ABSTRACT: Studies on sexual dimorphism in reference to vegetative features and on the sex ratio in different environmental conditions were carried out in lowland populations of dioecious species *Aruncus sylvestris*. It was shown that female individuals of *Aruncus sylvestris* produced higher flowering shoots, longer inflorescences, more leaves and longer and broader leaves than the male ones. All listed differences were statistically significant. In populations of *Aruncus sylvestris* growing in optimum ecological conditions (fertile and wet soil, half-light) the sex ratio M/F is close to 1:1. Changes of ecological conditions along the gradient of fertility, habitat moisture (estimated by the types of phytocoenoses) and intensity of solar radiation caused marked drift of sex structure towards the male domination. Light intensity was the crucial factor for the sex ratio in studied populations. Populations growing in half-light showed sex ratio M/F ≈ 1:1 irrespective of habitat. Those growing in shade and in full light showed significant shift to male dominance. The largest deviation from 1:1 sex ratio (M/F ≈ 1.0:0.2) was observed in extremely unfavorable habitat conditions (full sun irradiance, dry soil, strong competition from *Elymus repens* (L.) Gould). Obtained results demonstrated that female individuals of *Aruncus sylvestris* had markedly higher habitat requirements compared with the male ones.

KEY WORDS: *Aruncus sylvestris*, lowland populations, sex dimorphism, sex ratio

1. INTRODUCTION

Dioecious plants, in contrast to animals, comprise small percent of known species. There are only 5% of angiosperm dioecious plants but 75% of families include dioecious species (Westergaard 1958). Mechanisms decisive for sex determination were similar in plants and animals and they both passed the same course of evolution (Cronquist 1988, Irish and Nelson 1989, Dellaporta and Calderon-Urrea 1993, Charlesworth 2002, Gorelick 2005). Sex is usually determined genetically. Female individuals are as a rule homozygotic (sex chromosomes XX + autosomes), male individuals are heterozygotic with the genome XY + autosomes. Less frequently the male gender is determined by univalents X0 as e.g. in *Humulus* or by trivalents XY1Y2 like e.g. in *Rumex acetosa* (Zuk 1970, Negrutiu et al. 2001, Charlesworth 2002, Mosiolek et al. 2005). However, in most dioecious plants the presence of sex chromosomes was not found as yet. It is assumed that dioecism in vascular plants is rela-
tively new evolutionary trait (Charlesworth 2002, Nicolas et al. 2004). The presence of phenotypic (environmentally dependent) sex determination (Freeman et al. 1980, Nana- mi et al. 2004) and a large group of species of incomplete dioecism termed androdioecy (division into females and hermaphrodites) and much less frequent androdioecy (males and hermaphrodites) support this assumption (Delannay et al. 1981, Charlesworth 1993, 2002, Wolf et al. 2001).

Even typically dioecious species are able to sporadic production of hermaphroditic flowers (Dellaporta and Calderon-Urea 1993). It is possible since most dioecious plants produce flowers having vestigial organs of the opposite sex (Eckhart 1999, Mitchell and Diggle 2005). Diverse patterns of plants’ dioecism are closely associated with the occurrence of sex dimorphism and the sex ratio. Plants of incomplete dioecism (gynodioecy and androdioecy) do not show sexual dimorphism within vegetative organs (Kohn 1989, Alonso and Herrera 2001, Ramsey and Vaughton 2001, 2002, Ashman 2005). Distinct sexual dimorphism within vegetative organs was found, however, in most studied dioecious species (Ashman 2005). Differences between male and female individuals are limited to quantitative traits associated with uneven growth rate in individuals of opposite sexes. Female individuals as a rule are larger, have more leaves that are larger than males’ leaves i.e. have larger assimilation surface (Ashman 2005). Such growth differentiation is probably an evolutionary response to great energy expenditure for producing and ripening fruits. Male individuals in turn spent much more energy (at generally smaller reproductive effort) than female plant during the flowering period (pollen production) which, according to some authors (Korpelainen 1992) may lead to growth limitation in male individuals and may thus be one of the reasons for their smaller size. Apart from these differences in growth rates a phenomenon was observed which by some authors is named the ecological sex dimorphism. It consists in sex related habitat requirements (Bierzychudek and Eckhart 1988, Garcia and Antor 1995, Ortiz et al. 2002, Cvteković and Jovanović 2007). Female individuals as a rule occupy habitats optimum for the species and push male individuals to suboptimum habitats. This might be one of the factors affecting the sex ratio in populations of dioecious plants. Theoretically, the ratio of males to females in populations of chromosomally determined sex ratio should be 1:1 (Fisher 1930). Actually, sex ratio in the populations of dioecious plants rarely oscillates around this theoretical value. Remarkably, consequent shift to the domination of male or female individuals are sometimes sought not only in ecological but also in genetic factors (Taylor 1994).

