ABSTRACT. Based on the methodology developed by the author the developmental rhythm was observed in marsh helleborine *Epipactis palustris* (L.) Crantz growing under different habitat conditions. Twelve phases were taken into consideration, covering the most important and most readily observable stages in vegetative and generative development throughout the year. Results of observations are presented graphically in the form of figures and respective photographs were also taken. Data collected in situ were compared with the results found in the experimental garden. They supplied comparative material concerning synchronism of the course of phenological phases in marsh helleborine under in situ and ex situ conditions. In natural localities marsh helleborine exhibited differences in the course of phenophases depending on the type of habitat and as a consequence – depending on a specific plant community. These differences were retained after plants were transferred from these populations to the garden. Moreover, in marsh helleborine growing on plots in the garden an extension of the entire vegetation period and a shortening of the spring phase of the vegetative development were observed.

KEY WORDS: Orchidaceae, *Epipactis palustris*, marsh helleborine, phenology, botany, Poznań

INTRODUCTION

Phenological observations serve not only pure science, i.e. the determination of the specific character of development in individual plant species. They may also be a tool in the investigation of other phenomena, even as distant as climate change or analyses of the course of pollen seasons in allergenic species (Stosik 2010).

The rhythm of development in plants is determined mainly by biological characteristics of plants, although other factors are also of importance, such as temperature, soil moisture content and humidity affecting water balance in plants, insolation, mechanical damage, competition or even age in case of perennial plants (Łukasiewicz 1967).

According to Falińska (1974, 1975), observation of a different phenological behaviour within one species is a justification for a continuation of studies on this phenomenon in other disciplines, such as e.g. ecological genetics, experimental taxonomy or experimental population ecology. In the opinion of that researcher differences in the phenology of a cenopopulation may play a considerable role as an indicator of ecological variation within one geographical area.

The aim of this study was to compare the course of developmental phases in marsh helleborine growing in different habitats and to verify whether a change in habitat conditions is reflected in the development rhythm of this species.

MATERIAL AND METHODS

Marsh helleborine is a perennial, clonal species, which is characterised by a two-layer organisation, with the first layer formed by specimens (N), while the second layer comprises structural units of a specimen, i.e. rooted shoots, called modules or ramets (n) (Falińska 2002). Since in this species it is difficult to evaluate the volume and range of a single specimen, ramets, referred in this study also as shoots or plants, were subjected to observations and phenological analysis, as it was recommended by Falińska (2002).

All the observed populations of marsh helleborine were located in the city of Poznań. Analyses included four populations from two in situ localities, as well as two populations from the experimental garden.

The population denoted as RP was a population located at a natural locality in a north-eastern part of Poznań in the Dolina Różanego Potoku (ATPOL square BC-99). Marsh helleborine is found here on a humid moorgrass meadow represented by the association of *Selino carvifoliae-Molinietum caeruleae* Kuhn 1937 (syn. *Molinietum caeruleae* W. Koch 1926 nom. illeg.), from the *Molinion* association. In 2000 the population comprised 525 shoots (ramets), including 179 generative shoots (Wyrzykiewicz-Raszewska 2006).

Populations denoted as A1, A2 and A3 were populations localized on an escarpment of an excavation created in 1970 as a consequence of the construction of the Kiekrz-Zieliniec railway tracks. While digging this deep
PHOT. 1. Phase 3 – formation of inflorescences in *Epipactis palustris*

PHOT. 2. A freshly opened flower of *Epipactis palustris* with two pollinia visible in the stamen head

PHOT. 3. An inflorescence of *Epipactis palustris* with 20 flowers at different flowering phases

PHOT. 4. A cluster of flowering shoots of *Epipactis palustris*
pit soil layers were exposed, which were later spontaneously colonised by plants. As a result of this process communities were formed on the escarpment, resembling in their composition seminatural meadow communities. In the discussed habitat marsh helleborine is found in differently formed phytocenos which of xerothermic association of flower, calciphilous, meadow of Galietum borealis Nowiński 1928 (syn. Galio borealis-Molinion (W. Koch 1926) Philippi 1960) from the Molinion association. In populations A1 and A2 the association takes an almost typical form, while in population A3 the association reached the terminal form, evolving towards Arrhenatheretum elatioris (WYRZYKIEWICZ-RASZEWSKA 2006). It needs to be mentioned here that they are communities which are rarely found in the Wielkopolska region and which are under immediate threat of extinction (BRZEG and WOJTERSKA 2001). In 2002 population A1 comprised 1973 shoots, including 266 generative ones, population A2 consisted of 1615 and 298 shoots, respectively, while population A3 was composed of 1721 and 668 shoots (WYRZYKIEWICZ-RASZEWSKA 2006).

The discussed locality was also found in the north-eastern part of Poznań at the administrative boundary of the city with the village of Koziegłowy, belonging to the Czerwonak commune (ATPOL square BC-99).

Populations from the experimental garden (EG) originate from the population from Rożany Potok and population A1. They were denoted as RP-EG and A1-EG.

