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**First data on host-ant specificity of parasitic *Maculinea alcon* (DEN. & SCHIFF.)
(Lepidoptera: Lycaenidae) in Poland and eastern Europe**

Abstract: Host-ant specificity of *Maculinea alcon* and habitat requirements were studied on two sites (near Warsaw and in Polesie in eastern Poland) for the first time in Poland and also in this part of Europe. We found *M. alcon* full-grown larvae and pupae exclusively in *M. scabrinodis* nests – the commonest ant on both sites. Besides this species, *M. rubra*, *M. ruginodis*, *M. gallienii* were also observed. Our findings are important for its practical conservation of this threatened species in Poland. Observed and predicted changes in habitat management, especially abandonment, affect the structure of vegetation which is followed by changes in ant communities. We predict that overgrowing causes replacement of *M. scabrinodis* by other more hygrophilous *Myrmica* species and also makes the worse conditions for *G. pneumonanthe* – the host-plant of young *M. alcon* larvae.

Key words: *Maculinea alcon*, host-ant specificity, *Myrmica scabrinodis*, myrmecophily, endangered species, Poland

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INTRODUCTION

Advanced social parasitism observed in Alcon Blue *Maculinea alcon* (DEN. et SCHIFF.) and its close relative *M. rebeli* is one of the most fascinating phenomenon attracting attention of many scientists (THOMAS *et al.* 1989, ELMES *et al.* 1994, GADEBERG & BOOMSMA 1997, ALS *et al.* 2001, 2002). Females lay their eggs on exposed parts of flowers, buds and rarely upper leaves of specific host plants *Gentiana pneumonanthe*, although *G. asclepiadea* and *G. germanica* – are also reported as an alternative/or

additional food resources (MUNGUIRA & MARTIN 1999). After hatching the young larvae bore into flowers and feed on developing seeds. At the start of the fourth and final instar, caterpillars leave the plants and are adopted by ants of the genus *Myrmica* LATR. In the nests they spend next 10 months being fed by workers with insect prey and regurgitations, like ant larvae (THOMAS & ELMES 1998), which they achieve by the mimicry of ants chemicals (ELMES *et al.* 1991, AKINO *et al.* 1999, ALS *et al.* 2001), and also acoustical signals (DEVRIES *et al.* 1993). Other European species of the genus *Maculinea* VAN EECKE are also social parasites of *Myrmica* ants but only *M. rebeli* HIRSCHKE is 'cuckoo' feeder. *M. arion* (L.), *M. teleius* (BGSTR.) and *M. nausithous* (BGSTR.) prey on ant brood and are regarded as less advanced species (THOMAS & ELMES 1998).

Early studies carried out mainly in western Europe demonstrated that each of the European *Maculinea* species specialized in parasiting of different species of *Myrmica* and populations of *M. alcon* were supported by *M. ruginodis* (NYL.) (THOMAS *et al.* 1989). Further studies revealed the existence of geographical differentiation in the host specificity especially in case of *M. alcon* which had been detected in nests of three *Myrmica* species: in the Netherlands – *M. ruginodis*, in Sweden – *M. rubra* (L.) and in Spain, northern France and southern Netherlands – *M. scabrinodis* (NYL.) (ELMES *et al.* 1994, WARDLAW *et al.* 1998). In Denmark, on some sites *M. alcon* was raised by *M. rubra* and *M. ruginodis* simultaneously but never by *M. scabrinodis* (ALS *et al.* 2002). It was suggested that populations dependent on different ants can belong to different ecological subspecies or even cryptic species (ELMES *et al.* 1994).

Maculinea alcon is a Palaearctic species ranging from Southern Spain to Central Asia but showing a scattered pattern of distribution. Sometimes it is difficult to determine what is the exact range of this species, because in former times *M. alcon* was regarded as a joint species with *M. rebeli* (WYNHOFF 1998). However, because of morphological, ecological and physiological grounds they are separated into two species (THOMAS *et al.* 1989). *M. alcon* is a typical species of wet grasslands and inhabits hay meadows, wet heaths and fens where host-plants are present (MUNGUIRA & MARTIN 1999).