Aruncus sylvestris Kostel. (= A. dioicus (Walter) Fernald, family Rosaceae) is a perennial plant growing up to 2m high. It is a dioecious plant of unknown sex determination. Flowers, both male and female, contain vestigial organs of the opposite sex (Evans and Dickinson 1999). Exceptionally, male specimen can be found having monoclinous or both sexes’ flowers. Aruncus sylvestris is the holarctic species present in temperate zones of Asia, North America and in mountain areas of Europe – from the Pyrenees to the Alps to the Apennines to the Carpathians. In Poland the species is common in the Carpathians and the Sudetes, quite frequent on uplands and rare on lowlands (Zając and Zając 2001). The species is associated in Poland with fertile forests of the alliance Fagetalia sylvatica; it grows in partial shade and requires rich, wet and neutral soils (Zarzycki et al. 2002).

In this paper we present results of studies aimed at answering the following questions:

– what is the sex structure in lowland populations of dioecious plant Aruncus sylvestris,
– if and how the habitat might affect sex structure in these populations,
– is there sexual dimorphism within vegetative organs of Aruncus sylvestris?

2. STUDY SITES

We selected three most viable populations growing in Południowo-podlaska Lowland (central eastern Poland).

Population A occupied the forest NW of “Zabuże” village (52°19’23”N 23°01’13”E). Individuals of Aruncus sylvestris are spread over an area of ca 1500 m². They grow in
uniform conditions (strong shading, fertile, moderately wet soil) in a community of linden-oak-hornbeam forest (Tilio-Carpinetum). Tree layer of a coverage over 90% is mainly composed of Tilia cordata Mill., shrub layer of a coverage of 20% is built of Corylus avellana L. and Carpinus betulus L. Herb layer of a coverage of 80% is dominated, apart from Aruncus sylvestris, by Stellaria holostea L., Pulmonaria obscura Dumort., Galeobdolon luteum Huds. and Aegopodium podagraria L.

Population B – a forest south of “Korczeń” village (52°19′53″N 22°37′56″E). Individuals of Aruncus sylvestris grow on a high coverage on an area of ca 25 m². The population inhabits a fragment of linden-oak-hornbeam forest (Tilio-Carpintum) near the forest road. It grows on fresh soil and is side lightened. The coverage of tree layer composed of Tilia cordata and Quercus robur L. does not exceed 50%. Young individuals of Tilia cordata can be found in shrub layer of low coverage (ca 10%). Herb layer, apart from Aruncus sylvestris, is built of e.g.: Pulmonaria obscura, Galium schultesii Vest, Poa nemoralis L., Aegopodium podagraria, Rubus idaeus L. and Vicia cracca L.

Population C – forest south of “Wyczółki” village (52°10′47″N 22°26′11″E). The largest population of Aruncus sylvestris in the region (Poludniowo-podlaska Lowland) occurs on an area of ca 2 ha in 4 types of phytocoenoses. Due to habitat differentiation, population C was divided into 4 subpopulations:

- C₁ – subpopulation associated with the ecotone community and dominated by Elymus repens, which developed in a border zone between the forest Tilio-Carpinetum and a crop field. Aruncus sylvestris grows there in unfavorable conditions determined by intense insolation, dry soils and competition with Elymus repens.

