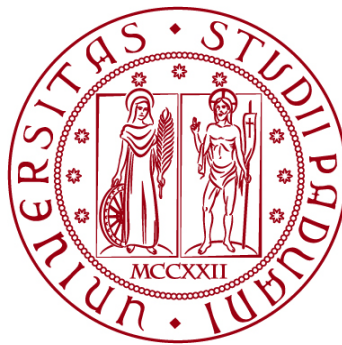


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TESI DI LAUREA

**The Rhopalocera of Monte Grave
(Dolomiti Bellunesi National Park),
with a focus on the ecology of *Phengaris arion***

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1. ABSTRACT

Diurnal butterflies play important ecological roles (Van Swaay et al., 2010), are good bioindicators (Sommaggio & Paoletti, 2018) and flag species (Bonelli et al., 2012). Over the past 50 years, many species have declined, due mainly to the habitat loss and habitat change (Balletto et al., 2015). These include *Phengaris arion*, a lycaenid with a complex life-cycle, closely related to its host ants and host plant (Genovesi et al., 2014). Its conservation is threatened by the destruction and deterioration of its habitat due to intensification or abandonment of agricultural practices (Griebeler & Seitz, 2002; Öckinger & Smith, 2006). This study had the objective to increase the knowledge of the Rhopalocera fauna of Monte Grave, a mountain area in northeastern Italy, partially included in the Dolomiti Bellunesi National Park. Specifically, determining the abundance, the spatial distribution, and the phenology of the species detected; trying to understand whether there are differences between communities in different portions of the study area; and comparing the species composition with that of past surveys. Another objective was to study in detail the population of *P. arion* established in this area. In particular, the abundance, the spatial distribution and the influence of some habitat variables on its presence. The surveys were carried out between May and September 2023. The Rhopalocera fauna was surveyed in three linear transects located in three different meadows, instead *P. arion* was searched during a specific session in the entire top portion of the study area. The habitat variables were collected in the plots where the species was detected and in the same number of random plots where the species was not detected. During the surveys, 44 species were found, 34 of which potentially reproduce in Monte Grave. The difference of the Rhopalocera communities in the three meadows was low and not significant. Temporal analysis showed intermediate similarity among the fauna of the years considered. Ten individuals of *P. arion* were found, dispersed throughout the investigated area. The plots in which the species was found showed positive correlation with thyme, flowering plants, exposure, slope and tree-shrub coverage; while negative with mean height, maximum height and coverage of the herbaceous layer. The differences between plots with *P. arion* and random plots were not significant. The diurnal butterfly communities and *P. arion* population of Monte Grave are related to semi-natural grassland environments, which require active and proper management to maintain their biodiversity (MacDonald *et al.*, 2000; Griebeler & Seitz, 2002).

2. RIASSUNTO

Le farfalle diurne svolgono importanti ruoli ecologici (Van Swaay *et al.*, 2010), sono buoni bioindicatori (Sommaggio & Paoletti, 2018) e specie bandiera (Bonelli *et al.*, 2012). Negli ultimi 50 anni molte specie hanno subito un declino, principalmente a causa della perdita e del cambiamento dei loro habitat (Balletto *et al.*, 2015). Tra queste si trova *Phengaris arion*, un licenide dal ciclo vitale complesso, strettamente legato alla sua pianta nutrice e formica ospite (Genovesi *et al.*, 2014). La sua conservazione è minacciata dalla distruzione e dal deterioramento del suo habitat, a causa dell'intensificazione o dell'abbandono delle pratiche agricole (Griebeler & Seitz, 2002; Öckinger & Smith, 2006). Questo studio ha avuto l'obiettivo di approfondire le conoscenze della fauna di ropaloceri del Monte Grave, un'area montuosa del Nord-Est Italia, parzialmente inclusa nel Parco Nazionale delle Dolomiti Bellunesi. In particolare, determinando l'abbondanza, la diffusione e la fenologia delle specie rilevate; cercando di capire se ci sono differenze tra le comunità di diverse porzioni dell'area di studio; confrontando la composizione in specie con quella di rilevamenti svolti in passato. Un altro obiettivo è stato studiare in modo dettagliato la popolazione di *P. arion* instaurata in questo territorio. In particolare, l'abbondanza, la diffusione e l'influenza di alcune caratteristiche dell'habitat sulla sua presenza. I rilevamenti sono stati svolti tra maggio e settembre 2023. La fauna di ropaloceri è stata rilevata in tre transetti lineari situati in tre prati diversi, mentre *P. arion* è stata cercata durante un'apposita sessione nell'intera porzione superiore dell'area di studio. Le variabili dell'habitat sono state raccolte nei plot in cui la specie è stata rilevata e in un numero uguale di plot casuali in cui la specie non è stata rilevata. Durante i rilevamenti sono state trovate 44 specie, 34 delle quali potenzialmente si riproducono nel Monte Grave. La differenza delle comunità di ropaloceri instaurate nei tre prati è risultata bassa e non significativa. L'analisi temporale ha mostrato una similarità intermedia tra la fauna degli anni considerati. Sono stati trovati 10 individui di *P. arion*, distribuiti in modo sparso nell'area indagata. I plot in cui la specie è stata trovata hanno mostrato una correlazione positiva con timo, piante in fiore, esposizione, pendenza e la copertura arboreo-arbustiva; mentre negativa con altezza media, altezza massima e copertura dello strato erbaceo. Le differenze tra le medie dei plot con *P. arion* e dei plot casuali non è risultata significativa. Le comunità di farfalle diurne e la popolazione di *P. arion* del Monte Grave sono legate agli ambienti dei prati semi-naturali, i quali necessitano di una gestione attiva e corretta per il mantenimento della loro biodiversità (MacDonald *et al.*, 2000; Griebeler & Seitz, 2002).

3. INTRODUCTION

3.1. About Rhopalocera

Butterflies are one of the best-known group of insects, with nearly 19,000 described species. The superfamily Papilionoidea includes seven families, distributed on all continents except Antarctica. Butterfly diversity is particularly high in the tropics, especially in the Neotropics, while only 496 species are present in Europe (Wiemers *et al.*, 2018). This group of arthropods plays important ecological roles such as pollination and as trophic resource for many other animal species (Van Swaay *et al.*, 2010). Butterflies respond rapidly to environmental changes, reflecting changes in other components of biodiversity and thus being excellent bioindicators for diverse terrestrial ecosystems. They occur in a wide variety of habitats, with a large number of species, comprising both generalist and specialist elements (Bonelli *et al.*, 2012). Other factors that make butterflies good bioindicators are their good taxonomic stability, easy identification, good autoecological knowledge of many species, and the high interest towards this group (Sommaggio & Paoletti, 2018). In fact, butterflies are one of the few invertebrate groups judged positively by public opinion, thus representing excellent flag species (Bonelli *et al.*, 2012).

Italy has one of the richest butterfly biodiversity in Europe, counting nearly 300 species, 37% of the entire Euro-Mediterranean fauna (Balletto *et al.*, 2015). Biodiversity is higher in northern Italy, particularly in the Alps and the Prealps. While endemic species are mainly found in the Apennines, small islands, Sicily, Sardinia and xerothermal areas of the Western Alps (Bonelli *et al.*, 2011).

The fauna of the Veneto region counts 170 species of butterflies, 164 of which are resident, three are regularly present, and another three species are irregular. The 98% of the resident species have populations within the hilly and mountainous areas, while only the 34% have populations in the plains (Bonato *et al.*, 2014).

In the Dolomiti Bellunesi National Park, 106 species were found, amounting the 62% of the species reported in the Veneto region and the 37% of the species reported in Italy. The most interesting species appear to be those typical of the high altitudes environments, e.g. *Pontia callidice* (Hübner, [1800]), *Erebia pandrose* (Borkhausen, 1788) and *Erebia gorge* (Hübner, [1804]) (Vettorazzo *et al.*, 2020).

The area of the Park and more in general all the hilly and mountain areas of the northeast Italy are involved in a project of citizen science, the “Neptis project”, which aims are to improve the knowledge about the Rhopalocera species of these territories trough volunteer observations (www.dolomitipark.it).

Most of the butterfly communities are associated with semi-natural habitat that often depend on anthropic management, particularly grazing or mowing activities. Therefore, they often suffer from the abandonment of marginal areas and the subsequent reforestation, but also from the overexploitation. In fact, among the main threats there are habitat loss and habitat change. These changes generate fragmentation and isolation, increasing the likelihood of the occurrence of

stochastic events. Climate change is also a contributing factor to the decline of butterflies (Balletto *et al.*, 2015; Bonelli *et al.*, 2012). As a result, over the past 50 years many European butterfly species have experienced a decline in their distribution and abundance (Öckinger & Smith, 2006).

In Europe, 9% of the butterfly species are considered threatened and more than 10% are considered near-threatened. In addition, 31% of butterfly populations are reported to be in decline (Van Swaay *et al.*, 2010).

In Italy, the endangered species are 18, amounting for 6.3% of the assessed species, while 5.6% are near-threatened. However, most populations are found to be stable (Balletto *et al.*, 2015).

Also within the Park and its proximity there are 12 species included in the European or Italian Red Lists. Five of them are also included in the Annexes of the Habitats Directive, i.e. *Euphydryas aurinia*, *Lopinga achine*, *Parnassius apollo*, *Parnassius mnemosyne* and *Phengaris arion*.

3.2. About *Phengaris arion*

Phengaris arion is a species that belongs to Lycaenidae family. It has a global range that occupies most of the Palearctic, with a distribution centered on Central and Eastern Europe. Its range reaches to the West, France, Norway and the UK and to the East, Southern Siberia, Mongolia and China. Across this large area it is restricted to locations that support the suitable habitat for its complex life-cycle (Hayes, 2015). Given its particular ecology, *P. arion* also attracts interest from part of the large public, making it a good umbrella species, so its protection allows for the conservation of a high diversity of other animal and plant species (Spitzer *et al.*, 2009).

This species emerges between June and July and lays eggs in the flower buds of its host plants, i.e. those of the genera *Thymus* L. and *Origanum* L.. The eggs hatch after 7-10 days, and the larvae spend about 3 weeks feeding on the host plant. At the fourth instar, the larvae leave the plant and fall onto the ground. Here they are adopted by foraging worker ants of the genus *Myrmica* Latreille, 1804, which transport it to their nest. To avoid aggression and integrate into the ant colony, the larva adopt morphological, chemical and acoustic mimicry. In the nest, the larvae prey and feed on the ant brood for about 9 months. Then they pupate and adults emerge a few weeks later (Hayes, 2015).

Its narrow specificity to both host plant and host ant has made this species very sensitive to very small changes in its habitat (Genovesi *et al.*, 2014). Throughout Europe it suffered a severe decline during the 20th century due to habitat destruction and deterioration, often due to intensification or abandonment of agricultural practices (Griebeler & Seitz, 2002). This species is included in Annex IV of the Habitats Directive, Annex II of the Bern Convention and also within the European

Red List, where it is considered threatened (Van Swaay *et al.*, 2010), and Italian Red List, where it is considered near-threatened (Balletto *et al.*, 2015).

In Italy it is widespread in the continental and peninsular part, while it is absent in the islands. It can be found from 200 to 2000 m a.s.l. (Bologna *et al.*, 2016). The alpine populations feed on *Thymus* spp. instead the mediterranean populations feed on *Origanum vulgare* L. (Bologna *et al.*, 2016).

In the Veneto region it has been found in the recent period in about 15 sites, concentrated in the northern part of the Dolomites and Prealps of Vicenza, the southern edge of the Lessini and the Dolomiti Bellunesi. The populations are found in a wide elevational range, from 250 m and 2060 m a.s.l., on sunny and dry meadows, managed in a low-invasive way (Bonato *et al.*, 2014). Two of the localities where this species occurs in the Veneto region are within the Dolomiti Bellunesi National Park, at Certosa di Vedana and Monte Grave (Vettorazzo *et al.*, 2020). At the latter site, the species has been known since 2013, when surveys of a monitoring project began in this area. During these, *P. arion* was detected in the top meadows of the mountain, which are under active management by the Park. The species was detected in 2013 with 2 individuals, in 2014 with 4 individuals, in 2018 with 2 individuals, but in 2019 no individuals were found (Timossi, 2020).

4. OBJECTIVES OF THE RESEARCH

The objective of this research was to improve the knowledge about the Rhopalocera fauna of Monte Grave, a mountain area of north-eastern Italy, also contributing to the Neptis project. First of all, determining the species composition and defining which of these are likely to reproduce and be established in this area. For the species surveyed, estimate the abundance and distribution and, when possible, some aspects of their phenology, such as flight period and number of annual generations. In addition, verify if there are differences between the communities in the different areas surveyed, located both inside and outside the boundary of the Dolomiti Bellunesi National Park. This research had also the objective to compare the species composition detected in this study with that detected in surveys of past years.

Another objective of this research was to verify the presence of the population of *Phengaris arion* settled in the Monte Grave area, to estimate its abundance and its spatial distribution. Finally, try to understand if certain habitat characteristics influence its presence and what implications there might be at the management level based on the results obtained.

5. STUDY AREA

5.1. Dolomiti Bellunesi National Park

The Dolomiti Bellunesi National Park (DBNP) is located in Belluno province, in Veneto region (North-East Italy)(Fig. 1). It was established in 1990, as a union of several State Natural Reserves created in the early 1970s (Schiara Occidentale, Monti del Sole, Val Scura, Piani Eterni-Erera-Val Falcina, Piazza del Diavolo, Vette Feltrine, Monte Pavione), whose boundaries were joined by the inclusion of new areas. In 1993, by Decree of the Republic President, the Park Authority was constituted, which has been responsible for the management of the protected area ever since. To date, its area comprises about 32,000 hectares and its boundaries almost coincide with those of the Special Protection Area (SPA) and Special Area of Conservation (SAC) IT3230083 "Dolomiti Feltrine e Bellunesi," established under the Birds Directive (79/409/EEC) and the Habitats Directive (92/43/EEC). In addition, since 2009 the Park area, together with the rest of the Dolomites, is included in the UNESCO World Heritage List. The Park includes medium-high mountain environments, where the highest elevation is reached by Monte Schiara (2565 m a.s.l.). The reliefs overlook, to the South, the Piave valley between Belluno and Feltre, and to the North, some slopes of Agordo and Val di Zoldo. The most important streams are the Cordévole, a tributary of the Piave River, and the Mis, a tributary of the Cordévole itself. They combine to form a dense hydrographic network along with numerous other streams (Stièn, Caoràme, Vesés, Falcina, Imperina, Vescovà, Ardo, Prampèra.). There are also two artificial lakes, the largest is Lago del Mis, located in the namesake valley, and Lago della Stua, located in Val Canzoi. Biogeographically, the protected area belongs to the Alpine region and represents a sector of the southeastern Alps of enormous environmental richness, expressed by high floristic, vegetational, and faunal biodiversity (A.A.V.V., 2017). The key events that have affected the geological and geomorphological history of the Park are: the accumulation during the Mesozoic Era of the sediments that make up the present sedimentary rocks, such as the Dolomia Principale, the most widespread rock within the Park; the collision during the Tertiary Era between the European and African plates, resulting in the uplift of the Alps and therefore also of the Dolomiti Bellunesi; and the shaping wrought by streams, glaciers and karstification, which are responsible for the great variety of present morphological landscapes (A.A.V.V., 2017).

The richness and rarity of the flora of the Dolomiti Bellunesi have been known since the 17th century. The floristic heritage of the Park includes more than 1300 species. Many of these are rare entities, located at the extreme of their range, of significant biogeographic value or disjunct range. Among the factors that play an important role in the floristic distribution, we certainly find the lithological matrix of the substrate, which leads to a differentiation of habitats; traditional agro-silvo-pastoral practices, such as mountain pasturing and mowing; and the transitional

geographical position. The reliefs, in fact, are close to the plains and thus influenced by thermophilic presences, and are located on the southern edge of the eastern alpine arc and thus subject to migrations of eastern-gravitational species that occurred in the arid-steppic phases of the postglacial period. Within the Park, 109 species were found to be most significant as listed within the national or regional Red List or Annexes II and IV of the Habitats Directive. The most threatened environments appear to be the marginal and anthropized ones, mainly due to the incursion of alien species, but also those no longer subject to human activities. In the latter, there can be found a development of dominant species of robust size and high establishment capacity, which leads to the simplification of the environment itself, resulting in the reduction of marginal niches. From the perspective of the Natura 2000 Network, of which the protected area is part, 34 habitats have been surveyed, referable to those included of Annex I of the Habitats Directive, eight of these are prioritized. The greater surface is occupied by habitat 8210 "Calcareous rocky slopes with chasmophytic vegetation", followed by 91K0 "Illyrian forests of *Fagus sylvatica* (*Aremonio-Fagion*)" and 4070* "Bushes with *Pinus mugo* and *Rhododendron hirsutum* (*Mugo-Rhododendretum hirsuti*)" (A.A.V.V., 2017).

The Park's fauna currently consists of 217 vertebrate and 901 invertebrate species, many of which are subject of specific surveys and monitoring activities. Lepidoptera Rhopalocera are represented by 106 species, about 40% of the species reported in Italy. While for Lepidoptera Heterocera, more than 600 species have been surveyed. Among beetles, 50 species of Carabidae and 47 species of Cerambycidae have been found so far. Other well-represented insect groups are Orthoptera, with 27 species, and Odonata, with 13 species (A.A.V.V., 2017; www.dolomitipark.it). The herpetofauna consists of 12 species of amphibians and 13 species of reptiles. Among the three species of tritons present within the Park, *Triturus carnifex* (Laurenti, 1768) is the most widespread. There are two species of salamanders, *Salamandra atra* Laurenti, 1768 and *Salamandra salamandra* (Linnaeus, 1758). *Bufo bufo* (Linnaeus, 1758) and *Rana temporaria* Linnaeus, 1758 are widespread, but *Bombina variegata* (Linnaeus, 1758) is also quite common. More localized within the protected area are *Bufo viridis* Laurenti, 1768, *Hyla intermedia* Boulenger, 1882 and *Rana dalmatina* Fitzinger, 1838. Among reptiles, the most widespread species are *Lacerta bilineata* Daudin, 1802 and *Podarcis muralis* (Laurenti, 1768), *Hierophis viridiflavus* (Lacépède, 1789) and *Natrix natrix* (Linnaeus, 1758). While more localized species are *Zootoca vivipara* (Lichtenstein, 1823) and *Natrix tassellata* (Laurenti, 1768). There are also three species of vipers in the Park, *Vipera aspis* (Linnaeus, 1758), *Vipera berus* (Linnaeus, 1758) and the rarer *Vipera ammodytes* (Linnaeus, 1758) (A.A.V.V., 2017; www.dolomitipark.it). With regard to ichthyofauna, the protected area includes the initial and infravalley traits of numerous watercourses, pertaining to the "Padano-Veneto District." In the classification of fish habitats they can be described as "trout zone," in which salmonids are the most represented group. Among the seven species surveyed within the protected area two are of community interest, *Salmo trutta marmoratus*

Cuvier, 1829 and *Cottus gobio* Linnaeus, 1758 (A.A.V.V., 2017). The avifauna is represented by 145 species, 115 of which are nesting. Currently, there are 18 species of diurnal raptors reported in the Park, such as *Aquila chrysaetos* (Linnaeus, 1758), *Circaetus gallicus* (Gmelin, 1788) and *Circus cyaneus* (Linnaeus, 1766); seven species of nocturnal raptors, such as *Aegolius funereus* (Linnaeus, 1758), *Bubo bubo* (Linnaeus, 1758), *Glaucidium passerinum* (Linnaeus, 1758); five species of woodpeckers, such as *Dryocopus martius* (Linnaeus, 1758), *Picus canus* J. F. Gmelin, 1788 and *Jynx torquilla* Linnaeus, 1758; five species of galliformes, such as *Alectoris graeca* (Meisner, 1804), *Tetrao urogallus* Linnaeus, 1758 and *Bonasa bonasia* (Linnaeus, 1758); and many other species (A.A.V.V., 2017). The teriofauna consists of 56 species, belonging to different orders. Among the Artiodactyla, *Cervus elaphus* Linnaeus, 1758, *Capreolus capreolus* (Linnaeus, 1758) and *Rupicapra rupicapra* (Linnaeus, 1758) are the more common. There are 16 species of bats, many of them are of community interest. Carnivora includes, e.g. *Canis lupus* Linnaeus, 1758, *Felis silvestris* Schreber, 1777, as well as small mustelids such as *Mustela erminea* Linnaeus, 1758. Finally, the Park is home of Lagomorpha, such as *Lepus timidus* Linnaeus, 1758; Rodentia, such as *Marmota marmota* (Linnaeus, 1758); and several species of micromammals (A.A.V.V., 2017; www.dolomitipark.it).

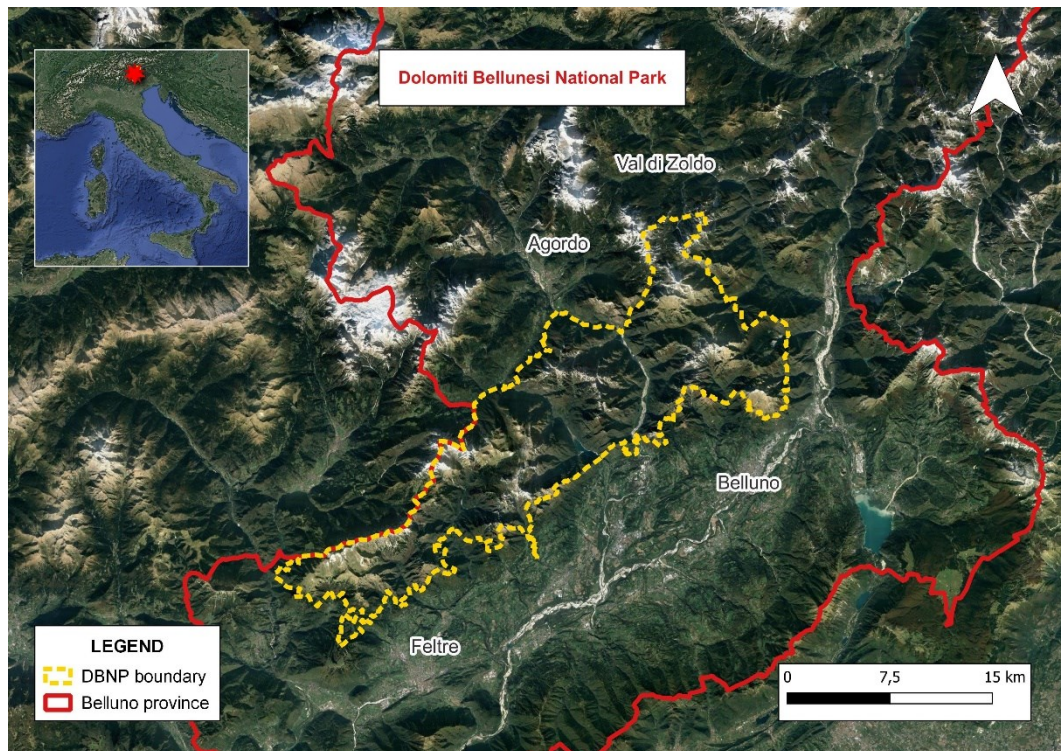


Figure 1. Map of the Dolomiti Bellunesi National Park.

5.2. Monte Grave

This study involved the area of Monte Grave (Fig. 2), located between the municipalities of Feltre and Cesiomaggiore, in Belluno province. It is only partially included in the Dolomiti Bellunesi National Park and the Natura 2000 Network SPA and SAC IT3230083 "Dolomiti Feltrine e Bellunesi", locating in the southwestern portion of the protected area. In this sector, the Park boundary is placed at a variable altitude, always above 1000 m a.s.l., and in a section coincided with the path "Sentiero tematico delle chiesette pedemontane". The mountain has an altitude of 1545 m a.s.l. and overlooks Val Belluna, to the South, and Val Canzoi, to the North. The geology of this area is characterized by the cretaceous Maiolica Formation, which is responsible for the calcareous soil (A.A.V.V., 2017; Canella *et al.*, 2020).



Figure 2. View of Monte Grave from Monte Miesna (Damiano Sartor).