M. alcon is believed to be fast declining species and like other *Maculinea* species thrives in seminatural, man-dependent habitats (VAN SWAAY & WARREN 1999). Abandonment of rural areas with the end of grazing and mowing of hay meadows causes ecological succession. Sites start to be unsuitable first for host-ants and only then for host-plants (ELMES *et al.* 1998). The famous story of *M. arion* is an example and warning for any conservational efforts. Any activities should be preceded by detailed studies of subtle habitat requirements (ELMES & THOMAS 1992, THOMAS 1995, NEW *et al.* 1995).

In the eastern part of European range knowledge on *M. alcon* is restricted to distribution and to the habitat description at the most. In Poland *M. alcon* was recorded from sparse localities mainly in south and eastern part of the country. Isolated populations occur in Biebrza Valley and Wielkopolska. *M. alcon* is believed to be endangered by extinction in Poland (BUSZKO 1997).

The aim of present studies was to identify host-ant of *M.alcon* in Poland and for the first time in the whole region. So far the only studies on *Maculinea* host-specificity in Poland concerned *M. teleius*, *M. nausithous* (STANKIEWICZ & SIELEZNIOW 2002) and *M. rebeli* (STEINER *et al.* in press). If we assume that *Maculinea* evolved in a steppe-like habitat in Asia (FIEDLER 1998) Polish sites could have been colonised earlier after the last ice age than western European ones which makes the knowledge on them even more important.

STUDY AREA, MATERIALS AND METHODS

Studies were conducted on two *M.alcon* sites in Poland, Augustówka and Sęków (Fig. 1).



Fig. 1. *M.alcon* sites in Poland used in the study. Pie charts reflect the proportion of *Myrmica* nests found within 2 m of the initial foodplant *G. pneumonanthe* and the area of pies is proportional to the sample size.

Augustówka (51°59' N 21°29' E) is situated on the territory of the Mazovian Lowland, 50 km east-south of Warsaw. Area with *G. pneumonantae* (about 200 clusters and single shoots) covers about 1ha and consists of two wet meadows separated by stripe of trees adjacent to mixed forest on one side and dirt road on the other.

Vegetation can be classified as a mosaic of unfertilised pasture *Nardo-Callunetea* (probably intensively grazed in the past) with *Nardus stricta*, *Carex pallescens*, *Potentilla erecta* and mainly degenerated *Molinion* meadow dominated by *Deschampsia caespitosa* with *Selinum carvifolia*, *Succisa pratensis*, *Salix rosmarinifolia* and *Thalictrum lucidum* (PAWLIKOWSKI 2002).

Sęków (52°04' N 19°03' E) is situated on the Bubnów calcareous fens in the Polesie National Park in eastern Poland. There are many local populations of *M. alcon* which constitute probably a metapopulation system. All area is very large (1500 ha) but all patches of *Molinion medioeuropaeum* with *G. pneumonanthe* are rather small and scattered among *Phragmition*, *Magnocaricion elatae*, *Salicetum pentandro-cinereae* and *Carici elongate-Alnetum* types of vegetation. Studies were performed on a meadow covering about 0,4 ha of land.

Both sites were investigated just before the flight period at the end of June and in the beginning of July 2002. After the identification of patches of host plants, the areas within 2 m of about 50 Gentians plants at Augustówka and 20 ones at Sęków were searched for *Myrmica* ants. Some nests were easy to find because of well-developed solaria while others were less visible and hidden in tufts of grass or moss. All were very carefully open and checked for *M. alcon* presence. There was no necessity of excavation and destruction of nests as it is known that full-grown larvae are carried by workers to upper chambers during the day and pupation takes places there as well (ALS *et al.* 2002). After counting they were immediately returned to nests and quick evacuation of larvae by ant workers was observed. Then nests were covered and arrangement of surrounding vegetation was restored as exactly as it was possible to minimize impact of investigation. Samples of a few workers from each nest were collected to identification in the laboratory according to RADCHENKO *et al.* 1997. and WARDLAW *et al.* 1998. On both sites average density of *Myrmica* nests was estimated using chosen at random 40 squares (1m² each) at Augustówka and 20 squares at Sęków was also performed.

