## Dispersal ability and habitat selection in *Melitaea telona kovacsi* Varga, 1967 and *M. phoebe* (Denis & Schiffermüller, 1775) (Nymphalidae) in steppe grassland

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Abstract. *Melitaea telona* is a protected species in Hungary. The known range has shown significant contraction over the last decades. In order to conserve this species it is important to understand its dispersal ability and patterns of movement. The, mobility of *M. telona* and the related *M. phoebe* were studied using capture-recapture methods. 307 *M. telona* and 139 *M. phoebe* individuals were marked and 12% recaptured during May 2009 in three study areas with ten sample sites. We estimated the densities of the larval food plant (*Cirsium pannonicum*) and of the most important nectar source (*Dianthus pontederae*). *M. telona* is moderately localized: the species is able to reach habitat patches several hundred metres distant. It tends to occur in high numbers where the food plant is abundant. In these sites, fewer *M. phoebe* were found, while there were higher numbers in the more degraded patches. Our data suggests that the density of the food plant is more important than the abundance of nectar sources in habitat selection by *M. telona*.

### Introduction

*Melitaea telona kovacsi* Varga, 1967 is a protected forest-steppe species in Hungary. It lives only in habitats where *Cirsium pannonicum* grows in significant density. Evidence from museum specimens suggest that in the 1960's this species was widely distributed in the central and northern hills of Hungary. Nowadays it is known from only a few localities in the Budai Hills, the region of the Aggtelek-Karst and the Borsodi Hills. Perhaps the most drastic decline has been observed in the Bükk Mts. There are many museum specimens from this region, but in the last few years we could not confirm the species' continuing presence here. The Aggtelek region seems to be the only locality where this species still has strong populations and has any chance of a long term conservation (Varga 2007; Varga et al. 2005). In this region there are many suitable habitats for the species in close proximity to each other. The situation in the Bükk Mts. is totally different with the known, suitable habitats sometimes tens of kilometres apart.

Habitat isolation depends on both the hostility of the environment in between and the organism's gap-crossing ability (Ricketts 2001; Schmitt et al. 2000; Tischendorf and Fahrig 2000; Wratten et al. 2003). Thus to understand isolation, one must consider the distance between all potential source populations and also the landscape between the habitat fragments. Landscape connectivity does not need to imply structural connectivity, but rather functional connectivity. Overall, depending on the species concerned, the landscape context may facilitate or impede movements of insects between habitat patches (Tscharntke and Brandl 2004). The mobility of the species can vary considerably even within a relatively small group like butterflies (Scott 1975).

We have carried out a capture-recapture survey on *Melitaea telona kovacsi* and *M. phoebe*. Our questions were: how localized are the populations, and how strong is

the connection between the habitat patches at different distances apart? In addition we were interested in other basic biological concerns: which plant species are the main nectar sources? What is the relationship between the supply of the larval food plant and the population size?

## Material and methods

*Target species. M. telona* Fruhstorfer, 1908 is distributed in the Ponto-Mediterranean belt. Its range extends from the eastern part of the Mediterranean Sea coast across Asia Minor to the north-western part of Balkan Peninsula and southern Italy including Sicily. The subspecies in the Carpathian basin has become isolated from the main distribution area and it has patchy habitats. In Hungary the only known food plant is *Cirsium pannonicum* while in other regions it feeds on different types of Asteraceae, mainly on *Centaurea* species, mostly local endemics (Russell et al. 2007). The species is univoltine, flying from mid-May to mid-June. The females lay their eggs near to the ground on the undersides of the food plant leaves. The young caterpillars live in a web, spreading out over the plant as they feed, leaving only the upper epidermis intact. When the warmer part of the summer arrives, the caterpillars enter diapause and overwinter in the third instar. The next spring they continue feeding, gregariously at first, but later becoming solitary. When fully developed, pupation occurs in the grass litter. The imago emerges one or two weeks later (Varga et al. 2005).