At lower elevations, Monte Grave is mainly characterized by beechwoods, referable to Natura 2000 habitat 91K0 "Illyrian forests of *Fagus sylvatica* (Aremonio-Fagion)". To be noted also the presence of *Carpinus betulus* L., *Ostrya carpinifolia* Scop. on the slopes most prone to runoff, and some secondary populations of *Picea abies* (L.) H.Karst. (Canella *et al.*, 2019; Cassol & Scariot, 2014). However, in this forest environment there are also small size meadows, located at about 1000 m a.s.l., at the localities "Case Cros" and "Casera al Pos" (Fig. 3). The latter site consists of a part of mesophilic and regularly mowed meadows, and a part of abandoned meadows dominated by *Molinia arundinaceae* Schrank and *Juniperus communis* L. (Cassol & Scariot, 2014). Higher elevations are also characterized by semi-natural environments, classified as habitat 62A0 "Eastern sub-mediterranean dry

grasslands (*Scorzoneretalia villosae*)" and in some areas as habitat 6210 "Semi-natural dry grasslands and scrubland facies on calcareous substrates (*Festuco-Brometalia*)" (Cannella *et al.*, 2020). The summit areas are those with the steepest slope and presence of rock outcrops, especially in the western part. Here in addition to grasslands there are some small populations of *Pteridium aquilinum* (L.) Kuhn. While in the eastern part is located a flat area, partly degraded by *Veratrum album* L. and *Asphodelus albus* Mill. (Cassol & Scariot, 2014).

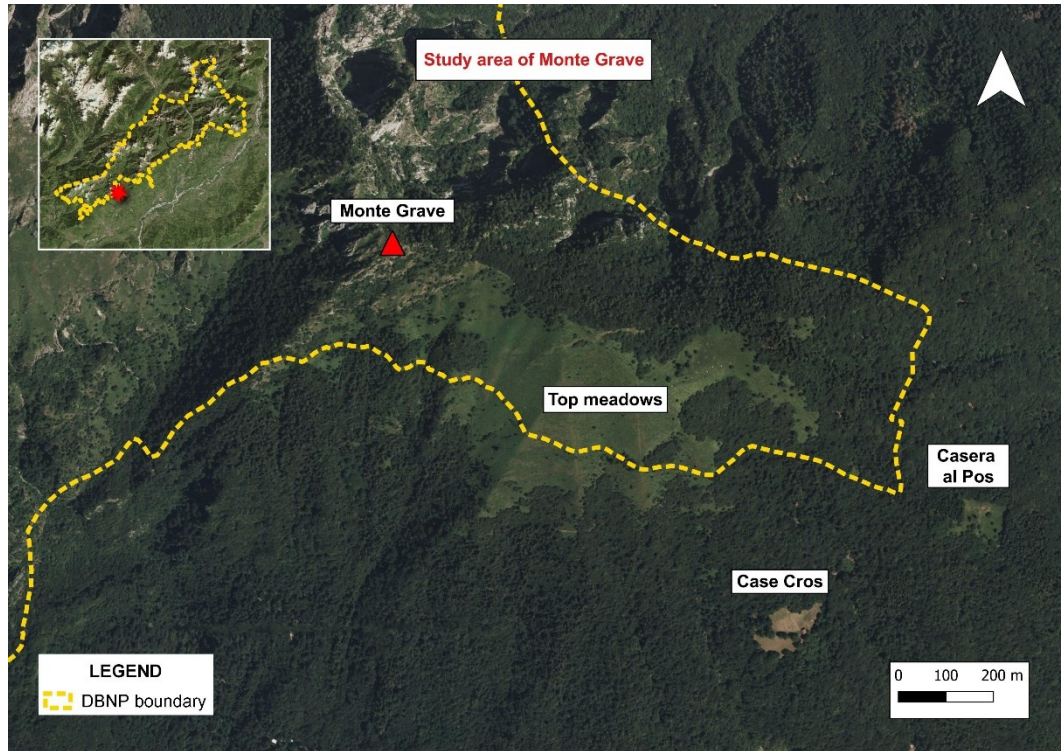


Figure 3. Map of study area of Monte Grave.

During the last century the meadows were managed through mowing for hay production, but this traditional management was abandoned between the 1960s and 1970s, with a consequent recolonization by shrubs and trees such as *Betula pendula* Roth, *Larix decidua* Mill., *Corylus avellana* L., *Sorbus aria* (L.) Crantz, and the increase of robust-sized graminoids (Cannella *et al.*, 2020). Three big wildfires between 1975 and 2012 limited the spread of woody species in the Monte Grave area (Cannella *et al.*, 2020). Since 2009, the Dolomiti Bellunesi National Park has adopted conservation actions consisting of woody species removal and periodic mowing of meadows, with the aim of restoring and maintaining the biodiversity of this area (Cannella *et al.*, 2020). In the first year, trees and shrubs were removed manually, while mowing is carried out biannually, between late August and early September. This action is carried out in the top meadows of Monte Grave, at a vegetation height of 10-12 cm above the ground, mowed material is not removed and additional treatments (such as fertilization or seeding) are not carried out

(Canella *et al.*, 2020). The mowed area changes based on the year, but normally includes always the easternmost and highest part of the top meadows (Fig. 4). Monte Grave is one of the sites included in the multi-year monitoring program for species of community interest in the Habitats Directive. It is also one of the sites of the long-term project "Monitoraggio della biodiversità in ambiente alpino", a project that involves six Italian alpine parks (Dolomiti Bellunesi National Park, Gran Paradiso National Park, Stelvio National Park, Val Grande National Park, Veglia Devero Regional Park and Orsiera Rocciavré Regional Park) to study the effects of the climate change on alpine biocenosis. This type of monitoring has been carried out since 2013 and consists of two years of surveys interspersed with four years of break (currently surveys have been carried out in the 2013-2014 and 2018-2019). The groups monitored by this project are: Lepidoptera Rhopalocera, Coleoptera Carabidae, Coleoptera Staphylinidae, Orthoptera, Hymenoptera Formicidae, Araneae, Aves; and vegetation surveys are also carried out (Cerrato *et al.*, 2017; Vettorazzo *et al.*, 2020).

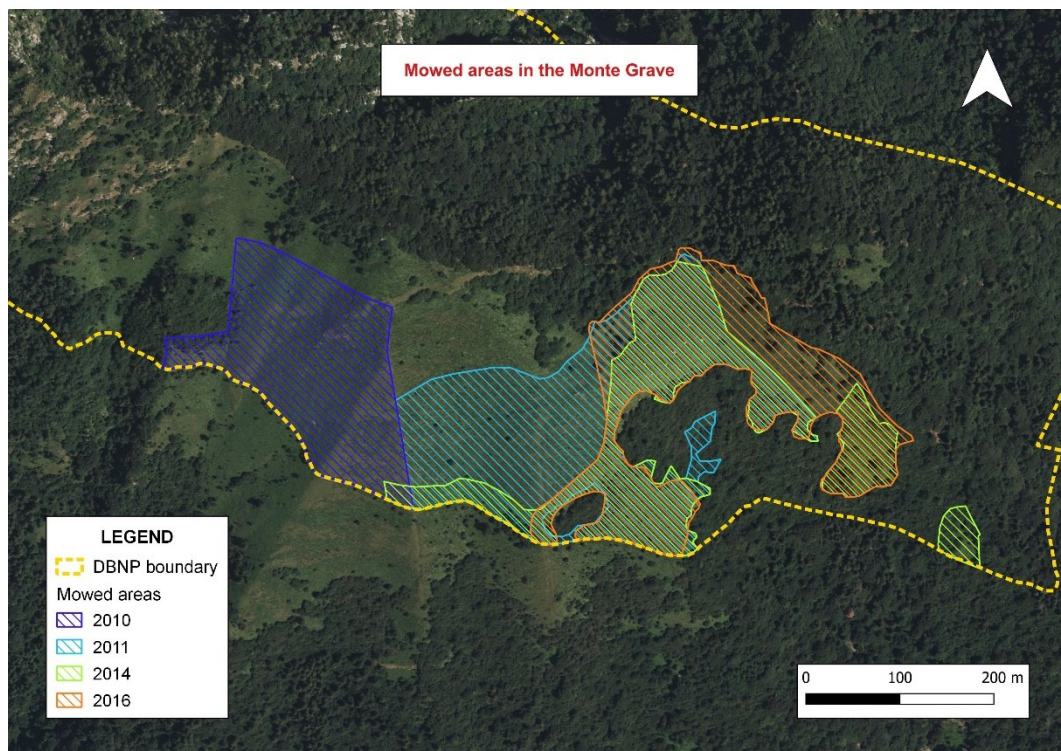


Figure 4. Some examples of mowed areas over several years in the Monte Grave.

6. MATERIALS AND METHODS

6.1. Sampling methods

6.1.1. Sampling of *Rhopalocera* fauna

Three linear transects were established in the study area, located in three different meadows, respectively at "Case Cros", "Casera al Pos" and the top meadows of Monte Grave (Fig. 3). From here on, to simplify, the transects will be called CCT (Case Cros Transect), CPT (Casera Pos Transect), and TMT (Top meadows Transect)(Fig. 5; Fig. 6).

The transects position was chosen in order to have the most long transect as possible respect the surface of the meadow itself. Also, in the top meadows the transect position coincides with the path "Sentiero tematico delle chiesette pedemontane", because of the steeper slope of this meadow it was preferred to have an already existent route to facilitate the surveys.

The coordinates, the elevation, the length, the position in relation to the DBNP boundary, the meadow area and the meadow mean slope of each transect are shown in table 1. Meadow area was considered as the area within the boundary of the meadow where the transect were located.

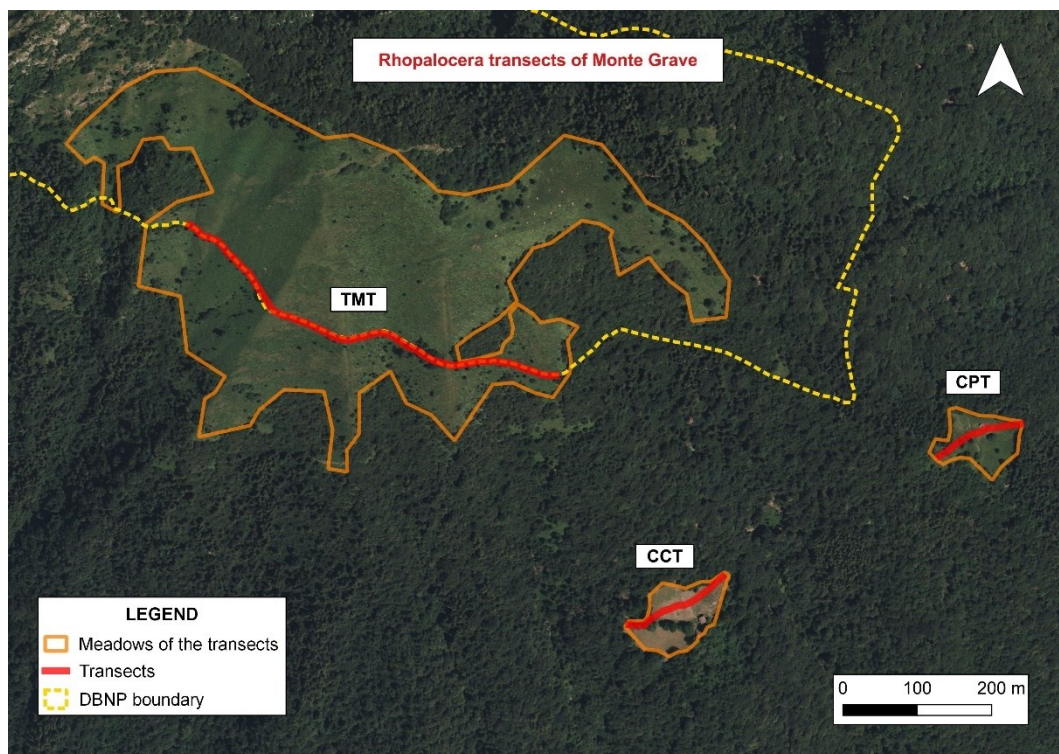


Figure 5. Location of the *Rhopalocera* transects of Monte Grave.

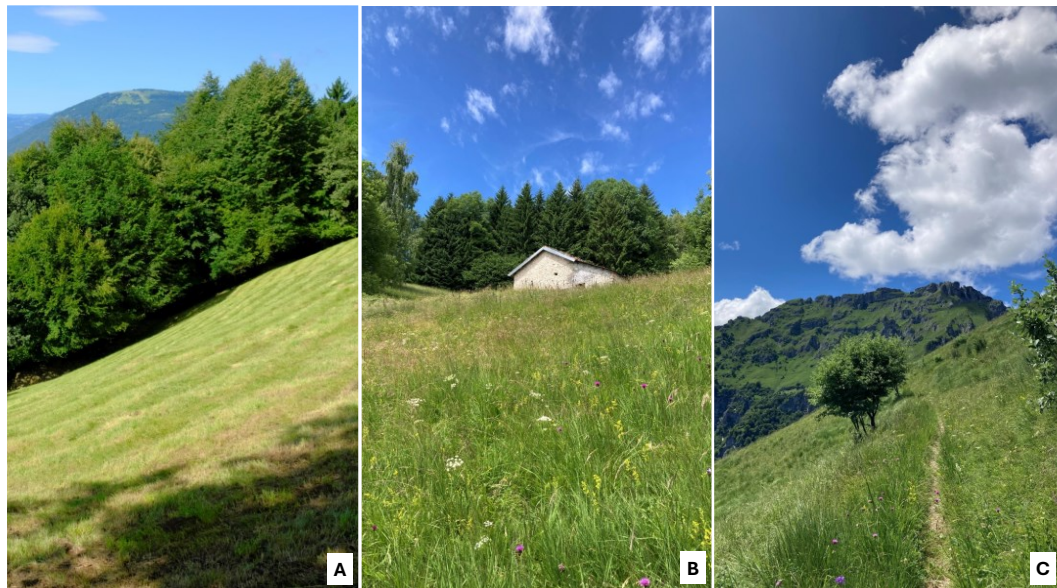


Figure 6. A) Environment of CCT (July 26); B) Environment of CPT (July 4th); C) Environment of TMT (July 4th).

Transect	Extreme points coordinates	Min and max elevation (m a.s.l.)	Length (m)	Position in relation to the DBNP boundary	Meadow area (ha)	Meadow mean slope (°)
Case Cros Transect (CCT)	46.08586 N 11.93493 E - 46.08532 N 11.33278 E	1011-1018	146	Outside	0.95	23
Casera Pos Transect (CPT)	46.08757 N 11.94020 E - 46.08722 N 11.93877 E	1055-1085	120	Outside but near the boundary (about 130 m)	0.74	18
Top meadows transect (TMT)	46.08836 N 11.93217 E - 46.09034 N 11.92589 E	1220-1290	570	On the boundary	21.27	31

Table 1. Extreme points coordinates, minimum and maximum elevation, length, position in relation to DBNP boundary, meadow area and meadow mean slope of each transect.

Each transect was surveyed between May and September 2023, with a maximum interval between two subsequent visits of 15 days. The surveys were carried out by the same person (Lorenza Zaetta). A total of 10 sessions were made, during which the three transects were surveyed on the same day, except on one occasion. On July 14th only TMT was carried out and so July 17th CCT and CPT were also carried out. Surveys were conducted under suitable weather conditions for butterfly activity, i.e. temperature above 13°C, mostly sunny and without strong wind (less than 5 on the Beaufort scale)(Pollard & Yates, 1993). Temperature was recorded through sensors placed at three different locations of the study area, while the other two variables

were estimated by sight in the field. In case of bad weather conditions, surveys were temporarily interrupted and resumed if weather improved, from the same point.

Transects were walked unidirectionally and at a constant speed, alternating the starting point and the direction between sessions. Adult individuals of *Rhopalocera* observed within an imaginary box of 5 m width, height, and distance from the detector were surveyed (Pollard & Yates, 1993).

The survey of the transects was not time constrained, but the start and end time of every session was recorded for each one. The average time of CCT was 49 minutes, of CPT was 24 minutes and of TMT was 80 minutes.

In addition, any notes on factors that may have had an influence on the presence of the butterflies were annotated (e.g. recent mowing activities in the meadows or particularly bad weather the days before the surveys).

All data were collected with the iNaturalist app, through the smartphone, and then saved to the Neptis project. The parameters collected for each record are:

- Day and time of the survey;
- Localization: with high accuracy (if possible less than 10 m);
- Species: identified directly in the field or later by looking at photos of the individuals;
- Number of individuals: if individuals of the same species were isolated from each other, one record was taken for each individual; if individuals of the same species were close to each other (approximately less than 10 m far from each other), one record was taken for the group of individuals, specifying the number;
- Photos: when possible and useful for identification; photos were taken with the smartphone, of the wing surfaces or other anatomical structures relevant for identification;
- Sex: it was identified by visual analysis of wing pattern or morphology of accessory structures of copulatory system by performing soft pressure on the abdomen and, if necessary, using a magnifying glass; this parameter was collected only for individuals that had been temporarily captured;
- Wing damages: when it was possible; was defined using a scores from 0 to 4 (from least to most damaged), either directly in the field or later, analyzing photos of individuals;
- Notes: e.g. characteristics of individuals important for identification or whether the specimen was collected.

Data were collected with three different survey methods:

- Observation without capture: used for all the species whose the identification was possible without capture, either directly or with the help of binoculars;
- Temporary capture: used in case of uncertainty in species identification and thus need for close examination. Individuals were captured by means of an

entomological net and held for the time necessary to make observations and take photos, and then released from the same point;

- Collection of specimens: only in the case of the cryptic species complexes *Pyrgus malvae/malvoides* and *Leptidea juvernica/sinapis*. Of these, one specimen was collected for each session in which the species were detected, in each transect. The specimens were placed in a tube with ethyl acetate and then moved into an envelope, with a label with relevant information (collector, date, code of record associated with the iNaturalist database) and stored in the freezer for future analysis.

The Dolomiti Bellunesi National Park has given the authorization of this research activity inside the protected area and ISPRA (Istituto Superiore Protezione e Ricerca Ambientale) has given the authorization to temporary capture the species of community interest, in relation to the research activities of the Neptis project.

A list of species potentially present in the study area was prepared before the survey sessions, based on Vettorazzo *et al.* (2020) and Bonato *et al.* (2014). Instead, "Le farfalle dell'Italia nordorientale: guida al riconoscimento" (Paolucci, 2010) was used for species identification. The present study follows the classification and the nomenclature of "An updated checklist of the European Butterflies (Lepidoptera, Papilionoidea)" (Wiemers *et al.*, 2018).

Whenever possible, individuals were identified to species level. However, some cryptic groups, which were difficult to distinguish morphologically, were considered as species complexes in this study, i.e. *Colias alfacariensis/hyale*, *Leptidea juvernica/sinapis*, *Melitaea athalia/celadussa*, *Pieris bryoniae/napi*, and *Pyrgus malvae/malvoides*.

Individuals of *C. alfacariensis* and *C. hyale* are often indistinguishable at the adult stage and both species are reported in the Veneto region, the first is resident and the second is more occasional due to its tendency of dispersal (Bonato *et al.*, 2014).

L. juvernica and *L. sinapis* can be distinguished by analysis of the copulatory system or by genetic analyses; in Veneto region, the first is present more frequently in mountainous areas, while the second is widespread from coastal to mountainous areas, however, the two species are often syntopic (Gallo *et al.*, 2015).

The evolutionary relationship between the two species *M. athalia* and *M. celadussa* is still complex and not clarified, and it has not yet been unambiguously defined whether they are distinct species (Tahami *et al.*, 2020).

Individuals of *P. bryoniae* and *P. napi* are difficult to distinguish, the first species is reported to occur at high elevations of mountainous areas, while the second species appears to be widespread at lower elevations in the Veneto region (Bonato *et al.*, 2014).

P. malvae and *P. malvoides* are distinguishable only by examining the copulatory system, both species are found to occur in the prealpine and alpine areas of northeastern Italy, while only the latter is also widespread in the Venetian plain (Koren *et al.*, 2013).

6.1.2. Sampling of *Phengaris arion*

In order to study the abundance and the spatial distribution of this species, a session was organized during its flight period and with good weather conditions, in the area located on the top meadows of Monte Grave, where the species had been reported in the past (Fig. 7; Fig. 8). This session was organized after the species was detected a first time during one of the periodic TMT surveys, on July 4th. Several detectors (Lucio Bonato, Enrico Carta, Damiano Sartor and Lorenza Zaetta) examined part of the area working in parallel, as complete as possible, searching for adult individuals of *Phengaris arion*. The individuals found were geolocated, using the iNaturalist app, and the same parameters of the species detected during the transects surveys were taken (wing damages, sex, etc.). The sex was identified by visual analysis of the accessory structures of copulatory system by performing soft pressure on the abdomen and using a magnifying glass. Each individual was marked with a dot on the lower surface of the left wing, using a yellow non toxic-ink marker, to have an estimation of the population numbers and the spatial distribution. This session was made on July 14th, between 10:30 and 16:30. The research area of *P. arion* has been defined in order to include all the points geolocated during the this session by all the detectors.

In a second session, an analysis of some characteristics of *P. arion* habitat was made to understand which of them have an influence on the presence of this species in the study area. These surveys were carried out in circular plots centered on the geolocated points of the surveyed individuals. Most variables were surveyed within a radius of 25 or 5 m, depending on the variable considered. In the case where the distance between points was less than 5 m, a single plot was established, because they were largely overlapping. The same analysis was carried out in an equal number of plots where the species was not detected and chosen randomly in the research area. The *P. arion* plots have been removed from the research area layer and the random plots were extracted from the remaining area using the QGIS tool "Random points in layer extension", indicating the number of points and the minimum plot distance of 50 m, so their areas did not overlap. The coordinates of the random points were obtained by the attribute table field operator of QGIS, through the expressions "\$y" and "\$x", respectively for longitude and latitude. During this second session, the presence of marked and unmarked individuals of *P. arion* was also checked throughout the research area. This session was made on July 26th, between 12:30 and 18:00.

The environmental parameters measured were:

- Elevation: was calculated using QGIS, with the "sample raster values" processing tool, using a DTM downloaded from the Veneto Geoportal (idt2.regione.veneto.it) and having cells of 5 by 5 m.;
- Exposure: was calculated using QGIS, with the geomorphological analysis tool "aspect" was created a raster with values from 0° to 360°, expressing the direction of the slope starting from North and continuing clockwise; the

tool "sample raster values" allowed to overlaid this raster with the layer of plot points, to obtain the exposure values of the them;

- Slope (25 m radius plot): was calculated using QGIS, the elevation of the highest and lowest points of the area of each plot was identified using the tool "sample raster values", the arctangent of the difference in elevation between these two points divided by the planimetric distance (50 m) was calculated, and the result was converted from radians to degrees;
- Thyme coverage (5 m radius plot): was estimated in m^2 and converted to percentage (Casacci *et al.*, 2010), the type of cover considered was foliar cover, the species of *Thymus* present in this area is *Thymus pulegioides* L. (Cassol & Scariot, 2014);
- Flowering plants coverage (5 m radius plot): was estimated in percentage (Spitzer *et al.*, 2009), the type of cover considered was foliar cover;
- Tree-shrub coverage (25 m radius plot): was estimated in percentage, in the field and then checked remotely (using QGIS and Google Earth Pro), the type of cover considered was canopy cover;
- Maximum height of herbaceous layer (5 m radius plot): was estimated using a meter, in cm (Vilbas *et al.*, 2015);
- Mean height of herbaceous layer (5 m radius plot): was estimated by sight, in cm;
- Herbaceous layer coverage: was estimated in percentage, on a 1 by 1 m square positioned so that it was as coincident as possible with the center of the plot, each square was photographed from above, the estimation was made in the field and looking at the photos, the type of cover considered was foliar cover;
- Presence of rocks (25 m radius plot): was estimated in the field, as presence or absence;
- Notes: regarding other important aspects of the plot environment.

The version of the software QGIS used in this study was 3.28 LTR.

