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The Habitat Network for Butterfly Communities of the Alta Murgia National Park (Apulia, Italy)

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Abstract: Habitat networks can help to make habitats more resilient and assist species survival in a fragmented landscape and changing climate. Butterflies are one of the main indicators of diversity due to their high sensitivity to environmental changes. In the context of sudden and unpredictable environmental changes, protection strategies for butterflies at risk of extinction should consider the exact distribution of these species, as well as the various threats to which each of them is subjected. About 290 species of butterflies are reported in Italian fauna, and 120 of them are recorded in Apulia (Southern Italy). In the Alta Murgia National Park (AMNP) (Apulia Region, Italy), screening was performed to study the relationships between area/landscape composition and diurnal butterfly community structure. Representative semi-natural habitats of Alta Murgia buffering productive crops were selected to set up transects/paths along dry grasslands, oak forests and pine forests. Monthly samplings were performed for one year. During the survey, 909 specimens from 53 species were collected. The highest values of butterfly's abundance and richness were recorded in dry grasslands. A strong positive correlation between butterfly abundance and air temperature was registered (Pearson correlation: r = 0.8; $p < 0.001$). *Melanargia arge*, endemic in central-southern Italy and considered threatened in Europe (Annexes II and IV—Habitats Directive), was registered in each habitat. The biodiversity indices (Chao 1, Shannon and Simpson) for each habitat were high and similar to those in protected areas of Sicily. The connected landscape is important for generalist or open-habitat specialists, and large remnants are key for disturbance-sensitive and threatened taxa. The presented evidence can provide useful information on butterfly conservation in the AMNP and for the management and conservation of characteristic landscapes of Alta Murgia.

Keywords: eco-mosaic landscape; phenology; biodiversity; Papilionoidea; biodiversity

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1. Introduction

The intensification of agriculture activities has caused a drastic decline in semi-natural rocky grasslands of high naturalistic value [1]. Many territories of Southern Italy are also affected by this phenomenon [2]. The soil profile has been drastically modified, especially by removing stones to obtain more areas suitable for crops. This land-use change has impacted the overall availability of natural habitats, their reciprocal connection and biodiversity [3]. In this context, more than 25,000 ha of semi-natural grasslands of the Alta Murgia National Park (AMNP), available fifty years ago, were converted to arable lands [4]. Since 2004, semi-natural dry grasslands of the AMNP have been considered of high naturalistic importance and have been protected [5]. The conservation of these habitats according to Habitats Directive 92/43/EEC contributes to the preservation of a wide range of rare, threatened or endemic species occurring in the EU. In the AMNP, the floristic composition of dry grasslands delineates a peculiar pseudo-steppe, characterized by the endemic grass *Stipa austroitalica*, which has a restricted range in Southern Italy and is listed as a priority species in Annex II of the Habitats Directive and under Appendix I of the Convention on the Conservation of European Wildlife and Natural Habitats (Bern Convention). Maintaining traditional rural systems such as those in the AMNP, which contribute to the biocultural diversity of the landscape, could provide useful ecosystem services and preserve biodiversity [6]. A biomonitoring network system can evaluate biodiversity conservation effectiveness in national parks, identifying anthropogenic pressures on natural ecosystems/landscapes and contributing to the design of effective management plans [7]. To assess the effectiveness of conservation initiatives or the global state of the environment in protected areas, butterflies stand as ideal indicators because they are relatively easy to observe in the field, they show mutualistic relationships with flowers, have a short life cycle [8] and are quite attractive to the general audience, facilitating citizen science surveys. Lepidoptera are considered early warning bioindicators due to their high sensitivity in a wide spectrum of ecological settings. In fact, they quickly respond to plant diversity, habitat complexity, altitude, climate, pollution and land-use systems [9–11]. Annual variation in climate conditions could be a determinant of between-year differences in butterfly phenology [12]. Nonetheless, studies documenting annual monitoring within butterfly populations are scarce [13]. Inventory and monitoring programs set in several countries in recent decades showed that climatic warming determines changes in the timing of flight periods and that their flight periods may have important consequences for species' population abundance through alterations of phenological patterns and disruptions in the synchrony of plant–insect or host–parasitoid interactions [12,14]. In some cases, changes related to management practices are more influential on butterfly populations than climate-related phenological advances [14]. The intensification of human land use, abandonment of traditional agropastoral activities or afforestation programs can strongly affect the suitability of the environment for diurnal Lepidoptera circadian activity, mobility, voltinism and the overwintering stage. It can also modify the structure of their populations [7,15].

