BABEȘ-BOLYAI UNIVERSITY
FACULTY OF BIOLOGY AND GEOLOGY
DOCTORAL SCHOOL OF INTEGRATIVE BIOLOGY

DOCTORAL THESIS

Scientific Supervisor
Prof. László RÁKOSY, PhD

PhD Student
Elena-Cristina COSTACHE

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Ecological and phenological changes in butterfly communities as a result of climate change and land use (by farmers)

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2025

Bărăi, Cluj – June 2022

„contimporan cu fluturii, cu Dumnezeu”

Lucian Blaga

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OUTLINE OF THE THESIS

Biodiversity is constantly changing.

Butterflies are probably the best-known group in the Insecta class. Although they are not the most species-rich order on the planet, with Coleoptera occupying first place with approximately 386,500 species (Stork, 2018), representatives of the order Lepidoptera have always attracted the attention of collectors, nature enthusiasts and entomologists – through their colour contrasts, unique wing patterns and graceful flight – being true symbols of beauty and richness, but also indicators of the state of ecosystems.

Over the past two decades, an increasing number of manuscripts have been published with the aim of raising awareness about the significant decline of insects on the two continents, Europe and North America.

OBJECTIVES

CHAPTER I

- To examine the effect of pesticides used in agriculture on 16 butterfly species in Transylvania, Romania
- To analyse the impact of climate change and land-use change on the decline of butterflies

CHAPTER II

- To investigate the impact of climate change on the phenology of 11 butterfly species in a small region in Transylvania, Romania.

CHAPTER III

- To compare abundance and diversity of butterfly species in different shrubs removal treatments
- To determine possible indicator butterfly species in different shrubs removal treatments

CHAPTER IV

- To identiy farmers' perceptions of the costs associated with biodiversity measures

- To assess the extent to which their viewpoints are influenced by local context and the specific biodiversity measures implemented

CHAPTER I

THE DECLINE OF BUTTERFLY POPULATIONS DUE TO CLIMATE AND LAND-USE CHANGE IN ROMANIA

Cristina COSTACHE, Andrei CRIȘAN, László RÁKOSY

The widespread use of pesticides, changes in the structure of the landscape due to industrial and agricultural uses, and climate change have massively affected insect populations. The decline in insect biodiversity and biomass is documented in some countries in Europe, Asia, and North America. Romania, compared to Western and Central European countries, is considered to preserve a less altered nature and high biodiversity, being often given as a positive example in terms of conservation of natural and seminatural structures of the landscape. Unfortunately, the reality is different. The negative trends highlighted in Central European countries are reflected almost at the same intensity in Romania. Biodiversity loss is one of the most alarming environmental issues nowadays. The magnitude and speed of biodiversity decline affect many taxonomic groups and bioregions differently (Pilotto et al., 2020).

Climate change affects not only the biomass and species richness of certain taxonomic groups (Hallmann et al., 2017) but also the interaction between species, community structure, and geographical distribution (Devictor et al., 2012). A good indicator of local or global climate warming is advance in phenology, studied in plants and animals (Macgregor et al., 2019).

Another factor that contributes massively to the decline of local and global biodiversity is the increase of land use intensity (Blüthegen et al., 2016). A special form of land use intensity is the continuous increase in the amount and toxicity of fertilizers and pesticides used in agriculture and forestry (Mineau and Whiteside 2013).

The Situation in Romania

Unlike southwestern, southern, and southeastern Romania, in Transylvania, where agricultural areas are small, they are not cultivated by important Romanian or European investors, who would turn the plots of land into a monoculture. Some risks for this region are large agricultural equipment and intensive grazing by sheep. Especially since Romania's accession to the European Union, the number of sheep has increased massively due to poorly designed or allocated subsidies. In addition, due to political involvement, grazing is carried out until winter, regardless of the rules imposed by ecologists. Thus, the consequences for pasture biodiversity are more or less negative. The population of diurnal and nocturnal butterflies has fallen sharply in recent years. The reasons are that many areas of pasture (including hay meadows) have intensive grazing, and small plots of land are gradually abandoned by villagers who move to cities (Rakosy 2013).

Materials and methods

We selected the data of the first observations noted in the bibliographic sources. These were entered into a database in Excel, then were processed in the form of a histogram chart.