- C₂ – subpopulation is present in the community Tilio-Carpinetum on fertile, moderately wet soil under heavy shading. The coverage of tree crowns (composed mainly of Carpinus betulus with the admixture of Alnus glutinosa (L.) Gaertn. and Fraxinus excelsior L.) ranges from 80 to 100%. Shrub layer built of Corylus avellana and young individuals of Carpinus betulus has a coverage of 70–90%. In relatively loose herb layer (the coverage of 50–70%) most frequent are: Brachypodium sylvaticum (Huds.) P. Beauv., Poa nemoralis, Melica nutans L., Asarum europaeum L., Daphne mezereum L., Galium schultesii and Polygonatum multiflorum (L.) All.

- C₃ – subpopulation associated with the community of ash-alder forest (Fraxino-Alnetum forest phase), which developed on the bank of a stream on very fertile and moist soil. Most individuals of Aruncus sylvestris are heavily shaded by a tree layer built of Alnus glutinosa of a coverage of 70–100% and abundant shrub layer mainly composed of Corylus avellana and Padus avium Mill. Herb layer of a coverage of 100% is dominated by: Mercurialis perennis L., Asarum europaeum and Carex acutiformis Ehrh.

- C₄ – subpopulation grows in the young, regularly cut community of ash-alder forest (Fraxino-Alnetum thicket phase). In comparison with the forest phase of this community (subpopulation C₃) there is much more light there due to a lack of tree layer and loose shrub layer of a coverage of ca 40%. The undergrowth is dominated by hydrophilous species of high light demands like: Cirsium oleraceum (L.) Scop., Lysimachia vulgaris L., Galium elongatum C. Presl. and very rare on the lowlands Aconitum variegatum L.

3. MATERIAL AND METHODS

3.1. Field studies

Field studies were carried out in the years 2006–2008 during the flowering period of Aruncus sylvestris. All flowering male and female shoots were counted in every population. To check the relationship between sex structure and habitat conditions the intensity of solar radiation was estimated in three degrees: full insolation, half-light and shade. Soil moisture was categorised as dried, fresh and wet. These parameters were determined for two small populations growing in a uni-
form habitat of population A and population B. Detailed studies on the differentiation of the sex ratio in relation to variable habitat conditions and on sexual dimorphism were performed in population C – the largest population of *Aruncus sylvestris* on Południowopodlaska Lowland. To do this, individuals of *Aruncus sylvestris* were inventoried and numbered by placing labels with subsequent numbers (from 1 to 106) on every clump. Spatial distribution of individuals within population was estimated by marking their position on a map of the object based on topographic map in the scale 1:25 000. Sex of each individual was determined and its distance from at least two closest neighbours was measured with measuring tape. All measurements were referred to two points to the nearby pole of low voltage energetic line. Soil moisture, solar radiation (full, half-light, shade) and the place of the clump in population (central, outskirts, separated clumps) were noted to estimate habitat conditions for each clump.

Twenty four phytosociological relevés (*Braun-Blanquet* 1964) were made with the consideration of variability of phytocoenoses inhabited by *Aruncus sylvestris*. To estimate whether sexual dimorphism really exists in the species the following parameters were measured (counted) separately in all flowering male and female individuals in the population:
- flowering and barren shoots,
- height of flowering shoots,
- length of inflorescences,
- number of leaves on flowering shoot,
- size of the middle leaf:
  - length with petiole,
  - length without petiole,
- maximum leaf width.

272 individuals were measured: 171 male and 101 female. All measurements were set up in the field charts. Identification of communities was made based on *Matuszkiwicz* (2001). Species names were adopted after *Mirek et al.* (2002).