About 290 species of butterflies are reported in Italian fauna and 120 species are recorded in Apulia [16,17]. Despite numerous reports throughout Apulia on Lepidoptera [18–21], butterflies in the AMNP are scarcely documented with details, while several studies have reported its high flora and fauna diversity [22–26]. The AMNP is part of the Natura 2000 network and is one of 431 prime butterfly areas in Europe [27], ensuring the continuity of migratory movements and genetic flows of species along biological corridors by the long-term vitality of natural habitats. Some butterflies are particularly affected by habitat loss and fragmentation, such as species at high trophic levels, food specialists, species with poor dispersal abilities, rare species and habitat specialists [28]. Generalist species provide ecosystem services such as pollination in agricultural landscapes and seed dispersal, but specialist species can play an important role in the resilience assessment of the ecosystem. Univoltine butterfly species generally exhibit fewer plastic responses to variable environmental shifts. Large biomonitoring studies often focus on butterflies because they include charismatic flagship species; monitoring species, which rapidly respond to environmental changes (e.g., *Zerynthia polyxena* (Denis and Schiffermüller), 1775), may indeed represent a suitable umbrella for the preservation of the habitats they dwell in [14]. Moreover, butterfly species show strong seasonality in the temporal activity window, with periods of flight limited to months with optimal climatic conditions for development and dispersal. However, the causes of variation in this phenological dimension across space and time are still poorly understood. In Central Europe, semi-natural calcareous grasslands have faced significant reductions and are highly fragmented but still of high conservation value as they rank as the most species-rich habitat in terms of flora and invertebrate fauna. Semi-open woodland represents an important ecotonal environment by including a mosaic of woodland and meadows and preserving wide open corridors along forest roads. The presence of clearings and ecotonal habitats in forest areas can support biodiversity [28]. Notwithstanding the landscape fragmentation, the ANMP natural mosaic, through dry grasslands/meadows, forest areas and arboreal–shrub–stone wall connection, allows the presence of several of butterfly species [29].

This study aims to survey diurnal butterflies in an alternation of dry grasslands/meadows, forest areas and linear elements of arboreal–shrub connection over the period of a year, describing the relative abundance, species richness, phenology and the relationship between species and habitats. Our research study mainly focuses on three issues as follows: (1) the status of butterfly diversity in the AMNP; (2) the variation in butterfly community diversity with habitat type and season; and (3) the assessment of endangered and protected species of butterflies. This last analysis, along with further data collection, can contribute to set management models in protected areas, ensuring agricultural income and resilience to environmental crises following extreme events (drought, erosion by corrivation, fire).

2. Materials and Methods

2.1. Study Area

The landscape of the Alta Murgia region is characterized by a gentle undulating countryside and doline-shaped depressions, with surface karstic phenomena represented by swallow holes. The substratum is formed by Cretaceous limestone dating back to approximately 130 million years ago and being up to 3000 m thick, usually covered with Pleistocene calcarenite. The climate area is Mediterranean, characterized by mild winters and hot dry summers [22], even though summer temperatures have increased and rainfall has decreased in recent decades [30]. The vegetation of the north-west Murge area consists, broadly, of three main physiognomic types as follows: the Mediterranean substeppic prairie (pseudo-steppe); the bushy and/or arboreal prairie; and the forest. The pseudo-steppe grasslands mainly occupy the summit and south-western part of the Alta Murgia with some aspects of garrigue and pseudogarrigues due to the presence of fruticose or suffruticose Camephytes and deciduous Nanofanerophytes, i.e., *Rhamnus saxatilis* Jacq. subsp. *infectorius* (L.). It is one of the largest sub-steppe areas in Italy with Festuco-brometalia herbaceous vegetation. Moving towards lower altitudes, the prairies shift through ecotonal areas with bushy and/or arboreal pastures to wooded phytocenoses, dominated by different species depending on the slope. The dominant species of the woods are *Quercus pubescens* Willd. s.l. and *Q. cerris* L., followed by *Q. ilex* and *Q. coccifera* L. The most typical and widespread spontaneous vegetation consists of steppe grasslands in *Stipa austroitalica* Martinovský ssp. *austroitalic*. This species is physionomically characterized, often dominating vast extensions of xerograminetes in which *Festuca circummediterranea* Patzke, *Koeleria splendens* Presl., *Bromus erectus* Hudson and *Scorzonera villosa* Scop. ssp. *columnae* (Guss.) Nyman are associated with high values in terms of coverage. These prairies have a high richness of vascular flora, with over 500 subgeneric taxa, including numerous endemic or sub-endemic plant species, inserted to varying degrees in the IUCN Red List of threatened species or, again, of particular phytogeographic interest.

In three habitats of AMNP with naturalistic interest [22,31], the following nine transects were selected (Figure 1; Table 1):

(1) Dry grasslands (afterwards referred as DGLs or DGL I/II/III): three sites characterized by an eco-mosaic of natural and semi-natural herbaceous priority habitat "Eastern sub-Mediterranean dry grasslands (*Scorzoneratalia villosae*)" and "Pseudo-steppe with grasses and annuals of the *Thero-Brachypodietea*", according to Habitats Directive 92/43/EEC;

(2) Oak forest paths (afterwards also referred as OFPs or OFP I/II/III): three sites characterized by herbaceous vegetation bordered by forests dominated by downy oak (*Q. pubescens*), a deciduous species belonging to the priority habitat "Eastern white oak woods", according to Habitats Directive 92/43/EEC;

(3) Pine forest paths (afterwards also referred as PFPs or PFP I/II/III): three sites characterized by herbaceous vegetation bordered by reforestation of conifers, mainly composed of Aleppo pines (*Pinus halepensis* Mill.).