The significance of differences in frequency *M. alcon* in *Myrmica* nests was tested by the Chi-Square Test.

RESULTS

Four *Myrmica* ant species were recorded on both investigated sites within foraging range of *G. pneumonanthe* plants (Fig. 1, Table 1). A total number of 77 *Myrmica* colonies was sampled. *M. scabrinodis*, *M. rubra* and *M. ruginodis* were present at Augustówka while *M. scabrinodis* and *M. gallienii* (BONDR.) at Sęków. *M. scabrinodis* was the commonest *Myrmica* species on both sites (85.2% and 82.6% respectively). Other non-*Myrmica* ants were represented only by *Lasius flavus* (FABR.) which was very rare in comparison to *Myrmica*.

M. alcon larvae and pupae were found exclusively in *M. scabrinodis* nests (Fig. 2). Every fourth nest contained *M. alcon* and a total number of 32 individuals was counted. Combined results Mean number of full-grown larvae and pupae in parasited nests was 2.5 (min 1, max 8) at Augustówka and 2.4 (1,3) at Sęków.

Table 1. The number of *Myrmica* nests within 2 m of *G. pneumonanthe* found on two *M. alcon* sites in Poland and the number of nests parasitized by *M. alcon*.

Site	Density of <i>Myrmica</i> nests per m ²	<i>Myrmica</i> ant species	Number of nests	% of all <i>Myrmica</i> nests	Number and % of nests with <i>M. alcon</i>	Number of <i>M. alcon</i> pupae and/or full-grown larvae
Augustówka	1.7	<i>M. scabrinodis</i>	46	85.2	12 (26)	30
		<i>M. rubra</i>	5	9.3	0	0
		<i>M. ruginodis</i>	3	5.6	0	0
Sęków	0.8	<i>M. scabrinodis</i>	19	82.6	5 (26)	12
		<i>M. gallienii</i>	4	17.4	0	0

The hypothesis that proportion of *M. alcon* larvae/pupae in *M. scabrinodis* nests is the same as proportion of *M. alcon* in non-scabrinodis ones was rejected ($\chi^2=2.95$, $p<0.1$) with Yates' correction.



Fig. 2. Larvae, prepupae and pupa of *M. alcon* in *M. scabrinodis* nest found on the site at Augustówka in Poland.

DISCUSSION

ELMES *et al.* (1994) proposed two hypothesis of geographical variation in host specificity of *M. alcon*: either different populations originated from different post-glacial refugia or they later shifted to new host-ants associated with gentian populations. ALS *et al.* (2002) suggested that *M. alcon* is adapted to the most common *Myrmica* ant species in habitats where *G. pneumonanthe* occurs and our results confirm that. We can not exclude that a fraction of *M. alcon* larvae is reared by other *Myrmica* ants but assuming existence of local *M. alcon* races dependent on specific *Myrmica* (ELMES *et al.* 1994) we suspect that *M. scabrinodis* is the only host-ant at least in eastern Poland. Interestingly the same ant is recorded as the host-ant first of all in southern Europe (ELMES *et al.* 1994).

We were recorded relatively small numbers of *M. alcon* individuals in ant nests in comparison to other authors (THOMAS & ELMES 1998, ALS *et al.* 2002) although 'cuckoo' feeding is more effective than predation (THOMAS & ELMES 1998). As the nests weren't excavated to census *Myrmica* populations (because of conservational reasons), it is almost impossible to interpret. Generally *M. scabrinodis* nests on *Maculinea* sites in Poland are smaller than *M. ruginodis*, *M. rubra* and *M. gallienii* ones but detailed studies haven't been carried out so far.

Host specificity of *M. alcon* and other *Maculinea* butterflies results mainly from differences in survival in various *Myrmica* nests (ELMES & THOMAS 1992), but the attractiveness of freshly emerged caterpillars could also differ (ALS *et al.* 2001). *M. scabrinodis* have been already recorded as a host-ant of all *Maculinea* species. *M. scabrinodis* is the main host ant of *M. teleius* in many localities in Europe (THOMAS *et al.* 1989) and of *M. nausithous* on one site in Spain (VAN SWAAY & WARREN 1999). *M. scabrinodis* is also recorded as the minor host of *M. rebeli* in France, Spain and Poland and the minor host of *M. arion* in England (THOMAS *et al.* 1989, STEINER *et al.*, in press).