### 3.2. Statistical data processing

Sex ratio *i.e.* the male to female ratio was calculated according to the formula $1:F/M$. It was estimated for the whole population and separately for individuals growing under different light conditions and in various types of phytocoenoses.

Differences between mean values of analysed traits (height of flowering shoots, length of inflorescences, number of leaves on flowering shoots, size of the middle leaf) for different sexes were verified with the t-test after preliminary checking their normal distribution. The significance of differences was verified at $P < 0.05$. Calculations were made with the Statistica 6.0 statistical package.

Chi-square test was used to analyse the preference of male and female individuals to photic conditions. Tabular value of $c^2_{tab}$ was taken from the tables for chi-square test for $\alpha = 0.05$ and $k = (r-1)(s-1)$ degrees of freedom. If the empirical value is $c^2_{emp} > c^2_{tab}$ at a given significance level then the hypothesis of independence of X and Y should be rejected. It means that X and Y depend on each other.

Similarities between populations and subpopulations growing in various phytocoenoses were estimated with the *W* and *F* statistics. The significance was verified with the *W* test at $\alpha = 0.05$ and *F* statistics with the Statistica 6.0 statistical package.

### Table 1. Analysis of variance – mean values and standard deviations of selected morphological traits in male and female individuals of *Aruncus sylvestris*. The data from all populations (n = 272) were used.

<table>
<thead>
<tr>
<th>Trait</th>
<th>Sex</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>male</td>
<td>mean</td>
<td>SD</td>
<td>female</td>
<td>mean</td>
</tr>
<tr>
<td>height of flowering shoots (cm)</td>
<td></td>
<td>147.48</td>
<td>28.10</td>
<td>191.53</td>
<td>46.59</td>
</tr>
<tr>
<td>length of inflorescences (cm)</td>
<td></td>
<td>42.30</td>
<td>12.90</td>
<td>53.60</td>
<td>19.22</td>
</tr>
<tr>
<td>number of leaves on flowering shoot</td>
<td></td>
<td>8.75</td>
<td>2.83</td>
<td>11.37</td>
<td>3.15</td>
</tr>
<tr>
<td>length of the middle leaf with petiole (cm)</td>
<td></td>
<td>46.17</td>
<td>10.44</td>
<td>58.59</td>
<td>10.31</td>
</tr>
<tr>
<td>length of the middle leaf without petiole (cm)</td>
<td></td>
<td>46.17</td>
<td>10.44</td>
<td>58.59</td>
<td>10.31</td>
</tr>
<tr>
<td>maximum width of the middle leaf (cm)</td>
<td></td>
<td>32.18</td>
<td>9.51</td>
<td>42.26</td>
<td>9.87</td>
</tr>
</tbody>
</table>

### Table 2. Similarities of communities (based on Matuszkiwicz 2001) and cophenetic similarity of subpopulations of *Aruncus sylvestris*.

<table>
<thead>
<tr>
<th>Population</th>
<th>Community</th>
<th>Cophenetic similarity</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td></td>
<td>1.000</td>
</tr>
<tr>
<td>B</td>
<td></td>
<td>1.000</td>
</tr>
<tr>
<td>C</td>
<td></td>
<td>1.000</td>
</tr>
</tbody>
</table>

It means that the communities of populations A, B, and C are identical.
noses were analysed with the cluster analysis (Statistica 6.0).

4. RESULTS

As checked with Student t-test (Table 1) statistically significant differences between male and female individuals were found for the following traits:
- height of the flowering shoots,
- length of inflorescences,
- number of leaves on the flowering shoot,
- length of the middle leaf with petiole,
- length of the middle leaf without petiole,
- maximum width of the middle leaf.

Higher values of all these features were noted in females.