To highlight changes in the phenology of the flight period for spring species of diurnal butterflies, we have studied the flight periods for 15 species from representative museums and private collections as well as personal data for the period 1920-2019 from the surroundings of Cluj. The twelve species with spring generations are: *Erebia medusa, Vanessa atalanta, Plebejus argus, Glaucopsyche alexis, Pieris napi, Leptidea sinapis, Iphiclides podalirius, Papilio machaon, Pyrgus malvae, Pieris rapae, Colias hyale, Vanessa cardui.*

The nine species with autumn generations are: *Iphiclides podalirius, Papilio machaon, Polyommatus icarus, Colias croceus, Colias hyale, Leptidea sinapis, Pieris napi, Pieris rapae, Coenonympha pamphilus*.

Results

Of the twelve species with the flight period at the beginning of spring, from which we took data for the nine species, we had sufficient data from the literature as well as recent data to make a graph of phenology (Figure 1.).

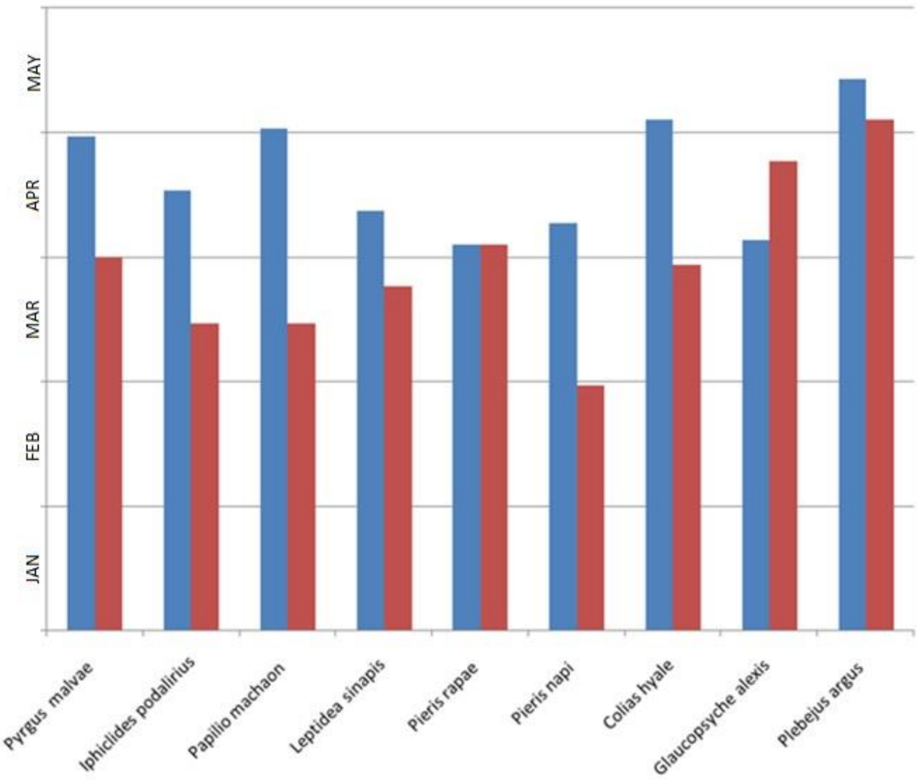


Fig. 1 Spring phenology, noting the first observation of the first specimens for each species, by calendar months (Blue bars - the first report in the period between 1920 and 1940; red bars - the first report in the period between 1990 and 2019)

We selected the first date when butterflies were observed in flight from old literature and collections and from recent observations made in the surroundings of Cluj-Napoca. The nine species with spring generations are: *Plebejus argus, Glaucopsyche alexis, Pieris napi, Leptidea sinapis, Iphiclides podalirius, Papilio machaon, Pyrgus malvae, Pieris rapae, Colias hyale*. We observed that phenology has changed during these almost 100 years, butterflies being observed by lepidopterologists 7 to 45 days earlier during the 1990 – 2019 period than 1920 - 1940. Those with significant differences were the first specimens of the species *Iphiclides podalirius,* which were observed about 37 days earlier in the recent period than in

1920-1940, and the first specimens of *Papilio machaon* - 45 days. The phenological changes may reveal adaptations to climate change in the last hundred years.

For the nine species, the flight period extended until the end of autumn, from which we took data for the three species, we had sufficient data from the literature as well as recent data to make a graph of phenology (Fig. 2).

