THE LONG-TERM DYNAMICS IN A POPULATION OF *Epipactis helleborine* (L.) Crantz (Orchidaceae, Neottieae) in South-Western Poland

ANNA JAKUBSKA¹, MAŁGORZATA DUDKIEWICZ²

 ¹ Department of Biodiversity and Plant Cover Protection, Institute of Plant Biology, University of Wrocław, Kanonia 6/8, 50-328 Wrocław, e-mail: Ajak@biol.uni.wroc.pl
² Department of Biometrics, Faculty of Agriculture and Biology, Warsaw Agricultural University, Nowoursynowska 159, 02-776 Warszawa, e-mail: malgorzata. dudkiewicz@omega.sggw.waw.pl

ABSTRACT: The research on occurrence fluctuation of Broad-leaved Helleborine *Epipactis* helleborine (L.) Crantz was carried out in the years 1997-2006, an area 12×18 m situated in an 'orchidaceous' beech forest on a slope of Shupiec Mt. 531.1 m a.s.l. (the Krowiarki Mts., Eastern Sudety Mountains). Population flux within the area was characterized by positions of individuals and phases of growth. We have implemented some mathematical and statistical methods (on the basis of Neyman's confidence intervals theory) to find out what was the pattern of population flux for this rhizome species and to estimate the rhizome's course and possible directions of population expansion.

KEY WORDS: Epipactis helleborine, 'occurrence fluctuation', orchids, rhizome

Introduction

Long term observation of plant populations are essential for the understanding of the dynamics and reasons of the occurrence fluctuations typical for numerous endangered plant species. Many of rhizomic species have unusual and interesting life-cycles with noticeable population flux, but the information on the structure and dynamics of the rhizome growth is still scarce and insufficient. The phenomenon of above-ground absence has been reported to be widespread among many species of orchids, especially: *Dactylorhiza incarnata* (Tamm 1972), *Orchis simia* (Willems 1982), as well as *Epipactis helleborine* (Light and MacConaill 1991). There are several hypotheses to explain why certain plants become 'absent' yet there have been few extensive below ground. Young (1949) suggests that the perennating bud of Broad leaved-Helleborine does not necessarily develop during the year immediately following appearance but may remain dormant for one or several seasons (Light and MacConaill 1991). According to Hutchings (1987) the mycorrhizae play an important part in the nutrition of the mature plant – this may explain why a plant is capable of passing a year or more underground

(Light and MacConaill 1991). A different and a very popular hypothesis suggests that the 'occurrence fluctuation' is the consequence of unfavourable climatic conditions, more precisely the weather records at the end of summer and in early autumn in the year preceding the growing season and in early spring in the given growing season (Wells 1981). In unfavourable temperature and moisture conditions in a given year, different orchids may subsist underground without developing aboveground organs (Ziegenspeck 1936). The knowledge of the causes of orchid populations' fluctuations and its range is of fundamental importance especially in assessing the degree to which a population is endangered, as it allows to estimate whether a substantial diminution in the number of population is the result of the 'occurrence fluctuation' or the beginning of the regression. The main impediment to conducting this kind of research is its long duration, as it is a long term investigation. The object of the research was an attempt to verify the changes in the number of ramets in successive growing seasons and whether the Broadleaved Helleborine indeed demonstrates cyclicity of mass occurrences and to establish which of the shoots are the most vulnerable to elimination.

Materials and methods

The 12×18 m area, situated in an 'orchidaceous' beech forest on a slope of Słupiec Mt. 531.1 m a.s.l. (the Krowiarki Mts., Eastern Sudety Mountains, Fig. 1), was selected for study in July 1997. The position of individuals was mapped. Between 1997 and 2006 population flux within the grid was characterized by a number of specimens as well as phases of growth. The observations have been conducted in field conditions during the peak of flowering period. The data from the year 2006 has not been considered in the study results as the area of study has been affected by a drought from May to August 2006, which directly caused both juvenile plants as well as ramets to dry out in the early stage of the growing season.

The results were obtained by employing mathematical methods. We used statistical methods (Neyman's confidence intervals theory) together with spatial representation of population growth rate estimated by two quantities: proportion of juvenile specimens in each experimental square and average increase of the number of specimens in every square per year.