Sex ratios in studied populations of *Aruncus sylvestris* were quite variable (Table 2, Fig. 1). In population B the male to female ratio was 1:0.94 which was close to the theoretical value. The population is compact, grows on a small area of 21m² near a forest road in side but not direct insolation (half-light). In the two other populations the sex ratio was markedly shifted towards the domination of male individuals. The population A grows on an area of 1000m² in full shade under compact tree stand of *Tilia cordata*. Population C is dispersed on over 2 ha. *Aruncus sylvestris* grows there in differentiated habitat conditions. Population as a whole is dominated by male individuals in the proportion of 1:0.65 but the ratio varies greatly in particular subpopulations that inhabit various types of phytocoenoses and grow under different photic conditions.

The ratio of male to female individuals growing in half-light (i.e. in conditions optimum for the species) varied around 1:1. In both full solar radiation and shade male individuals definitely prevailed (Table 3). These regularities were statistically confirmed with the chi-square test. Significant relationship was demonstrated between the sex ratio and insolation: $c^2_{emp} = 24.30 > c^2_{0.05} = 5.991$

Sex ratios estimated for subpopulations growing in various phytocoenoses in the site of population C showed an increase of

Table 2. Sex ratio in three lowland populations of *Aruncus sylvestris*. A – unfavorable site (shade), B – favorable site (half-light), C – site with different habitat conditions. Details in the text.

<table>
<thead>
<tr>
<th>Population</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\sigma^a$</td>
<td>37</td>
<td>36</td>
<td>168</td>
</tr>
<tr>
<td>$\sigma^b$</td>
<td>20</td>
<td>34</td>
<td>110</td>
</tr>
<tr>
<td>$\sigma^a / \sigma^b$</td>
<td>-</td>
<td>-</td>
<td>3</td>
</tr>
<tr>
<td>Sex ratio</td>
<td>1:0.54</td>
<td>1:0.94</td>
<td>1:0.65</td>
</tr>
</tbody>
</table>

Fig. 1. Cluster analysis of sex ratio values for three lowland *Aruncus sylvestris* populations. A,B,C – see Table 2 and text.
females’ contribution from very low (sex ratio = 1:0.20) in the ecotone community with *Elymus repens* (subpopulation C1), through shady forests *Tilio-Carpinetum* (subpopulation C2) and fertile wet forest *Fraxino-Alnetum* (subpopulation C3) to a thicket form of *Fraxino-Alnetum* with often logging of older trees (subpopulation C4), where the sex ratio of *Aruncus sylvestris* achieved the value of 1:1 (Table 4).

Cluster analysis (Fig. 2) showed that subpopulations C1 and C2 of distinct male domination are similar to each other and significantly different from the subpopulations C3 and C4 where the sex proportions were more or less balanced.

Comparison of subpopulation C4 of *Aruncus sylvestris* growing in *Tilio-Carpinetum* with those growing also in *Tilio-Carpinetum* (population A and population B) revealed that population B growing in better light conditions near forest road at the outskirts of the community differed significantly from two other populations (Fig. 3).

### 5. DISCUSSION

Three hermaphroditic individuals of distinct prevalence of male flowers were found in studied lowland populations of *Aruncus sylvestris*. Similar phenomenon is found quite often in dioecious plants (Dellaporta and

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### Table 3. The number of male and female individuals of *Aruncus sylvestris* in different light conditions.

<table>
<thead>
<tr>
<th></th>
<th>full light</th>
<th>half-light</th>
<th>shade</th>
</tr>
</thead>
<tbody>
<tr>
<td>population</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>♂</td>
<td>37</td>
<td>86</td>
<td>36</td>
</tr>
<tr>
<td>♀</td>
<td>11</td>
<td>86</td>
<td>34</td>
</tr>
<tr>
<td>♂/♀</td>
<td>1</td>
<td>2</td>
<td>-</td>
</tr>
<tr>
<td>Sex ratio</td>
<td>1:0.30</td>
<td>1:1</td>
<td>1:0.94</td>
</tr>
</tbody>
</table>

### Table 4. The number of male and female individuals in various subpopulations of largest *Aruncus sylvestris* population C. C1 – ecotone community with *Elymus repens*, C2 – *Tilio-Carpinetum*, C3 – *Fraxino-Alnetum* forest phase, C4 – *Fraxino-Alnetum* thicket phase.