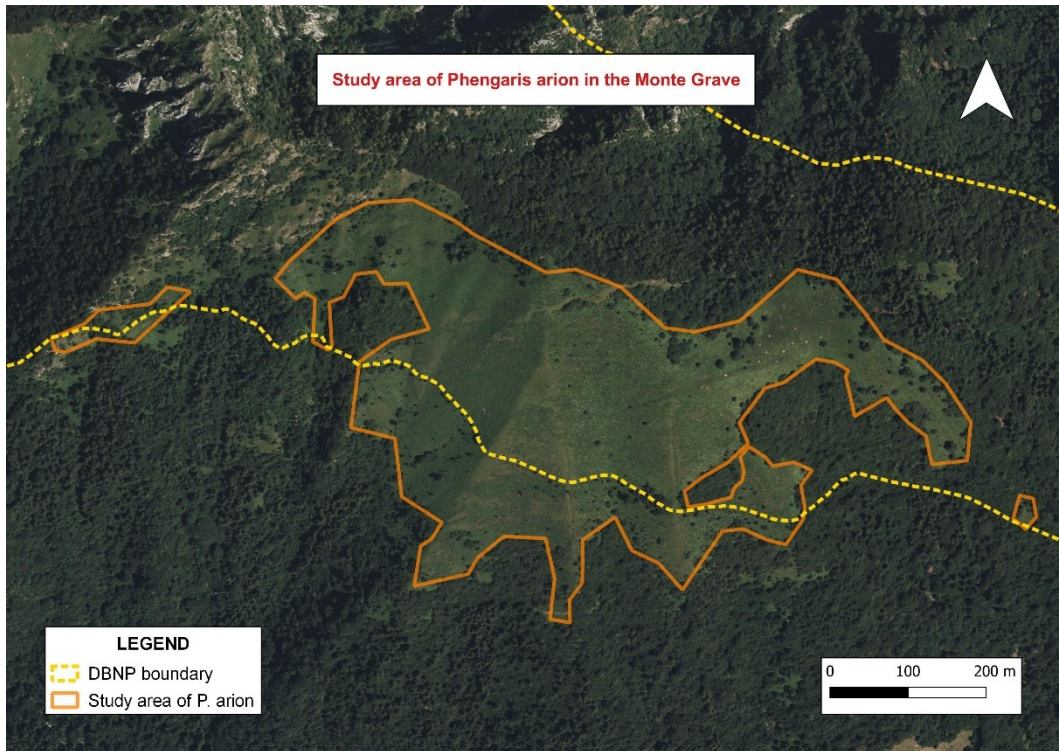


Figure 7. Study area of *Phengaris arion* in the Monte Grave.



Figure 8. Environment of the top meadows of Monte Grave (August 23rd).

6.2. Analysis methods

6.2.1 Rhopalocera fauna analysis

Species that have been considered to potentially reproduce in the Monte Grave area are those whose the presence and reproduction are ascertained at elevations involved in this study, the data obtained were congruent with the species flight period and do not have dispersal tendency; based on information about the different species for the Veneto region (Bonato *et al.*, 2014). Applying a very stringent criterion, species of which a single individual was found during all the flight season were excluded, as they could be the result of isolated dispersal events.

For each species, the maximum number of individuals and the arithmetic mean of individuals detected during all the sessions were considered as abundance indexes (Bonato *et al.*, 2009). Instead, the number of transects in which the species were detected was used as diffusion index (Bonato *et al.*, 2009).

For species that potentially reproduce in the study area, some aspects of their phenology were investigated. The flight period was intended as the time interval between the first and last detection of a species. For the species which it was possible, the number of annual generations completed in the study area was estimated. To make this evaluation, the peaks of the number of individuals, spaced at least one month apart, were considered as the period of maximum concentration of flying individuals of a generation (Bonato *et al.*, 2009). The results obtained were compared with the information about phenology of each species for the Veneto region.

Overall, time trends of the number of individuals and number of species during the flight season were investigated, both for individual transects and for the total of the transects data. For a better graphic representation, the data of July 14th and 17th were considered as belonging to the same date because the transects were surveyed in these two different days for the same session, as previously mentioned.

To estimate the diversity of the three transects and compare it to each other, Shannon index (H), Simpson index (D), and the evenness index based on Shannon index (E_H) were calculated (Magurran, 2004), about the data obtained during the entire flight season in each transect, considering both the totality of the species surveyed and only the species that potentially reproduce in the study area.

$$H = - \sum p_i * \ln p_i$$

$$D = 1 - \sum p_i^2$$

Where p_i is the relative abundance of the i -th species.

$$E_H = \frac{H}{H_{max}} = \frac{H}{\ln S}$$

Where S is the total number of the species.

Diversity indexes were calculated with RStudio, thanks to the "vegan" package.

Finally, the Sørensen similarity index (C_s) was calculated to compare the species composition of the three communities (Magurran, 2004).

$$C_s = \frac{2a}{2a + b + c}$$

Where a is the number of species in common at the two sites, b is the number of species found only at the first site, and c is the number of species found only at the second site.

Since TMT is much longer than the other transects, to understand if its length influences the number of the species detected and also if there are any differences among different portion of this transect, subsamples of it were made in order that they had about the same length of the other transects (Fig. 9). The transect was divided in four subsamples of the same length (142.5 m) and the number of species detected in each one was calculated.

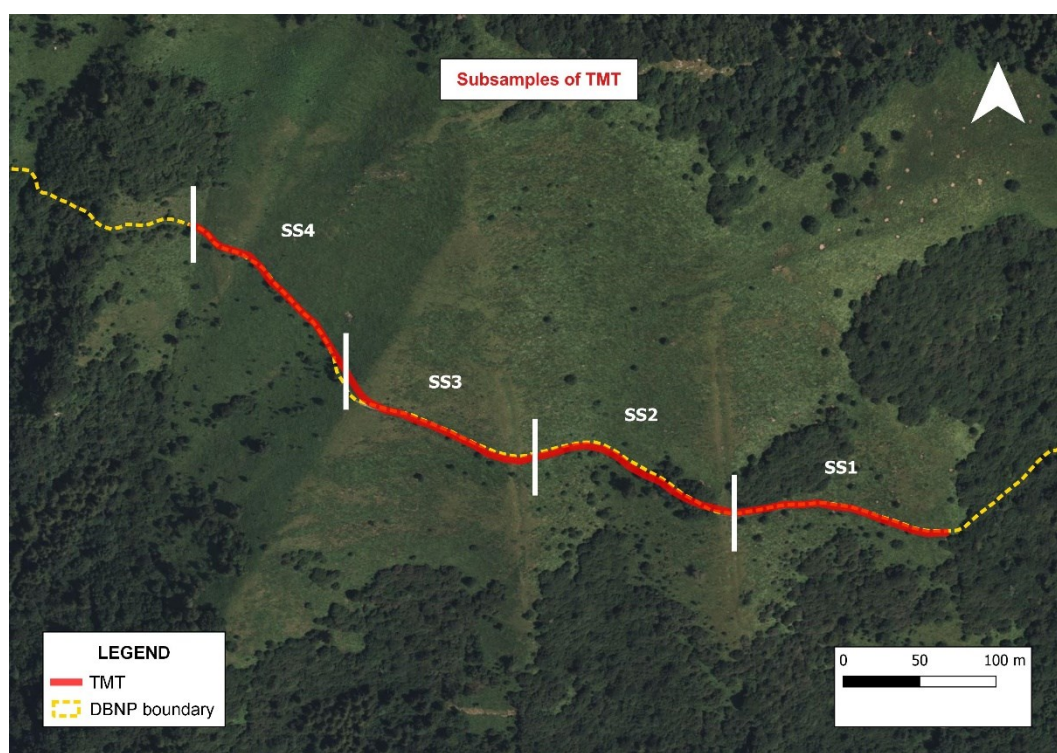


Figure 9. The subsamples of TMT (SS1, SS2, SS3 and SS4).

Sørensen's index was also used to compare the species composition of Monte Grave meadows between this study and past surveys, specifically in the periods 2013-2014

and 2018-2019, on the occasion of the surveys of the project "Monitoraggio della biodiversità in ambiente alpino".

The methodology used in this monitoring consist in three plots (MGA, MGB and MGC), at successive interval of 200 m of elevation. The diameter of these plots correspond to a linear transect of 200 m, that was surveyed on the way out and on the way back. The surveys were carried out between May and September, for a total of five sessions per year (Vettorazzo *et al.*, 2020).

The comparison was made between 2023 and the two biennia, to understand if there are difference between the actual fauna and that of about five and 10 years ago. For each comparison, only data with an identification at species level and detected in the same seasonal interval (using a tolerance of one week) were included in these analyses. The plots of the past surveys were located in slightly different areas than those chosen in this study (Fig. 10). Specifically, MGA plot basically coincides with CPT, while in the top meadows two plots, MGB and MGC, had been established in portions at higher elevations than TMT. The comparisons were made both considering the whole area and the two meadows separated. CCT were not subject to this type of analysis, as this area had not been investigated in the past.

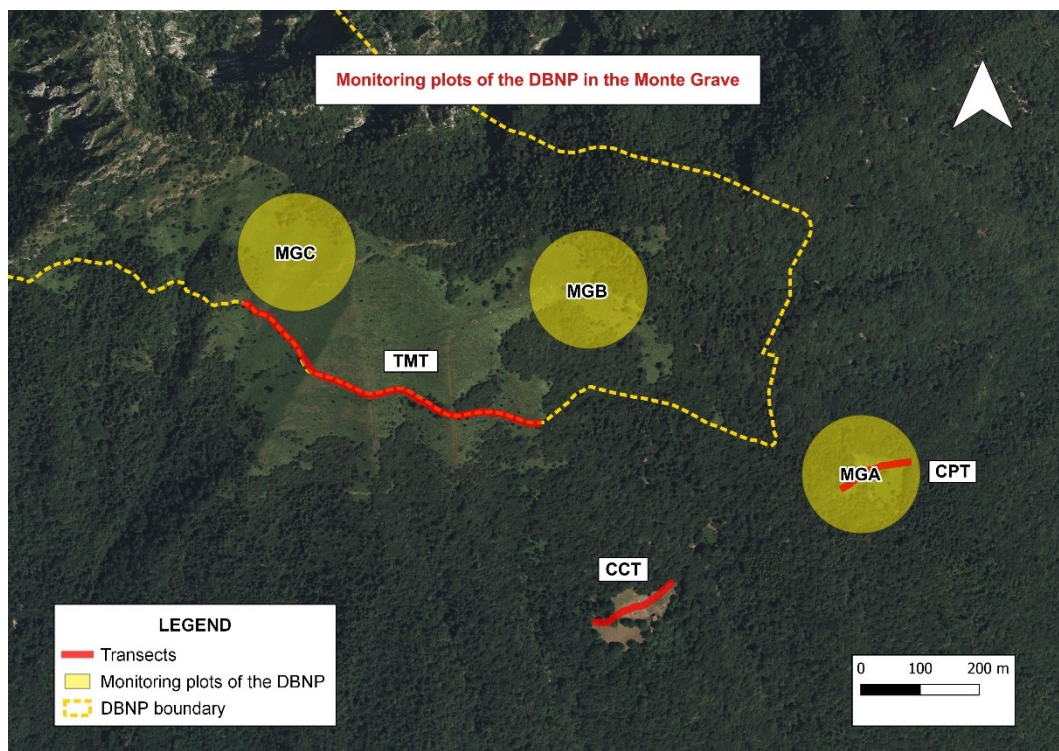


Figure 10. Position of the monitoring plots of the DBNP (MGA, MGB and MGC) in the Monte Grave.

The confidence interval was calculated for each index, using the bootstrap method (Magurran, 2004). This method creates new datasets, created by randomly sampling the original dataset with replacement (Hastie *et al.*, 2009; James *et al.*, 2013). The

bootstrap samples were used to compute the indexes, and these results are used to calculate the confidence interval. For each index, the respective function was created to be implemented on the Rstudio "boot" package, the number of replications chosen was 1000, and the 95% confidence interval was calculated thanks to "conf.int" function. This procedure was repeated for each index calculated for different transects and for different years (in the case of the temporal analyses).

6.2.2. *Phengaris arion* analysis

To assess the spatial distribution of *Phengaris arion* in the study area the nearest neighbor method was used. The ratio of mean observed distance to mean expected distance (R) provides a value of how much the population distribution deviates from a casual distribution. The mean observed distance (\bar{r}_A) was calculated by arithmetic mean of the distance of the closest *P. arion* individual to each individual itself, using the QGIS tool "line measurement". The expected mean distance (\bar{r}_E), assuming a casual distribution, was calculated by the formula of Clark & Evans (1954).

$$R = \frac{\bar{r}_A}{\bar{r}_E}$$

$$\bar{r}_A = \frac{\sum r}{N}$$

Where r is the distance of each individual from its nearest neighbor and N is the number of individuals.

$$\bar{r}_E = \frac{1}{2\sqrt{\rho}}$$

Where ρ is the density of the observed distribution, expressed as the number of individuals per unit area. The area considered was the research area of the July 14th session.

To understand which habitat characteristics influence the presence of this species and how, a principal component analysis was performed, using nine of the surveyed variables (elevation, exposure, slope, thyme coverage, flowering plants coverage, tree-shrub coverage, maximum height of herbaceous layer, mean height of herbaceous layer, herbaceous layer coverage). This analyses was done on the standardized data, using the "pr.comp" function of Rstudio.

A t test and a Wilcoxon-Mann-Whitney test were made between the first principal component values of *P. arion* plots and the random plots, to understand if the

difference between them are significant. This analyses was also repeat for the second principal component.

The version of Rstudio used in this study is the 2023.12.0+369 "Ocean Storm".

The preliminary data of the study of this species were also used to contribute to a report on the monitoring of Rhopalocera species of community interest present in the Dolomiti Bellunesi National Park, that is reported in the appendix.

7. RESULTS

7.1. Rhopalocera fauna of Monte Grave

7.1.1. Species detected: time trend, abundance, diffusion and phenology

A total of 504 individuals and 44 species of Lepidoptera Rhopalocera were detected in the transects of Monte Grave. All the detected individuals were identified to species level, except three of the genus *Polyommatus* Latreille, 1804 and one of the genus *Pyrgus* Hübner, 1819.

The detected species belong to five families: HesperIIDae, Lycaenidae, Nymphalidae, Papilionidae and Pieridae. Nymphalidae is the most represented group, with 20 species, about 45% of those detected, followed by Lycaenidae with nine species, HesperIIDae and Pieridae both with seven species, and finally Papilionidae with only one species.

The species of which the highest number of individuals was detected, considering the total data, is *Melanargia galathea*, which also has the highest mean number of individuals, amounting to 7.5, and the highest maximum number of individuals, amounting to 27; followed by the species *Erebia aethiops* and *Coenonympha arcania* (Fig. 11). While the species of which only one individual was detected are several, i.e. *Celestrina argiolus*, *Coenonympha gardetta*, *Heteropterus morpheus* and *Issoria lathonia*.

For what concern the single transects, 253 individuals and 34 species were detected in TMT, the most frequently detected species in it were *C. arcania* and *E. aethiops*. In CPT 113 individuals and 23 species were detected, the most detected species in it was *M. galathea*. Finally, 138 individuals and 28 species were detected in CCT, the most detected species in it was *Melitaea athalia/celadussa*.

It is important to mentioning some species that were found only outside the transect standardized sessions, i.e. *Colias alfacariensis* Ribbe, 1905/*Colias hyale* (Linnaeus, 1758), *Glaucopsyche alexis* (Poda, 1761), *Lasiommata petropolitana* (Fabricius, 1787), *Lycaena phlaeas* (Linnaeus, [1760]), *Nymphalis antiopa* (Linnaeus, 1758), *Pararge aegeria* (Linnaeus, 1758) and *Polygonia c-album* (Linnaeus, 1758).

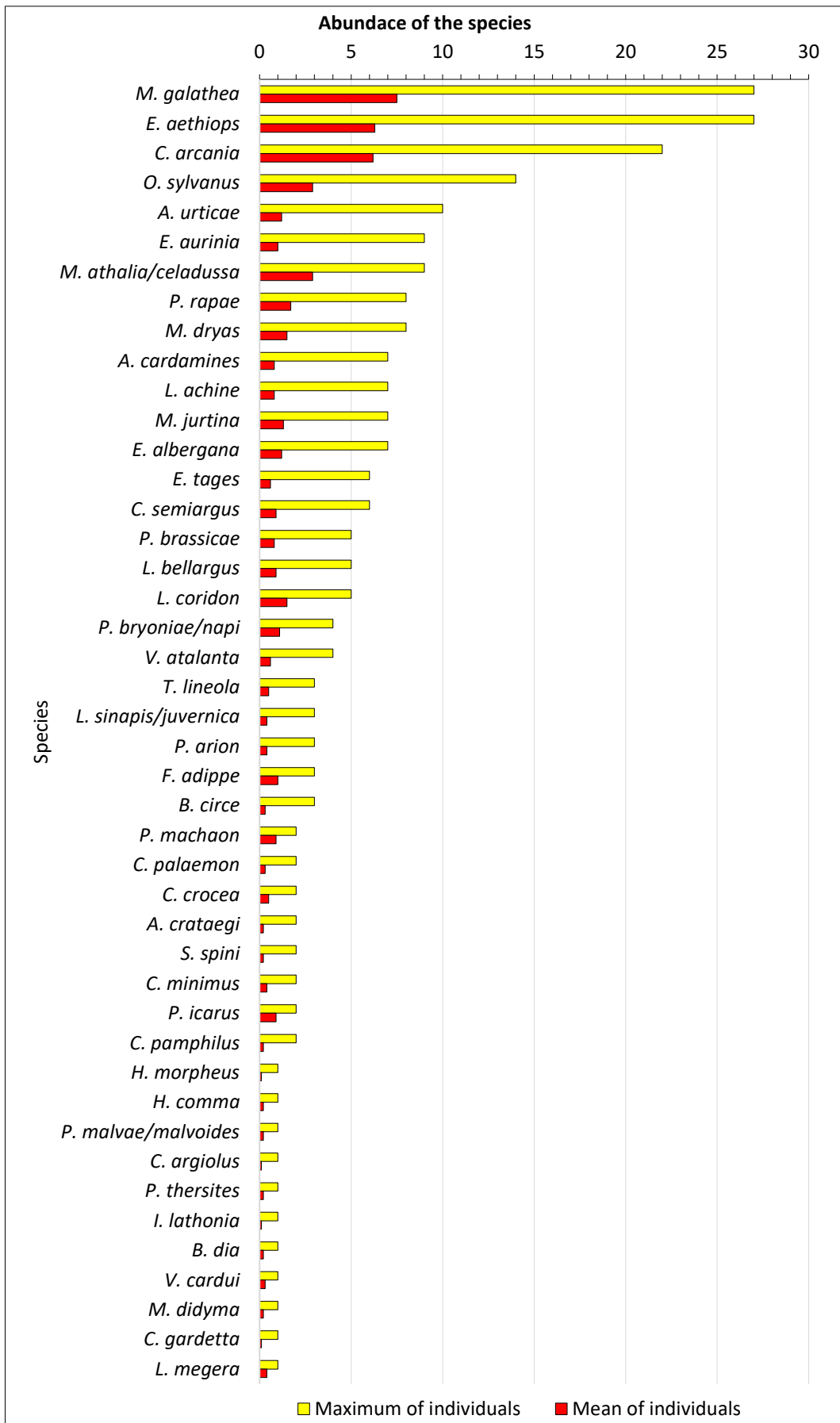


Figure 11. Maximum and mean of individuals of the species in descending order of abundance.

The trends of the number of individuals and the number of species during the flight season are similar. Starting on May 25th, there is a slight decline on June 6th, followed by an increase until July 4th, after which there is a more pronounced decline, with an slight increase on August 23rd, followed by a further decline, as far as the number of individuals is concerned, and a more constant trend, as far as the number of species is concerned (Fig.12). The day of which the highest number of individuals and species were detected is July 4th, with 115 individuals and 21 species. Instead, the day in which the least number of individuals were detected are September 5th and September 19th, both with 19 individuals, and the day in which the least number of species were detected is August 10th, with five species. A similar trend can also be observed for the single transects (Fig. 13; Fig. 14; Fig. 15).

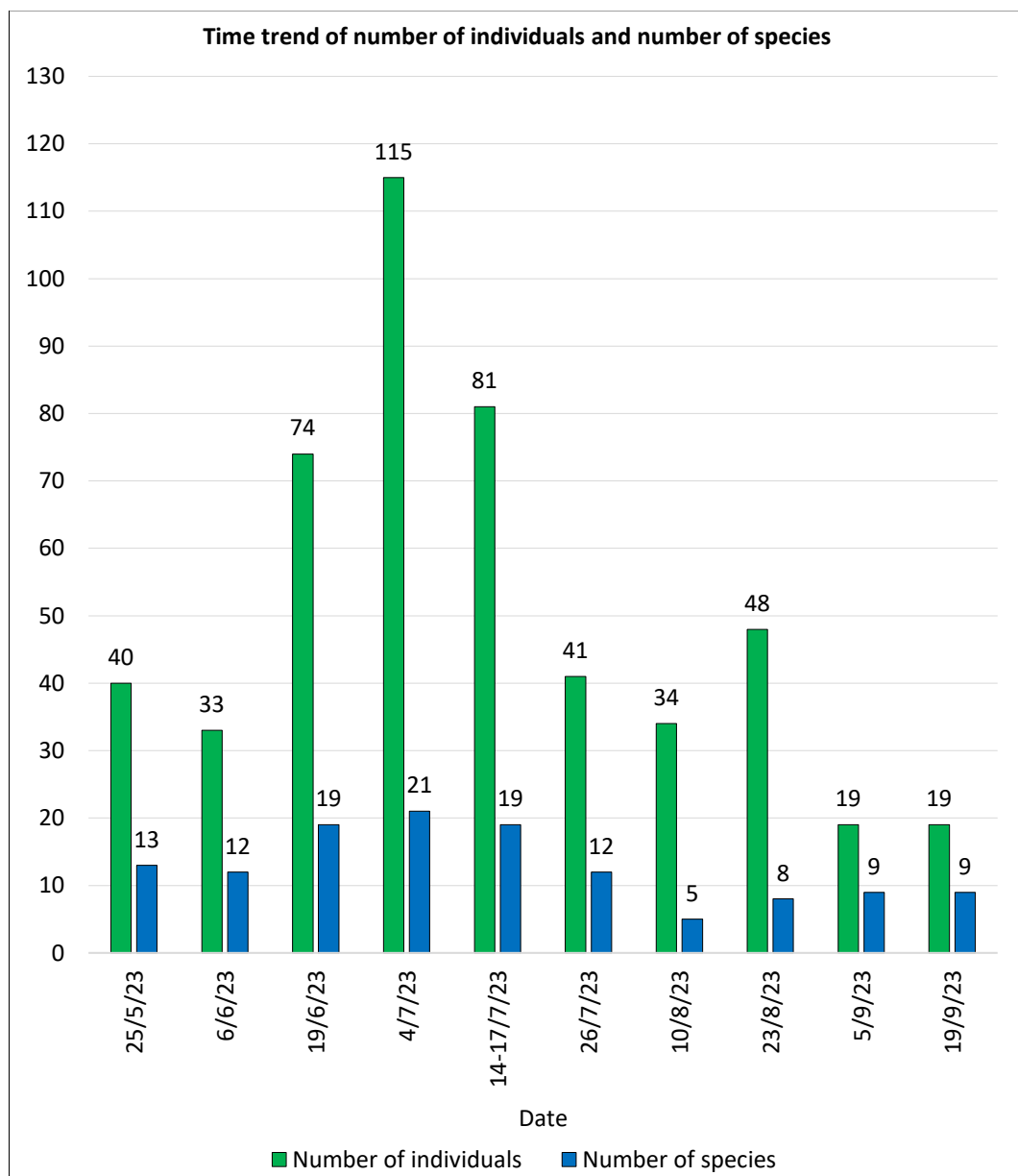


Figure 12. Time trend of the number of individuals and number of species of all the transects.