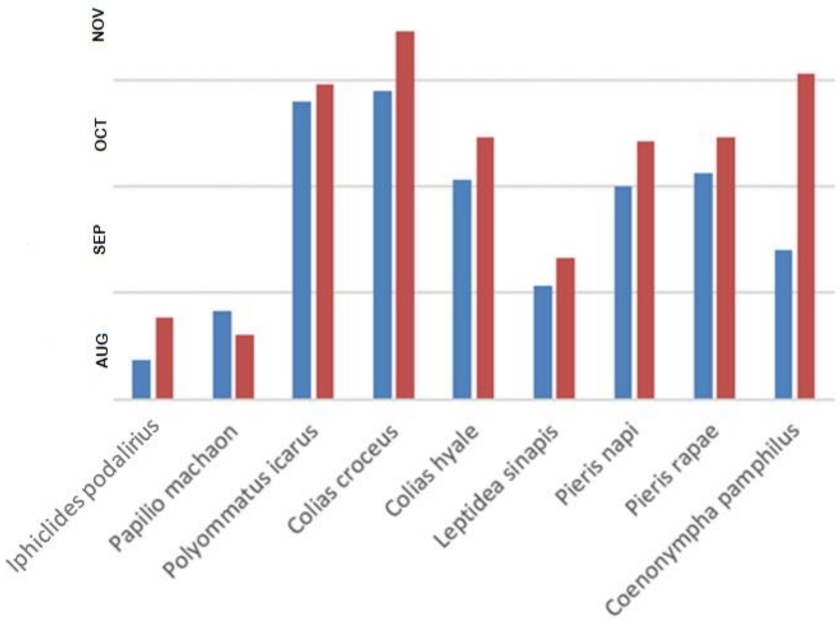


Fig. 2 Autumn phenology, noting the last observation of the last specimens for each species, by calendar months (Blue bars - the last report in the period between 1920 and 1940; red bars - the last report in the period between 1990 and 2019)

We selected the last date when butterflies were observed in flight from old literature and collections and from recent reports made in the surroundings of Cluj-Napoca. The nine species with autumn generations are: *Iphiclides podalirius*, *Papilio machaon*, *Polommatus icarus*, *Colias croceus*, *Colias hyale*, *Leptidea sinapis*, *Pieris napi*, *Pieris rapae*, *Coenonympha pamphilus*. We observed that phenology has changed during the past 100 years, butterflies being observed by lepidopterologists 20 to 30 days later during 1990-2019 period than 1920-1940. Those with significant differences were the last specimens of the species

Coenonympha pamhilus, which were observed about 30 days later in the recent period than in 1920-1940, and the first specimens of *Colias croceus* and *Polyommatus icarus* - 20 days. These phenological changes may reveal adaptations to climate change in the last hundred years

Using the data regarding butterflies from the surroundings of Cluj from the period 1920-2019, we could show that 16 butterfly species have disappeared and 29 species have gone through a massive population decline. On the other hand, in the Red List of Romanian Lepidoptera, 25.11% species are considered endangered.

CHAPTER II

Long-term phenological shifts in butterfly species from Transylvania, Romania – a case study

Cristina COSTACHE[1], László RAKOSY [2,3], Demetra RAKOSY[4]

Insects react rapidly but very diversely to climate change through population oscillations, areal regression, latitudinal or altitudinal expansion, changes in voltinism, changes in life cycle, changes in flight period, changes in sedentary or migratory behaviour, changes in habitat or food preference, etc. All these reactions have regional geographic valences, determined by geological, climatic, biological and anthropogenic factors.

Phenological shifts in organisms serve as valuable bioindicators of climate change [21, 22]. This is particularly true for butterflies, which serve as valuable indicators for studying climate change due to their sensitivity to climatic and environmental variables, rapid responses, and extensive history of research [17, 20, 23, 24]. As poikilothermic organisms, which evolved obligatory mutualisms with plants both as adults and as larvae, butterflies, are highly influenced by variations in temperature and precipitation [4,25]. The situation in countries, such as Romania is no better than in Central European countries [43]. Due to the lack of extensive data provided by volunteers through citizen science, the phenological response of insects to climate change in countries that do not benefit from a large monitoring network is not known. In order to change the situation and to bring more data and knowledge, we aimed to make use of the existing data – from historical collections and present day observations - on butterflies from the most studied area in Romania, located in Transylvania, around the city of Cluj-Napoca.