Prior permissions of the Ministry of Environment and of the Provincial Nature Conservation Officer in Poznań were obtained before sample collection to the garden.

Plants were transferred in the autumn 1999 for sample A1-EG and in the autumn 2000 for sample RP-EG. They were placed in separate plots in insolated sites. Cultivation measures comprised abundant watering of plots during the vegetation period and manual removal of appearing weeds.

In 2004 population A1-EG comprised 139 shoots, including 110 generative ones, while population RP-EG as many as 182 shoots, of which 167 were generative (WYRZYKIEWICZ-RASZEWSKA 2009).

The experimental garden belongs to the Department of Botany, Poznań University of Life Sciences and it is located in the district of Jeżyce, in the area of Solacz (ATPOL square BC-98).

Preliminary observations and developed methods, making it possible to follow the development of marsh helleborine, lasted for three years preceding the proper year of observations. Although ŁUKASIEWICZ (1984) suggested uniformization of phenological observation methods to ensure comparability of results, the species specific characteristics of plants are not always possible to determine using the previously developed methods.

Observations of the course of phenological phases were conducted in 2004, at regular 3- to 4-day intervals throughout the entire vegetation period of marsh helleborine, while during flowering and seed dispersal it was even on a daily basis, following the recommendations by MITKA and TUMIDAJOWICZ (1992).

The following developmental phases were observed:

A. Vegetative development
1. emergence of aboveground shoots in the spring – onset,
2. unfolding of leaves – onset,
3. flowering – onset (the first flowers open) (Phot. 2),
4. unripe fruits – onset (from overblowing of the first flowers),
5. the last flower bud opens,
6. completion of flowering (the last flower is overblown),
7. opening of capsules and dispersal of seeds – onset,
8. opening of capsules and seed dispersal – completion.

B. Generative development
3. inflorescence formation – onset (Phot. 1),
4. flowering – onset (Phot. 3),
5. unripe fruits (Phot. 4),
6. the last flower bud opens,
7. completion of flowering (the last flower is overblown),
8. opening of capsules and dispersal of seeds – onset,
9. opening of capsules and seed dispersal – completion.

In marsh helleborine, in the flowering period, on one plant (ramete) the course of several phases is observed simultaneously (flower buds, open flowers, overblown flowers, set unripe fruits), smoothly evolved from one to another (Phot. 3). This considerably hindered the determination which phase predominates at a given moment, particularly when many plants in a very big cluster are being observed at the same time (Phot. 4).

In the determination of the ripe fruit phase (opening of capsules) an additional difficulty is connected with the absence of marked symptoms of their ripening.

In view of the above problems it was impossible to present the results of observations quantitatively, i.e. to determine the percentages of shoots (rametes) at a given phase. Results are presented in the form of figures, on which only the sequence of symptoms and the duration of the most important and most readily observable developmental phases were marked (Fig. 3).

The paper (Tables 1 and 2) contains meteorological data from the years 2003 and 2004 in relation to the multi-annual means, as well as the course of phenological seasons in the city of Poznań (Fig. 1).

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Typical temperature: 4.4°C, 8.9°C, 15.0°C, 16.9°C, 18.9°C, 15.0°C, 9.4°C, 3.5°C, −1.7°C

Fig. 1. The course of phenological phases in Poznań among to data from 1958-1997 (GORSKA-ZAJACZKOWSKA and WOJTOWICZ 1998)
The spring resumption of vegetation, consisting in the appearance of the apical parts of aboveground shoots (phase 1, Fig. 2) in marsh helleborine took place in the second half of April, the earliest on \( \text{phase} \ 1 \), in pop-ulation A, while the latest on \( \text{phase} \ 1 \).\( \text{phase} \ 2 \) in population RP (Fig. 1). The appearance of leaves (phase 2) was defined as the time, in which leaf blades unfold. In most locali-ties the onset of this phase occurred 21 days after the emergence of shoots, except for locality RP-EG, where it took place already after 10 days. Developing leaves gradually grow bigger and form a rosette just above the ground (Fig. 2).

The next, third phase (Fig. 2), initiating generative development, is connected with the appearance of inflorescence primordia inside well-developed leaf rosettes (Phot. 1). It may be considered as the onset of formation of immature flower buds. Generally it took place in the first half of June, except for population RP – the end of June, after 43 (A1-EG) – 57 (RP) days from the onset of vegetation (Fig. 3).

From 12 (A1) to 27 days (RP-EG) passed from the appearance of inflorescences to the flowering of the first flowers (phase 4, Fig. 3). In that time successive

![Phase 1](Image1) ![Phase 2](Image2) ![Phase 3](Image3) ![Phase 4](Image4)

**Fig. 2. Developmental phases in *E. palustris* from spring resumption of vegetation to flowering**
Fig. 3. The sequence of appearance and duration of phenological phases in *E. palustris* in populations A1, A2, A3, RP and in the experimental garden A1-EG, RP-EG, in 2004.
The course of flowering in 49 plants of *E. palustris* A1-EG, in 2003

![The course of flowering in 49 plants of *E. palustris* A1-EG, in 2003](image-url)
changes were observed in the plant habit, consisting in the strong elongation of stems, as a result of which leaves were becoming increasingly distant from one another, losing the form of a rosette, while the inflorescence axis gradually straightened, with flower buds increasingly becoming directed towards one side, forming a one-sided raceme (Fig. 2).

