The influence of *Ambrosia trifida* on vegetative production of *A. artemisiifolia*

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**SUMMARY**

*Ambrosia artemisiifolia* (common ragweed) and *A. trifida* (giant ragweed) are very important weed species that are invasive in Serbia and are often found in agricultural regions. When these weeds are present at high densities, crop yields can be significantly reduced or even completely destroyed. Unlike *A. artemisiifolia*, *A. trifida* is locally present in the Central Bačka region (Vojvodina province), and it is expected that its area of distribution will expand in the future. Starting from the assumption that future distribution of *A. trifida* could take on larger proportions than now, the aim of this study was focused on examining the interaction between these two species.

Experiments were conducted using the replacement design model, in which *Ambrosia trifida*/*Ambrosia artemisiifolia* per m², were planted as density ratios of 10/0; 8/2; 4/6; 6/4; 2/8, and 0/10, in a completely randomized block system with four replications. The vegetative parameters (height and dry mass) of *A. artemisiifolia* were measured in July, August and September over a period of two years (2016 and 2017), and the results were statistically analysed in the Statistical Package for the Social Sciences (SPSS 23).

In July 2016, the average height of *A. artemisiifolia* was in the range between 35.00 and 50.40 cm, in August it was from 68.00 to 95.50 cm, and between 83.75 and 99.80 cm in September. In the following season (2017), the corresponding values ranged from 56.19 to 78.50 (July), 98.38 to 125.50 cm (August) and 111.19 to 148.50 (September).

An increase in the number of *A. artemisiifolia* plants and decrease in *A. trifida* counts per m² caused an increase in the dry mass of *A. artemisiifolia* per plant. The dry mass of *A. artemisiifolia* ranged from 4.22 to 6.11 g/plant (July), 8.96 to 10.27 g/plant (August) and 7.04 to 19.53 g/plant (September). In the following season, these values ranged from 9.62 to 14.60 g/plant, 14.37 to 28.90 g/plant, and 23.43 to 40.47 g/plant in July, August and September, respectively.

Minimum values of vegetative parameters were recorded in the treatment with 2 plants, and maximum in the treatment with 10 *A. artemisiifolia* plants/m². This means that interspecific competition is more pronounced in this ragweed species than intraspecific competition.

**Keywords:** Ambrosia, common ragweed, giant ragweed, competition, vegetative parameters
INTRODUCTION

Invasive species are considered to be one of the main factors contributing to global change nowadays, making them the prime focus of various studies in the fields of ecology and related sciences (Fumanal et al., 2007; Pyšek et al., 2009; Essl et al. 2009). When they grow in the same habitat, invasive weeds compete for resources and the more competitive plants suppress other species, which leads to a reduction in the number of plants of the endangered population. Invasive species are more prone to suppress other plants, resulting in changes in biodiversity and plant community structure (Shine et al., 2010). Their expansion has a detrimental effect both on agricultural production and biodiversity of a given area, and many of them also have a negative impact on human health due to allergens that they produce (Šikoparija et al., 2009 Gerber et al., 2011).

Both *Ambrosia artemisiifolia* and *A. trifida* belong to a group of alien, harmful and invasive weed species (Kömives et al., 2006; Webster et al., 1994).

*A. artemisiifolia* (common ragweed) is native to the North American continent, whence it was introduced to Asia, Australia and Europe (Chun et al., 2010; Cunze et al., 2013). Common ragweed belongs to a group of invasive weed species harmful to the economies of many parts of Europe, including the Balkans (Kömives et al., 2006). Aside from being a very important weed, it is also listed as an invasive species in Serbia, where it is often present in rural areas (row crops, orchards and vineyards, alfalfa crops, abandoned fields, field edges and ruderal land), as well as urban areas. At high density, it acts as a strong competitor (for light, water, nutrients and space) against other plant species, and is able to cause huge yield losses of many crops (Kömives et al., 2006). *A. artemisiifolia* can withstand strong competition, which allows it to survive in environments that are less favourable for growth, and to achieve high spread rates (Raynal & Bazzaz, 1975). According to Weber and Gut (2005), only a few species (*Senecio inaequidens, Erigeron canadensis* and *Reynoutria japonica*) in Europe have higher spread rates than common ragweed. The fact that it is an extremely vigorous grower is also indicated by data on its abundance per unit area, i.e. some authors state the number of over 1000 plants/m² and even up to 4000 plants/m² (Mataruga, 2004).