We constructed first the 95% confidence interval for proportion of juvenile specimens in the whole population on the strength of the sample data according to the following formula:

$$P\{(\hat{p} - z_{\alpha}\sqrt{\frac{\hat{p}(1-\hat{p})}{n}}) \le p \le (\hat{p} + z_{\alpha}\sqrt{\frac{\hat{p}(1-\hat{p})}{n}})\} = 1-\alpha \qquad (1)$$

where:

p – proportion of the juvenile from for whole population of *E. helleborine* in Krowiarki Range treated as structure coefficient, that is the fraction of elements distinguished selected from the whole population

 \hat{p} – is an estimation of the proportion of juvenile form in the population calculated from sample data as m/n, where m is the number of juvenile specimens and n is the number of plants in whole population (the population in the test area for 12 squares together). For

big probes this estimator has asymptotically a normal distribution.

 α – significance level of statistics (0.05)

 z_{α} – 1.96 – test t value for 0.05 significance level and infinite number of elements in the probe - identical with normal distribution.

The second feature we analyzed was the average population growth calculated as a sum of differences between the number of plants in given year and in the previous season per year in the j experimental square as follows:

$$\overline{\Delta}_{j} = \frac{\sum_{i=1}^{k} (n_{i} - n_{i-1})}{k}$$
(2)

where:

 n_i – number of specimens observed in year i

 \dot{n}_{i-1} – number of specimens observed in previous year

k – length of the observation term (8 years)



Fig. 1. Study site: 'orchidaceous' beech forest on a slope of Słupiec Mt. 531.1 m a.s.l. (the Krowiarki Mts., Eastern Sudety Mountains) SW Poland.

Results

Total number of specimens observed during 8 years of experiment amounted to 1052 and total number of juvenile forms was 148. Figure 2 was plotted to sum up the results obtained according to formula (2) and consist of 72 graphs representing the annual population flux for each square separately. The squares where the occurrence of the species was confirmed every observation year were marked with circles. Finally we connected the neighbouring circles with solid lines to roughly estimate the probable rhizome course, which is only the sketch providing the basis for further analysis. More sophisticated and precise methods for approximate the shape and development of *E. helleborine* rhizome were presented by Jakubska *et al.* 2006.

The spatial distributions of the results obtained for every square of the test field are shown in Fig. 3. We applied the circle diameter to compare individual squares. We joined the graphical representation of average population growth rate with the spatial representation of the test area. The blanks correspond with the squares where the occurrence of the *E. helleborine* was not confirmed within the whole observation period and no data were obtained.

Using the formula (1) we calculated the 95% juvenile specimens proportion's confidence interval limits as (0.1197; 0.1618). Comparing the structure coefficients estimated for each square separately with this value, we have plotted Fig. 4. We established 3 grades scale: below lower limit, between confidence limits and above upper limit.

Discussion

The scheme resulted from Fig. 2 suggests that the population consisted of two separate rhizomes at least. The pattern of fluctuation stated for the whole population is mostly valid for the individual squares as well. We observed one distinct decrease in population number just after the year 2001 and two periods of 'population boom' in 2000 and 2003. In the years 1998-2000 there was a perceptible tendency of population growth. This year's observations will be crucial to definitely establish the direction of last three years' trend and to eventually confirm or reject the three years long cycle hypothesis. Analysis of Fig. 3 suggests a stronger growth potential of the rhizomes occupying the north part of the habitat. Particularly in the squares 2/1, 2/2, 2/4 and 2/5 the number of specimens continually increased. This data partially agreed with juvenile form proportion coefficient in this area, which allowed drawing the conclusion about northeast direction of population spread. The next interesting feature noticed during the graph analysis was the intense fluctuation in the bottom and middle part of experiment area, which agrees with the older parts of the rhizome. It could suggest that the population flux concerns the mature individuals as well, which could be connected with the sensibility of the perennating bud stadium. We performed the comparison of structure coefficient for juvenile specimens in each square with the calculated confidence interval for the whole population in the experimental area. The majority of values (except three for the square 3/2, 8/3 and 9/3) were situated beyond the confidence interval, what could confirm the instability and non random trends in population spatial distribution and growth. The problem of socalled 'occurrence fluctuation' is of great importance particularly when describing the condition of the population as it is not certain whether a single observation of a significant reduction in the size of a population is a symptom of its gradual decline or if it is one of the characteristic features of this population. So far the nature of this process has not been explained. It is very likely that what underlie this phenomenon are the biochemical



Fig. 2. Bar charts representing annual population flux in experimental squares (observation period 1997-2006). Each panel of the graph corresponds to one 2×2 m square in the experimental area. On the x axis are marked subsequent years of observation, on the y axis – numbers of observed ramets. Circles mark the squares where the specimens were observed continuously; dotted and solid lines connecting the circles trace the estimated course of the rhizomes.