Thus, we analysed the response of selected butterfly species to climate changes—defined as temperature averages, snow cover on the ground, and precipitation—and the potential impact of these changes on their annual phenology.

Materials & methods

The historical collections and present day observations were used to extract information of the date of first emergence per year for all butterfly species recorded in the area of Cluj-Napoca. From the total of 95 butterfly species known to occur in the area [**Error! Reference source not found.**] we selected 16 species, based on data availability, the number of generations per year, overwintering stage and the period of emergence. For species which emerge from the pupa in spring 12 species selected: *Erynnis tages* (Linnaeus, 1758), *Pyrgus malvae* (Linnaeus, 1758), *Iphiclides podalirius* (Linnaeus, 1758), *Papilio machaon* (Linnaeus, 1758), *Leptidea sinapis/juvernica* (Linnaeus, 1758), *Anthocaris cardamines* (Linnaeus, 1758), *Pieris napi* (Linnaeus, 1758), *Pieris rapae* (Linnaeus, 1758), *Colias alfacariensis/hyale* (Ribbe, 1905), *Glaucopsyche alexis* (Poda, 1761), *Plebejus argus* (Linnaeus, 1758), *Erebia medusa* ([Denis & Schiffermüller], 1775). The five butterfly species with long flight periods which extend towards autumn were: *Pieris rapae* (Linnaeus, 1758), *Colias hyale/alfacariensis* (Ribbe, 1905), *Colias crocea* (Geoffroy in Fourcroy, 1785), *Polyommatus icarus* (Rottemburg, 1775) and *Coenonympha pamphilus* (Linnaeus, 1758). Finally, a migratory species, *Vanessa atalanta* (Linnaeus, 1758) was chosen, which is not believed to overwinter as an adult in Romania, but which may do so if climatic conditions change. Each observation was annotated with species name (genus, species) and date of observation (day, month, year). To standardize the data, only the first date the species was observed was selected, with a single record.

For the studied period (1921-2013), we used monthly average data for temperature (°C) and total monthly precipitation (l/m2), as these two climatic parameters are essential for influencing the emergence dates of butterflies

Results

From the eleven butterfly species which emerge in spring, five species exhibited phenological shifts in more recent records (post-1973), as evidenced by a concentrated cluster of observations occurring earlier in the season (Fig. 1). The observations of the species *Pieris napi*, *Colias hyale* and *Leptidea sinapis/juvernica*, show a drastic shift, with peak observations moving from May to April and mid-March over the same time frame (Fig. 2). Data after 1973 being more numerous, the trend change is more evident for the last 40 years. Not all species, however, show clear phenological shifts between 1973 and 2023, with some species exhibiting smaller or less consistent changes. For example: *P. malvae* and *E. medusa* show only moderate shifts in their phenology (Fig. 2). *G. alexis* and *E. tages* appear to show relatively stable emergence times, with peak first observations remaining from late April to mid-May (Fig. 2 c, d)