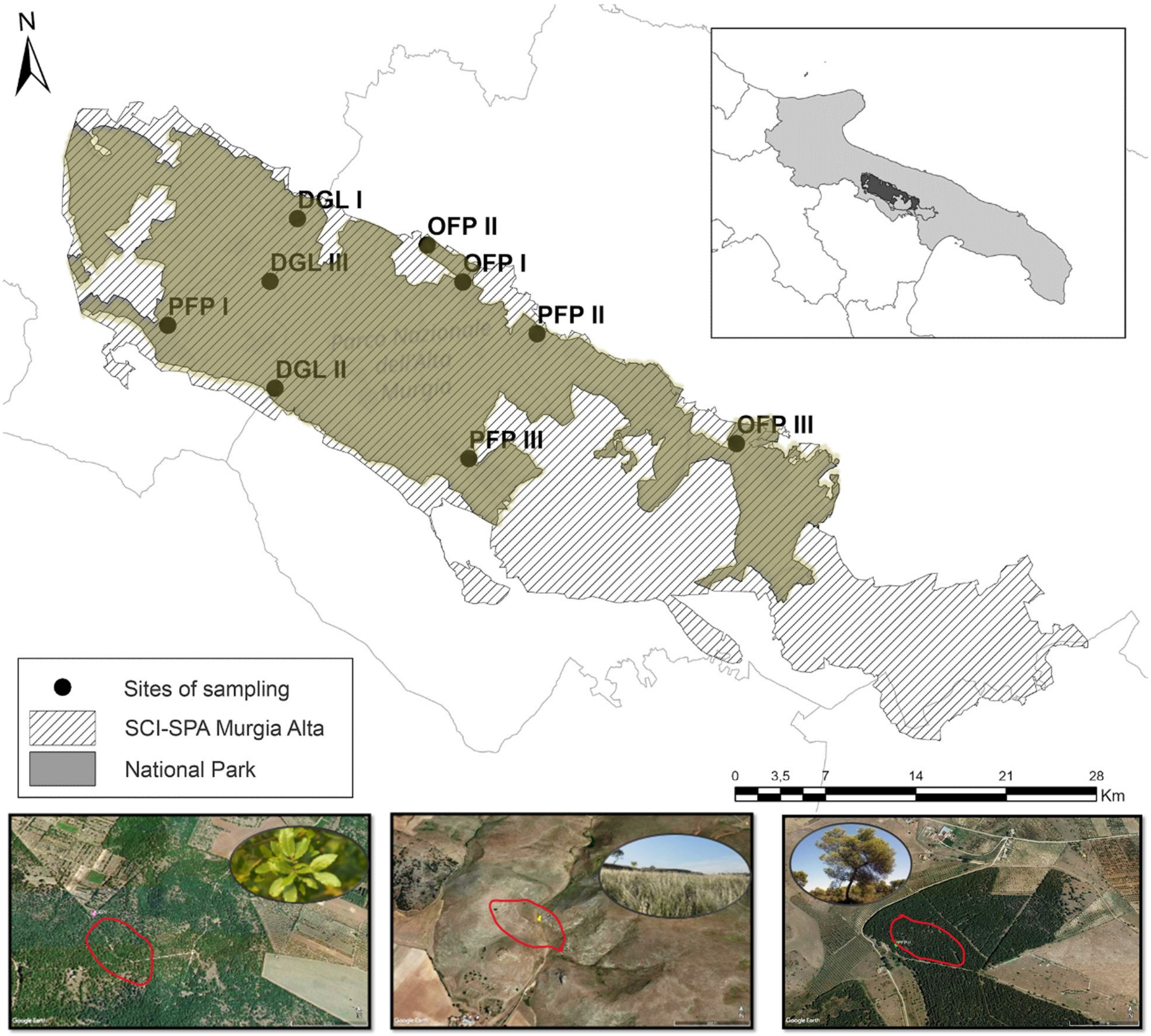

Figure 1. Geographical positions of the sampling study sites in the Alta Murgia National Park and details of three kinds of habitat (OFP; DGL; PFP) on a Google Earth™ satellite image of Italy.

Table 1. Sampling sites, their habitat characteristics and geographical position.

Site	Habitat/N. Site	LAT N	LONG E	Altitude (m a.s.l.)
Bosco Scoparella	Oak forest OFPI	41°01′41.86″	16°25′56.51″	250
Lama delle Grotte	Oak forest OFPII	41°03′21.72″	16°7-10′09.55″	379
Monte Cucco	Oak forest OFPIII	40°54′00.44″	16°40′09.71″	428
Foresta di Acquatetta	Pinewood PFPI	41°00′55.91″	16°09′33.09″	595
Bosco Rogadeo	Pinewood PFPII	40°59′17.17″	16°29′46.04″	379
Pulicchio di Gravina	Pinewood PFPIII	40°54′20.91″	16°25′26.63″	555
Castel del Monte	grasslands DGLI	41°04′54.69″	16°17′09.69″	540
Rocca del Garagnone	grasslands DGLII	40°57′56.88″	16°15′09.11″	528
Monte Savignano	grasslands DGLIII	41°02′10.16″	16°15′21.92″	539