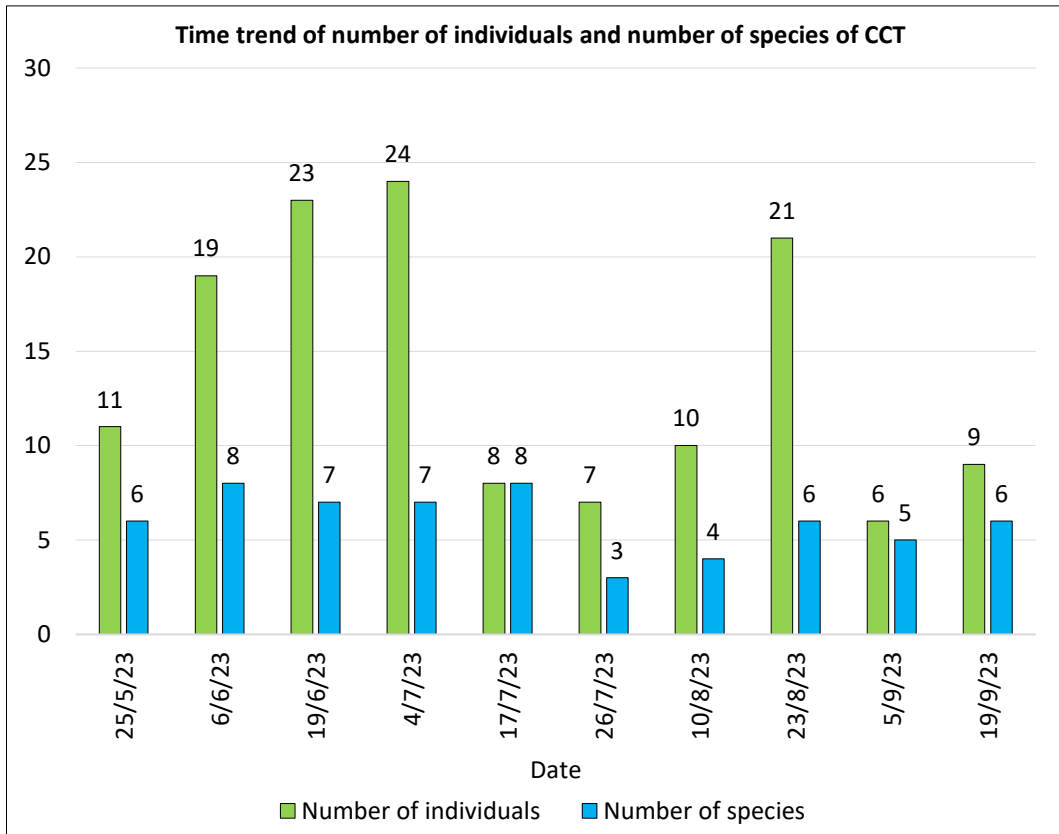


Figure 13. Time trend of number of individuals and number of species of CCT.

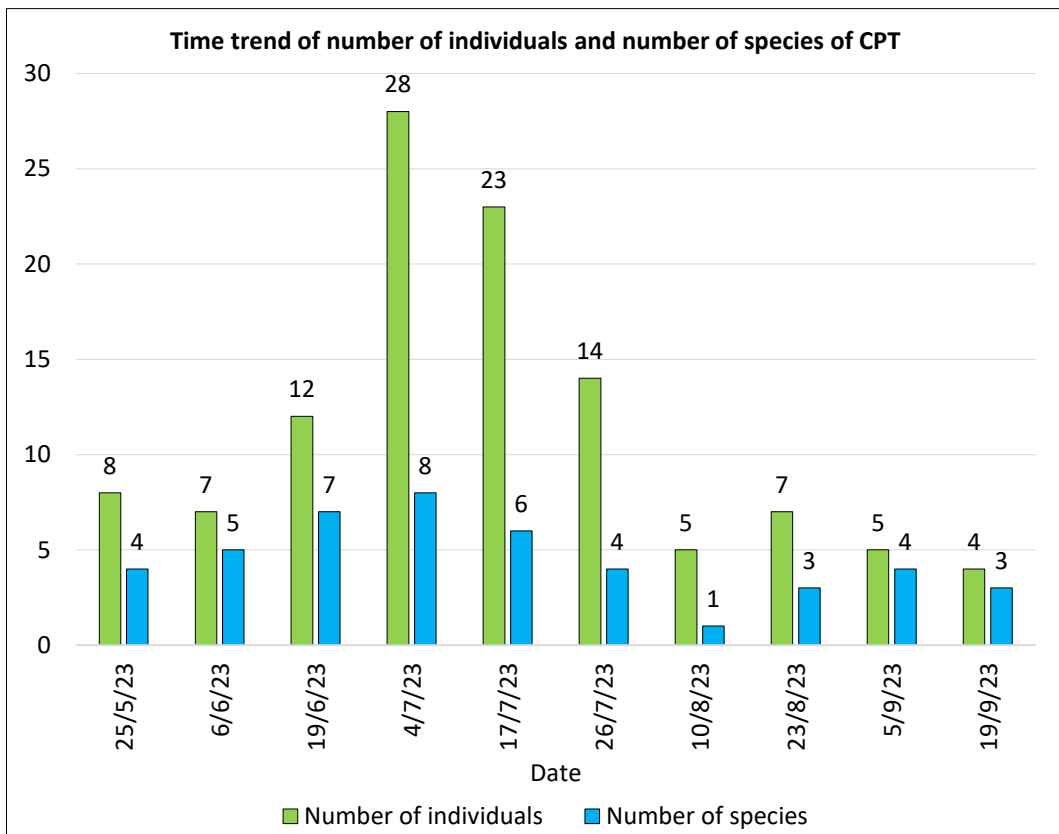


Figure 14. Time trend of number of individuals and number of species of CPT.

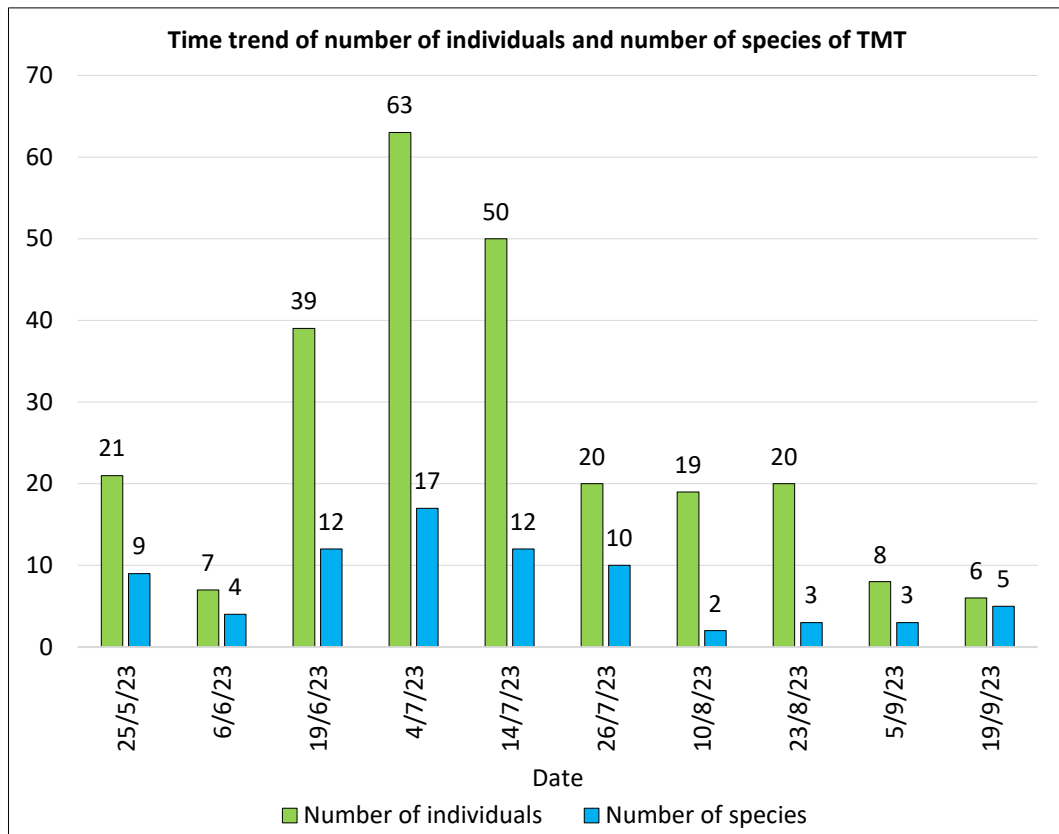


Figure 15. Time trend of number of individuals and number of species of TMT.

Regarding the diffusion of the species throughout the entire study area of Monte Grave, about 32% were detected in all the transects, 29% in two transects and 39% in only one. Most of the species detected in all transects belonged to Nymphalidae family, such as *Aglais urticae*, *C. arcania* and *E. aethiops*; but also to Hesperidae, such as *Erynnis tages*, Pieridae, such as *Pieris rapae* and only one species of Lycaenidae, i.e. *Lysandra bellargus*. Most of the species detected in only one transect belong to Lycaenidae family, such as *Cupidus minimus*, *Cyaniris semiargus* and *Polyommatus icarus*; but also to Nymphalidae, such as *Boloria dia*, *Brintesia circe* and *Erebia albergana*; to Hesperidae, such as *Carterocephalus palaemon*; and to Pieridae, such as *Anthocharis cardamines*.

The species detected that, for their ecological and phenological characteristics, were considered to be potentially present in the study area with populations that reproduce are 34, about 77% of the total species detected. The species *C. argiolus*, *C. gardetta*, *H. morpheus* and *I. lathonia* were excluded because only one individual of each was detected throughout the flight season. *B. dia* is less frequent at the altitude considered in this study (Bonato *et al.*, 2014), furthermore, the period when individuals were detected (only in September) does not suggest the possibility that it reproduce in this area. *B. circe* is more common at lower altitude, with single individuals occurring above 1000 m a.s.l. (Bonato *et al.*, 2014). *Colias crocea* was excluded because this species reproduce preferentially in the plains, has a remarkable flight ability and tendency to dispersion (Bonato *et al.*, 2014).

Lasiommata megera is a species that becomes rarer as it rises in altitude (Bonato *et al.*, 2014) and therefore the detections made could be of isolated individuals. *Vanessa atalanta* and *Vanessa cardui* are species with remarkable dispersion ability and whose reproduction is known mainly in the plains (Bonato *et al.*, 2014).

For species that potentially reproduce in the study area, when possible, some aspects of their phenology were defined. The earliest species, those that were detected since the beginning of transects surveys, are nine. Some examples of them are *A. urticae*, *A. cardamines*, *C. palaemon*, *Papilio machaon* and *P. icarus*. For 14 species, which is the majority, the flight period began in June, while for five species in July. The later species, which started their flight period in August, are two, i.e. *Hesperia comma* and *Minois dryas*. While the end of the flight period for five species was in June, for 13 in July, for four in August and, finally, for eight it extended until September.

The duration of the flight period was very variable among the species. Some species were found in only two successive sessions, such as *A. urticae*, *C. palaemon* (both on May 25th and June 6th), *P. arion* (on July 4th and July 14th) and *Thymelicus lineola* (on July 4th and July 17th). All the other species had a wider flight period, some were detected more continuously while others were detected more discontinuously. Only the species *P. icarus* was reported to have a flight period extended from May 25th to September 19th, which is the entire period of the surveys. Only one detection was reported for four species, i.e. *Aporia crataegi* (on July 14th), *Coenonympha pamphilus* (on June 6th), *E. tages* (on May 25th) and *Satyrium spini* (on July 4th).

Regarding the number of annual generations completed in the study area, 27 species are found to have only one generation and seven species two or more generations. Among the species that could have two generations, some had a trend easier to interpret, such as *M. athalia/celadussa*, while others, due to the few data collected, are more difficult to define. *L. juvernica/sinapis* was found in two surveys, one individual on June 19th and three individuals on August 23rd. Given the time interval between the detections and what is known in the literature about this species, the individuals from the two detections were considered to be of two distinct generations. The same reasons also led to *L. bellargus*, *P. malvae/malvoides* and *P. bryoniae/napi* being considered bivoltine. The detection of individuals of *P. machaon* has been enough continuous over time and does not show definite peaks. There are two periods when the number of individuals was higher, i.e. in late June and late July, given the temporal proximity between them it is difficult to be certain that the individuals belong to different generations, so it can be assumed that in the study area this species completes one or two generations. Also *P. icarus* was detected relatively continuously during the survey period, the trend with a peak in late May and in middle September allows to assume that there are two generations, however the presence of some isolated detections in late July could be due to a third generation. Species that were detected only once were considered univoltine because the period of detection was congruent with the known period of maximum

concentration of flying individuals for the Veneto region at the study area elevations.

The abundance, diffusion and phenology aspects of each species detected are reported in the table 2.

Species	TNI	MX	MN	F	CCT	CPT	TMT	FP	R	NAG
Papilionidae										
<i>Papilio machaon</i> Linnaeus, 1758	9	2	0.9	2	-	X	X	25.V-23.VIII	X	1-2
Hesperiidae										
<i>Heteropterus morpheus</i> (Pallas, 1771)	1	1	0.1	1	-	-	X	-	-	-
<i>Carterocephalus palaemon</i> (Pallas, 1771)	3	2	0.3	1	-	-	X	25.V-6.VI	X	1
<i>Ochlodes sylvanus</i> (Esper, 1777)	29	14	2.9	3	X	X	X	19.VI-26.VII	X	1
<i>Hesperia comma</i> (Linnaeus, 1758)	2	1	0.2	1	X	-	-	23.VIII-19.IX	X	1
<i>Thymelicus lineola</i> (Ochsenheimer, 1808)	5	3	0.5	2	X	X	-	4.VII-17.VII	X	1
<i>Erynnis tages</i> (Linnaeus, 1758)	6	6	0.6	3	X	X	X	25.V	X	1
<i>Pyrgus malvae</i> (Linnaeus, 1758)/ <i>Pyrgus malvoides</i> (Elwes & Edwards, 1897)	2	1	0.2	2	X	-	X	6.VI-26.VII	X	2
Pieridae										
<i>Leptidea juvernica</i> Williams, 1946/ <i>Leptidea sinapis</i> (Linnaeus, 1758)	4	3	0.4	2	X	X	-	19.VI-23.VIII	X	2
<i>Colias crocea</i> (Geoffroy, 1785)	5	2	0.5	2	X	-	X	-	-	-
<i>Aporia crataegi</i> (Linnaeus, 1758)	2	2	0.2	1	-	-	X	14.VII	X	1
<i>Pieris brassicae</i> (Linnaeus, 1758)	8	5	0.8	2	X	-	X	19.VI-17.VII	X	1
<i>Pieris rapae</i> (Linnaeus, 1758)	17	8	1.7	3	X	X	X	25.V-17.VII	X	1
<i>Pieris bryoniae</i> (Hübner, [1806])/ <i>Pieris napi</i> (Linnaeus, 1758)	11	4	1.1	3	X	X	X	19.VI-19.IX	X	2
<i>Anthocharis cardamines</i> (Linnaeus, 1758)	8	7	0.8	1	-	-	X	25.V-19.VI	X	1
Lycaenidae										
<i>Satyrrium spini</i> ([Denis & Schiffermüller], 1775)	2	2	0.2	1	-	-	X	4.VII	X	1
<i>Celastrina argiolus</i> (Linnaeus, 1758)	1	1	0.1	1	-	-	X	-	-	-
<i>Phengaris arion</i>	4	3	0.4	1	-	-	X	4.VII-14.VII	X	1

(Linnaeus, 1758)										
<i>Cupido minimus</i> (Fuessly, 1775)	4	2	0.4	1	-	-	X	16.VI-4.VII	X	1
<i>Cyaniris semiargus</i> (Rottemburg, 1775)	9	6	0.9	1	-	-	X	4.VII-26.VII	X	1
<i>Lysandra bellargus</i> (Rottemburg, 1775)	9	5	0.9	3	X	X	X	6.VI-19.IX	X	2
<i>Lysandra coridon</i> (Poda, 1761)	15	5	1.5	2	X	-	X	4.VII-10.VIII	X	1
<i>Polyommatus thersites</i> (Cantener, 1835)	2	1	0.2	2	X	-	X	25.V.-19.VI	X	1
<i>Polyommatus icarus</i> (Rottemburg, 1775)	9	2	0.9	1	X	-	-	25.V-19.IX	X	2-3
Nymphalidae										
<i>Issoria lathonia</i> (Linnaeus, 1758)	1	1	0.1	1	X	-	-	-	-	-
<i>Fabriciana adippe</i> ([Denis & Schiffermüller], 1775)	10	3	1	3	X	X	X	19.VI-26.VII	X	1
<i>Boloria dia</i> (Linnaeus, 1767)	2	1	0.2	1	X	-	-	-	-	-
<i>Vanessa cardui</i> (Linnaeus, 1758)	3	1	0.3	3	X	X	X	-	-	-
<i>Vanessa atalanta</i> (Linnaeus, 1758)	6	4	0.6	2	-	X	X	-	-	-
<i>Aglais urticae</i> (Linnaeus, 1758)	12	10	1.2	3	X	X	X	25.V-6.VI	X	1
<i>Euphydryas aurinia</i> (Rottemburg, 1775)	10	9	1	1	-	-	X	25.V-19.VI	X	1
<i>Melitaea didyma</i> (Esper, 1778)	2	1	0.2	2	X	X	-	6.VI-4.VII	X	1
<i>Melitaea athalia</i> (Rottemburg, 1775)/ <i>Melitaea celadussa</i> Fruhstorfer, 1910	29	9	2.9	3	X	X	X	25.V-5.IX	X	2
<i>Coenonympha pamphilus</i> (Linnaeus, 1758)	2	2	0.2	2	X	X	-	6.VI	X	1
<i>Coenonympha gardetta</i> (Prunner, 1798)	1	1	0.1	1	-	-	X	-	-	-
<i>Coenonympha arcania</i> (Linnaeus, [1760])	62	22	6.2	3	X	X	X	6.VI-10.VIII	X	1
<i>Lopinga achine</i> (Scopoli, 1763)	8	7	0.8	3	X	X	X	19.VI-4.VII	X	1
<i>Lasiommata megera</i> (Linnaeus, 1767)	4	1	0.4	2	-	X	X	-	-	-
<i>Melanargia galathea</i> (Linnaeus, 1758)	75	27	7.5	3	X	X	X	19.VI-26.VII	X	1
<i>Minois dryas</i> (Scopoli, 1763)	15	8	1.5	3	X	X	X	23.VIII-19.IX	X	1

<i>Brintesia circe</i> (Fabricius, 1775)	3	3	0.3	1	-	X	-	-	-	-
<i>Maniola jurtina</i> (Linnaeus, 1758)	13	7	1.3	2	X	X	-	19.VI-5.IX	X	1
<i>Erebia albergana</i> (Prunner, 1798)	12	7	1.2	1	-	-	X	19.VI-26.VII	X	1
<i>Erebia aethiops</i> (Esper, 1777)	63	27	6.3	3	X	X	X	26.VII-5.IX	X	1

Table 2. Species detected and relative total number of individuals (TNI), maximum number of individuals (MX), mean number of individuals (MN), frequency among the transects (F), presence in which transects (CCT, CPT and TMT), flight period (FP), reproduction (R) and number of annual generation (NAG); the species that probably do not reproduce in the study area are marked in yellow.

7.1.2. Differences between areas of Monte Grave

Regarding the diversity among the three areas surveyed in Monte Grave, considering the totality of the species, the first element that can be consider is the number of species. TMT result to be the transect with the highest number, amounting to 34 species, followed by CCT, with 28 species, and finally CPT, with 23 species.

The difference between the indexes of the transects is very small. However, Shannon index and Simpson index also show the highest values for TMT and the lowest values for CPT (Tab. 3; Tab. 4). Species that were only found in TMT and not in the other transects were 12, i.e. *A. cardamines*, *A. crataegi*, *C. palaemon*, *C. argiolus*, *C. gardetta*, *C. minimus*, *C. semiargus*, *E. albergana*, *E. aurinia*, *H. morpheus*, *P. arion* and *S. spini*. The species detected only in CCT were four, i.e. *B. dia*, *H. comma*, *I. lathonia* and *P. icarus*. While the species *B. circe* was the only one detected exclusively in CPT. The evenness index finds the highest value in CCT, followed by TMT and CPT (Tab. 5). In CCT, the relative abundance of species ranges from a minimum of 0.007 to a maximum of 0.188. In particular, for nine species out of 28, only one individual was detected in this transect and thus they have the lowest relative abundance. While the most abundant species was *M. galathea*, with 22 individuals, and *M. athalia/celadussa*, with 26 individuals out of 138, which together account for about 35% of the individuals found in CCT. In CPT, the relative abundance of species ranges from a minimum of 0.008 to a maximum of 0.348. For six species, only one individual was detected in this area, while for the rest of the species less than 15 individuals were found, except for the species *M. galathea*, with 39 individuals detected, which alone accounts for 34.5% of the total number of individuals of this transect. In TMT, the relative abundance of species ranges from a minimum of 0.003 to a maximum of 0.169. For eight species, only one individual was detected, while for 24 species less than 15 individuals were surveyed. The most abundant species in this transect were *O.*

sylvanus, with 22 individuals, *C. arcania* and *E. aethiops*, both with 43 individuals, only the latter two species account for 34% of the total number of individuals of TMT.

The 95% confidence intervals of the diversity and evenness indexes show partial overlap between transects (Fig. 16; Fig. 17; Fig. 18). In particular, in Shannon and Simpson indexes, the intervals of CCT and TMT overlap the most, while those of CPT and TMT overlap the least. The interval of CPT is the widest, while those of CCT and TMT are tighter and about the same width. The Simpson index turns out to have the tightest intervals. For the evenness index, the overlap between the intervals is even more pronounced, while their widths, as for the other indexes, turn out to be larger for CPT and similar between CCT and TMT.

Transect	H	95% confidence interval
CCT	2.787	2.498 - 2.939
CPT	2.487	2.003 - 2.757
TMT	2.925	2.611 - 3.107

Table 3. Shannon index of the totality of the species (H) and its confidence interval, for all the transects.

Transect	D	95% confidence interval
CCT	0.908	0.874 - 0.932
CPT	0.847	0.754 - 0.896
TMT	0.916	0.881 - 0.938

Table 4. Simpson index of the totality of the species (D) and its confidence interval, for all the transects.

Transect	EH	95% confidence interval
CCT	0.836	0.752 - 0.882
CPT	0.793	0.641 - 0.879
TMT	0.829	0.737 - 0.884

Table 5. Evenness index of the totality of the species (EH) and its confidence interval, for all the transects.

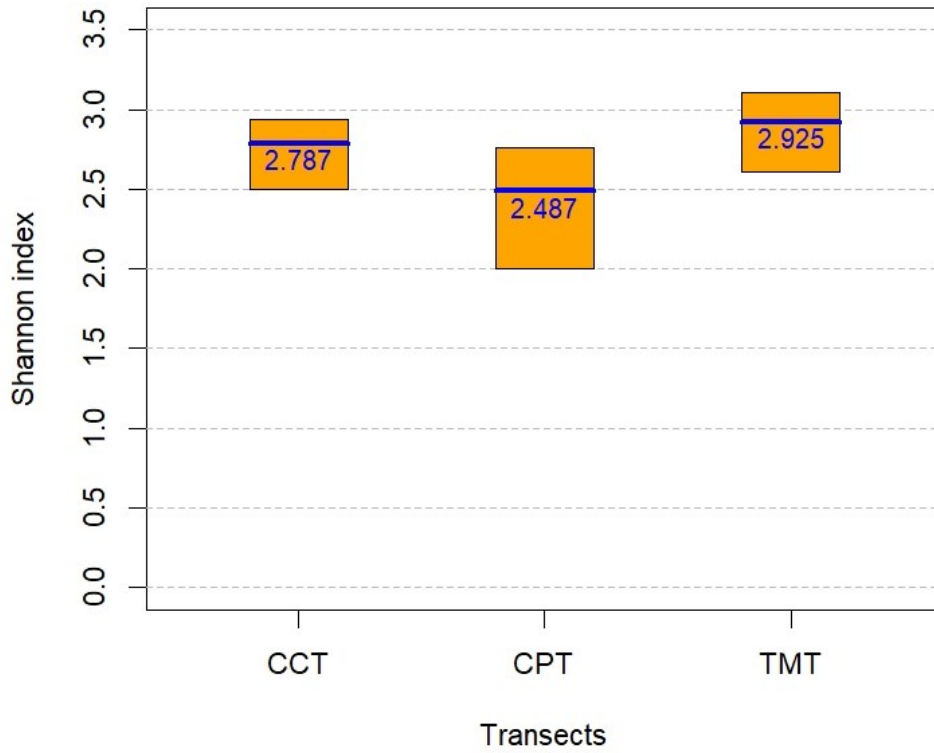


Figure 16. Value of Shannon index of the totality of the species and its 95% confidence interval, for all the transects.