Unlike *A. artemisiifolia*, *A. trifida* (giant ragweed) is currently present in Serbia only locally, along roadsides in rural areas and between settlements in the Central Bačka region of Vojvodina Province. It is also commonly found on field edges, as well as in sunflower, maize, soybean and sugar beet fields. *A. trifida* forms a large vegetative mass and belongs to the group of strong competitors for both natural resources (light, water and nutrients) and space. Rapid growth and development, as well as pronounced plasticity allow it to develop and occupy large surface areas, where it absorbs water and nutrients from the soil and shades out other plant species, which enables it to make better use of solar radiation (Webster et al., 1994).

Considering that *A. trifida* is becoming naturalized in Serbia (Vrbičanin et al., 2004; Vrbičanin, 2015), even though it is now only locally present in the area of Central Bačka (Kucura, Savino Selo, Despotovo, Ruski Krstur, Ravno Selo, etc.) (Vrbičanin et al., 2004; Malidža & Vrbičanin, 2006; Vrbičanin et al., 2015), this research was performed to study its invasive potential and its capacity to suppress *A. artemisiifolia* when it is associated with it. Looking at the vegetative and generative potential of *A. trifida*, it is highly likely that this ragweed species will spread in the future. The focus of this research was to study the inter- and intraspecific influence of *A. artemisiifolia* and *A. trifida* when they coexist in the same habitat. The goal was to examine the vegetative production of *A. artemisiifolia* when growing in association with *A. trifida*.

MATERIALS AND METHODS

The experiment was performed in April 2016 and 2017 on a piece of land in the village of Dobrić (Šabac) (44° 41’N 19° 34’E) where nothing had been sown in the previous two years. Since the presence of *A. trifida* was not detected in the area of Western Serbia, seeds of this plant were obtained from a location in Centralna Bačka region (45°30’N 19°31’E). After collection in the previous season, the seeds were stored under controlled conditions (in a refrigerator at *t = 4 °C*). Considering that *A. artemisiifolia* was already present in the test area in high numbers (>100 plants/m²), it was not necessary to sow it. Up to 100 seeds of *A. trifida* were sown depending on treatment.

The experiment was set up on the principle of completely randomized block design with four replications, using the model of replacement series
(“Replacement design”) (Kropff & van Laar, 1993). A total of five treatments were conducted, i.e. five different density ratios of A. trifida/A. artemisiifolia: 2/8, 4/6, 6/4, 8/2 and 10/0 (total number of plants was 10/m²). The given plant density was maintained by thinning every 7 to 10 days over the season. When establishing any given number of A. trifida and A. artemisiifolia, care was taken to ensure that their distribution per unit area was uniform (that the plants of both ragweeds were at an equal distance from each other) and that samples of both species were in the same stage of development.

Plot size was 6 m², and each plot was divided into six sub-plots of 1 m². Vegetative parameters, i.e. plant height (cm) and dry mass (g) of A. artemisiifolia, were measured on four sub-plots during the vegetative season three times (in July, August and September). Plant height was measured on experimental plots with a wooden pull-out meter, while dry mass was measured on precise scales in the laboratory.

The results of texture and chemical analysis of powdered clay soil regarding the basic agrochemical properties [pH in KCl and H₂O, P₂O₅ (mg/100 g), K₂O (mg/100 g) and humus (%)] are presented in Table 1.