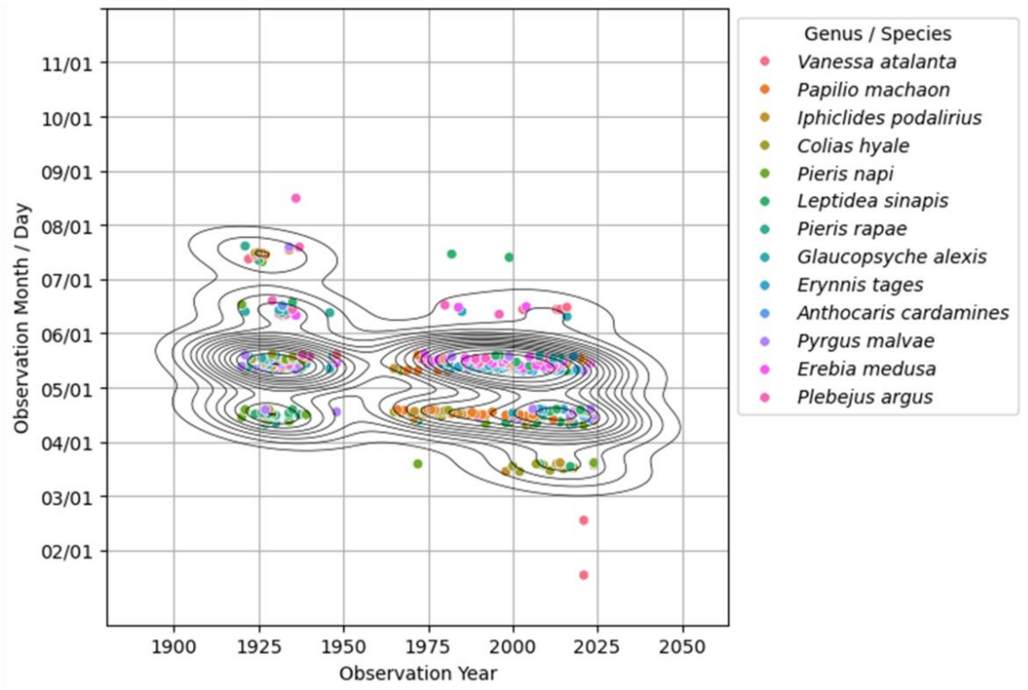


Fig. 3 Phenological trends in the butterfly species with spring generations from 1920 to 2023. Individual species are represented by different colours in the scatterplot and are part of the cumulative data analyses for phenological shifts. Lines represents the KDE (kernel density estimation) fit through the data, representing a 2D distribution of the data.

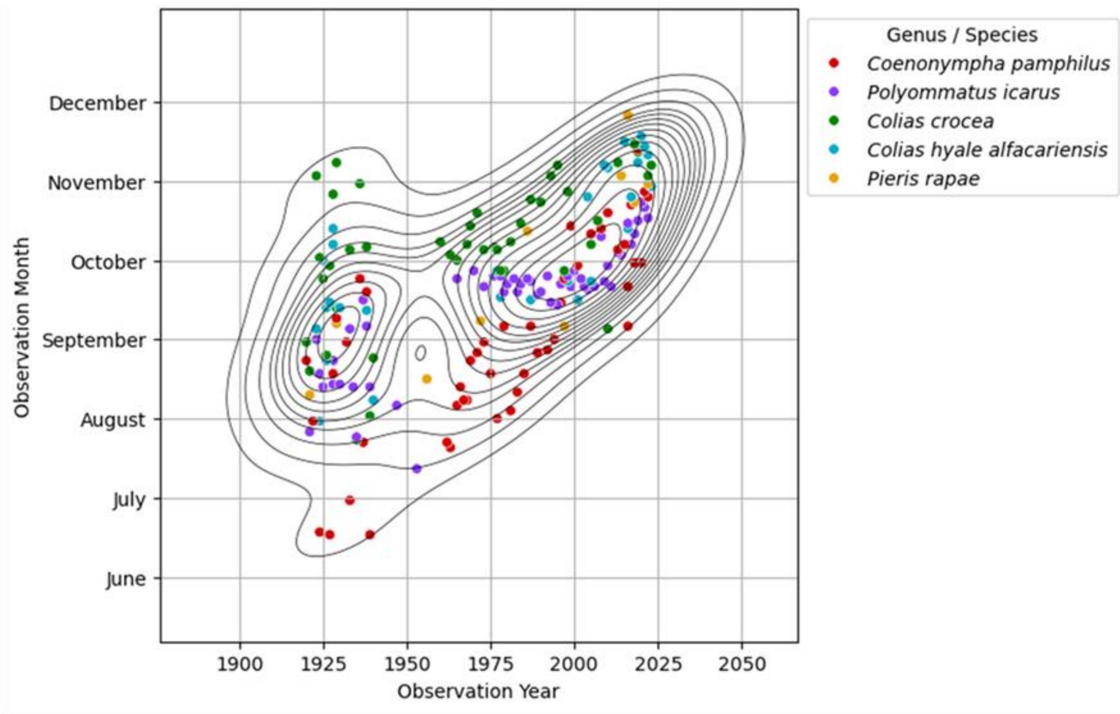


Fig. 4 Phenological trends in butterfly species with autumn generations from 1920 to 2023. The individual species are represented by different colours in the scatterplot and are part of the cumulative data analysed for phenological shifts. Lines represents the KDE (kernel density estimation) fit through the data, representing a 2D distribution of the data

The five species of butterflies chosen to capture the possible shift in the phenology for multivoltine butterflies, showed a real prolongation of the flight period towards the end of October and beginning of November. The shift seems to have started around the 1970s-1980s and had a more pronounced shift in recent years (after the 2000s) (Fig. 3). For *C. crocea*, *P. icarus* and *C. pamphilus,* the extending of the flight period towards the end of October is documented for the whole period 1973-2023 (Fig.4 a, b, e). *P. rapae* extends its flight period by more than 35 days between 1973-2023 (Fig 4d). The most profound phenological shift is recorded in the *C. hyale/alfacariensis* complex, which extended its flight period from mid-September to early November between 1980-2023 (Fig. 4c).