Melitaea phoebe (Denis & Schiffermüller, 1775) has the widest distribution in the tribus Melitaeini, with many described subspecies. Its range is nearly continuous, extending from North Africa over Eurasia to the Far East. It is widely distributed in Hungary, and was found at all of the sample sites. It is known to use several larval food plant species: Centaurea, Carduus, Cirsium spp. and, like all member of the tribus Melitaeini, is able to absorb iridoid glycosides from the plant (Wahlberg 2000). It is possible to feed the caterpillars with Cirsium pannonicum. Moreover, we collected young caterpillars from this plant and later realised they were Melitaea phoebe. Generally it is only possible to separate the caterpillars once they have reached the 4<sup>th</sup> instar, based on the coloration of the head capsule. M. phoebe has a black and M. telona has a brick red larval head capsule (Russell et al. 2007). The young caterpillars feed together like *M. telona*, but M. phoebe is bivoltine. The first brood flies nearly at the same time as M. telona and the second flies from the end of June/beginning of July to the end of August/beginning of September. To summarise the comparison of these two species, we can say that while *M. phoebe* is a widely distributed euryoecious species, *M. telona* is a narrowly distributed euryoecious species, *M. telona* is a narrowly distributed euryoecious species. uted food plant specialist.

Even though these two species are very similar; we are able to separate them with a high degree of accuracy based on the morphology of wings and the shape of the tips of the antennae (Varga 2007; Varga et al. 2005).

*Data sampling*. Our study areas were in three well-known *M. telona* localities: Szőlőhegy near to Jósvafő, and Zabanyik and Borház-tető between Tornakápolna and Varbóc in the Aggtelek-Karst area. The vegetation of the study areas belongs to the alliance *Polygalo* 

*majori-Brachypodietum pinnati.* The choice of these study areas was based on three main considerations: the presence of the focal species, a variety of distances between the sample sites, and a variation in the density of the food plant in the different sites. The Szőlőhegy study area has been extensively cultivated for a long time, most typically for fruit production, using widely spaced fruit trees with seasonally mown herbaceous vegetation. Thanks to this sensitive management, the vegetation on this hill has become

very similar to that of forest steppe. We had five sample sites here.

- A: is good condition steppic grassland with *Stipa tirsa*, *S. joannis* and *Iris pumila*. *Cirsium pannonicum* has a high density here. It is rich in dicots (*Jurinea mollis*, *Inula ensifolia*, *Polygala major*, *Scorzonera purpurea*, many tall forb Asteraceae and Apiaceae etc).
- B: with degraded vegetation, dominated by grasses. There is little evidence of the food plant here.
- C: formerly used as a forestry hayfield. Nowadays, this site is mown irregularly by the staff of the national park. It is characterized by many different grass and herbaceous species, including some tall forbs and polycormon-forming species (*Echium maculatum*, *Peucedanum cervaria*, *Centaurea* spp., and *Inula hirta*, *I. ensifolia*, *Thymus* spp., *Dorycnium germanicum*)
- L: is cut every year. Some Fabaceae are abundant here, e.g. *Onobrychis arenaria*, *Hippocrepis comosa* and *Vicia tenuifolia*.
- N: is a slightly bushy patch, poor in dicotyledonous plants.

On the Zabanyik hill we can see the remains of fruit trees, indicating its former use as an orchard, similar to the Szőlőhegy. Compared with the Szőlőhegy, this hill is drier and warmer. Probably the species-rich patches are edaphically treeless. However, close to the top we can see some old Italian pubescent oaks (*Quercus virgiliana*) with a species-rich fringe vegetation (*Anemone sylvestris, Cytisus procumbens, Dracocephalum austriacum, Euphorbia polychroma*). We had three sample sites here.