2.2. Butterfly Surveys and Species Identification

The phenological model was based on butterfly observations collected using a standard protocol, including repeated visits to the same sites. Between August 2016 and July 2017, monthly samples were collected during 9 transect surveys, using an entomological net. The survey team recorded all diurnal butterflies [32]. For each sampling session, fieldworkers recorded all the diurnal butterflies detected in an imaginary 5 m^3 box along a 200 m long fixed linear transect. The duration of each sampling session was about 30 min [33]. Counts were conducted when the sun warmed up and allowed for the flight of butterflies. Transects were geo-referenced on a Garmin Foretrex^{TM} 101 Personal Navigator, XGFE101. About 80% of the butterflies captured were identified in the field and immediately released; the remaining specimens were retained and identified in a laboratory by the examination of genital apparatuses. The most common keys were adopted for identification [34–42]. Air temperatures recorded in five weather stations near the study sites were provided by Acrotec (CIMA Technology Foundation, Savona, Italy).

2.3. Statistical Analysis

To characterize the biodiversity and community structure in each habitat, Chao 1 richness, Fisher's alpha, Shannon (based natural logarithms), Simpson (inverse) and Bray–Curtis indices and the number of species, singletons (species with only one surveyed individual), and doubletons (species with two surveyed individuals) were calculated using Estimates software [43,44]). Differences in the each considered biodiversity index among the three habitats were studied via applying the non-parametric Mann–Whitney test using Statistica 10 software [45]. Data matrix (53 species by 3 habitats) based on the population abundance of each species at each habitat was subjected to multivariate analyses. The effects of season, habitat and sites on butterfly abundance were evaluated using the generalized linear model (GLM), fitted through the SPSS software by assuming Poisson distribution for the response variable (i.e., abundance data). The air temperatures were also correlated with monthly mean values of butterfly abundances using the Pearson coefficient [46]. Relationships between species, grouped into five families, and AMNP habitats were analyzed by principal components analysis (PCA) based on the variance–covariance matrix, using the PAST software [47].

3. Results

Within the nine active transects in the AMNP, 907 adult butterflies were detected, and 53 species were identified (Table S1). The most abundant family was Nymphalidae (51%), followed by Pieridae (18%), Lycaenidae (18%), Papilionidae (8%) and Hesperiidae (5%).

3.1. Habitat Selection

Species accumulation curves (SACs) were used to estimate the number of species in the main natural habitats of the Alta Murgia National Park and are reported in Figure 2. Evidence found by the Chao 1 abundance-based estimator indicates the adequacy of the survey carried on in representing the fauna in the habitats considered. In all curves, the number of recorded species approximates an asymptotic trend, within all three different habitats at about 150 observed individuals.

The upper and lower bound curves corresponding to the 95% confidence interval of the Chao 1 richness estimate obtained for the DGLs (the habitat with the highest abundances) encompassed the accumulation curves drawn for the two remaining habitats. Mann–Whitney U tests showed that the composition of butterfly communities, cumulating the species recorded throughout the 12-month study period, was similar ($p > 0.05$) for each pair of habitat kinds (Table 2. In DGL, the most abundant species were *Hipparchia statilinus* (Hufnagel), *Melanargia russiae* (Esper), *Papilio machaon* L., *Pieris rapae* (L.) and *Pontia edusa* (F.). In PFP, they were *Lasiommata megera* (L.), *P. machaon*, *Polyommatus icarus* (Rottemburg), *M. russiae* and *P. rapae*. In OFP, they were *H. statilinus*, *L. megera*, *P. icarus* and *Maniola jurtina* (L.).