Discussions

In the present study an increase of +2°C over the past 40 years was found, aligning with global observations of anthropogenic climate change during the latter half of the 20th century and early 21st century [27].

In the absence of a standardized monitoring program for butterflies in Romania, we used all available historical and current data for one of the best studies areas of the country (Cluj-Napoca and its surroundings, Transylvania) from 1920-2023 to assess whether diurnal species have undergone phenological changes due to climate change. The results show a phenological shift towards March-February for butterflies hatching from pupae in spring and a shift of the flight period towards October-November for species active in late summer. At the same time, an increase in voltinism was observed, with some monovoltine species becoming bivoltine and some bivoltine species becoming consistently trivoltine. In the case of the migratory species *Vanessa atalanta*, it has also been shown that it can successfully overwinter in Transylvania under the winter conditions of the last 10-15 years.

Conclusions

This is one of the few studies which has combined historical and present-day data to assess how the phenology of butterflies' changes in response to climate change in Eastern Europe. This region has been long considered a safe haven for biodiversity due to the comparatively lower rates of land-use change. However, this view needs to be challenged, considering the results of an increasing number of studies, which highlight that while Easter Europe may face in part different challenges as Western Europe, land-use and climate change are still negatively impacting its once well-preserved biodiversity. And while our study highlights how climate change may impact species differently; further research is needed across a broader range of environmental gradients to accurately assess the response of butterfly species. Only in this way will it be possible to prioritize species for conservation action.

CHAPTER III

The role of shrubs in maintaining butterflies' biodiversity in grasslands

Cristina COSTACHE, Flaviu BODEA, Ionuț TĂUȘAN, László RÁKOSY

Introduction

Shrubs are an essential component of the agricultural and grassland landscapes of many farms and semi-natural grasslands around the world, and their management plays an important role in enhancing habitat biodiversity. In this article, we show how shrub-removal management practices affect butterfly and plant communities in a small region of Transylvania, Romania.

Even though it is recommended in the agri-environment work package that farmers should maintain 5-15% of shrubs after hedgerow management to preserve insect diversity, this is still not well understood by some farmers and is not being properly implemented, resulting in unintended negative consequences such as declines in biodiversity, soil erosion, and the degradation of landscape features (Cremene et al. 2005).

In this study, we aim to show to measure the diversity of flowering plants and butterflies and butterfly indicator species where hedgerow conservation measures such as shrub removing have been implemented in 15 sites near Cluj-Napoca, Transylvania

Materials & methods

We surveyed butterfly communities in early summer and late summer - in May and August in 2022 and 2023 across 15 sites (1 reserve), in the surroundings of Cluj-Napoca, Transylvania. We ensured that daytime sampling is randomized across sites. The sampling design was chosen as four linear 50m long belt transects of 1.5m width (7min 30sec sampling time per transect) positioned parallel to the field edge at approximately 1m, 10m, 20m and 30m from the field edge, covering a total area of 300m2. We recorded flowering vascular plant diversity using 10 quadrats of 50x50 cm (0.25 m2) per site. Plant diversity was surveyed on the same day as the butterfly transects. To estimate flower cover in shrubby vegetation, hedgerows, we used the same methodology as described above to estimate herbaceous vegetation.

