La Mancha, 2018). Most of them have a preference for cold waters, have a short summer emergence period and are specialised in one or two substrates. On the other hand, the least vulnerable species, *Mesophylax aspersus*, is a common and widely distributed species that appears in Europe, Northern Asia, North Africa and even the Canary Islands. It is a species unlikely to be endangered by GCC since it inhabits areas below 1000m and along a longitudinal gradient, from epirithral to epipotamal stream zones (Graf et al., 2008). Furthermore, its life cycle is synchronised with the drying period and adult aestivate in caves (Salavert et al., 2008).

Our results agree with those obtained in other studies (Conti et al., 2014; Hershkovitz et al., 2015) in the sense that, regarding vulnerability to GCC of species, Trichoptera is a heterogeneous group, with very highly vulnerable species but also tolerant species, unlike other groups like Plecoptera. The fact that all of the ten most vulnerable species were only found in the Pyrenees, and that in the three Pyrenean subareas over 28% of species were highly or very highly vulnerable, is consistent with some of the findings of Hershkovitz et al. (2015). They found that the Pyrenees were one of the “hot-spots” of GCC-vulnerable species along with other mountainous ecoregions such as the Alps and the Dinaric Alps. However, our results also showed a higher percentage of highly or very highly vulnerable Trichoptera species in both mountain ranges (38.8% in the Pyrenees and 13.5% in Sierra Nevada) than the 10% of endangered species addressed by Hering et al. (2009) in high mountain areas. This difference could be due to the differences between the spatial scales of the studies, bioregions defined by Illies (1974) against a mountain range, that is a bioregion on its own, and a mountain range inside the Ibero-Macaronesian bioregion. However, it could also mean that vulnerability of high mountain Trichoptera has been underestimated and that, despite their high diversity, a significant amount of species could be threatened by GCC.

Trichoptera is a species-rich group in the Iberian Peninsula compared with other regions of Europe (e.g. England), and have with similar numbers to other countries of the Mediterranean basin (e.g. Italy, Bonada et al., 2004). The main reason of this high diversity is that the Iberian Peninsula acted as glacial refugia and speciation area during the Pleistocene (Hewitt, 1999; Ribera & Volger, 2004). Trichoptera is, however, a group characteristic of more humid and rainier climates than the Mediterranean, which dominates in the Iberian Peninsula. For example, De Moor & Ivanov (2007) reported that 47-77% of all worldwide species of Trichoptera are found in the tropical areas of Oriental or Neotropical ecoregions. In addition, Trichoptera, despite being much more diverse in southern Europe, is a group of major importance in rivers in central Europe with temperate climates (Bonada et al., 2012). The Mediterranean climate of the Iberian Peninsula implies the presence of many intermittent streams, where very few Trichoptera species are found and, when found, they are presented in lower abundances than in central Europe (Bonada et al., 2012). Therefore, the higher number of Trichoptera species, together with the low appropriateness of their environmental conditions imposed by the Mediterranean climate, and the low occurrence of high mountain areas, make Trichoptera a group much more vulnerable in the Iberian Peninsula than previously thought. Our results, in comparison to those found for other areas in Europe (Hering et al., 2009), highlight this statement.

The use of the TVA provided insightful information for high mountain Trichoptera. The most influential traits in determining the total vulnerability of species were “Stream zonation preference” and “Temperature preference”. Thus, species with preference for cold or very cold temperatures and headwater zones tended to be overall more endangered, meaning that, in general, the further upstream a species lives the more vulnerable it is. This finding is consistent with the upstream–downstream gradient Conti et al. (2014) found in their research. Although the obvious correlation between these two traits, it is important to acknowledge that not all species with preference for very cold water are restricted to headwaters and thus, that their upstream migration could be possible, as observed by Sáinz-Bariáin et al. (2016) for species such as *Lepidostoma basale* and *Polycentropus kingi*. On the other hand, water warming could have further consequences for Trichoptera, other than upstream migration, such as a decrease in body size (Cogo et al., 2020) or fecundity reduction (Bovill et al., 2019), that could further threaten the survival of species. Despite we used a higher number of traits in our TVA than others, this also highlights the need to consider further traits. Indeed, despite there is a vast information on traits of Trichoptera (e.g. see www.freshwateecology.info), many species remain poorly assessed, which could bias the results and underestimate the vulnerability of some species. For example, *Anitella iglesiasi*, an Iberian endemic species highly vulnerable due to its low dispersal capacity and low genetic flux between its populations (Múrria et al., 2020) has the same total vulnerability score as *Ceraclea dissimilis* which is an eurytherm species widely distributed through Europe (Graf et al., 2008).