Agricultural improvements, abandonment causing successional changes, isolation and fragmentation of habitats, land drainage and afforestation are main threats indicated by national compilers in Europe (VAN SWAAY & WARREN 1999). In Poland overgrowing is probably the crucial negative factor in case of *M. alcon*. *M. scabrinodis* is polytopic mesothermophilous species of humid habitats which requires great insolation but is very tolerant of soil moisture. This ant occurs in open areas (meadows, pastures, pet bogs) and also in sunny patches in forests (CZECHOWSKI *et al.* 2002). We suspect that ecological succession causes replacement of *M. scabrinodis* by other more hygrophilous and preferring taller type of vegetation *Myrmica* species. Unfortunately no studies monitoring changes in ant communities have been undertaken so far, but observations from a site of *M. teleius* (another wetland species) in Warsaw show such influence. Density of *M. scabrinodis* is lower and share of *M. rubra* and *M. ruginodis* is higher on meadows which have been abandoned for years in comparison to extensively grazed or mowed ones (SIELEZNIOW & STANKIEWICZ unpublished). Reduction of insolation by increasing shade affects also initial foodplant *G. pneumonanthe* which niche is probably narrower than niche of *M. scabrinodis*.

Both investigated *M. alcon* sites and probably many others in Poland are without management at the moment and we predict unfavourable processes in the future. Rate

of changes will depend on soil conditions but conservation measures should be taken immediately. Further studies are urgently need for evaluation of Action Plan for *M. alcon* in Poland. Detailed ecological research should be undertaken as well as the exploration of known *G. pneumonanthe* sites (ZAJĄC & ZAJĄC 2001) to increase basic knowledge on *M. alcon* distribution. Otherwise many sites not described so far can imperceptibly disappear because of environmental changes. *M. alcon*, *G. pneumonanthe* (both protected by law in Poland) could be used as indicators of high biodiversity and 'umbrella species' in conservation of grassland ecosystems.

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STRESZCZENIE

[Tytuł: Specyficzność pasożytniczego motyla *Maculinea alcon* (DEN. & SCHIFF.) (Lepidoptera: Lycaenidae) względem mrówek-gospodarzy: pierwsze dane dla Polski i Europy Wschodniej.]

Modraszek alkon (*Maculinea alcon*) jest wyspecjalizowanym gatunkiem motyla zagrożonym wyginięciem w Polsce i Europie. W cyklu życiowym występują dwa kluczowe elementy: larwy zaczynają swój rozwój na specyficznej roślinie żywicielskiej – goryczce wąskolistnej (*Gentiana pneumonanthe*), a kontynuują w gniazdach mrówek z rodzaju *Myrmica*, gdzie są karmione przez robotnice. Specyficzność modraszka alkona względem gospodarzy oraz uwarunkowania środowiskowe były badane na dwóch stanowiskach w Polsce: w okolicach Warszawy oraz na Polesiu. Były to pierwsze tego typu obserwacje nie tylko w Polsce, ale i w całej wschodniej Europie. Na stanowiskach stwierdzono występowanie czterech gatunków mrówek z rodzaju *Myrmica*: *M. scabrinodis* (NYL.), *M. rubra* (L.), *M. ruginodis* (NYL.) i *M. gallienii* (BONDR.). Wyrośnięte gąsienice i poczwarki alkona były znajdowane wyłącznie w gniazdach najpopularniejszej mrówki z tego rodzaju – *M. scabrinodis*, która była wykazana jako gospodarz motyla we Francji, Hiszpanii i południowej Holandii. Dalsze badania są konieczne dla określenia, czy w Polsce mamy do czynienia z geograficzną zmiennością interakcji motyl–mrówka, czy też *M. scabrinodis* jest jedynym gospodarzem *M. alcon*. Poznanie tego elementu biologii jest niezwykle istotne w kontekście ochrony tego gatunku, jako że obserwowane i przewidywane zmiany środowiskowe wpływają na strukturę roślinności, a w konsekwencji zmiany składu gatunkowego mrówek.