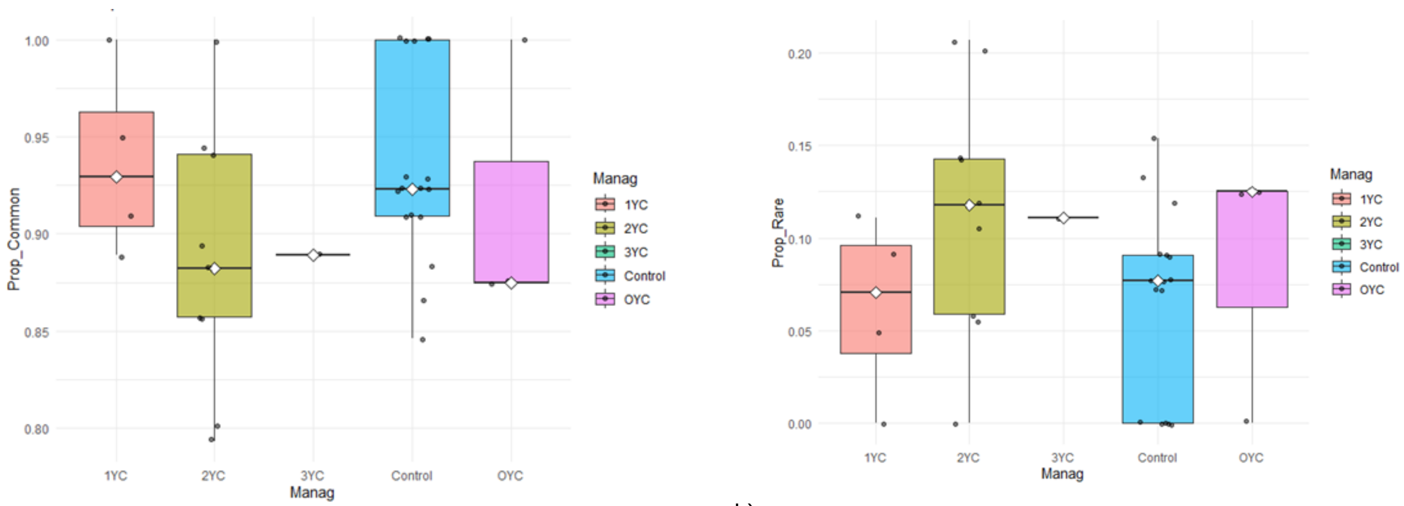
We classified hedgerows according to the trimming management type into four groups: 1YC-hedgerows that were cut one year prior to first year sampling; 2YC- hedgerows cut two years prior to first year sampling; 3YC-hedgerows removed three years prior to first year sampling;

OYC-hedgerows that were removed more than 3 years prior to our first year sampling and control where hedgerows were never trimmed.

Results

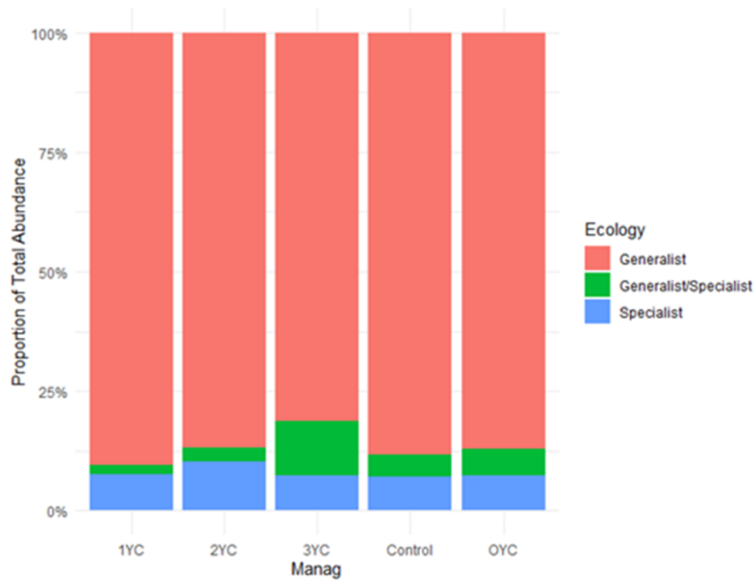
The analysis of frequency categories revealed no statistically significant differences (Kruskal-Wallis test, p = 0.37) in the occurrence of common butterfly species across different grassland management regimes. However, a higher frequency of common species was observed in the control plots, followed by 1YC and 2YC plots (Fig. 3a).

A similar pattern was identified for rare species (Fig 3b). Although the highest proportions of rare species were recorded in 2YC and OYC sites, followed by 1YC and control plots, the differences across management types were not statistically significant (Kruskal-Wallis test, p = 0.37).



a) b)

Proportion of common species a) and rare species b) in different shrub managed plots using the Kruskal-Wallis test, p=0.37



Proportion of butterfly species traits abundance by management type (generalist – red bars, generalist/specialist – green bars, specialist – blue bars)

When examining the distribution of ecological categories, generalist species were the most prevalent, followed by specialist species and last were those with mixed traits (generalist/specialist).