Fig. 3. Average population growth in the experimental squares calculated as the sum of differences between the number of plants in given year and in the previous season normalised by the length observation period (8 years). Circles diameters in cm equals the values calculated for a given square (as show the scale above). Circles colours indicate positive (white) or negative (black) value. White squares mark the areas, where zero resultant growth was observed.

square 1/1	square 1/2	square 1/3	square 1/4	square 1/5	square 1/6
square 2/1	square 2/2	square 2/3	square 2/4	square 2/5	square 2/6
square 3/1	square 3/2	square 3/3	square 3/4	square 3/5	square 3/6
square 4/1	square 4/2	square 4/3	square 4/4	square 4/5	square 4/6
square 5/1	square 5/2	square 5/3	square 5/4	square 5/5	square 5/6
	62		64	<u>^</u>	
square 0/1	square orz	square 0/3	Square 0/4	square 0/3	square 0/0
square 7/1	square 7/2	square 7/3	square 7/4	square 7/5	square 7/6
square 8/1	square 8/2	square 8/3	square 8/4	square 8/5	square 8/6
square 9/1	square 9/2	square 9/3	square 9/4	square 9/5	square 9/6
< <u>0 1197</u>					
>0,161847					
within confidence interval					

Fig. 4. Graph representing the juvenile forms distribution along the experimental area. Dark squares correspond with the fields where the proportion of juvenile forms was higher then upper limit of confidence interval estimated for the whole population data.

processes that take place in the rhizome. Also we cannot exclude the possibility that temperature differences are of importance here, especially when we define their limits, which are the optima for the species. Undertaking this type of research seems to be wellgrounded as it allows proving whether small changes in the sizes of studied populations are a sign of their decline or a characteristic feature of these taxa.

Phenological observations of 31 species of orchids have been carried out for many years in California by Coleman (1991) and also in Poland. 30 year long studies conducted in Lower Silesia (SW Poland) by Sarosiek (1993) showed that the phenomenon of 'occurrence fluctuation' refers to such species as Cephalanthera longifolia, Dactylorhiza majalis, Dactylorhiza sambucina, Neottia nidus-avis, Gymnadenia conopsea, Platanthera bifolia, Epipactis helleborine. The results of Sarosiek's studies show that the sizes of populations of these species reduced in time, yet a significant reduction of the population size was not always a sign of its decline in a given growing period. Fluctuations in the population size do not always result from drastic changes in the natural environment but are likely to be related to a factor of undefined nature. The results of our study show that the 'occurrence fluctuations' of Epipactis helleborine (L.) Crantz are much smaller than it could be expected. The lack of shoots appearance may be conditioned by perennating buds being eaten by soil Arthropods or by mechanical damage to a fragment of a rhizome, e.g. in consequence of windfallen trees. The shoots occurrence of Broad-leaved Helleborine also heavily depends on the amount of rainfall. In seasons of poor rainfall a substantially decreased frequency of juvenile plants was observed.

Another eliminating factor may be deep damages to soil caused by falling blown down trees with their complex root systems or by animals digging in search for food, e.g. boars.

Breaking the continuity of the rhizome in this way usually leads to elimination of this fragment of genet, which has been confirmed in the course of studies. Identification of frequent elimination of new ramets shows that they are particularly sensitive. It cannot be excluded that the observed elimination of top parts of the rhizome is the beginning of degradation of natural environment. Vanishing of various orchid species is exactly very often the first signal of the degradation of natural communities, and may have serious biocenotic and ecological consequences (Sarosiek 1990). Despite the development of new techniques, it is still difficult to work out an optimal research method, which would be safe for rhizome orchids at the same time (Jakubska et al. 2006). A great problem for discovering the possible causes of the 'occurrence fluctuation' is the fact that orchids are rare plants and are protected. Any interference in the study area may pose a threat to the future of that population.

This paper should be treated as an introduction to further long-term studies aiming to evaluate the range and the causes of population flux in the studied population of *Epipactis helleborine*.

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Streszczenie

Wieloletnie badania dynamiki populacji Epipactis helleborine (L.) Crantz (Orchidaceae, Neottieae) w południowo-zachodniej Polsce

Artykuł przedstawia wyniki badań dynamiki populacji kruszczyka szerokolistnego *Epipactis helleborine* (L.) Crantz, które prowadzono w latach 1997-2006 na powierzchni eksperymentalnej o wymiarach 12×18 m położonej w "buczynie storczykowej" *Cephalanthero-Fagenion* na stoku Słupca (531,1 m n.p.m.) w Masywie Krowiarek (pd.-zach. Polska).

Celem badań było określenie zakresu zmian liczebności oraz poziomu tzw. fluktuacji pojawu *Epipactis helleborine*. Prezentowane opracowanie graficzne i statystyczne zebranych wyników oparto na estymacji przedziałowej według teorii przedziałów ufności Neymana.