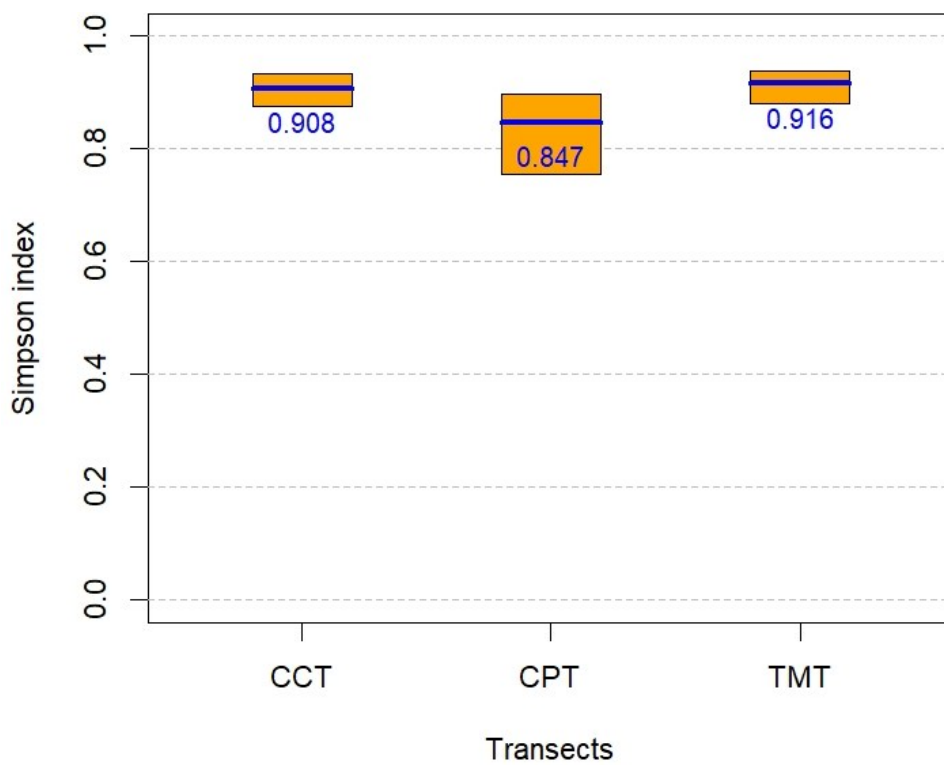


Figure 17. Value of Simpson index of the totality of the species and its 95% confidence interval, for all the transects.

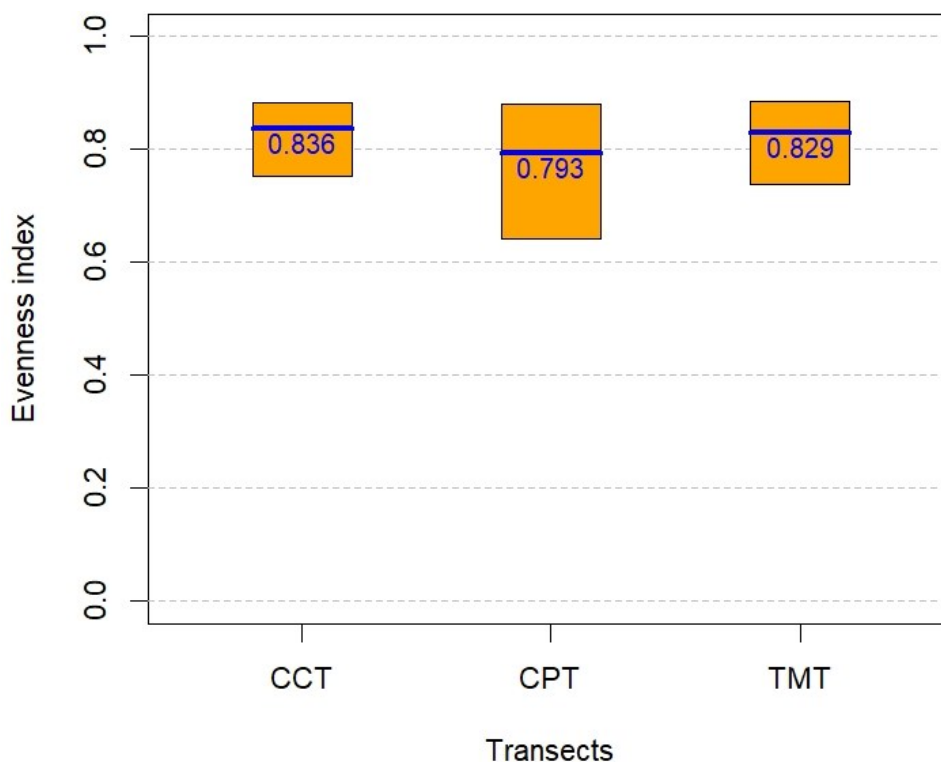


Figure 18. Value of evenness index of the totality of the species and its 95% confidence interval, for all the transects.

The Sørensen index shows that the most similar transects are CCT and CPT, while the most different transects are TMT and CCT, although the value is very close to that between CPT and TMT (Tab. 6).

There are 19 species in common between CCT and CPT, amounting to about 68% of CCT species and about 83% of CPT species. The common species include mainly members of the Nymphalidae family, e.g. *A. urticae*, *C. arcania* and *E. aethiops*; but also Hesperidae, i.e. *E. tages*, *O. sylvanus* and *T. lineola*; and Pieridae, i.e. *P. bryoniae/napi* and *P. rapae*. There are 17 species in common between CPT and TMT, amounting to about 74% of CPT species and about 50% of TMT species. In these transects, the species in common belong, not only to the families just mentioned, but also to Lycaenidae, i.e. *L. bellargus*, and Papilionidae, i.e. *P. machaon*. Finally, there are 19 species in common between TMT and CCT, amounting to about 56% of TMT species and about 68% of CCT species. Again, many of the species in common are members of the Nymphalidae family, e.g. *F. adippe*, *L. achine* and *M. galathea*; but also Pieridae, i.e. *C. crocea*, *L. juvernical/sinapis* and all the species detected of the genus *Pieris* Schrank, 1801; Hesperidae, i.e. *E. tages*, *O. sylvanus* and *P. malvae/malvoides*; and Lycaenidae, i.e. *L. bellargus*, *L. coridon* and *P. thersites*.

The 95% confidence intervals of the Sørensen index also show overlap, in particular between CPT-TMT and TMT-CCT (Fig. 19). Unlike the other indexes, all the intervals appear to have about the same width.

Transect	CS	95% confidence interval
CCT-CPT	0.745	0.620 - 0.894
CPT-TMT	0.596	0.465 - 0.752
TMT-CCT	0.612	0.490 - 0.769

Table 6. Sørensen index of the totality of the species (CS) between transects and its confidence interval.

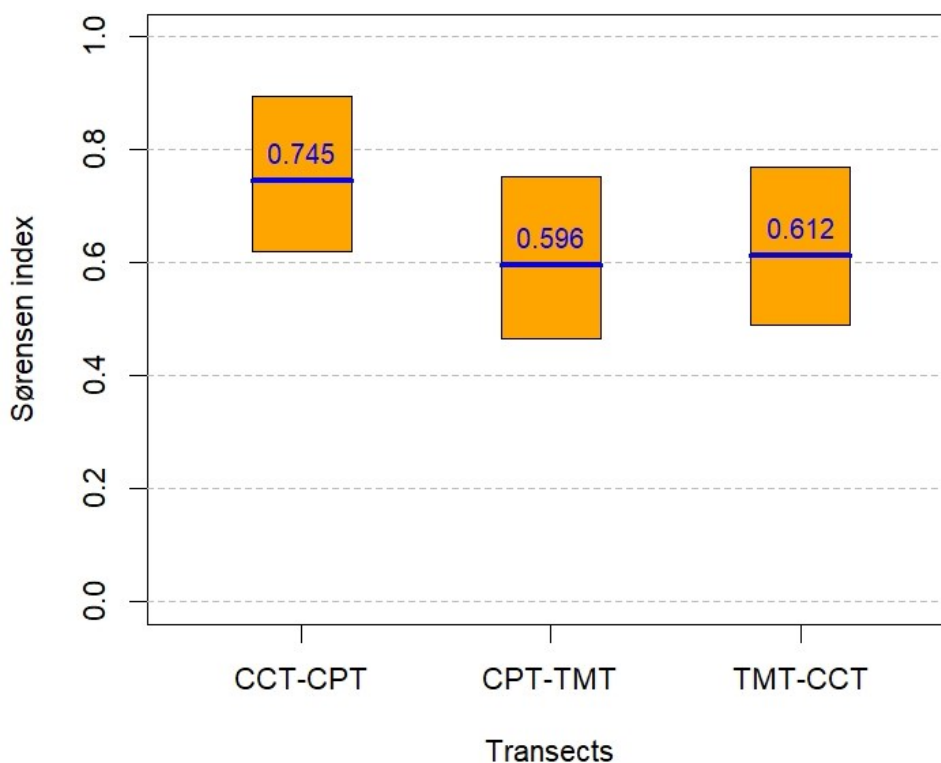


Figure 19. Value of Sørensen index of the totality of the species and its 95% confidence interval, between all the transects.

Considering only the species that potentially reproduce in the study area, the number of the species detected in CCT is 24 (86% of all the species detected in this transect), in CPT is 19 (83% of all the species detected in this transect), and in TMT is 27 (79% of all the species detected in this transect).

The results of the diversity indexes are very similar of those calculated for the totality of the species, but a little bit lower. The Shannon and Simpson indexes have higher values for TMT, followed by CCT and CPT (Tab. 7; Tab. 8). The evenness index has an higher value for TMT, instead of CCT like for the totality of the species, (Tab. 9) followed by CCT and CPT.

The 95% confidence interval of the diversity and evenness indexes show partial overlap between transects, in the same way as the intervals of the totality of the species (Fig. 20; Fig. 21; Fig. 22). Also the width of the intervals results very similar to other ones.

Transect	HR	95% confidence interval
CCT	2.655	2.391 - 2.835
CPT	2.300	1.821 - 2.555
TMT	2.781	2.486 - 2.964

Table 7. Shannon index of the species that potentially reproduce in the study area (HR) and its confidence interval, for all the transects.

Transect	DR	95% confidence interval
CCT	0.899	0.864 - 0.930
CPT	0.824	0.720 - 0.889
TMT	0.908	0.871 - 0.931

Table 8. Simpson index of the species that potentially reproduce in the study area (DR) and its confidence interval, for all the transects.

Transect	EHR	95% confidence interval
CCT	0.835	0.752 - 0.892
CPT	0.781	0.618 - 0.867
TMT	0.843	0.754 - 0.899

Table 9. Evenness index of the species that potentially reproduce in the study area (EHR) and its confidence interval, for all the transects.

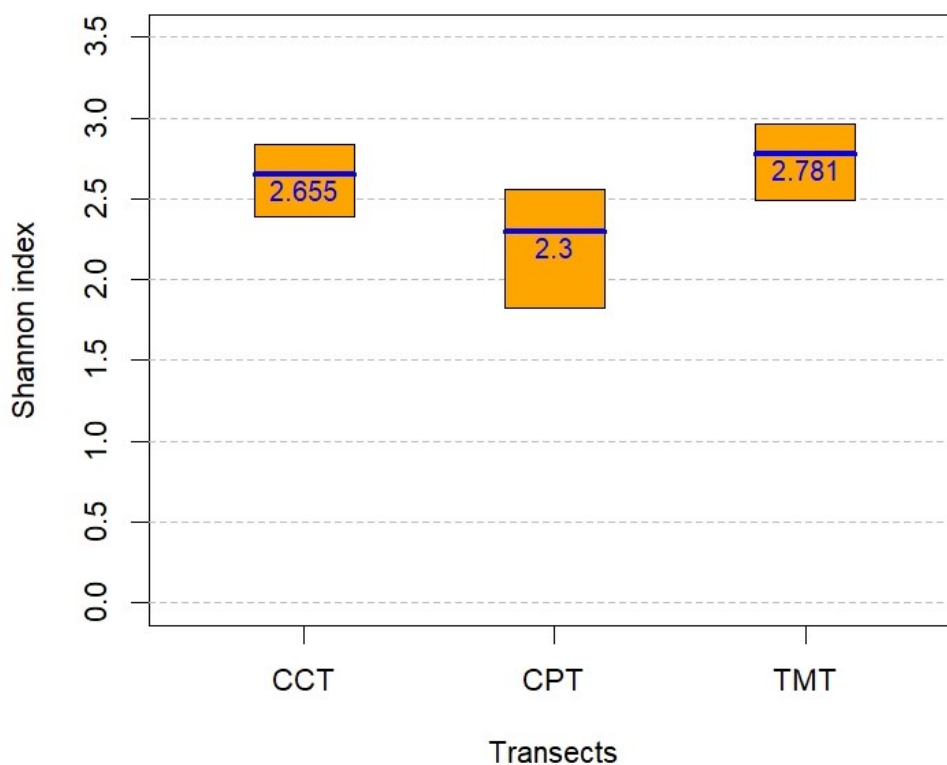


Figure 20. Value of Shannon index of the species that potentially reproduce in the study area and its 95% confidence interval, for all the transects.

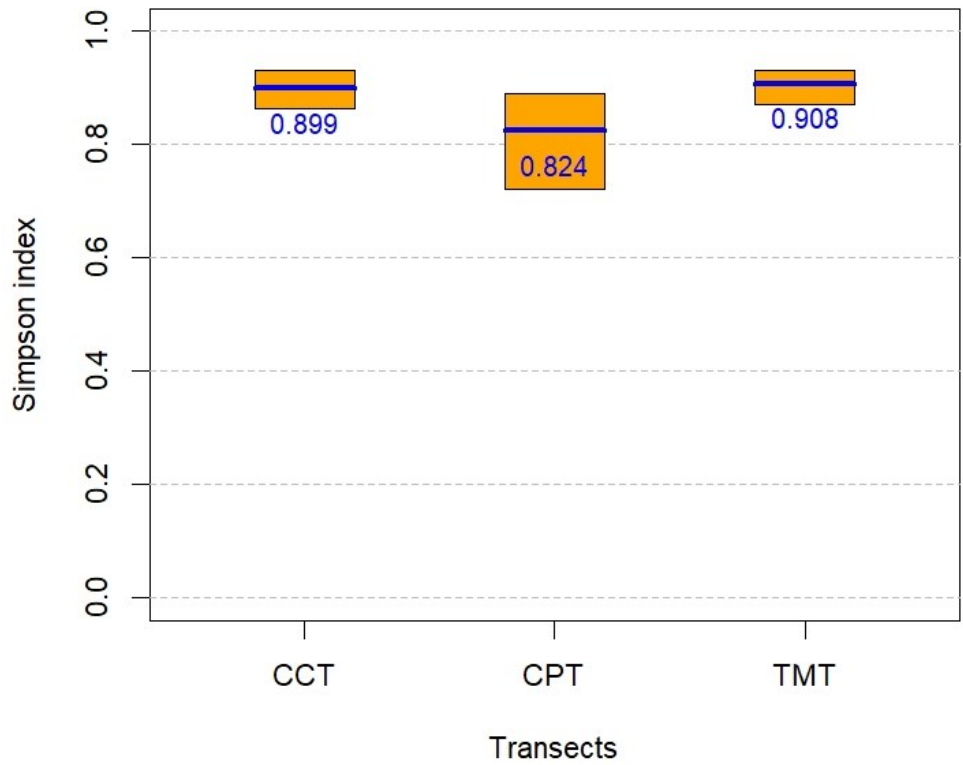


Figure 21. Value of the Simpson index of the species that potentially reproduce in the study area and its 95% confidence interval, for all the transects.

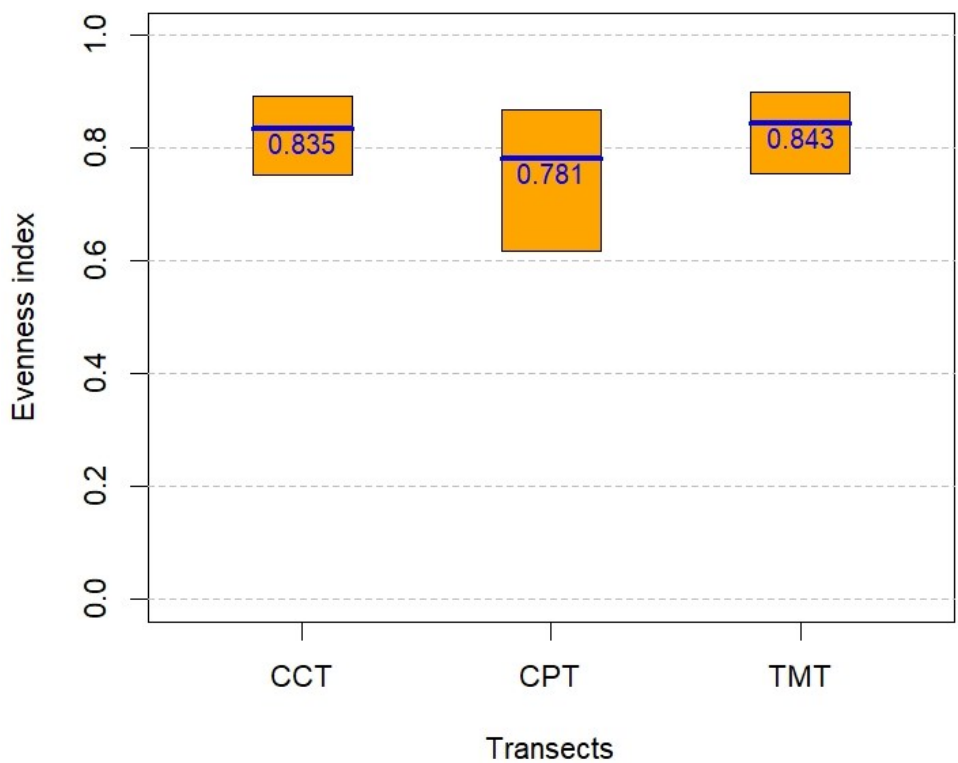


Figure 22 . Value of evenness index of the species that potentially reproduce in the study area and its 95% confidence interval, for all the transects.

Considering only the species that potentially reproduce, the Sørensen index shows the highest value for TMT-CCT and the lowest for CPT-TMT (Tab. 10), the values are a little bit higher than those of the totality of the species.

The 95% confidence intervals of the Sørensen index also show overlap, in particular between CPT-TMT and TMT-CCT (Fig. 23), as for the totality of the species.

Transect	CSR	95% confidence interval
CCT-CPT	0.837	0.719 - 0.974
CPT-TMT	0.608	0.451 - 0.796
TMT-CCT	0.666	0.526 - 0.811

Table 10. Sørensen index of the species that potentially reproduce in the study area (CSR) and its confidence interval, between all the transects.

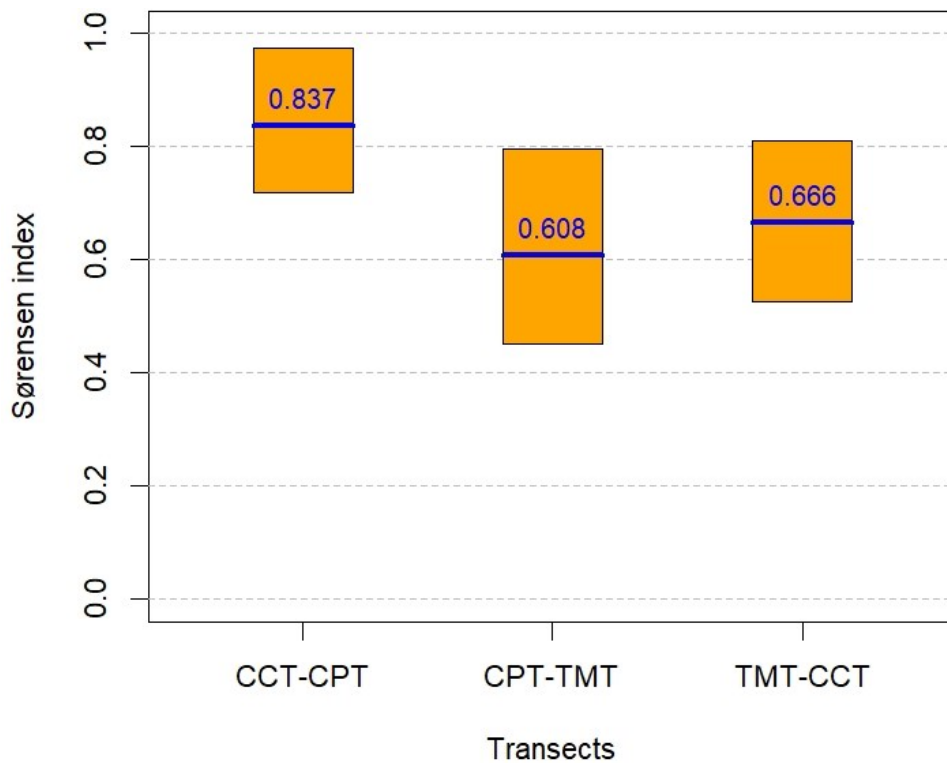


Figure 23. Value of Sørensen index of the species that potentially reproduce in the study area and its 95% confidence interval, between all the transects.

The analyses of the four subsamples of TMT shows that the first has the highest number of species, and the fourth has the lowest number (Tab. 11). The mean of all the subsamples of TMT is 19.5.

The species that have been detected only in the first subsample are five: *H. morpheus*, *L. achine*, *M. dryas*, *P. malvae/malvoies* and *V. cardui*; in the second subsample are two: *C. gardetta* and *M. athalia/celadussa*; in the third subsample are two: *C. argiolus* and *S. spini*; and finally in the fourth subsample are two: *A. crataegi* and *P. thersites*. Instead, the species that have been detected in all the

subsamples are seven: *A. urticae*, *A. cardamines*, *C. arcania*, *E. aethiops*, *M. galathea*, *O. sylvanus* and *P. coridon*.

Subsamples	SS1	SS2	SS3	SS4
Number of species	26	17	19	16

Table 11. Number of species of each subsamples of TMT.

7.1.3 Temporal analysis

In the surveys of the Rhopalocera fauna performed in the Monte Grave in 2013-2014 were detected 35 species, in 2018-2019 were detected 34 species, while during the surveys of the present study 37 species were detected, considering only those detected in the areas and in the time interval involved in this analysis.

The Sørensen index calculated between the species detected in the biennia and those detected in 2023 shows intermediate and similar values both for 2013-2014 and 2018-2019, with confidence intervals of almost the same width (Tab. 12).

Comparing 2023 data with 2013-2014 data, 20 species were detected both years, some examples of members of different families are *B. circe*, *L. coridon*, *O. sylvanus* and *P. rapae*. Seven species were detected only in 2013-2014, such as *Boloria euphrosyne*, *Phengaris alcon* and *Pyrgus alveus*. Comparing 2023 data with 2018-2019 data, 21 species were detected both years, e.g. *F. adippe*, *T. lineola* and *A. cardamines*. Ten species were detected only in 2018-2019, such as *Erebia ligea*, *Hamearis lucina* and *Nymphalis polychloros*. The species that were detected in the surveys of this study and in both biennia are 13, e.g. *P. machaon*, *P. brassicae*, *P. arion*, *C. arcania*. Species that were detected, at least in one year, during past surveys but were not in this study, taking into account only the data surveyed in the areas involved in this analyses, are 18. Species that, instead, were detected in this study but were not in the past surveys are 10 (Tab. 13). Furthermore, it can not be excluded that the unidentified *Pyrgus* individual detected during this study is an individual of *P. alveus*.

Biennia	Number of species	Sørensen index	95% confidence interval
2013-2014	35	0.547	0.420 - 0.701
2018-2019	35	0.558	0.424 - 0.698

Table 12. Number of species, Sørensen index between 2023 and 2013-2014 data, Sørensen index between 2023 and 2018-2019 data, and the respective 95% confidence interval.