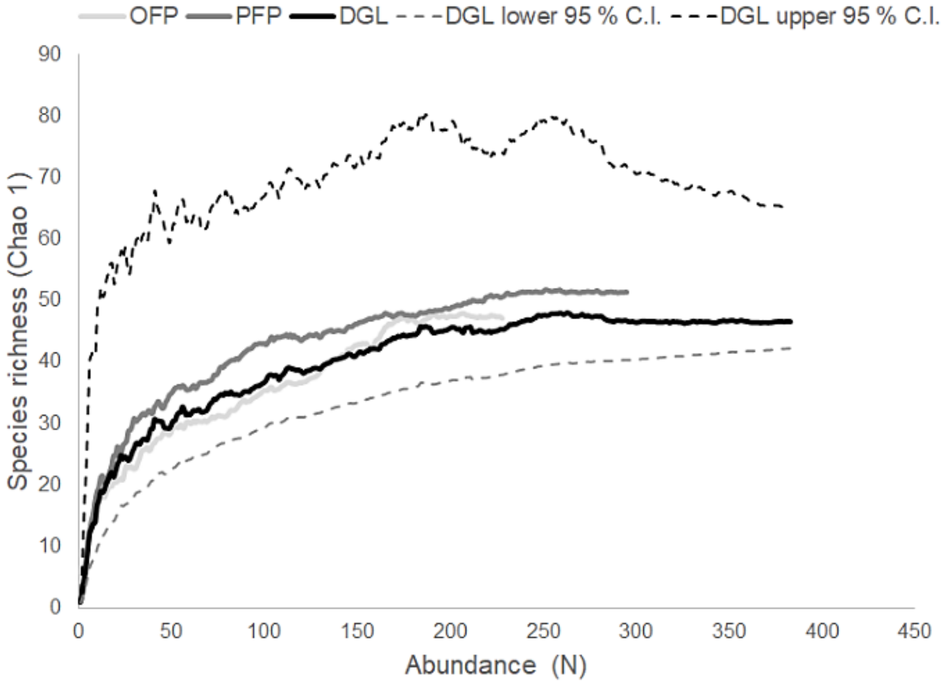

Figure 2. Cumulative (12 months) number of species plotted for the cumulative number of individuals observed in each habitat (DGL, OFP and PFP); dashed lines are the upper and lower end of 95% confidence interval of DGL, the most populated habitat of the AMNP.

Table 2. Butterflies' diversity (means ± SE) detected in the three habitats (DGL, OFP and PFP) (cumulative data for 12 months).

Estimator	DGL	OFP	PFP
Absolute abundance	128 ± 17.21	76 ± 11.15	98.33 ± 3.93
Species richness	25 ± 2.52	18 ± 1.73	23 ± 0.58
Number of singletons	8 ± 0.58	7.33 ± 1.45	7.66 ± 1.20
Number of doubletons	4.33 ± 1.45	2.67 ± 1.20	5 ± 0.58
Chao 1 richness	30.85 ± 1.34	25.71 ± 3.74	27.83 ± 2.05
Fisher's alpha diversity	9.36 ± 0.95	7.88 ± 1.41	9.48 ± 0.50
Shannon's diversity (n log)	3.98 ± 1.26	2.42 ± 0.14	2.67 ± 0.09
Simpson's index (inv.) diversity	10.59 ± 0.85	8.36 ± 1.14	11.81 ± 1.34

Species richness, Shannon index and Chao-1 diversity registered the highest values in the DGL habitat. The frequency of singletons, i.e., species represented by single individuals, was low in all habitats during the butterfly surveys. Meanwhile, the Simpson and Fisher diversity values were highest in the PFP habitat due to the small number of abundant species and the large proportion of "rare" species (Table 2).

The three habitats appeared to be differentiated according to the presence of some exclusive species as follows: *Cupido minimus* (Fuessly), *Melanargia galathea* (L.) and *Pseudophilotes baton* (Bergstrasser) in OFPs; and *Euchloe ausonia* (Hubner), *Gonepteryx cleopatra* (L.), *Pieris mannii* (Mayer) and *Spialia sertorius* in PFPs; *Carcharodus flocciferus* (Zeller), *Colias alfacariensis* Ribbe, *Lampides boeticus* (L.), *Melitea trivia* (Denis et Schiffermuller) and *Polyommatus coridon* (Poda) in DGLs. On the contrary, some other species were present just in forest paths (*Limenitis reducta* Staudinger, *Nymphalis polychloros* (L.), *Pararge aegeria* (L.) and *Pyronia ceciliai* (Vallantin)) according to Bruno [48] and Villa et al. [40]. In Figure 3, the distribution of butterflies by PCA showed high proximity of most species on the origin of the biplot with PC1 = 79.01% and PC2 = 18.91%. The butterfly families exhibited overlap among their distribution (ellipses).