<table>
<thead>
<tr>
<th>Table 1. Texture and chemical properties of soil in the experimental field</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Texture properties</strong></td>
</tr>
<tr>
<td>Soil texture</td>
</tr>
<tr>
<td>Coarse sand (0.2-2)</td>
</tr>
<tr>
<td>Fine sand (0.2-0.5)</td>
</tr>
<tr>
<td>Total sand (0.05-2)</td>
</tr>
<tr>
<td>Physical clay (&lt;0.05)</td>
</tr>
<tr>
<td>Powder (0.002-0.05)</td>
</tr>
<tr>
<td>Clay (&lt;0.002)</td>
</tr>
<tr>
<td><strong>Chemical properties</strong></td>
</tr>
<tr>
<td>pH KCl</td>
</tr>
<tr>
<td>H₂O</td>
</tr>
<tr>
<td>P₂O₅ (mg/100 g)</td>
</tr>
<tr>
<td>K₂O (mg/100 g)</td>
</tr>
<tr>
<td>Humus (%)</td>
</tr>
</tbody>
</table>

**Statistical analysis**

To calculate the effects of treatments (different plant ratios) on the measured vegetative parameters of A. artemisiifolia, the analysis of variance (ANOVA) was used in the statistical program SPSS 23, and differences between means were tested using the LSD test at the levels of significance of: * p<0.05, ** p<0.01 and ns p>0.05.

**RESULTS**

In general, the values of vegetative parameters were higher in 2017 due to more favourable meteorological conditions, primarily the amount and distribution of precipitation (Figure 1). Vegetative production of A. artemisiifolia plants, growing under different ratios of two Ambrosia species during July, August and September, had a similar trend in both years. With increasing number of A. artemisiifolia and decrease in A. trifida/m², the height and dry weight of A. artemisiifolia increased.
In 2016, the average height of *A. artemisifolia* plants ranged between 35.00±2.50 and 50.40±1.78 cm in July, 68.00±2.88 and 95.50±2.11 cm in August, and 83.75±2.33 and 99.80±3.70 cm in September (Figure 2). The highest *A. artemisifolia* plants were recorded at its maximum density (0/10 *A. trifida/A. artemisifolia*), and the lowest in the treatment with 2 plants/m² (8/2 *A. trifida/A. artemisifolia*). However, differences between the other treatments, i.e. the different ratios of *A. artemisifolia* plants in association with *A. trifida*, were not statistically significant (Table 2).
In 2017, the height of *A. artemisiifolia* plants ranged from 56.19 ± 2.90 to 78.50 ± 4.10 cm (July), 98.38 ± 4.83 to 125.50 ± 4.58 cm (August) and 111.19 ± 3.50 to 148.50 ± 5.42 cm (September). Minimum value was recorded in the treatment with the lowest number of *A. artemisiifolia* plants (8/2 *A. trifida/A. artemisiifolia*) and maximum with 10 *A. artemisiifolia* plants/m² (0/10 *A. trifida/A. artemisiifolia*) (Figure 2). Significant differences (*P* ≤ 0.05) were found between treatments with 4 *A. trifida* versus 8 *A. artemisiifolia* plants/m², as well as treatments with 10 *A. trifida* compared to 6 and 8 *A. artemisiifolia* plants/m² (Table 3).

### Table 2. Significance of differences between treatments regarding vegetative parameters of *A. artemisiifolia* (*P* - value), 2016

<table>
<thead>
<tr>
<th><em>A. trifida/A. artemisiifolia</em></th>
<th>Height plants</th>
<th>Dry mass plants</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>July</td>
<td>August</td>
</tr>
<tr>
<td>6/4 to 8/2</td>
<td>0.6228</td>
<td>0.2938</td>
</tr>
<tr>
<td>4/6 to 6/4</td>
<td>0.1108</td>
<td>0.6443</td>
</tr>
<tr>
<td>2/8 to 8/2</td>
<td>0.0755</td>
<td>0.1404</td>
</tr>
<tr>
<td>0/10 to 8/2</td>
<td>0.6441</td>
<td>0.9979</td>
</tr>
</tbody>
</table>

* p<0.05, ** p<0.01 and ns p>0.05

### Table 3. Significance of differences between treatments regarding vegetative parameters of *A. artemisiifolia* (*P* - value), 2017