Species	2013-2014	2018-2019	2023
Papilionidae			
<i>Iphiclides podalirius</i> (Linnaeus, 1758)	X	X	-
<i>Papilio machaon</i> Linnaeus, 1758	X	X	X
<i>Parnassius apollo</i> (Linnaeus, 1758)	X	X	-
Hesperiidae			
<i>Heteropterus morpheus</i> (Pallas, 1771)	-	X	X
<i>Carterocephalus palaemon</i> (Pallas, 1771)	-	-	X
<i>Ochlodes sylvanus</i> (Esper, 1777)	X	-	X
<i>Thymelicus sylvestris</i> (Poda, 1761)	X	X	-
<i>Thymelicus lineola</i> (Ochsenheimer, 1808)	-	X	X
<i>Erynnis tages</i> (Linnaeus, 1758)	-	X	-
<i>Pyrgus malvae</i> (Linnaeus, 1758)/ <i>Pyrgus malvoides</i> (Elwes & Edwards, 1897)	-	-	X
<i>Pyrgus alveus</i> (Hübner, [1803])	X	-	-
Pieridae			
<i>Leptidea juvernica</i> Williams, 1946/ <i>Leptidea sinapis</i> (Linnaeus, 1758)	-	X	X
<i>Gonepteryx rhamni</i> (Linnaeus, 1758)	X	X	-
<i>Colias crocea</i> (Geoffroy, 1785)	-	-	X
<i>Aporia crataegi</i> (Linnaeus, 1758)	-	-	X
<i>Pieris brassicae</i> (Linnaeus, 1758)	X	X	X
<i>Pieris rapae</i> (Linnaeus, 1758)	X	-	X
<i>Pieris bryoniae</i> (Hübner, [1806])/ <i>Pieris napi</i> (Linnaeus, 1758)	-	X	X
<i>Anthocharis cardamines</i> (Linnaeus, 1758)	-	X	X
Riodinidae			
<i>Hamearis lucina</i> (Linnaeus, 1758)	-	X	-
Lycaenidae			
<i>Satyrium spini</i> ([Denis & Schiffermüller], 1775)	-	-	X
<i>Celastrina argiolus</i> (Linnaeus, 1758)	-	-	X
<i>Phengaris alcon</i> ([Denis & Schiffermüller], 1775)	X	-	-
<i>Phengaris arion</i> (Linnaeus, 1758)	X	X	X
<i>Cupido minimus</i> (Fuessly, 1775)	X	X	X
<i>Cyaniris semiargus</i> (Rottemburg, 1775)	X	X	X
<i>Lysandra bellargus</i> (Rottemburg, 1775)	X	X	X
<i>Lysandra coridon</i> (Poda, 1761)	X	-	X
<i>Polyommatus thersites</i> (Cantener, 1835)	-	-	X
Nymphalidae			
<i>Issoria lathonia</i> (Linnaeus, 1758)	-	X	-
<i>Argynnis paphia</i> (Linnaeus, 1758)	X	X	-
<i>Speyeria aglaja</i> (Linnaeus, 1758)	X	-	-
<i>Fabriciana adippe</i> ([Denis & Schiffermüller], 1775)	-	X	X
<i>Boloria euphrosyne</i> (Linnaeus, 1758)	X	-	-
<i>Apatura iris</i> (Linnaeus, 1758)	X	X	-

<i>Vanessa cardui</i> (Linnaeus, 1758)	X	X	X
<i>Vanessa atalanta</i> (Linnaeus, 1758)	X	X	X
<i>Aglais urticae</i> (Linnaeus, 1758)	-	X	X
<i>Nymphalis polychloros</i> (Linnaeus, 1758)	-	X	-
<i>Euphydryas aurinia</i> (Rottemburg, 1775)	X	X	X
<i>Melitaea didyma</i> (Esper, 1778)	-	-	X
<i>Melitaea athalia</i> (Rottemburg, 1775)/ <i>Melitaea celadussa</i> Fruhstorfer, 1910	-	-	X
<i>Coenonympha pamphilus</i> (Linnaeus, 1758)	X	-	X
<i>Coenonympha gardetta</i> (Prunner, 1798)	-	-	X
<i>Coenonympha arcania</i> (Linnaeus, [1760])	X	X	X
<i>Lopinga achine</i> (Scopoli, 1763)	X	-	X
<i>Pararge aegeria</i> (Linnaeus, 1758)	X	-	-
<i>Lasiommata maera</i> (Linnaeus, 1758)	X	X	-
<i>Lasiommata megera</i> (Linnaeus, 1767)	-	-	X
<i>Melanargia galathea</i> (Linnaeus, 1758)	X	X	X
<i>Hipparchia fagi</i> (Scopoli, 1763)	X	-	-
<i>Minois dryas</i> (Scopoli, 1763)	X	X	-
<i>Brintesia circe</i> (Fabricius, 1775)	X	-	X
<i>Maniola jurtina</i> (Linnaeus, 1758)	X	X	X
<i>Erebia ligea</i> (Linnaeus, 1758)	-	X	-
<i>Erebia pluto</i> (Prunner, 1798)	X	-	-
<i>Erebia albergana</i> (Prunner, 1798)	X	X	X
<i>Erebia aethiops</i> (Esper, 1777)	X	X	X

Table 13. Species detected in the 2013-2014, 2018-2019 and 2023 surveys.

Considering the data from the two meadows separated, the one at Casera al Pos (CP) and the one at top meadows (TM), the number of species detected in 2013-2014 in CP is 20 and in TM is 27. Instead, the number of species detected in 2018-2019 in CP is 19 and in TM is 30.

The Sørensen index between 2013-2014 and 2023 data has a value a little bit higher for CP respect TM (Tab. 14), instead between 2018-2019 and 2023 data the values are equal between the two areas (Tab. 15). Furthermore, the values of the indexes in 2013-2014 are a little bit lower respect those of 2018-2019.

2013-2014	Number of species	Sørensen index	95% confidence interval
CP	20	0.487	0.294 - 0.697
TM	27	0.432	0.245 - 0.622

Table 14. Number of species, Sørensen index between 2023 and 2013-2014 data for the two meadows, and the respective 95% confidence interval.

2018-2019	Number of species	Sørensen index	95% confidence interval
CP	19	0.517	0.378 - 0.688
TM	30	0.517	0.378 - 0.688

Table 15. Number of species, Sørensen index between 2023 and 2018-2019 data for the two meadows, and the respective 95% confidence interval.

7.2. *Phengaris arion* population of Monte Grave

7.2.1. Population characteristics

During the surveys conducted in the Monte Grave, the species *Phengaris arion* was detected during two sessions, on July 4th and on July 14th. A total of 10 individuals were detected, four during the surveys of TMT (three on July 4th and one on July 14th), and the remaining six during the research session conducted on July 14th.

The individuals detected were seven males, one female and of three individuals the sex was not identified. Regarding the latter, the sex was not identified analyzing the individuals in the field, like reported in the methods, and the photos were insufficient to recognize subsequently the sex, based only on wing characters, due to the low sexual dimorphism (Verity, 1943).

Individuals marked on July 14th were not recaptured, neither in the same session or in the next one. Although the individuals detected on July 4th had not been marked, comparison of the photos of the wing surface between all the individuals detected made it possible to say that they are different individuals than those detected during the session of July 14th, due to the high morphological variability among them. The elements that were useful for this distinction were the shape and size of the dark spots, the thickness of the dark band on the edge of the upper surface of the wings, and the presence of particular signs of damages in the wings.

Wing damages of the detected individuals varied widely, but on average lower values were assigned to individuals detected on July 4th (values from 1 to 2) than to those detected on July 14th (values from 1 to 4).

7.2.2. Population spatial distribution

Individuals of *Phengaris arion* detected in TMT were found mostly in the central portion of the path, while in the research area individuals were found widespread in different sectors. The research area of the July 14th session corresponds to about 7 hectares, which is about 32% of the total area of the top meadows of Monte Grave (Fig. 24).

The nearest neighbor analysis shows that the mean expected distance was 42 m, while the mean observed distance was 87 m. The greatest nearest neighbor distance between individuals was 214 m (between individual 3 and 6), while the shortest was 4 m (between individual 7 and individual 10). The density of individuals per unit area surveyed was 0.0144 per *ha*. The ratio of mean expected distance to mean observed distance had a value of 2.1, indicating a dispersion of the detected individuals. All the individuals were detected within the Dolomiti Bellunesi National Park or in the immediate proximity of its boundary.

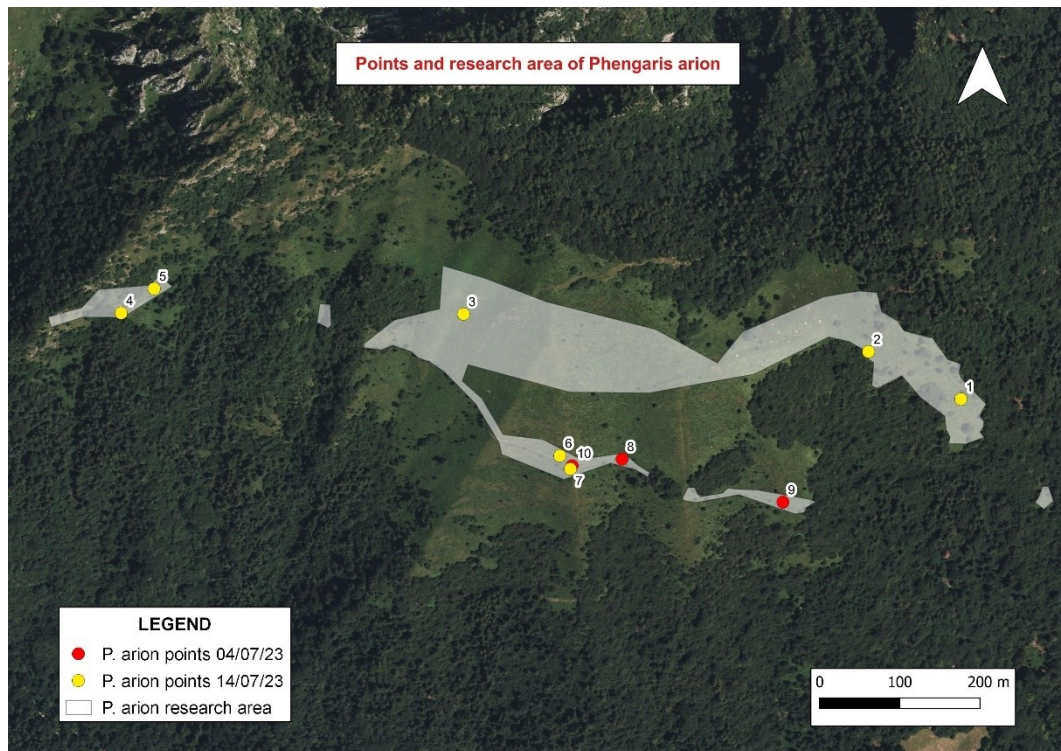


Figure 24. Points and research area of *Phengaris arion* (numbers are not in order of detection).

7.2.3. Influence of habitat variables on *Phengaris arion*

Nine plots were considered instead of 10, given the distance between point 7 and point 10 of 4 m that made the plots almost coincident. Thereby, nine random plots were established in which the species was not detected, for a total of 18 habitat plots sampled (Fig. 25).

Among the data collected in the plots, elevation ranged from a minimum of 1224 m a.s.l. to a maximum of 1454 m a.s.l., on average *P. arion* plots were at lower elevations than random plots. Exposure ranged from 98° to 220° starting from North and continuing clockwise, all the plots therefore are exposed in a range from South-East to South-West, with an higher mean for the *P. arion* plots than the random plots. The slope ranged from 7° to 38°, again with an higher mean for the *P. arion*

plots than the random plots. The thyme coverage was not found to be very high, neither in the *P. arion* plots nor in the random plots, ranging from 0% to 8%. The flowering plants coverage ranged from 1% to 20%, the mean of *P. arion* plots was 12% while that of random plots was 6%. The tree-shrub coverage ranged from a minimum of 1% to a maximum of 40%, with similar values in both *P. arion* and random plots. The maximum height of herbaceous layer ranged from 110 cm to 190 cm, while the mean height ranged from 40 cm to 100 cm; the means of these parameters for the *P. arion* plots and the random plots were very similar. The herbaceous layer coverage also had a similar mean in the two plot types, it ranged from 75% to 100%. Finally, the presence of rocks was found on four plots out of 18, including three *P. arion* plots and one random plot.

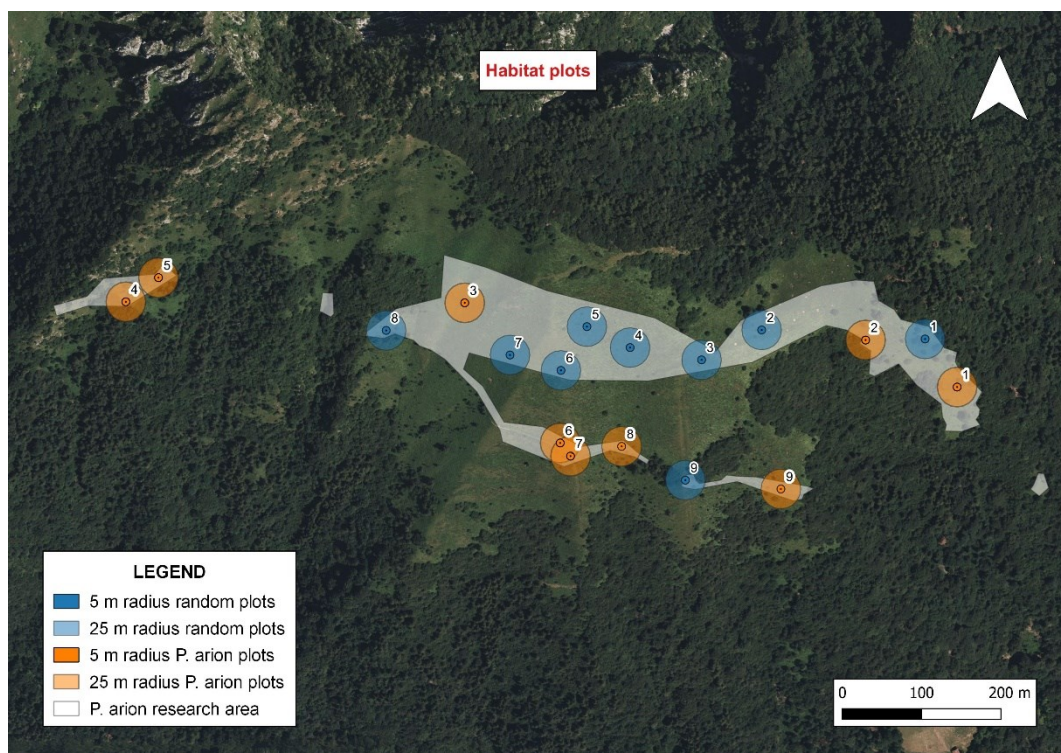


Figure 25. Random and *P. arion* habitat plots.

In the principal component analysis conducted on the nine habitat variables, the first two principal components together explain 64% of the overall variance of the 18 plots (Tab. 16).

The first principal component explains 43% of the variance and its eigenvalue is 3.911. This component is positively correlated mainly with the maximum height of herbaceous layer (factor load 0.401) and the mean height of herbaceous layer (0.442). The variables that are negatively correlated with this component are the exposure (-0.400), the slope (-0.387), the thyme coverage (-0.387) and the flowering plants coverage (-0.331).

The second principal component explains 20% of the variance and its eigenvalue is 1.862. This component is positively associated mainly with the tree-shrub coverage

(0.667) and also with the flowering plants coverage (0.404), and instead negatively correlated with the exposure (-0.340).

The other components explain less than 12% of the overall variance and possess eigenvalues slightly higher or lower than one (Fig. 26), for these reasons they were not considered.

The arrangement of the habitat plots in the first two principal components shows some plots of *P. arion* aggregated with each other (Fig. 27). In particular, plots 1, 3, 4, 5, 6, and 7 are shifted toward negative values of the first component, i.e. where the variables thyme coverage, flowering plants coverage, slope and exposure are higher; while maximum height, mean height and coverage of the herbaceous layer are lower. Instead, plot 8 is the only one of these shifted toward positive value of this component, i.e. the opposite conditions from those above. Plots 2 and 9 are close to the zero value of first component. Considering the second component, almost all the *P. arion* plots are shifted towards positive value or very close to the zero. Plots 2, 4 and 5 have the higher values than the others and thus associated especially with higher tree-arbustive coverage. Instead, the plot 8 has the lowest value on this component. The random plots show no particular aggregations and, based on the data collected, appear to be located at points with very different values of the two components. Plots 1, 2, 3 and also but less 4 and 5 are associated with positive value of the first component, and so to higher maximum height, mean height and coverage of the herbaceous layer, while lower coverage of thyme and flowering plants, exposure and slope. However, plots 6, 7, 8 and 9 are associated with negative values, and so opposite conditions. Considering the second component, the plots 1, 2, 8 and 9 are shifted towards positive values of it, and the other plots towards negative ones.

The results of this analysis may have been influenced by the data distribution of the variables.

Both t test and the Wilcoxon-Mann-Whitney test of the first principal component resulted that the difference between *P. arion* plots and random plots was not significant, the p-value of the first was 0.1639 and of the second was 0.2581. The same tests resulted not significant also for the second principal component, the p-value of t test was 0.5724, and the p-value of Wilcoxon-Mann-Whitney test was 0.4363.

	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9
Standard deviation	1.977	1.364	1.017	0.881	0.744	0.587	0.551	0.354	0.288
Proportion of variance	0.434	0.206	0.115	0.086	0.061	0.038	0.033	0.013	0.009
Cumulative proportion	0.434	0.641	0.756	0.842	0.904	0.942	0.976	0.990	1.000

Table 16. Standard deviation, proportion of variance and cumulative proportion of variance of the principal components.

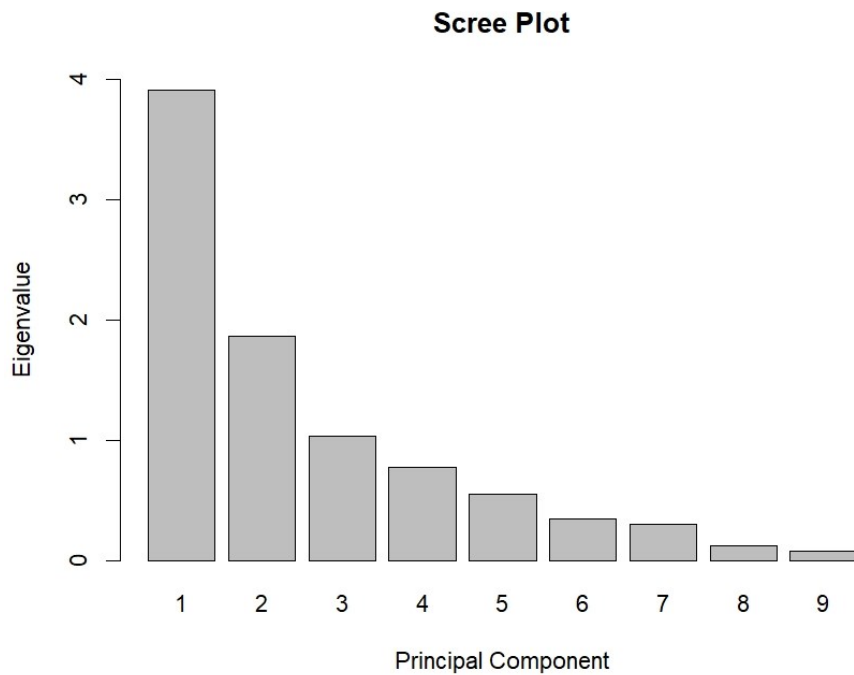


Figure 26. Scree plot of the principal component analyses.

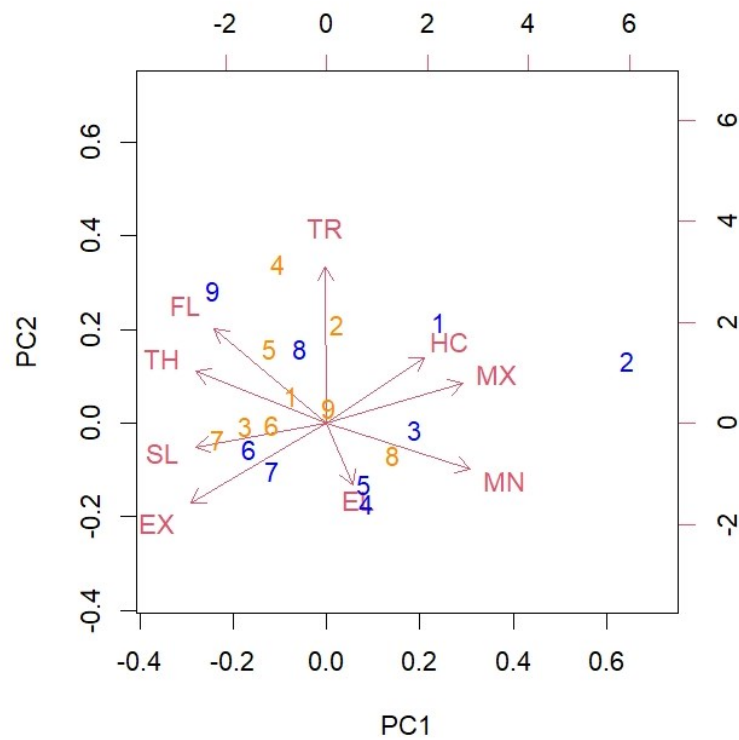


Figure 27. Biplot of the first and the second principal components (PC1 and PC2), the orange number are the *P. arion* plots and the blue number are the random plots; EL=elevation, EX=exposure, SL=slope, TH=thyme coverage, FL=flowering plants coverage, TR=tree-shrub coverage, MX=maximum height herbaceous layer, MN=mean height herbaceous layer, HC=herbaceous layer coverage.

8. DISCUSSION AND CONCLUSIONS

8.1. Rhopalocera fauna of Monte Grave

The surveys carried out in Monte Grave during this study aimed to improve the knowledge of the Lepidoptera Rhopalocera fauna of this area partially included in the Dolomiti Bellunesi National Park and, more generally, of this mountainous sector of the southeastern Alps, contributing to the Neptis project.

A total of 44 species were detected, amounting for about 41% of the species reported within the Park (Vettorazzo *et al.*, 2020), 26% of the Lepidoptera Rhopalocera fauna of Veneto region (Bonato *et al.*, 2014) and 15% of the Italian Rhopalocera fauna (Balletto *et al.*, 2014).

The number of species that were found for the different Rhopalocera families reflects the Italian situation, with a greater number of Nymphalidae species, followed by Lycaenidae, Hesperidae, Pieridae, and, finally, Papilionidae (Balletto *et al.*, 2015).

The most abundant species, considering the entire study area, was *Melanargia galathea*, (Fig. 28) which is among the most frequently reported species within the Park (Vettorazzo *et al.*, 2020). This species is most frequent between 300 and 1400 m a.s.l., and it prefers meadows with tall and dense grasses, on sloping and dry soils (Bonato *et al.*, 2014), probably finding an ideal site in the Monte Grave. Two other species of which high numbers of individuals have been detected are *Erebia aethiops* and *Coenonympha arcania*, also among the most frequently reported species within the Park (Vettorazzo *et al.*, 2020). Both are well adapted to mountainous areas and prefer meadows close to forests (Bonato *et al.*, 2014), congruent with the type of environments of the study area. Some species, however, were detected only once during the entire flight season. *Celestrina argiolus* is a more frequent species in the plains but in the Alps its reproduction is known up to 1300 m a.s.l., this lycaenid is adapted to different types of environments united by the presence of woody and shrubby areas and at the larval stage it feeds on different types of woody and herbaceous plants (Bonato *et al.*, 2014). Therefore, it would be interesting to understand whether the only detection made of this species, is due to altitudinal rarefaction or was accidentally underestimated during the surveys. *Coenonympha gardetta* lives in mountainous areas above 1200 m a.s.l., colonizing herbaceous areas, with few shrubs and trees, on partly rocky soils (Bonato *et al.*, 2014). Since this species was detected only once, it could be a dispersing individual from higher areas close to the Monte Grave, such as the adjacent Monte San Mauro, or this species may have been underestimated during sampling because of its morphological similarity to *C. arcania* (Schmitt & Besold, 2010). In the Veneto region, *Heteropterus morpheus* is mainly widespread at low elevations, preferring meadows with tall and dense grasses, in warm and moist sites (Bonato *et al.*, 2014). Given the elevations and type of environment included in this study, thus the individual detected may have been in dispersion. *Issoria lathonia* occurs over a

wide elevation range and is capable of colonizing diverse environments (Bonato *et al.*, 2014). Given the high mobility of this species, it would be interesting to understand if it only occasionally visits the Monte Grave area or if it is a more stable presence. Further surveys in this area would be useful also to confirm the presence of the species that have only been detected outside of standardized surveys, i.e. *Colias alfacariensis/hyale*, *Glaucopsyche alexis*, *Lasiommata petropolitana*, *Lycaena phlaeas*, *Nymphalis antiopa*, *Pararge aegeria* and *Polignia c-album*.