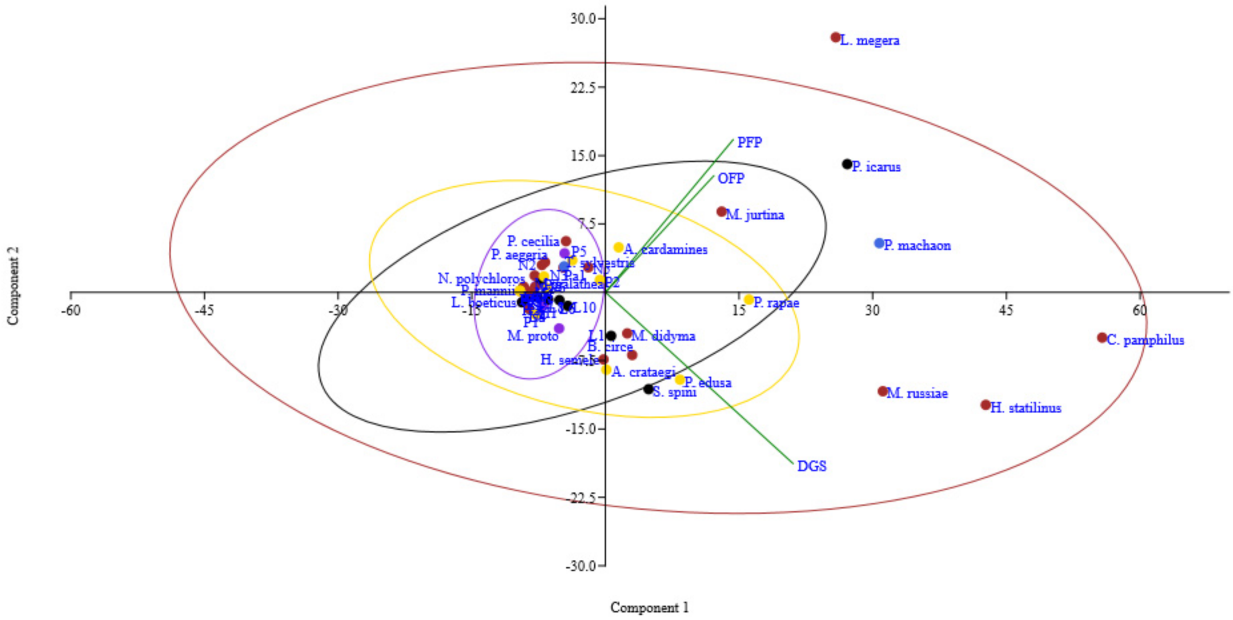

Figure 3. Results of principal component analysis (PCA) showing the differentiation of species by the first two principal axes with 95% confidence ellipses of 5 butterfly families. The orientation of an ellipse depends on the correlation coefficient between the variables. The position of each species in biplot space indicates the strength of the relationship between species and habitat types (green vectors). Axes 1 and 2 show the PCA of the distribution of species and habitat parameters.

The vectors of PFP and OFP were closely overlapped, indicating a strong mutual correlation in the two-dimensional plane of the principal components. The two forest habitats had high PCA scores for *M. megera*, *P. icarus* and *M. jurtina*, while dry grasslands had a high value for *P. edusa* and *M. russiae.*

3.2. Seasonal Population Dynamics

Butterfly abundance was affected by sampling time ($F_{11,94} = 9.36$; $p < 0.001$). The higher seasonal frequencies of butterfly captures were recorded during the months of Spring 2017, with a peak in May (N = 200) (Figure 4; Table 3). No butterfly was detected in December 2016 and only a few individuals were detected in January and February 2017 in forest paths.

A positive correlation between the monthly butterfly abundance and monthly mean temperature of the air was pointed out (Pearson correlation: r = 0.8; $p < 0.001$). The most abundant families, Lycaenidae and Nymphalidae (70% of total abundance), increased as the temperature increased. Abundance in the butterfly community was affected by habitat ($F_{2,94} = 3.48$; $p = 0.035$); in DGL and PFP, abundance was significantly higher than in OFP (HSD Tukey's test: $p < 0.05$). No significant difference was observed in butterfly abundance between the transect replicates of the same habitat ($F_{2,92} = 0.81$, $p = 0.445$).

The month of first record and monthly abundance were reported for each recorded butterfly species in Table 3 to focus on phenological advancements (emerging earlier). Some of the recorded species are of conservation concern: according to the European Red List of Butterflies [49], *N. polychloros* (the "blackleg tortoiseshell") is classified as a vulnerable species, while *M. trivia*, *C. flocciferus* and *H. statilinus* are near threatened. Finally, *M. arge* (the "Italian marbled white") is a European endemism listed in the Habitats Directive 92/43/EEC [50].

Noticeably, the study sites appeared very relevant in terms of the abundance of *Coenonympha pamphilus*, *L. megera* and *P. icarus*, which are reported to be in strong or moderate decline in the European Union [51] and can indicate the environmental quality of the AMNP territory. Few specimens of *Lycaena phlaeas* (L.) and *Ochlodes sylvanus* (Esper), which are in moderate decline in the EU, were recorded in the study area and need to be monitored to assess actual information regarding their population trends in order to improve the efficiency of conservation measures.

Figure 4. (**A**) Abundances of the five butterfly families (colored bars) and monthly mean of air temperature (2016–2017; sharped line). Different letters on the top of bars indicate differences between monthly butterfly abundance (HSD Tukey's test, $p < 0.05$). (**B**) Changes of butterfly Families during 12 months in three habitat types (below radar graphs).

Table 3. Checklist of butterfly species, their temporal variation, total abundance (N) and first record of flight (1st record) in PNAM. The flight period is evidenced in yellow cells. Habitat types: GR, grasslands, meadows, stony/rocky slopes, dry grasslands, open habitat, pastures; SCR, scrubby; WE, woodland edges/clearings; DW, deciduous woodlands; OAKW, oak woodlands; PINEW, pine woodlands.