<table>
<thead>
<tr>
<th><em>A. trifida/A. artemisiifolia</em></th>
<th>Height / plant</th>
<th>Dry mass / plant</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>July</td>
<td>August</td>
</tr>
<tr>
<td>6/4 to 8/2</td>
<td>0.7431</td>
<td>0.7912</td>
</tr>
<tr>
<td>4/6 to 6/4</td>
<td>0.0674</td>
<td>0.1137</td>
</tr>
<tr>
<td>2/8 to 8/2</td>
<td>0.2688</td>
<td>0.3298</td>
</tr>
<tr>
<td>0/10 to 8/2</td>
<td>0.0038</td>
<td>0.0153</td>
</tr>
<tr>
<td>2/8 to 8/2</td>
<td>0.0566</td>
<td>0.1087</td>
</tr>
</tbody>
</table>

* p<0.05, ** p<0.01 and ns p>0.05
In 2016, increase in the number of *A. artemisiifolia* plants versus *A. trifida* caused an increase in the dry mass of *A. artemisiifolia*/plant. Dry mass values ranged from $4.22 \pm 0.23$ to $6.11\pm0.89$ g/plant (July), $8.96\pm1.43$ to $10.27\pm0.90$ g/plant (August) and $7.04\pm0.64$ to $19.53\pm1.16$ g/plant (September). Minimum value was measured in the treatment with 2 *A. artemisiifolia* plants/m² and maximum with 10 plants/m² (Figure 3). Statistically significant differences ($P\leq0.05$) were confirmed between treatments with 10 *A. artemisiifolia* plants/m² compared to 2, 4, 6 and 8 *A. artemisiifolia* plants/m²; treatments with 2 compared to 4 and 6 plants, and also between treatments with 8 and 6 *A. artemisiifolia* plants/m² (Table 2).

The values of dry mass/plant were higher in 2017 in all assessments. The dry mass of *A. artemisiifolia* ranged from $9.62\pm0.59$ to $14.60\pm0.35$ g/plant (July), $14.37\pm0.44$ to $28.90\pm0.49$ g/plants (August) and $23.43\pm1.45$ to $40.47\pm0.94$ g/plant (September). *A. artemisiifolia* plants produced the highest dry mass/plant in the treatment with 10 *A. artemisiifolia*/m², and the lowest at the lowest density per area unit (8/2 *A. trifida/A. artemisiifolia*) (Figure 3). Statistically significant differences between treatments are shown in Table 3. ($0.01 \leq P \leq 0.05$) (Table 3).

Based on the Pearson correlation coefficient (Pcc), a high positive correlation between plant height and dry weight ($0.528^{**}; 0.518^{**}$) was confirmed both in 2016 and 2017 (Figures 4 and 5).

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![Figure 3. Dry mass of *A. artemisiifolia* plants in treatments with 2, 4, 6, 8 and 10 plants/m²](image3.png)

![Figure 4. Correlative dependence between the height and dry mass of *A. artemisiifolia* plants in 2016](image4.png)
DISCUSSION

In general, a large number of factors (biotic and abiotic) can affect the behaviour of weeds, their appearance, abundance and distribution, and thus the manifestation of inter- and intraspecific competition (Gibson et al., 2017; Adeux et al., 2019). Also, competition largely depends on meteorological conditions (temperature, precipitation and insolation). The results of this research showed that meteorological conditions greatly influenced the vegetative production of ragweed plants. Differences between the two years were more pronounced in terms of precipitation amounts and distribution, i.e. 2016 was drier and 2017 richer in precipitation (Figure 1). This statement is in accordance with results of previous studies. Nelson et al. (2006) emphasized the importance of optimal conditions for germination, growth and development of plants, and their fruiting, for their competitive potential. Consequently, a noticeable precipitation deficit during 2016 caused lower vegetative production of common ragweed plants at all A. trifida/A. artemisiifolia density ratios. Most often, weed species in competitive relationships undergo various changes under the conditions in which they grow, depending, among other factors, on the density of plants per unit area (Adeux et al., 2019). According to many authors, vegetative production is a very important indicator of competition between plants (Tollenaar et al., 1994; Patterson, 1995). Also, the ability of species to compete for resources is influenced by their very morphological characteristics. Among the most indicative morphological features in that sense are plant height and weight (Bertholdsson, 2005).