When comparing the two mountain ranges, the Pyrenees had higher species total vulnerability and higher percentages of highly or very highly vulnerable species. This could partly be due to the fact that while the Pyrenees still preserve several glaciers and other glacial masses, Sierra Nevada, with no glaciers left, is at a later stage of deglaciation (Andrés et al., 2018; Gómez Ortiz et al., 2014; Palacios et al., 2016). This could mean that Sierra Nevada could have already undergone a selection of species more tolerant to global warming, besides potential adaptation of existing populations through microevolutive processes. However, the vulnerability of species in Sierra Nevada should not be overlooked since some of its species, such as *Wormaldia granada*, are endemic of this mountain range or even have only been found in one of the subareas (Universidad de Castilla-La Mancha, 2018). In addition, the dimensions of Sierra Nevada, much smaller than any Pyrenean subarea, could make endemic species in this region to be more vulnerable due to their restricted distribution. Furthermore, while the Pyrenees extend through an area that ranges between Mediterranean climate at its eastern border and a more temperate climate in its western border, Sierra Nevada is located in a completely Mediterranean climate area. Thus, since the Mediterranean area is expected to be one of the regions most harshly affected by GCC (IPCC, 2014), species in Sierra Nevada will possibly have to face harder conditions than those in the Pyrenees, what would make them more vulnerable.

Despite the use of TVA offers an overall look of species vulnerability using a wide variety of traits, as shown in our study, other aspects should be considered to have a better estimation of species vulnerability. Firstly, information on the genetic diversity of the populations of the most vulnerable species could reveal they potential microevolutionary adaptations to GCC (Bálint et al., 2011) and the effective population size (i.e. population trend; Miller et al., 2003). Secondly, a more complete characterization of current drought resistance and dispersal ability would be very needed, since these two are traits that could help species survive GCC but are still unknown for most Trichoptera species (Graf et al., 2008). For this purpose, a measure of dispersal ability could be the maximum female wing length, since females with larger wings are able to disperse further (Hoffsten, 2004). However, small species might also passively disperse with the wind. Alternatively, intraspecific genetic diversity and structure could be used as a measure of potential dispersal as in Múrria et al. (2020). Finally, besides using information on potential traits, even if fuzzy coded, information on real (i.e. in situ) intra-species trait variability would be highly recommended to detect microevolutive differences among populations and to have a more reliable trait information than obtained from literature (Bonada & Dolédec, 2018).

In terms of regional conservation, special attention should be paid to Central Pyrenees and Eastern Pyrenees subareas since they are the ones with greatest number of species and highest percentage of highly and very highly vulnerable taxa. This coincides with the work of Davis et al. (1994, as cited in Médail & Quezél, 1999) that found the Pyrenees to be a Center of Plant Diversity to preserve in the Mediterranean, and with the boundaries of the Mediterranean climate drawn by Médail & Quezél (1999) that match the boundaries of Central Pyrenees and Eastern Pyrenees. Thus, part of the Pyrenees under Mediterranean climate conditions looks like specially interesting to preserve. On the other hand, further samplings in the Western Pyrenees would be also needed as they could reveal several other GCC-vulnerable species not detectable with the existing records. Recent studies have reported that even in streams from relatively well-sampled areas, such as Central Pyrenees, rare and limited distribution species can be found (Guareschi et al., 2018). In this way, our results provide a valuable contribution for the management and conservation of high mountain streams in the Iberian Peninsula. For this purpose, an observatory network to periodically monitor the most vulnerable species in headwater streams of these areas would be necessary.

Acknowledgements

Thank you very much to Núria Bonada and Raúl Acosta for their unvaluable support, advice and involvement in this work. Thanks also to Carmen Zamora-Muñoz for her support when assessing the vulnerability of species of Sierra Nevada. This study was supported by the ERANet-Lac GLOBIOS ("Global observatory network for freshwater biodiversity in high mountain streams") project (Ref: PCI2019-103495) funded by the Spanish Ministry of Science, Innovation and Universities. Thanks also to the IberArtro project (Ref: 030569-SKAZ) founded, by Junta de Castilla-La Mancha, Ministry of Education, Culture and Sports that was the main source of the fine-scale data about species distribution and occurrence.

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APPENDIX 1

Appendix 1: Percentage of species (only species that could be scored are taken into account) in each trait that fell into each vulnerability category.

Parameter	Taxa (%) with very high vulnerability in relation to scored taxa			Taxa (%) with high vulnerability in relation to scored taxa			Taxa (%) with medium vulnerability in relation to scored taxa			Taxa (%) with low vulnerability in relation to scored taxa		
	Total	Pyrenees	Sierra Nevada	Total	Pyrenees	Sierra Nevada	Total	Pyrenees	Sierra Nevada	Total	Pyrenees	Sierra Nevada
Distribution	24	20	30	5	5	0	61	65	57	10	10	14
Endemicity	51	50	24	23	25	3	12	10	14	14	14	59
Stream zonation preference	7	7	3	29	27	35	24	24	27	40	41	35
Altitude preference	0	0	0	0	0	0	4	3	3	96	97	97
Microhabitat specialist	54	55	51	39	38	38	8	7	11	0	0	0
Temperature preference	1	1	0	31	31	23	29	28	45	40	40	32
Feeding specialist	21	20	14	29	32	38	44	44	41	6	4	8
Emergence period	66	70	54	24	20	35	1	1	0	9	10	11
Emergence length	42	41	38	2	1	5	6	5	5	50	53	51
Life cycle	5	6	0	88	87	97	4	4	0	3	3	3
Persistence	27	29	3	23	25	5	49	46	86	1	0	5
Max female wing length	9	10	5	34	34	35	34	35	27	23	21	32
Species lifespan	18	17	23	0	0	0	18	17	18	64	66	59