However, when these categories were analysed separately, distinct patterns emerged. For generalist butterfly species, statistically significant differences were found across management regimes (Kruskal-Wallis test, $p < 0.05$), with higher abundances recorded in both 2YC and control plots. For generalist/specialist species, the highest abundance was found in 3YC sites. While the most specialist species were found in 2YC sites.

Conclusions

The studied sites, each managed under different regimes, host distinct butterfly and plant communities. These communities are not merely richer or poorer versions of a single species pool but rather consist of entirely different species assemblages. Consequently, rather than experiencing a gradual gain or loss of species across sites, there is a complete turnover in species composition. This suggests that the observed differences are not solely due to relative abundance but reflect fundamentally different combinations of species shaped by specific environmental and management-related factors.

Thus, the agri-environmental schemes aiming at preserving butterfly diversity should promote landscape and hedgerow management heterogeneity and offer incentives to conserve grasslands and agricultural lands with shrubs.

CHAPTER IV

Farmers’ perceived economic and non-economic costs of their biodiversity measures

Verena Scherfranz[a], Henning Schaak[a], Jochen Kantelhardt[a], Karl Reimand[a], Michael Braito[b], Flaviu V Bodea[c], Cristina Costache[c], Razvan Popa[c], Reinier de Vries[d], David Kleijn[d], Aki Kadulin[e], Indrek Melts[e], Amelia S C Hood[f], Simon Potts[f], Lena Schaller[a]

Introduction

Farmers' willingness to continue participation in their agri-environmental program and maintain biodiversity measures in the long term is shaped by the nature of costs they perceive during implementation. Research emphasizes the need to account for both financial and non-financial costs, but holistic assessments which both put these costs into relation and account for farmers' varied perceptions remain lacking.

Methods

To capture the plurality of perceived costs, as well as the plurality of viewpoints farmers have of these costs, we applied Q-methodology across four European study areas. Building upon scientific literature and expert interviews, we defined a Q-set comprising 41 cost aspects from four dimensions, i.e. financial, management-related, emotional and social costs. 34 farmers with different socio-demographic and farming background Q-sorted these cost aspects.

Results

Factor 1: governance-related uncertainty

Farmers sharing the viewpoint represented by Factor 1 emphasize the problem of unstable or unclear regulations, or other forms of governance-related uncertainty coming with their biodiversity measure. More than any other group, they feel too much insecurity due to changing requirements (Q20:+4[13]). Like Farmer RO-5[14], arguing that “*It's not really explained. […] We are in the fog every year*”, Farmer RO-3 reasons that “*policies change from year to year, and we have to adapt on the fly, and we can't make an exact plan*”. Farmer EE-6 relates uncertainty to potentially not receiving money when applying for grants, even in return for major investments: "*And then, how do you build your activities, like, on sand*?" In addition, worries are expressed that lease contracts might be cancelled despite ongoing obligations to maintain the biodiversity measure on the leased land (RO-3), funding might be stopped because of lacking state budget (EE-6), or payments are too dependent on unclear outcomes rather than controllable efforts (EE-5).

Factor 2: unproductiveness

Factor 2 highlights a perceived discrepancy between farmers' own, more production-oriented idea of farming and the need to reduce productiveness when implementing a biodiversity measure. This, on the one hand, results in emotional costs, including loss of identity: More than in any other group, farmers agree strongly that their farmland now looks less appealing to them (Q29:+3*). Farmer NL-10, while acknowledging his measure's results, argues: *"I prefer to see a straight field, like what is being mown every four weeks"* and continues: *"The school I went to, the agricultural school, they say you have to produce. Potatoes, milk, beet; and this has nothing to do with production*". Similarly, they agree relatively strongly that the work associated with their biodiversity measure is not part of a farmer's job (Q19:+2).

Conclusions

In this study, using Q-methodology across four European countries, we showed a plurality of farmer perspectives, with some respondents most affected by governance-related uncertainties, others by perceived unproductiveness, inadequate support, administrative complexity, undercompensation, or social non-conformity. These results demonstrate the inadequacy of one-size-fits-all approaches in policy making and we suggest the need for context-specific, farmer-centered strategies.

The findings suggest that successful implementation of biodiversity measures depends not only on financial incentives but also on addressing broader systemic and perceptual barriers.

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