- G: This site is very near the oak forest on the south-east slope of the hill. It is a species-rich semi-natural grassland.
- H: Facing Szőlősardó on the south-west slope of the hill, this site is drier and a little poorer in species. *Stipa joannis* and *S. tirsa* are characteristic here.
- I: is very bushy and poor in species. The south-east and north-east parts border *Pinus nigra* plantations.

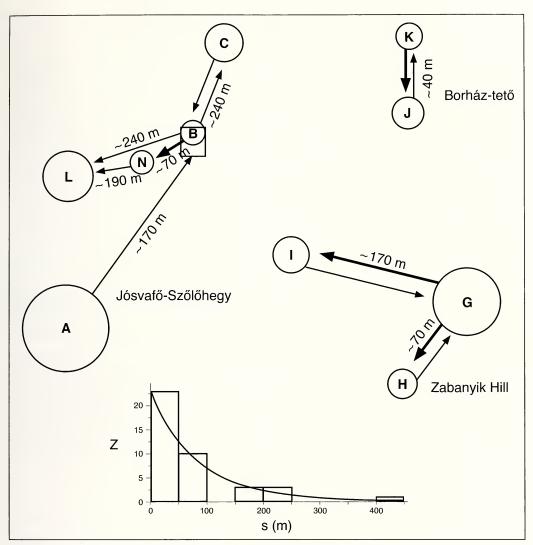
The Borház-tető used to be a fruit production area too. Nowadays we can only see a few relict vines which bear witness to this activity. The north-east slope with *Bromus erectus* has been burned every year. We chose two sample sites here on the western slope very close to each other:

- J: is a slightly bushy and dry patch with clastic soil. Some parts of this are very rich in *Stipa tirsa* and *S. joannis*.
- K: is near to the forest with many bushes; the dominant grass species is *Brachypodium pinnatum*.



Fig. 1. Topography of sample sites, as well as the observed ratios of *M. phoebe* (black) and *M. telona* (white).

*Methods*. Our data were collected through capture-recapture surveys on three or four occasions (Tab. 1) in 2009 May 15-25 from ten sample quadrats (sample sites) with dimensions  $50 \times 40$  m. We worked on five sites simultaneously between 9:00 and 13:00 for 3 hours, only in good weather conditions. Captured butterflies were marked with a water



**Fig. 2.** The observed movement events. The size of the circles is commensurate with the number of butterflies, and the thickness of the lines with the frequency of the movement events. The distances are the measured distances between the middles of the sample quadrats.

resistant XF marker pen. We wrote a code on the underwing, consisting of a letter and a number. This code enabled butterflies to be recognized on recapture and their movement pattern deduced. The following information was registered on the field sheets: the sex, the species, and the activity before capture (or recapture), the latter being important in identifying nectar sources. *M. phoebe* and *M. telona* are very similar, so all specimens were re-identified on recapture to double-check. Misidentifications were not noticed. From the daily distribution of the marked animals we concluded that the sampling was made at the second part of the flight period (Tab. 1). It may have been better if we had started the survey before the peak of the flight period. However, it came earlier than we had anticipated because of the hot and dry weather conditions.

	Α		L		Ν		В		С	
date	t	ph								
V.16	28	4	-	-	-	-	10	8	22	0
V.20	24	5	16	3	7	9	11	5	8	4
V.22	12	1	1	1	3	5	0	4	6	4
V.25	6	1	7	6	1	4	1	1	4	0
sum	70	11	24	10	11	18	22	18	40	8
	J		K		G		Н			I
	t	ph								
V.15	14	13	9	1	27	0	-	-	-	_
V.18	11	22	8	11	15	9	8	2	13	2
V.21	3	5	4	0	15	4	6	0	4	3
V.25	1	1	0	1	2	0	0	0	0	0
sum	29	41	21	13	59	13	14	2	17	5

**Tab. 1.** Marked individuals of *M. telona* (t) and *M. phoebe* (ph) with date. Some of the sites we could not sample on the first day (-).