According to some authors, plant height is a parameter that largely contributes to suppressing other plants (Cosser et al., 1997; Piliksere et al., 2013). Mason et al. (2007) asserted that the interaction between plant height and other species characteristics could contribute to more efficient use of natural resources, so it often happens that plants growing at higher density can have better growth and higher yield. Also, the height of plants greatly affects the extent to which they can use solar energy. For example, under conditions of moderate soil moisture and nutrient content, competition for light between Abutilon theophrasti and soybean is one of the most important factors that affect plant height (Lindquist & Mortensen, 1999). Accordingly, A. theophrasti makes better use of light energy, which provides it with a competitive advantage over soybean crop (Ackey et al., 1990). According to Irwin and Aarsen (1996), the height and branching of a shoot in common ragweed can often vary depending on habitat conditions in which it grows, and especially on humidity (Bollinger et al., 1991). In Europe, the average height of A. artemisiifolia plants ranges from 1.5 to 2 m (Šilc, 2002), while in Serbia the height ranges from 20 to 150 cm, and sometimes up to 2 m (Vrbičanin et al., 2007). In general, the results of this study indicate that variable ratios of A. trifida/A. artemisiifolia affected the height of A. artemisiifolia plants.
The height of *A. artemisiifolia* plants ranged from 35.00 to 99.80 cm (from July to September) in 2016, and the growth trend remained the same in the following season, except that plants generally increased growth due to more favourable weather conditions (from 56.19 up to 148.50 cm) (Figure 2). However, even though the raised number of *A. artemisiifolia* plants in association with *A. trifida* increased the height of *A. artemisiifolia* plants, this cannot be attributed to competition for light because the total number of plants per unit area was constant (*A. trifida + A. artemisiifolia = 10 plants/m²*). Thus, the reason for the increased height of *A. artemisiifolia* plants that goes with the increase in its ratio in association with *A. trifida*, can be connected to absent intraspecific competition, i.e. when the number of *A. trifida* increases, the number of *A. artemisiifolia*/m² (with the same total number) drops, and the interspecies competition comes to the fore. Similar to these results, Oljača et al. (2000) found that with an increase in the number of *Datura stramonium* from 1 to 10 plants per meter length (1, 3, 6, and 10) in the inter-row space of corn, the height of *D. stramonium* had the following upward trend: 116 cm, 117.7 cm, 119.7 cm and 141.7 cm.

Although competition is a process that can manifest in several ways, it is most common for a more competitive species to act on a less competitive one by effecting its lower mass production (Bussan et al., 1997). Based on height and dry weight, stronger interspecies competition was confirmed in *A. artemisiifolia* in our experiment (Figure 3). In 2016, maximum dry weight/plant of *A. artemisiifolia* was 19.53 g (10 *A. artemisiifolia*/m²). Dry mass/plant in 2017 in all treatments was higher compared to the previous season, which was characterized as drier of the two years (Figure 1). The highest dry mass of *A. artemisiifolia* was recorded in monoculture (40.47 g/plant), and the lowest in the treatment with 2 *A. artemisiifolia*/m² (23.40 g/plant) (Figure 3). This fact can partly explain the success of common ragweed’s invasiveness, as it survives even when there is a great physical pressure within the population, leaving abundant offspring, which is a prerequisite for its further spreading and colonization of new habitats. *A. artemisiifolia* is a species that has increased plasticity and adapts better to stressful conditions such as nutrient, water and physical space deficits (Leskovšek et al., 2012). However, there is also an opposite view to be found in literature. Vidotto et al. (2007) found that with the number of *A. artemisiifolia* increasing from 4 to 25 plants/m², the dry weight of these plants fell from 687 to 140 g/plant (4.9 x), which was associated with nutrient deficiency. In contrast, and similar to our results, *Brassica napus* produced a higher mass/plant in monoculture compared to the situation when it grew alongside *Malva paraviflora* at different ratios, namely: 25/75%, 50/50% and 75/25%. In a treatment with *B. napus*/M. paraviflora 25/75% abundance ratio, the dry mass of *B. napus* was reduced by 68%, compared to monoculture (Bakhtiari & Saeedipoor, 2014).