	Month												N	1st Rec.	Habitat
	i	ii	iii	iv	v	vi	vii	viii	ix	x	xi	xii			
Hesperiidae															
Carcharodus alceae (Esper)								4	1				5	VIII	GR/SCR
Carcharodus flocciferus (Zeller)								1					1	VIII	GR/WE
Muschampia proto (Ochsenheimer)								6	1				7	VIII	GR/SCR
Ochlodes sylvanus (Esper)					1			1	1				3	V	GR

Table 3. *Cont.*

	Month												N	1st Rec.	Habitat
	i	ii	iii	iv	v	vi	vii	viii	ix	x	xi	xii			
Pyrgus malvoides (Elwes et Edwards)			1	2	1								4	III	GR
Spialia sertorius (Hoffmansegg)					1								1	V	GR
Thymelicus sylvestris (Poda)					11								11	V	GR/WE
Lycaenidae															
Aricia agestis (Denis et Schiffermüller)					1		2	4	9				16	V	WE
Callophrys rubi (L.)			1	1									2	III	DW/SCR
Celastrina argiolus (L.)			1		1			1	2				5	III	WE/SCR
Cupido minimus (Fuessly)				2									2	IV	GR
Lampides boeticus (L.)						1							1	VI	GR
Leptotes pirithous (L.)						1			1				2	VI	GR
Lycaena phlaeas (L.)	1		1			1		1	4				8	I	GR/WE
Polyommatus bellargus (Rottemburg)				2				1	1				4	IV	GR
Polyommatus coridon (Poda)								1	1				2	VIII	GR
Polyommatus icarus (Rottemburg)				27	3	20		3	16				69	IV	GR/WE
Polyommatus thersites (Cantener)								4	1	1			6	VIII	GR/WE
Pseudophilotes baton (Bergstrasser)				2									2	IV	GR
Satyrium ilicis (Esper)					10								10	V	OAKW
Satyrium spini (Denis et Schiffermüller)					21								21	V	GR/WE
Nymphalidae															
Brintesia circe (F.)					6	9	2	3					20	V	GR/WE
Coenonympha pamphilus (L.)			21	28	14	21	5	4	9	8			110	III	GR/WE
Hipparchia semele (L.)					9			2	3				14	V	PINEW
Hipparchia statilinus (Hufnagel)					1	28	33	19	5				86	V	GR
Issoria lathonia (L.)						1		1	1				3	VI	GR
Lasiommata megera (L.)	1	5	9	15	3	1		6	9	12	10		71	I	GR
Limenitis reducta Staudinger					1	1	2	1	1				6	V	DW/WE
Maniola jurtina (L.)					19		5	9	10				43	V	GR/WE
Melanargia arge (Sulzer)				9	1								10	IV	GR
Melanargia galathea (L.)						5							5	VI	GR
Melanargia russiae (Esper)					61	3							64	V	GR
Melitaea didyma (Esper)				3	11			6					20	IV	DW/WE/GR

Table 3. *Cont.*

	Month												N	1st Rec.	Habitat
	i	ii	iii	iv	v	vi	vii	viii	ix	x	xi	xii			
Melitaea trivia (Denis et Schiffermüller)				2									2	IV	GR
Nymphalis polychloros (L.)			1					1					2	III	DW/WE
Pararge aegeria (L.)			4	3									7	III	DW/WE
Pyronia cecilia (Vallantin)						12		1					13	VI	GR/WE
Vanessa atalanta (L.)	4								3	6	2		15	I	GR
Vanessa cardui (L.)								1	3				4	VIII	GR
Papilionidae															
Iphiclides podalirius (L.)			1	1		3	2	2	1				10	III	GR
Papilio machaon L.		2	17	10	8	6		5	15	5	1		69	II	GR/WE
Pieridae															
Anthocharis cardamines (L.)			20	2									22	III	GR
Aporia crataegi (L.)				11	3								14	IV	GR/WE
Colias alfacariensis Ribbe			1	1				1					3	III	GR
Colias crocea (Fourcroy)			1	3	4	1		2	2	2	1		16	III	GR
Euchloe ausonia (Hubner)				1									1	IV	OAKW
Gonepteryx cleopatra (L.)			1										1	III	GR/WE
Gonepteryx rhamni (L.)			8	2	1	1				1			13	III	GR/WE/SCR
Pieris brassicae (L.)			2						2	2			6	III	WE/SCR
Pieris mannii (Mayer)									1				1	IX	GR
Pieris napi (L.)			2			1		1	1	1			6	III	GR/SCR
Pieris rapae (L.)	1		16	11	6	1		5	1	2			43	I	GR/WE
Pontia edusa (F.)			2	6	2		8	2	7				27	III	GR/CUL

4. Discussion

During the present survey, butterfly phenology was highly correlated with temperature; a low presence of butterflies was registered in all habitats in late autumn and winter, probably due to low temperatures and limited daylight. The highest abundance peaked in spring, which was followed by a decline in summer, more evident in dry grasslands than in forest paths. Cormont et al. [52] observed the same trends and related the frequency of butterfly flight to temperature and radiation incidence. This trend can be explained by the larger insolation degree of the DGL, which influences the abundance and distribution of flowering plants that are actively sought by butterfly adults for their nutrition and oviposition [32,53,54]. The distribution of butterfly communities was non-random, but likely oriented to some ecological traits in the habitat (i.e., characteristics of host plant species). Some species of Papilionoidea found in the AMNP have larval stages that are tightly linked to plants present in meadows and forest clearings, such as *Melanargia* spp. on Poaceae and *P. machaon* on Apiaceae. Others, such as *Iphiclides podalirius* (L.), *Limenitis reducta* Staudinger and *Aporia crataegi* (L.), have larvae linked to trees and shrubs [40,48,55,56].