The flowering phase was defined as a period from the opening of the first flowers (4) to the overblowing of the last (7). For individual plants (ramets) flowering time depended on the number of flowers in the inflorescence and amounted to e.g. 6-13 days at nine flowers, 14-17 days at 20-24 flowers in the raceme (Fig. 4). In turn, for the entire populations, this time ranged from 20 (RP) to 27 (A2) days, while for A1-EG it was even 42 days. Flowering in marsh helleborine started the earliest, i.e. the 20th June, in the experimental garden, in plot A1-EG, while it was the latest in the locality in Różany Potok, i.e. on the 18th July (Fig. 3).

The description of flowering for a single flower: the flower bud, which is about to open, changes its position from overhanging to perpendicular to the inflorescence axis. The opening flower retains this position, facilitating the landing of pollinators. This state lasts for five days, after which the flower peduncle overhangs again. Although the flower is still in good condition and open, it is already most frequently devoid of the pollinium and it is pollinated, which results in a change of its position. This was considered the onset of fruit setting (phase 5). In the course of the next 5-7 days the flower begins to show symptoms of ageing, consisting in the gradual closing of the perianth and a change in the appearance until it dies back completely. The perianth does not fall, but it remains on the forming fruit (Phot. 3, 4).

During flowering in the course of one day in a single inflorescence a total of 0-7 flowers opened, although most frequently there were one or two flowers (Fig. 4).

The phase of unripe fruits lasted from 38 (A2) to 41 (A1, A1-EG) days, only in the Różany Potok population it was much longer, i.e. as many as 52 days. The symptom of ripe fruits (8) was their opening. Opening of capsules and dispersal of seeds occurred in the second half of August and in the beginning of September, lasting from 13 (RP-EG) to 21 days (A1-EG) (Fig. 3).

Marsh helleborine entered the period of autumn leaf discoloring to yellow-green colour (phase 10) in the second half of September, while dying back of plants (phase 12), manifested by their browning, ended in the second decade of October, after 173 (A2, RP) to 188 (A1-EG) days of vegetation (Fig. 3).

An analysis of the course of phenophases in marsh helleborine in the three populations, i.e. A1, A2 and A3, located in the anthropogenic habitat showed that differences in their developmental cycles were not very big. This is most probably a consequence of more or less identical habitat conditions found in the investigated escarpment (WYRZYKIEWICZ-RASZEWSKA et al. 2003, 2004), as well as the affiliation of marsh helleborine to plant communities belonging to the same association – Galietum borealis.

In turn, marsh helleborine from population RP initiated vegetation much later and most phases were also realised with a delay in relation to the other analysed populations. In this locality marsh helleborine is found in the Selino carvifoliae-Molinetum caeruleae association. The phenomenon of the effect of community type, in which a given species is found, was stressed by FALINSKA (1975). Moreover, in the opinion of that author deviations in the seasonal rhythm reveal several different ecological processes, such as the phenomena of succession, community degeneration or changes in abiotic systems of ecosystems.

The response of marsh helleborine transferred to the experimental garden consisted mainly in the considerable extension of the entire vegetation period – by 12 days for A1-EG plants and by 11 days for RP-EG plants. However, a considerable shortening of the spring vegetative development phase was found. Spring development of marsh helleborine from RP-EG was shorter by 18 days than that of helleborine from population RP, while in marsh helleborine from A1-EG it was by 13 days in relation to that of marsh helleborine from population A1.

Marsh helleborine coming from Różany Potok under the garden conditions retained the trait of delayed onset and completion of the vegetation period, although the phases of generative development were considerably accelerated (Fig. 3).

We also need to stress here the very long period of flowering in A1-EG plants, amounting to as many as 42 days, while in locality A1 it was only 23 days, especially that it was for a much bigger number of plants. No similar variation was observed in the length of flowering between plants from RP and RP-EG. However, we need to cite here data from the previous year, i.e. 2003, when pilot observations of phenological phases were conducted for A1-EG plants (Fig. 4). Flowering lasted then for exactly the same number of days as in 2004, despite different weather conditions observed in both years (Tables 1, 2). MIKOŁAJCZYK (1962) was of an opinion that flowering date is determined by genetic traits, while environmental factors play a considerable modifying role, whereas BYSZEWSKI (1969) indicated that first of all environmental conditions are causes of the differences.

This study confirms the involvement of both genetic factors and environmental conditions in the course of phenophases in E. palustris. Further studies need to be conducted in order to verify which of the factors has a greater effect on the developmental cycle in this species.

**REFERENCES**