**CONCLUSION**

The results of this experiment showed that the highest average height of *A. artemisiifolia* plants was in the treatment with 10 *A. artemisiifolia* plants/m², and the lowest with 2 *A. artemisiifolia* plants/m² (8/2 *A. trifida/A. artemisiifolia*). Also, an increase in the number of *A. artemisiifolia* plants/m², versus *A. trifida*, caused an increase in the dry weight of *A. artemisiifolia*/plant.

In general, the lowest values of vegetative parameters were found in treatments where the number of *A. artemisiifolia* plants was the lowest, compared to *A. trifida*. With increasing number of plants per area unit, vegetative parameters increased, and the maximum value was recorded in treatments with the highest number of *A. artemisiifolia* (10 plants/m²). It infers that interspecific competition is more pronounced than intraspecific competition in *A. artemisiifolia*. Increasing numbers of *A. artemisiifolia* plants per area unit did not suppress their growth, and this facilitates its successful invasion of new habitats, where it acts as a strong competitor and a great potential danger to crops. Considering all of the above, maximum responsibility of experts is needed, along with education of agricultural producers about the danger of this invasive weed species, the damage it can do to crops, as well as all available control methods, emphasising prevention.

**ACKNOWLEDGEMENT**

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REFERENCES


Uticaj *Ambrosia trifida* na vegetativnu produkciju *A. artemisiifolia*

**REZIME**

*A. artemisiifolia* (pelenasta ambrozija) i *A. trifida* (trolisna ambrozija) predstavljaju dve veoma važne vrste korova koje se nalaze u invaziji u Srbiji u poljoprivrednim regionima. Kada se jave u većoj brojnosti, prinosi useva mogu biti značajno umanjeni ili potpuno uništeni. Za razliku od *A. artemisiifolia*, *A. trifida* je lokalno prisutna u Centralnoj Bačkoj (Vojvodina) i očekuje se da se u budućnosti može još više proširiti. Polazeći od pretpostavke da bi u budućnosti distribucija *A. trifida* mogla da zauzme veće razmere nego sada, cilj ovih istraživanja bio je usmeren na ispitivanje interakcije između ovde dve vrste.

Eksperiment je izveden primenom modela zamenjujućih serija gde su vrste *A. trifida/A. artemisiifolia* po m² bile u zadatom odnosu 10/0; 8/2; 4/6; 6/4; 2/8 i 0/10, u potpuno slučajnom blok sistemu u četiri ponavljanja. Vegetativni parametri (visina i suva masa) *A. artemisiifolia* izmereni su u julu, avgustu i septembru tokom dve godine (2016. i 2017.), a rezultati su analizirani u statističkom paketu SPSS 23.

U julu 2016. godine prosečna visina *A. artemisiifolia* bila je u rasponu između 35,00 i 50,40 cm, u avgustu od 68,00 do 95,50 cm i između 83,75 i 99,80 cm u septembru. U sledećoj sezoni (2017) te vrednosti kretale su se od 56,19 do 78,50 cm (jul), 98,38 do 125,50 cm (avgust) i od 111,19 do 148,50 cm (septembar).

Povećanje broja biljaka *A. artemisiifolia* i smanjenje *A. trifida* po m² prouzrokovalo je povećanje suve mase *A. artemisiifolia* po biljci. Suva masa *A. artemisiifolia* bila je u rasponu od 4,22 do 6,11 g/biljci (jul), 8,96 do 10,27 g/biljci (avgust) i 7,04 do 19,53 g/biljci (septembar). U sledećoj sezoni ove vrednosti kretale su se od 9,62 do 14,60 g/biljci, 14,37 do 28,90 g/biljci i 23,43 do 40,47 g/biljci u julu, avgustu i septembru.

Minimalne vrednosti vegetativnih parametara zabeležene su u tretmanu sa 2 biljke, a maksimalne u tretmanu sa 10 biljaka *A. artemisiifolia/m²*, što znači da je kod ove vrste izraženija interspecijalska u poređenju sa intraspecijalskom kompeticijom.

**Ključne reči:** *Ambrosia*, pelenasta ambrozija, trolisna ambrozija, kompeticija, vegetativni parametri