<table>
<thead>
<tr>
<th>subpopulations</th>
<th>C1</th>
<th>C2</th>
<th>C3</th>
<th>C4</th>
</tr>
</thead>
<tbody>
<tr>
<td>♂</td>
<td>30</td>
<td>46</td>
<td>23</td>
<td>68</td>
</tr>
<tr>
<td>♀</td>
<td>6</td>
<td>17</td>
<td>18</td>
<td>69</td>
</tr>
<tr>
<td>Sex ratio</td>
<td>1:0.20</td>
<td>1:0.40</td>
<td>1:0.78</td>
<td>1:1.01</td>
</tr>
</tbody>
</table>

Fig. 2. Cluster analysis of sex ratio values in various subpopulations of largest *Aruncus sylvestris* population C. C1, C2, C3, C4 – see Table 4.
Sexual dimorphism in *Aruncus sylvestris* Calderon-Urea 1993). The ability of male individuals to produce fruits may have great biological importance especially in fragmentary populations. No explanation of the mechanism of sex determination in *Aruncus sylvestris* nor any remark on a possibility of changing sex in a real individual were found in published data.

Interesting results were obtained in studies on sexual dimorphism in *Aruncus sylvestris*. It was demonstrated that female individuals were higher, produced longer inflorescences and had longer and wider leaves than male individuals. This is quite common phenomenon in dioecious plants. Results similar to ours were obtained for other dioecious plants like e.g.: *Asparagus officinalis* (Dzhaparidze 1967, Machon et al. 1995), *Cannabis sativa* and *Pistacia mutica* (Dzhaparidze 1967), *Rumex acetosella* (Lovett Doust et al. 1987), *Rumex acetosa* (Korpelainen 1992), *Lindera benzoin* (Cipollini and Whigham 1994), *Maireana pyramidata* (Leigh and Nicotra 2003) and many others. Higher growth parameters and thus larger assimilation apparatus in females is probably associated with high energetic costs spent for fruit production (Korpelainen 1992, Ashman 2005). In tree species, however, high energetic expenditures may cause growth inhibition in females (Sakai et al. 2006). Strong female shoots of great mechanical strength play an additional important function in spreading seeds, whose large amounts are stored on dried shoots till early spring.

In studied lowland populations of *Aruncus sylvestris* the sex ratio varied from distinct domination of male individuals in unfavourable conditions (excessive or insufficient solar radiation or dried soils) to balanced sex ratio of 1:1 in optimum conditions (half-light, fertile and wet habitats). Consequent deviation from the theoretical sex ratio in dioecious plants of chromosome type of sex determination might be genetically influenced and inherited from father. Such type of determination of the sex ratio was found in *Silene alba* (Taylor 1994), and slightly different – in *Salix viminalis* (Alström-Rapaport et al. 1997). The species shows primary, at the embryonic stage, deviation towards female domination. It is possible that the sex ratio is similarly determined in dioecious plants of the genus *Rumex*, in which consequent deviation to female domination was also found (Zarzycki and Rychlewski 1972). However, according to other authors (Putwain and Harper 1972, Korpelainen 1992) the deviations are associated with different habitat requirements of male and female individuals. Occupation of different ecological niches by males and females seems to be the most frequent reason for the deviation of the sex ratio from theoretical 1:1 value (Faliński 1980,
Meagher 1981, Waser 1984, Bierzychudek and Eckhart 1988, Garcia and Antor 1995, Rottenberg 2000, Ortiz et al. 2002, Cvetković and Jovanović 2007). Our results seem to exclude the possibility of genetic determination of the sex ratio in Aruncus sylvestris. Observed deviations of the sex ratio towards male domination were positively correlated with worsening habitat conditions of this species which clearly shows higher environmental requirements of female individuals in this species, more so, that the sex ratio in populations growing in optimum conditions was close to 1:1.

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6. REFERENCES


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