Among the species detected in this study, three are found to be on the Italian and European Red List of Butterflies and on the Annex II or IV of the Habitats Directive. *Euphydryas aurinia* is assessed as "vulnerable" at the Italian and European levels (Balletto *et al.*, 2015; Van Swaay *et al.*, 2010). This species is mainly linked to wet meadows, in the Veneto region it is more present in the northern alpine sector and most colonies are found around 2000 m a.s.l.; in the past it was also present in the plains, although in many of these sites it is extinct, probably due to habitat changes (Bonato *et al.*, 2014). Within the Dolomiti Bellunesi National Park, it is known in five sites, both in meadows subject to natural grazing by wild ungulates and also in mowing meadows actively maintained by the Park (Vettorazzo *et al.*, 2020). During the surveys of this study, it was detected only in TMT, between May 25th and June 19th, with a total of 10 individuals. *Lopinga achine* is "near threatened" at the Italian level and "vulnerable" at the European level (Balletto *et al.*, 2015; Van Swaay *et al.*, 2010). In the Veneto region, this species is widespread in mountain forests of the prealpine and alpine sectors (Bonato *et al.*, 2014). In the Park, populations of this species are numerous and substantial, currently not subject to particular pressures or threats (Vettorazzo *et al.*, 2020). During the surveys of this study, it was found in all the three transects, between June 19th and July 4th, with a total of eight individuals. Finally, the other species of conservation interest is *Phengaris arion*, which will be discussed in detail below. The presence of these butterflies in the Monte Grave area denotes the importance of its environments for the conservation of these species, both those within and outside the Park.

Regarding the temporal trend of the number of individuals, the July 4th and August 23rd sessions represent the days of highest concentration of flying adults. This bimodal trend, as mentioned earlier, can also be seen for the number of species, although in a less pronounced way. Considering the individual transects, in CCT the number of individuals undergoes a drastic decline between July 17th and 26th, this could be due to a mowing done in this meadow near these two sessions. This decline is also seen, to a lesser extent, in the number of species on July 26th. A similar situation is seen in the trend in the number of individuals and number of species of CPT on August 10th, again a recent mowing of the meadow had been annotated. Mowing of the meadows during the summer period can cause a decrease in nectar resources given by flowers, which are necessary for the foraging, consequently also affecting the abundance of butterflies (Feber *et al.*, 1996). It is important to take these observations into account when interpreting the overall trend

of the number of individuals and species in the entire study area. The second peak, at the end of August, could therefore be given by a recovery after mowings made in CCT and CPT due to the restoration of flower resources; but it could also be given by the presence of adults of the later species, such as *Minois dryas*, and the second generations of some species, such as *Melitaea athalia/celadussa*. Most likely the trend is determined by the concomitance of all these factors.

The spread of species in the study area shows a diversified situation, 32% of species were detected in all transects, 29% in two and 39% in only one. Interestingly, six of the nine species of lycaenids were detected in only one transect. This could be due that some members of this family have low mobility and forest edges with tall trees may act as a barrier for their dispersal (Kallioniemi *et al.*, 2014). Members of other families, were found more frequently on more than one transect. Some species of Nymphalidae and Pieridae, e.g., are more mobile and more prone to dispersal (Kallioniemi *et al.*, 2014). These landscape-level observations should be taken into account in environment management.

The 77% of the detected species potentially reproduce in the study area and so they are closely related to the environments of Monte Grave, both those outside and those within the protected area. Therefore, it is very important to safeguard these environments in order to conserve the species related to them. Regarding the phenological aspects of these species, it would be interesting in the future to anticipate the start of surveys to have a more precise estimation of the flight period of the species and detect eventually earliest generations. Most of the species were found to be univoltine. Some of these are known to have only one generation throughout their range, such as *Anthocharis cardamines*, *Cupido minimus* and *Carterocephalus palaemon*. While others, such as *Aglais urticae*, *Cyaniris semiargus*, *Erynnis tages* and *Pieris brassicae* at low altitudes develop more than one generation but at high altitudes tend to have only one (Bonato *et al.*, 2014). In fact, as altitude rises the length of the suitable season decreases and consequently the number of annual generations that butterflies are able to complete (Nylin, 2009). Further surveys could also improve knowledge of species that have been excluded, particularly those that were at the limit of their altitudinal range of reproduction, and that new data might instead suggest reproduction in situ.

Considering both the totality of the species detected and only those species that potentially reproduce in the study area, TMT appears to possess the greatest diversity in species, followed by CCT and CPT. In contrast, the evenness was higher in CCT and lower in CPT in the first case, while higher in TMT and lower in CPT in the second case. This is probably due to the fact that in the second case, many species of which only one individual had been detected were excluded, which in the first case went to lower the value of evenness. Finally, CCT and CPT were found to be the most similar transects in both cases. However, all these differences are very small, in fact the values of all the indexes are very close to each other. Consequently, there is a partial overlap of the confidence intervals of them, which shows that the differences are not statistically significant. Interestingly, CCP is

often found to have a wider interval than the other transects, indicating greater uncertainty of the parameter estimation (Whitlock & Schluter, 2010), and this could be due to its smaller sample size than the others. Despite this, it is worthwhile to make some observations about the different transects. First, about the length of the transects, which was defined according to the size of the respective meadow. While the lengths of CCT and CPT are similar, both transects in fact were located in two small meadows surrounded by forest, TMT is considerably longer, as it was located in meadows that occupy the entire summit portion of Monte Grave. Subsamples of the latter showed, on average, lower species number than the entire transect, thus suggesting that the length of the transect, and thus the size of the meadow, is a factor influencing the number of TMT species. In the study of Öckinger & Smith (2006), there is evidence of a relationship between species richness and meadow size, probably given the greater variety of sub-habitats in the latter. Therefore, it is interesting to note the variability in the number of species among different TMT subsamples, which might suggest the presence of environmental differences within the transect itself, which would be investigated with further studies. Another factor that might influence the communities of Lepidoptera Rhopalocera is the type of management. The CCT and CPT meadows were mowed during the year of the surveys for this study, in late July and middle August, respectively. The study by Feber *et al.* (1996) shows that butterfly numbers are higher in meadows that are mowed in spring or autumn, because summer cuts remove nectar resources at the time of greatest butterfly abundance, as well as having destructive effects on the eggs and larvae of many species. Furthermore, the precise methods used for mowing of these two meadows are not known. Instead, the portion of the Monte Grave top meadows that falls within the boundary of the protected area is mowed biennially in late August or early September. Postwar social and economic changes have led to a gradual abandonment of traditional agricultural practices, particularly in mountain areas (MacDonald *et al.*, 2000). The abandonment of meadow mowing practices led to the natural colonization of these by woody and shrubby species, resulting in a loss of animal and plant typical of meadow environments (MacDonald *et al.*, 2000) due to the contraction of their habitat (A.A.V.V., 2017). Within the Park, species most threatened by these changes include, e.g., several species of orchids, with regard to plant communities, and birds such as tetraonids and corncrake, with regard to animal communities (A.A.V.V., 2017). The active meadow management practices implemented by the Park, have been shown to have a positive effect on the plant communities of Monte Grave, particularly on the species of conservation interest *Gladiolus palustris* Gaudin (Cannella *et al.*, 2020). Specific studies on the effects of mowing on butterfly communities have not been conducted in the study area, but other studies have shown their positive effects on this group of animals (Bubová *et al.*, 2015; Kubo *et al.*, 2009; Stefanescu *et al.*, 2005; Skórka *et al.*, 2007). For what concerns the active management, moreover, moderate grazing appears to be less impactful than mowing on butterflies because it is more gradual and less intrusive (D'Aniello *et al.*, 2011). Otherwise, also

rotational mowing seems to be good management to butterfly communities conservation (Bubová *et al.*, 2015). Other aspects that affect *Rhopalocera* communities and should be taken into account to understand differences within the study area, are habitat quality, as vegetation height, flower abundance and richness, and landscape composition (Öckinger & Smith, 2006). Regarding the latter, in addition to the area of the meadows, their shape and connectivity should be taken into account (Sitzia *et al.*, 2018). The importance of the conservation of environments of the Monte Grave meadows and their proper management, highlights how protected areas are often confined to higher elevations, leaving out those at lower elevations with greater biodiversity (Sergio & Pedrini, 2008). Although the area involved in this study is located between 1000-1300 m a.s.l., the Park boundary in this area is still located at very high elevation, always above 1000 m a.s.l., thus excluding most of the Monte Grave environments. The study of Vettorazzo *et al.* (2020) has already shown the importance of the environments close the Park boundary for the butterfly communities, highlighting the inadequacy of the actual perimeter for their conservation.

Comparison of the surveys made in this study with those of the past surveys showed intermediate and practically coincident similarity values for both biennia, considering the entire Monte Grave area, while higher values for the 2018-2019 biennium, considering the two meadows separated. It should be emphasized that although the data were made as comparable as possible, the methodology used are not coincident and this may have affected the results of this analysis. However, it is interesting to make some considerations about some species. The species of community interest, *E. aurinia* and *P. arion* were detected during both biennia and during this study, confirming a probable stability of their populations and underscoring the importance of these environments for their conservation. The first was detected both in MGA and MGB in the past surveys, and only in TMT in surveys of this study. The second was detected only in the transects of the top meadows both in the past surveys and this study. *L. achine* was also detected during past monitoring but not with the same continuity. This species was detected only in the top meadows in the past surveys and in all the transects in this study. On other hand, the species *Parnassius apollo* was not found during this study, unlike in the two biennia. This could be due that the monitoring surveys were carried out in higher elevation portions of the study area, where there are more rocky outcrops, an ideal environment for plants of the genus *Sedum* L. and *Sempervivum* L. on which the caterpillars of this species feed (Bonato *et al.*, 2014). Further investigations will also be able to confirm the presence of the other species detected in the past surveys but not in this study.



Figure 28. *Melanargia galathea* (Damiano Sartor)

8.2. *Phengaris arion* population of Monte Grave

The species *Phengaris arion* (Fig. 29) has been known in the Monte Grave top meadows for a decade. During the surveys of the "Monitoraggio della biodiversità in ambiente alpino" project, the species has always been detected, except for 2019, with a maximum number of four individuals. The results of this study, therefore, represent the highest number of individuals detected in this area. It is important to note that the methodology used during past monitoring were different from those used in this study, and this may have affected the number of individuals detected. In the past surveys, individuals were detected between late June and late August, whereas in this study between early and middle July. Given the short flight period of this species (Bonato *et al.*, 2014), it would be interesting to carry out the surveys more frequently to study better this population.

Interestingly, almost all of the individuals whose sex was identified were males, which might suggest greater mobility of the latter. Some studies, however, have shown less mobility and dispersive behavior of males than females of this species and other members of the genus *Phengaris* Doherty, 1891 (Bonelli *et al.*, 2013; Skórka *et al.*, 2013; Ugelvig *et al.*, 2011). The higher mobility of females appears to be related to oviposition, laying eggs in different habitat fragments maximizes egg survival success (Skórka *et al.*, 2013). Instead, the males tend to stay in proximity of the sites where the receptive females emerge (Skórka *et al.*, 2013).

The nearest-neighbor analysis showed that the individuals detected were dispersed throughout the research area, a positive result regarding the spread of this species in the Monte Grave. Further investigations could expand the research area and verify its presence also in the surrounding areas of Monte Grave, such as the near Monte San Mauro. In fact, this species has been considered sedentary for a long

time (Griebeler & Seitz, 2002) but now there is more evidence that it can move several kilometers between patches of suitable habitat (Hayes, 2015; Radchuk *et al.*, 2012; Ugelvig *et al.*, 2011). An interconnection between populations is important to counter local extinctions and maintain genetic diversity (Hayes, 2015). Further investigation could study the mobility of this species in the Monte Grave area, including analyzing any differences between sexes. It would also be interesting to understand whether other populations of *P. arion* are present in the territories surrounding the study area and any connections between them.

The principal component analysis carried out on the nine environment variables showed a positive relationship of most plots where *P. arion* was detected with thyme coverage and flowering plants coverage. Many studies have shown a higher occurrence of individuals of this species in meadows with greater cover of its host plant (Casacci *et al.*, 2011; Griebeler & Seitz, 2002; Spitzer *et al.*, 2009; Vilbas *et al.*, 2015) and with greater flower richness, i.e. greater nectar resources (Spitzer *et al.*, 2009). Heterogeneous vegetation with the presence of some shrubs also seems to have a positive influence on this species (Spitzer *et al.*, 2009), which may be the reason why most plots where *P. arion* was found were found positively correlated with tree-shrub coverage. The heterogeneity, both at landscape and local level, is very important to desynchronise the population dynamics, contributing to the resilience of the species in the entire system (Spitzer *et al.*, 2009). A steeper slope and a southern or southwestern exposure also seem to be correlated with a higher occurrence of this species (Spitzer *et al.*, 2009), as is also seems from the analysis of this study. A southern exposure, in fact, has a positive influence on the host ants of this species because it provides to a warmer local climate (Thomas *et al.*, 1998). In contrast, these plots showed, almost always, a negative relationship with maximum and mean height and coverage of the herbaceous layer. This could be due that both the presence of *Thymus* and the *Myrmica* nests require low height and cover of the turf (Casacci *et al.*, 2011; Griebeler & Seitz, 2002; Thomas *et al.*, 1998; Thomas *et al.*, 2009; Vilbas *et al.*, 2015). However, *Myrmica* ants require different sward structure based on the local climate because it influences the microclimate of the sites (Hayes, 2015). In particular, in the warmer region of Europe, the suitable microclimate occurs under taller vegetation, explaining why many sites were former pastures freshly afforested, which could however become unsuitable in the long term (Spitzer *et al.*, 2009). In the Italian Alps the greater tolerance of taller vegetation can be due to more pronounced daily and monthly temperature fluctuations that might pushed *Myrmica* species to become more adapted to broader niches and tolerant of marked changes of sward height (Casacci *et al.*, 2011). In this study the elevation does not seem to be a driver of the individuals of this species, probably cause elevation range its very small.

The random plots, on the other hand, were found to have much more diverse values of the environmental variables considered, with no aggregation of any kind.

The elevation does not result an important factor in this analysis, maybe because it does not ranged a lot in the area considered.

The differences between the two groups were not found to be statistically significant. This could be due in part to the low sample size. In addition, the plots were close to each other, almost all in the same meadow, so the differences within it are not expected to be very high and maybe almost all this area is suitable for this species.

The plots where *P. arion* was detected were found to have rocks more times than the random plots. This variable was collected because thyme is also often found in rocky environments (Argenti *et al.*, 2019) and therefore the presence of outcropping rocks in the study area could favor it, especially considering that the height of the vegetation detected in all the plots was found to be much higher than the ideal height for this plant, and also for the host ants, which is between 10 and 20 cm in the Alps (Casacci *et al.*, 2011). Therefore, it would be interesting to do further studies to quantify the rocks present and check for a correlation between these two variables. The results obtained showed the further importance of active management practices in these meadows. Mowing or grazing are conservation measures that have a positive impact on this species, as they create suitable habitat for the host plant and host ants associated with *P. arion* (Griebeler & Seitz, 2002). Moreover, a proper mowing also increases the plant species richness (Cannella *et al.*, 2020), thus providing more trophic resources for the adults of this species and also for the others butterflies. Further surveys could investigate the distribution and the abundance of *Myrmica* ants in the study area to better understand the conservation status of this lycaenid.

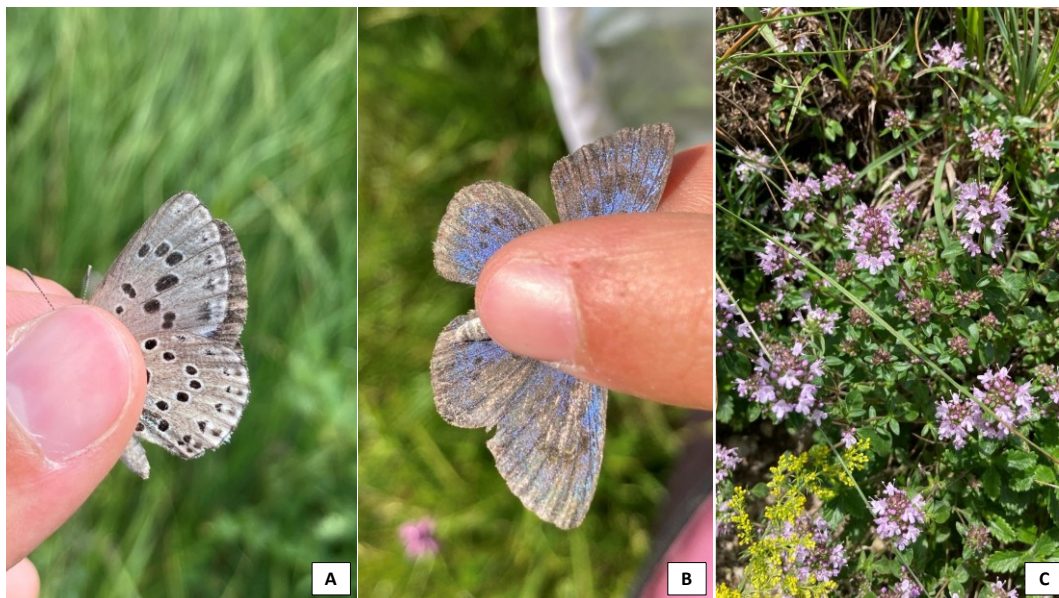


Figure 29. A: lower surface of *P. arion* wings (July 14th); B: upper surface of *P. arion* wings (July 14th); C: *Thymus pulegioides* (July 4th).

In conclusion, the grasslands of Monte Grave have an high biodiversity of butterflies, also hosting several species of conservation interest. In absence of active management this semi-natural environments would be easily invaded by dominant and woody species, causing a contraction and a loss of this habitats (MacDonald *et al.*, 2000). Proper management, therefore, is essential for the maintenance of the flora and fauna communities associated with this environments, as the Rhopalocera (A.A.V.V., 2017; Bubová *et al.*, 2015). A proper mowing is also important for the conservation of the species *Phengaris arion*, since its host plant and host ants benefit from it (Griebeler & Seitz, 2002). Further study could investigate more the Rhopalocera communities and the *Phengaris arion* population of the Monte Grave, to a better knowledge and conservation of its biodiversity.

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10. APPENDIX

Monitoraggio di alcune specie di Ropaloceri in Direttiva Habitat

a cura di Enrico Carta, Lorenza Zaetta, Lucio Bonato

Nell'estate 2023 sono state svolte ricerche su alcune specie di Ropaloceri nel Parco Nazionale Dolomiti Bellunesi, nell'ambito del piano pluriennale di monitoraggio delle specie presenti in Direttiva 92/43/CEE "Habitat".

Delle 5 specie di Ropaloceri presenti nel Parco ed elencate nell'allegato IV della Direttiva, le ricerche sono state focalizzate su *Phengaris arion*, *Parnassius mnemosyne* e *Lopinga achine*. Si tratta di specie di interesse comunitario, per le quali è necessaria una protezione rigorosa. Per queste tre specie, le ricerche sono state svolte in alcune aree precedentemente selezionate per il monitoraggio e gli obiettivi sono stati i seguenti: (i) verificare o confermare la presenza delle specie; (ii) determinare il loro periodo di volo; (iii) stimare alcuni indicatori semi-quantitativi per monitorare la consistenza delle popolazioni.

Metodi

Per ognuna delle 3 specie sono stati considerati percorsi-campione (transetti) all'interno di aree già definite nell'ambito della pianificazione del monitoraggio pluriennale delle specie animali presenti nel Parco Nazionale Dolomiti Bellunesi (Tab. 1).

Per *Phengaris arion* è stata selezionata l'area dei prati sommitali del Monte Grave, a circa 1300 m s.l.m. (Fig. 1). Si tratta di una delle poche aree in cui in passato è stata segnalata la specie all'interno del Parco Nazionale.

Per *Parnassius mnemosyne*, l'area scelta è collocata in prossimità di Pian d'Avena, presso la località Stalle Boa, a circa 800 m s.l.m. (Fig. 2).

Per *Lopinga achine* invece, è stata selezionata un'area in Val Canzoi, lungo la strada principale immediatamente a valle della località Fraina, a circa 500 m s.l.m. (Fig. 3).

Specie target	Sito di monitoraggio	Punto di inizio del transetto	Punto di fine del transetto	Lunghezza transetto	Quota di inizio	Quota di fine
<i>Phengaris arion</i>	Monte Grave	46.088367 N 11.932173 E	46.090349 N 11.925898 E	570 m	1220 m	1290 m
<i>Parnassius mnemosyne</i>	Stalle Boa	46.060305 N 11.839802 E	46.062237 N 11.842629 E	330 m	812 m	850 m
<i>Lopinga achine</i>	Val Canzoi	46.103561 N 11.943993 E	46.110073 N 11.935868 E	1000 m	566 m	616 m

Tab. 1 Localizzazione dei siti di monitoraggio e dei rispettivi percorsi-campione.



Figura 1 Area di monitoraggio e transetto per Phengaris arion, sul Monte Grave.



Figura 2 Area di monitoraggio e transetto per Parnassius mnemosyne, in località Stalle Boa

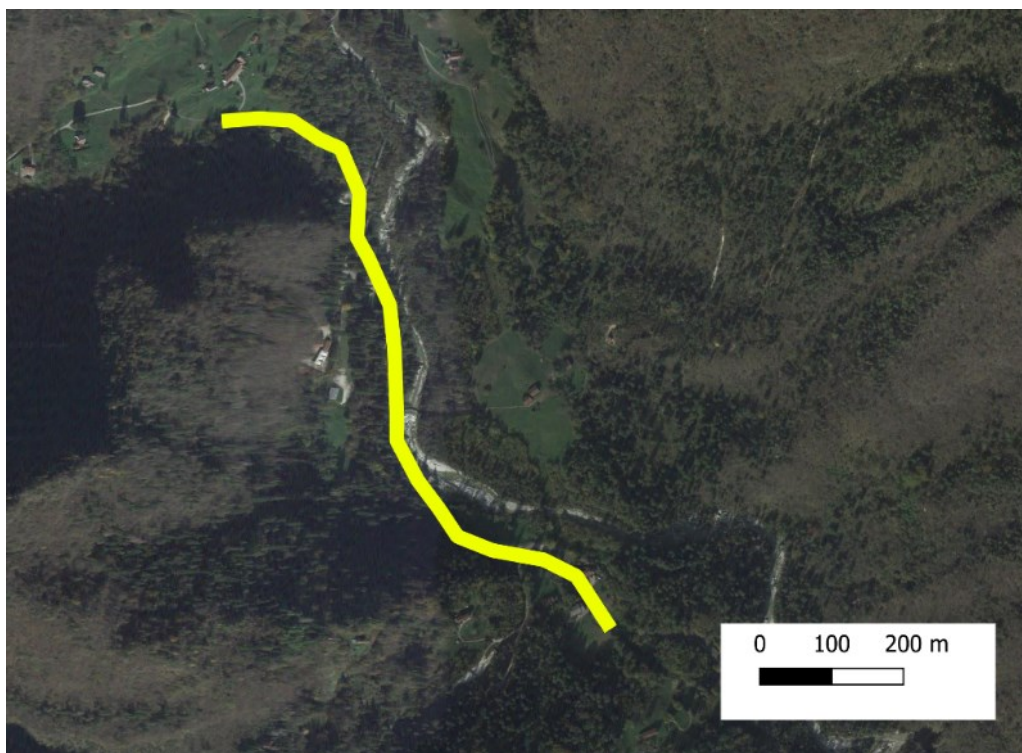


Figura 3 Area di monitoraggio e transetto per *Lopinga achine* in Val Canzoi

Il monitoraggio ha riguardato esclusivamente gli adulti. Si è adottata una metodologia ibrida tra le due indicate nel documento tecnico “Pianificazione del monitoraggio delle specie animali presenti nel Parco Nazionale Dolomiti Bellunesi e inserite negli allegati della Direttiva 92/43/CEE Habitat” (ovvero il transetto semiquantitativo a cadenza settimanale e cattura-marcatura-ricattura a giorni alterni). Si è quindi effettuato il transetto semiquantitativo di Pollard & Yates, ripetuto ogni 10-15 giorni, associando ad esso la cattura temporanea e marcatura degli individui. Tutte le ripetizioni sono state svolte da uno stesso rilevatore per ogni transetto: Lorenza Zaetta per il sito del Monte Grave, per il monitoraggio di *P. arion*; Enrico Carta per i siti di Stelle Boa e Val Canzoi, per il monitoraggio rispettivamente di *P. mnemosyne* e *L. achine*.