Climate changes drive phenological changes in butterflies, and there is considerable variation in the use of species' traits to account for predicted future phenological responses [57]; however, it is unclear whether this is due to the low explanatory power of species' traits or to limited species numbers and study durations.

During the survey, several species were common to different habitats, e.g., species spread in meadows were found also in forest paths. The presence of species typical of grasslands in oak and conifer forests might be due to paths and cleared areas interspersed between the tree canopies that showed some similar vegetational characteristics between these two habitats [58,59]. Species usually occurring in a wide range of habitats can move through the landscape more easily than those that are habitat specialists. Xerophilous species, such as *P. edusa*, C. *pamphilus*, *H. statilinus* and *M. russiae*, are inextricably linked to habitats rich in Poaceae, such as *Poa*, *Brachypodium*, *Stipa*, *Cynosurus*, *Anthoxanthum*, *Lamarchia*, *Briza*, *Festuca*, etc., which represent the feeding plants of their larvae [16,18,40]. They are also considered widespread or habitat generalist species [60,61]. Dominant species characterized each habitat of the AMNP, and they were consistent with those found in the Murgia Materana Regional Park, close to the AMNP and with many environmental similarities [62]. Some dominant species of the grasslands differed from those of the forests, as has been previously reported [63].

The current results are in accordance with Fisher et al. [64], who stated that no community has equally common species but only few species can be abundant, while most of them are represented by a few individuals. Our results display a rich diversity of butterflies in each habitat. The biodiversity indices (Chao 1, Fisher's alpha, Shannon's index and Simpson's index) for each of them were generally high and similar to the highest values recorded in protected areas of Sicily [65]. Singletons in the butterfly surveys would have a low chance to reproduce and could play a significant ecological role in local rarefaction or extinction of populations. Here, the number of singletons was low in the total butterfly community. Hosting a rich butterfly fauna with target species based on the conservation EU priorities justifies the inclusion of the ANMP in the "Prime Butterfly Areas". The monitoring of these species can represent efficient support in biodiversity conservation and environmental planning [66]. These species, together with others that are extremely localized, deserve to be protected by (a) safeguarding their habitats; (b) limiting or inhibiting the use of plant protection products in sites hosting both larvae and adults; and (c) spreading the host plants for larvae and adults. Although it is advisable that butterfly research in selected sites should be continuous for at least three years (i.e., [67]), these preliminary results already yield plans for further monitoring to support important conservation policies for xerocalcicole grasslands and South Mediterranean European forests within the EU Natura 2000 ecological network. The present contribution to the knowledge of butterfly diversity of the ANMP points out the significance of both dry grasslands and woodlands to safeguard the butterfly species, including those of conservation concern [53,68]. It is widely acknowledged that the most specialized species usually have the highest susceptibility to habitat disturbance. Therefore, preservation of the different habitats is crucial for butterfly conservation as it ensures suitable conditions for specialized species to thrive [69]. Furthermore, assessing the properties of butterfly communities at different spatial and temporal scales can be used as an important connection strategy among sites of community importance in the EU Natura 2000 network and to study the global impacts of climate change. From this point of view, the ANMP represents a significant chance in conservative policies and biodiversity heritage. In this sense, of interest is the recording of *Zerynthia cassandra* Geyer in areas very close to some of those currently surveyed (Panzarino, Loverre and Mastronardi, personal observations). This butterfly is endemic to Italy, and its biodiversity value is demonstrated by its protection status under the Habitats Directive 92/43/EEC (Annex IV) and the Bern Convention (Appendix II) as *Z. cassandra* sensu lato.

5. Conclusions

Habitat loss and fragmentation are among the main threats to species inhabiting dry grassland. The butterfly communities of the Alta Murgia National Park showed high diversity, which is reasonably ascribable to the existence of the protected area within suitable environmental conditions and adequate trophic sources. The complexity of the eco-mosaic of the park represents a "core area" in an ecological network with heterogeneous environments. Throughout the three habitats, DGL presented a very high abundance and richness of butterflies, pointing out the relevance and role of semi-natural dry grassland in fauna conservation. Hosting a rich butterfly fauna with "target species" justifies the inclusion of the ANMP in the "Prime Butterfly Areas".

The first flight of some species (i.e., *Pyrgus malvoides* (Elwes et Edwards), *Thymelicus silvestris* (Poda)) was registered earlier than in the references, suggesting possible phenological advances linked to global warming but needing further inquiry. In this context, ecological connectivity should be favored to enhance species displacement capabilities, which could be critical to face global changes.

This survey indicates that habitat specialist species may suffer from habitat fragmentation. Therefore, the species–area relationship is important for ensuring long-term biodiversity preservation [68]. This evidence can be used to design incisive strategies, ensuring connections among sites of community importance in the EU Natura 2000 network, as well as to study the global impact of climate change on biotic communities by monitoring butterfly flight periods.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/land12051039/s1. Table S1: Abundance of butterfly communities in three different habitats.

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