The coordinates of the sample sites were measured with GPS, and were matched with Google Earth to a satellite image. The centres of the sample quadrats were connected with lines and measured. The value was used as the distance between the quadrats. Movement patterns were drawn from the field sheets data.

After the capture-recapture survey, we estimated the density of *Cirsium pannonicum* and *Dianthus pontederae*. Five  $2 \times 2$  m quadrates were marked randomly at each sample site, and the two plant species were counted in these sample sites. We calculated the density of plants per m<sup>2</sup>. The relation between the observed numbers of butterflies and the density of the nectar source and the larval food plant were analysed using Pearson's correlation.

## Results

307 *M. telona* and 139 *M. phoebe* were marked; 12% of *M. telona* and 14% of *M. Phoebe* were recaptured. The sex ratio was 56% male and 44% female in the case of *M. telona* and 70% male and 30% female in case of *M. phoebe* individuals.

The most *Melitaea* individuals were observed in the sites "A" and "G", and the fewest in sites "N" and "B" (Tabs 1 and 2). *M. telona* and *M. phoebe* were observed at every sample site but in very varying proportions (Fig. 2). The biggest differences were in the Szőlőhegy. The ratio changed from 92% to 36%. The smallest differences were observed in the Borháztető 46% and 44%, respectively, but in this place the two sample sites were very close to each other. On the Zabanyik Hill this ratio varied between 93% and 77%, respectively.

Feeding *M. telona* individuals (51% males, 49% females) were observed on 64 occasions during the capture-recapture survey. This sex ratio is not significantly different from the sex ratio of the marked butterflies. Feeding was recorded 62 times on *Dianthus pontederae* (96%), once on *Ajuga reptans* and once on *Polygala major*.

We found a significant (p < 0.05) but slightly different level of correlation between the density of *Dianthus pontederae* and *Cirsium pannonicum* and the observed number of

**Tab. 2.** The sum of observed *M. telona* (N t) and *M. phoebe* (N ph) individuals on the last three days, as well as the density of *C. pannonicum* (C.p.) and *Dianthus pontederae* (D.p.) in the sample sites.

	N t	N ph	C.p./m <sup>2</sup>	D.p./m <sup>2</sup>
Α	42	7	18.9	6.2
L	24	10	5.4	3.2
N	11	18	2.2	1.75
B	12	10	0	1.05
C	18	8	5.35	1.85
J	15	28	0.15	1.3
K	12	12	4.75	0.05
G	32	13	18.65	4.95
Н	14	2	4.95	2.55
Ι	17	5	0.35	6.75

individuals: r=0.7 for the nectar source, and r=0.9 for the larval food plant. The connection between the two sources is not statistically significant. If we compare the ratio of *M. telona* and *M. phoebe* with the density of the food plant we can see *M. phoebe* never has a significant ratio where *Cirsium pannonicum* grows densely (Tab. 2).

From the 37 movement events discernible for *M. telona* we conclude that the individuals can fly several hundred metres (Fig. 2). The longest registered distance was 420 m. The greater the distance between two sample sites, the less likelihood there is of observing

flights between them. Naturally these distances are just approximate values, calculated from a map. However, as butterflies do not fly in straight lines, in reality their routes will surely be much greater than our estimated values.