Nel passato il monitoraggio era già stato svolto anche nel 2020, per le stesse specie target, da un altro operatore che aveva utilizzato come metodo il transetto semiquantitativo di Pollard & Yates.

Tutte le sessioni sono state effettuate in condizioni meteorologiche favorevoli, con copertura nuvolosa inferiore al 70%, vento assente o moderato (valori 0 o 1 della scala Beaufort) e temperatura superiore ai 20° C. L'unica eccezione, per esigenze logistiche, è stata il 14 giugno in Val Canzoi, in cui si è operato con condizioni di nuvolosità inizialmente più estesa. Ciò nonostante, è stato possibile rilevare *Lopinga achine* in modo efficace, poiché molti individui erano in attività. Tutti i transetti sono stati svolti tra le 9.30 e le 16.

Per ogni sessione, si è cercato di contare tutti gli individui adulti della specie-target presenti: per *Phengaris arion* e *Lopinga achine* sono stati considerati solo gli individui presenti entro una distanza di 2,5 m dall'operatore, mentre per *Parnassius mnemosyne* è stato possibile rilevare e conteggiare anche individui a distanza maggiore, all'interno dell'area prativa. Il rilevamento è stato effettuato o tramite

cattura temporanea, utilizzando un retino entomologico (con marcatura), o tramite osservazione diretta senza cattura, mediante binocolo, macchina fotografica o ad occhio nudo (senza marcatura, ma con la possibilità di riconoscere eventuali individui già marcati). La cattura temporanea, con conseguente manipolazione e marcatura degli individui, è stata possibile grazie a specifiche autorizzazioni ministeriali.

Gli individui catturati temporaneamente sono stati marcati. La marcatura è stata realizzata scrivendo con un pennarello di colore giallo un numero progressivo sulla pagina inferiore dell'ala posteriore sinistra. Tale marcatura è stata applicata per evitare doppi conteggi, sia all'interno della stessa sessione, che in giorni differenti. Per stimare il numero minimo di individui rilevati in una sessione, sono stati conteggiati sia gli individui marcati sia eventuali altri individui che non era stato possibile catturare ma per i quali era stata osservata a distanza la mancanza di marcatura (quando applicata a tutti gli individui precedentemente rilevati nella stessa sessione) e che potevano essere verosimilmente distinti tra loro.

Per ciascuna specie, e quindi per ogni transetto, sono state svolte in totale 4 sessioni, ad una distanza temporale di 10-15 giorni: per *Phegaris arion* tra fine giugno e fine luglio; per *Parnassius mnemosyne* tra metà maggio e inizio luglio, coerentemente con il periodo di volo della specie, e diversamente da quando indicato nel piano di monitoraggio citato; per *Lopinga achine* tra metà giugno e fine luglio (Tab. 2).

<i>Phengaris arion</i>	<i>Parnassius mnemosyne</i>	<i>Lopinga achine</i>
19 giugno	21 maggio	14 giugno
4 luglio	7 giugno	26 giugno
14 luglio	24 giugno	8 luglio
26 luglio	8 luglio	21 luglio

Tab. 2: date delle sessioni di monitoraggio nei tre transetti

Nello svolgimento delle attività di campo, per *Parnassius mnemosyne* e *Lopinga achine* si è utilizzata una scheda di raccolta dati (Tab. 3) e sono stati registrati alcuni parametri meteorologici (copertura nuvolosa, velocità del vento secondo la scala Beaufort e temperatura dell'aria), sia all'inizio che alla fine del transetto. È stata inoltre registrata l'ora di inizio e di fine di ogni transetto effettuato.

Codice individuo	Ora	Distanza dal transetto stimata a vista	Sesso	Usura	Comportamento	Foto si/no	Marcato Si/No	Note

Tab. 3 Tabella di raccolta dati utilizzata durante le sessioni

Tutti gli individui che si sono riusciti a catturare temporaneamente tramite retino sono stati manipolati, oltre che per effettuare la marcatura, per valutarne il sesso e il grado di usura delle ali. Per valutare il sesso, per *Parnassius mnemosyne*, sono state considerate la forma e il colore dell'addome, nonché la presenza o meno di peluria grigia abbondante, caratteristica dei maschi ed assente invece nelle

femmine. È stata inoltre rilevata l'eventuale presenza di sphragis, una struttura cerosa rilasciata dai maschi sull'addome delle femmine durante l'accoppiamento, con la funzione di prevenire successivi accoppiamenti della femmina con altri maschi. Per valutare il sesso in *Lopinga achine* ed in *Phengaris arion* è stata invece valutata a vista la morfologia delle strutture accessorie dell'apparato riproduttore sporgenti presso la punta dell'addome, effettuando una leggera pressione sull'addome degli animali e utilizzando una lente di ingrandimento quando necessario.

L'usura delle ali è valutata secondo una scala da 0 (non usurato) a 4 (molto usurato). Per quanto riguarda *Phengaris arion*, oltre alle sessioni standardizzate nel transetto, è stata svolta un'indagine di un'area più ampia, posta nei prati sommitali del Monte Grave e comprendente il transetto stesso (Fig. 5). L'indagine è stata svolta il giorno 14 luglio, dalle 10:30 alle 17:00 circa, da parte di diversi rilevatori (Lucio Bonato, Enrico Carta, Damiano Sartor e Lorenza Zaetta) che hanno perlustrato contemporaneamente diverse parti dell'area, focalizzandosi sulla ricerca di individui adulti di questa specie.

Risultati

Phengaris arion (Linnaeus, 1758)

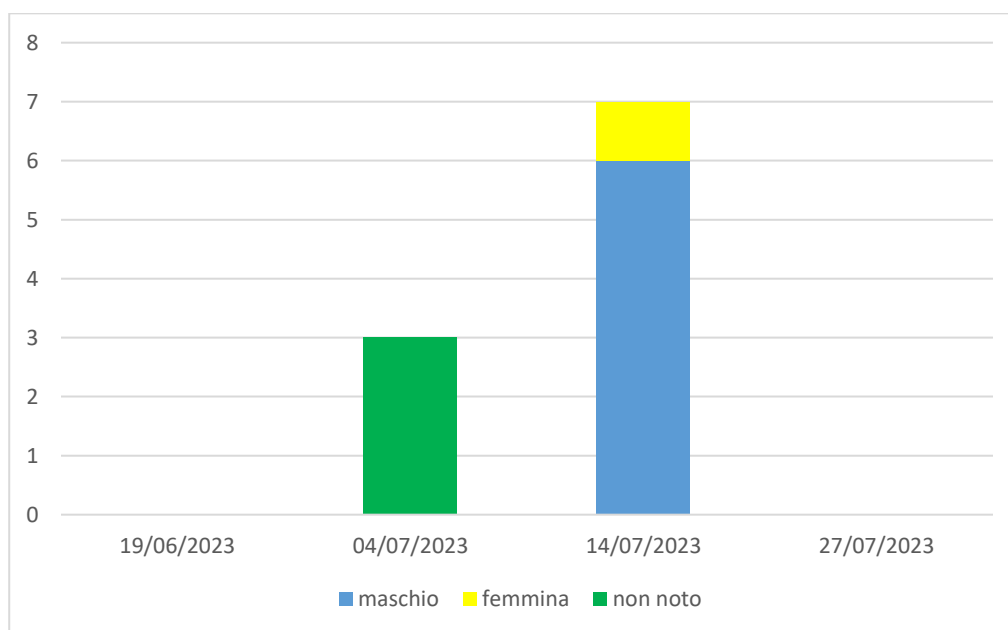


Figura 4 Numero di individui di *Phengaris arion* rilevati nelle quattro sessioni di monitoraggio, distinti per sesso

Nel sito del Monte Grave, durante le 4 sessioni, sono stati rilevati 4 individui di *Phengaris arion*, mentre durante la perlustrazione dell'area ne sono stati rilevati 6, per un totale di 10 individui (Fig. 4). Di questi, 7 erano maschi, 1 femmina e di 3 non è stato identificato il sesso, e le foto non sono state sufficienti per riconoscerlo a posteriori, basandosi solamente sui caratteri delle ali. Sono stati marcati solo gli individui rilevati in data 14 luglio e questi individui non sono mai stati ricatturati né nella stessa sessione né in quella successiva. Anche se gli individui rilevati il 4

luglio non sono stati marcati, il confronto delle foto della superficie alare ha consentito di escludere che siano stati ricatturati durante la sessione successiva. Gli elementi che sono risultati utili per tale distinzione sono la forma e la grandezza delle macchie scure, lo spessore della banda scura sul bordo della superficie superiore delle ali e la presenza di particolari segni di usura.

L'usura delle ali è risultata molto variabile, ma sono stati assegnati in media valori inferiori agli individui rilevati il 4 luglio (valori da 1 a 2) rispetto a quelli rilevati il 14 luglio (valori da 1 a 4).

Lungo il transetto, la specie è stata rilevata maggiormente nella porzione centrale del percorso (Fig. 5). Per quanto riguarda l'area perlustrata, invece, sono stati trovati individui sparsi in diversi settori (Fig. 5).

La specie è nota nell'area del Monte Grave dal 2013. Nel corso dei rilievi per il progetto "Biodiversità in ambiente alpino" del 2018 sono stati rilevati due individui mentre nel 2019 nessuno. I dati raccolti in passato sono, comunque, difficilmente confrontabili a causa della diversa metodologia utilizzata e della diversa area esaminata.

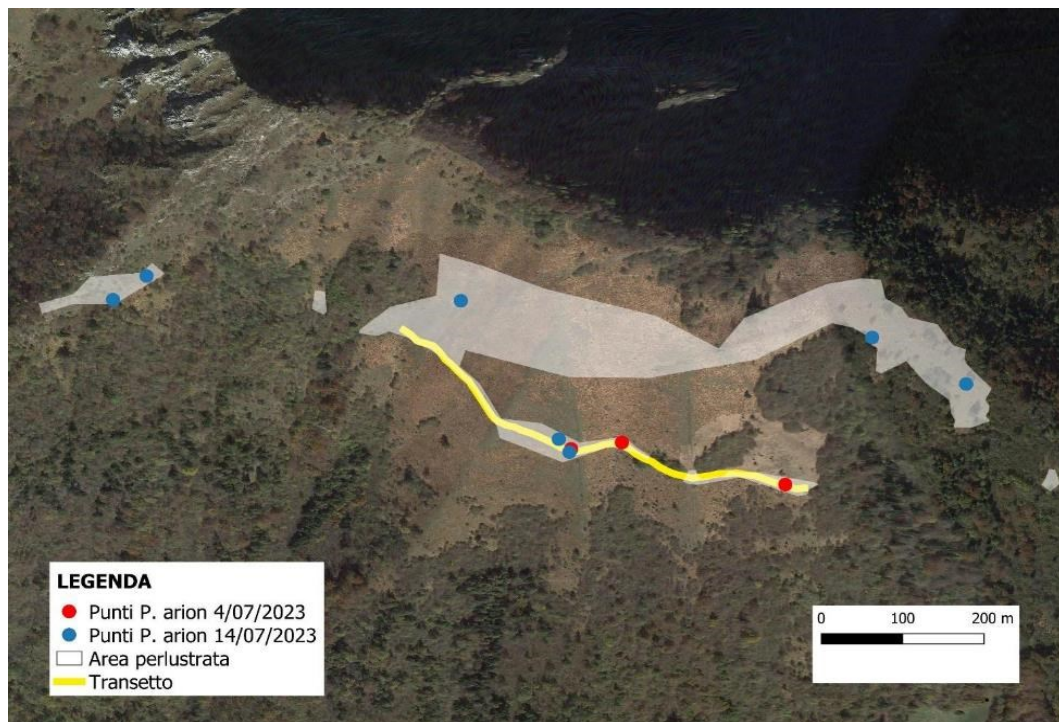


Figura 5 Punti dove è stata rilevata *Phengaris arion*, localizzazione del transetto e dell'area più ampia perlustrata.

Parnassius mnemosyne (Linnaeus, 1758)

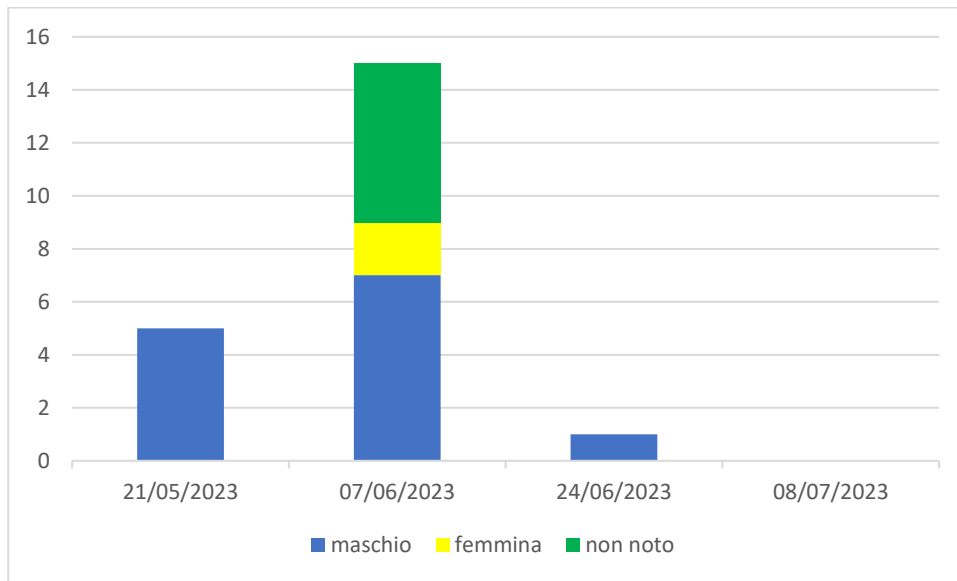


Figura 6 Numero di individui di *Parnassius mnemosyne* rilevati per ogni sessione di monitoraggio, distinti per sesso

Nel sito di Stalle Boa, nel corso delle 4 sessioni, sono stati rilevati in totale 21 individui. Di questi, 18 sono stati distinti perché catturati temporaneamente e marcati con numero progressivo. Gli altri 3 sono stati distinti perché sono stati osservati in una stessa sessione (07/06/2023), non presentavano la marcatura e due di essi erano in accoppiamento. La specie è stata rilevata nel corso delle prime tre sessioni (21/05, 07/06, 24/06), e non è stata rilevata nell'ultima (08/07) (Fig. 6).

L'usura delle ali è risultata un parametro piuttosto variabile nel corso delle prime due sessioni, in cui sono stati rilevati animali con ogni grado di usura. Nella terza sessione invece, l'unico individuo rilevato presentava ali molto usurate.

La maggior parte degli individui sono stati rinvenuti nella porzione più settentrionale del transetto, in ambiente di ecotono tra prato e bosco misto (Fig. 7). La specie è stata rinvenuta nelle immediate vicinanze anche al di fuori dell'area di studio, il 7 e il 24 giugno 2023, in un simile ambiente di ecotono lungo la strada sterrata che collega il sito di monitoraggio a Pian D'Avena, a circa 350 m a Sud-Ovest da quelli rilevati.

In ognuna delle tre sessioni in cui la specie è stata rinvenuta, tutti gli individui sono stati rilevati in un raggio di circa 20 m.

Individui marcati non sono mai stati ricatturati nelle sessioni successive.



Figura 7 Punti in cui è stato rinvenuto *Parnassius mnemosyne* nei transetti. Data la vicinanza tra loro degli individui trovati, ad ogni sessione è stato registrato un solo punto, di raggio di circa 20 m. In azzurro gli individui del 21 maggio, in rosa quelli del 7 giugno e in verde quello del 24 giugno

Il monitoraggio di *Parnassius mnemosyne* nel Parco Nazionale Dolomiti Bellunesi era già stato svolto nel 2020, lungo un transetto diverso ma molto vicino a quello del 2023 (a circa 450 m di distanza a Nord, in prossimità del punto con coordinate 46.0661904 N, 11.841969 E). In quell'anno erano state svolte 3 sessioni, a distanza di circa una settimana una dall'altra, tra il 18 maggio e il 3 giugno. La specie era stata rinvenuta in tutte le sessioni in numerosità compresa tra 3 e 8 individui. Data la differente località, sebbene vicina, e la diversa metodologia applicata (anche in quel caso è stato svolto il transetto di Pollard & Yates, però senza marcare gli individui e probabilmente considerando solo individui entro 2.5 m dal percorso), risulta difficile un confronto tra i risultati dei rilevamenti svolti in passato e quelli svolti nel 2023.

Lopinga achine (Scopoli, 1763)

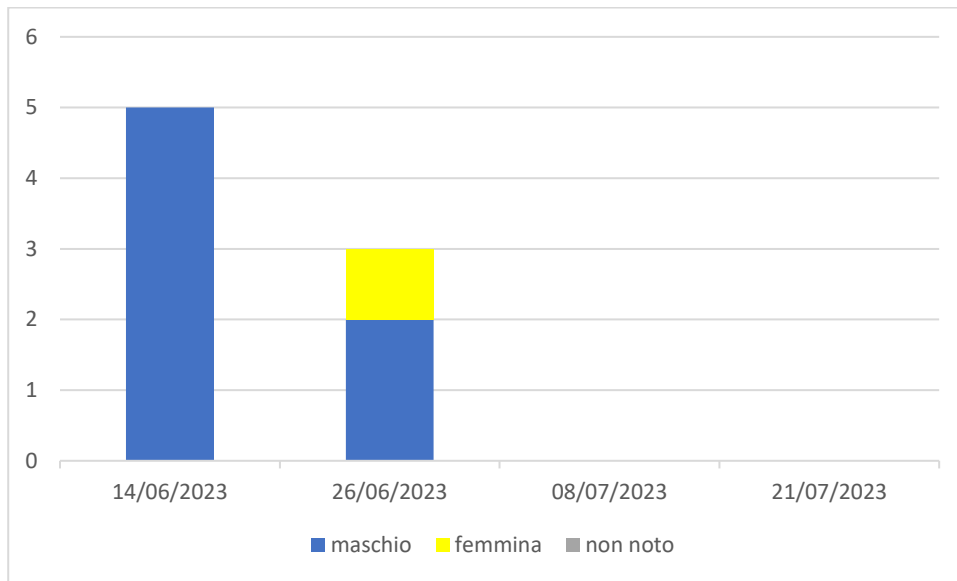


Figura 8 Numero di individui di *Lopinga achine* rilevati nelle quattro sessioni di monitoraggio, distinti per sesso

Per *Lopinga achine* sono stati registrati in totale 8 individui, 7 maschi e una femmina (Fig. 8). La specie è stata rinvenuta esclusivamente nelle prime due sessioni di monitoraggio. Probabilmente, forse data la bassa quota della località, il periodo di volo è risultato più precoce rispetto ad altri siti del Parco Nazionale. Quindi, per i monitoraggi futuri, sarà utile iniziare i rilevamenti prima nel corso della stagione. Va sottolineato come la specie sia stata rinvenuta non distante, sempre in Val Canzoi, anche prima dell'inizio delle sessioni di monitoraggio (prima osservazione; 1 giugno 2023, a circa 200 m a Est dal transetto). Inoltre, nei giorni in cui si sono svolte le sessioni, è risultata presente anche al di fuori del transetto, talvolta anche quando non rinvenuta all'interno del transetto (es. 8 luglio 2023). L'usura delle ali è risultata variabile nella prima sessione di monitoraggio, con valori compresi tra 0 e 3, e piuttosto elevata nella seconda, con valori tra 2 e 4. Individui marcati non sono mai stati ricatturati nelle sessioni successive. La specie è risultata presente lungo tutto il percorso, con una distribuzione apparentemente uniforme, tranne che nel tratto di 200 m più meridionale (Fig. 12).

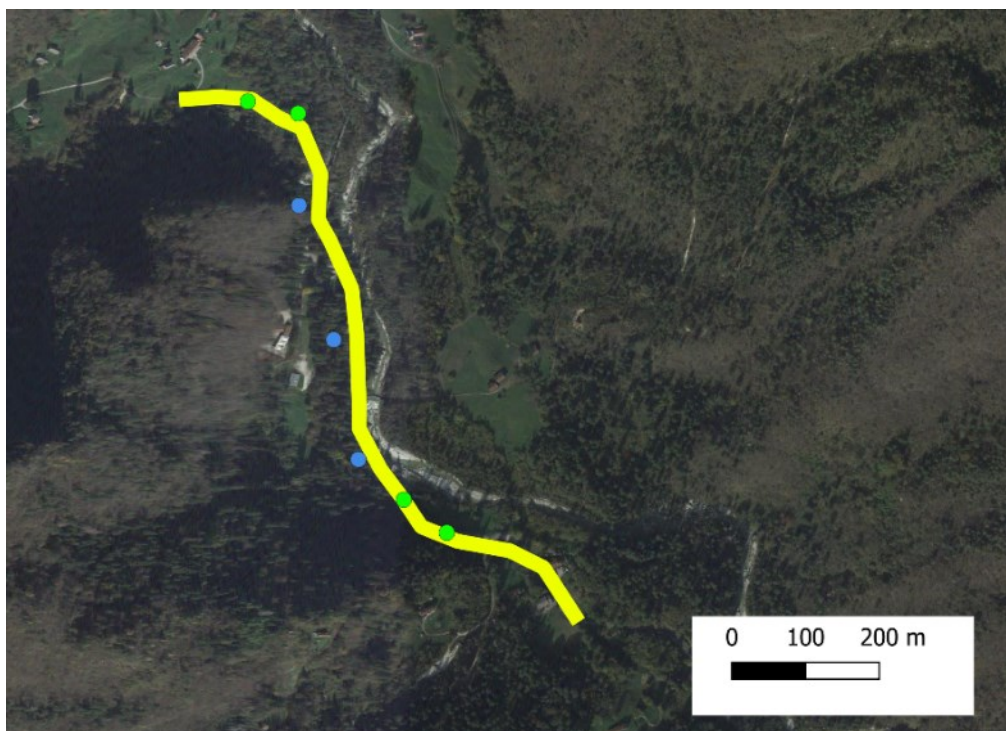


Figura 12 Localizzazione dei punti in cui è stata rinvenuta *Lopinga achine* lungo il transetto: in verde quelli del 14 giugno e in blu quelli del 26 giugno

Nel corso del 2020 il monitoraggio era stato svolto lungo lo stesso percorso, ma limitatamente a quattro tratti, ciascuno della lunghezza di 100 m, intervallati da altrettanti tratti dove non era stato svolto. I 4 transetti erano codificati come Cesiomaggiore 1, 2, 3 e 4. Erano state svolte tre sessioni, dal 29/06 al 12/07. Nel corso di quelle sessioni era stato rinvenuto solo un individuo, nel primo tratto a Sud del percorso, in data 7 luglio. Verosimilmente, anche in quelle occasioni, vale quanto affermato in precedenza sulla fenologia locale della specie, probabilmente più precoce che nel resto del Parco Nazionale.

Pur considerando che lo sforzo complessivo di ricerca è stato maggiore nel 2023 (4 sessioni e lunghezza del transetto 1000 m) rispetto al 2020 (3 sessioni e lunghezza complessiva del transetto 400 m), l'abbondanza lineare della specie è risultata apparentemente maggiore nel 2023 (circa 2.0 indd/km) rispetto al 2020 (circa 0.8 indd/km).