#### Discussion

The closely related Melitaea cinxia Linnaeus, 1758 can survive for 12-13 days in cloudy cool weather (Hanski et al. 2006). Similar results were obtained by us in an air-conditioned laboratory environment, using overwintered M. phoebe and M. telona caterpillars collected from the sample sites in spring. These butterflies hatched in the laboratory, and were fed on Spiraea sp. flowers with a honey and water mixture as a supplement, and were able to survive more than 14 days at stable temperatures of 22° C. During the field work, the warm, very dry weather probably reduced the butterflies' life expectancy. Because the intervals between samplings were usually 2 days (maximally 4 days), we think that the low level of recaptures cannot be explained simply by a high level of mortality caused by the dry weather. Rather by the mobility of butterflies – because all the sample sites are in large areas with suitable vegetation for *M. telona*, this species can easily move out from the sample sites. From our results we can see that the butterflies can move between patches several hundred metres apart. This ability could be very important for the long term survival of populations, especially in habitats which are subjected to deliberate partial or complete burning in spring. These fires can be extremely dangerous for *M. telona* – they could possibly kill all the individuals in a patch, because the species feeds, overwinters, and pupates in the litter at ground level. However if there are patches where the species can survive within flying distance, then recolonization is possible and the species will not necessarily become extinct. An important question for the conservation of M. telona is its relationship with M. phoebe. In our survey, M. phoebe occurred at all the sample sites. Moreover, in some quadrats, greater numbers of M. phoebe were recorded than of M. telona. These were the sites with degraded vegetation. M. phoebe has the ability to develop on Cirsium pannonicum but interestingly M. phoebe tends to be found in lower numbers where that plant is abundant. We have information from studies of strong and stable populations over several decades. Almost certainly *M. phoebe* has always occurred in these places or nearby, but cannot displace *M. telona*. We think the main condition for the long-term survival of *M. telona* is the high density of the larval food plant.

An additional exciting question is the possibility of hybridisation. Enzyme electrophoresis has not shown any evidence of hybridisation between these two species (Pecsenye et al. 2007), but this does not mean that they do not mate. It is possible that the hybrids are sterile or have a lower viability (or perhaps they do not hatch from eggs). It might be that there is so-called 'reproductive interference' between these two species (Gröning and Hochkirch 2008) and that this could explain the extinction of *M. telona* from some of its former known localities and the continued occurrence *M phoebe* in these places. Further work is needed to resolve this question.

Our survey shows there is a strong connection between the target species observed numbers and the density of *Cirsium pannonicum* and *Dianthus pontederae*. In spite of the presence of many other flowering plants like *Campanula sibirica*, *Cytisus procumbens*, *Genista tinctoria*, *Helianthemum ovatum*, *Inula ensifolia*, *Lotus corniculatus*, *Onobrychis arenaria*, *Polygala major* and so on, feeding was recorded on mostly (96%) *Dianthus pontederae*.

Many plants such as *Jurinea mollis*, *Centaurea scabiosa* or *Cirsium pannonicum* were flowering later and mostly in very low numbers because the extremely dry spring. We have recorded butterflies feeding on these plants several times in the last few years. So it could be a mistake to conclude from our findings this year that this species is a nectar source specialist – an impression supported by the fact that *M. phoebe* individuals were also only recorded on *D. pontederae*, when we know that this species also lives in habitats where pink flowers do not occur (for example several weedy associations). In order to get more complete information about the nectar sources, we should have to repeat the survey in a year with more average weather.

The situation with the larval food plant is totally different. The target species, *M. telona*, is a true specialist. The species occurs only where *C. pannonicum* also occurs. One very interesting site is "I", where the nectar source has a significant density  $(6.75/m^2)$  but the marked butterflies' numbers were much lower than we expected (Tab. 2). The larval food plant also had a very low density here  $(0.35/m^2)$ . If we delete this sample site from the dataset, we get a much stronger correlation (p<0.01, r=0.892) between nectar source and larval food plant, so this data is very valuable in our few samples because it indicates the secondary importance of the nectar source.

In conclusion, contrary to our expectations, *M. telona* has a greater mobility than we would have expected from its insular distribution and its specialisation (in nature) on one larval food plant. Future surveys will have to take this into consideration. If we want to increase the number of recaptures to get more accurate information about the population size or the dispersion ability, we must achieve a better coverage of the habitat with sample sites. It is better to choose habitats where there is a high density of food plants, not just because we observed the largest number of *M. telona* in these places but because we also observed the lowest number of *M. phoebe*.

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