Effects of nicosulfuron on biological production of weedy sunflower (*Helianthus annuus*)

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**SUMMARY**

Field experiments were conducted to quantify the effects of nicosulfuron on biological production of weedy sunflower (*Helianthus annuus*). Plants of this weed species were sown in plots sized 5x4.2 m with inter-row spacing of 24 cm, and 70 cm distance between rows. Plants with two pairs of developed leaves were treated with nicosulfuron using the next rates: 0, 10, 20, 40, 60 and 80 g a.i. ha⁻¹. Vegetative parameters (plant height, fresh weight and leaf area) were recorded five times during vegetation, including the first measurement just before herbicide application and four measurements at intervals of about two weeks. Head and seed production (number of heads per plant, head weight, head diameter, weight of seeds per head, weight of seeds per plant, number of seeds per plant) were determined after seed maturity.

The application of nicosulfuron caused growth reduction of weedy sunflower in comparison with the control. All application rates of this herbicide reduced vegetative (height, fresh weight, leaf area) and some generative (number of heads per plant, weight of seeds per plant, number of seeds per plant) parameters, which decreased with increasing rates of nicosulfuron. Effects on the remaining generative test parameters (head weight, head diameter, weight of seeds per head) were not analogous to the effects on other parameters.

**Keywords**: nicosulfuron; generative production; vegetative production; weedy sunflower

**INTRODUCTION**

Herbicides constitute an essential tool for weed management in conventional agriculture, and acetolactate synthase (ALS) inhibitors are widely used. Those herbicides have become popular due to their effectiveness, relatively low use rates, and low mammalian toxicity (Schmidt et al., 2004). Today, ALS inhibitors are used to control weeds effectively both pre- and post-emergence in corn and other crops. One of them is nicosulfuron, a selective post-emergence sulfonylurea herbicide that provides excellent control of many perennial and annual grasses and some broadleaf weed species (Baghestani et al., 2007; Nosratti et al., 2007). Although, broadleaf species like *Amaranthus retroflexus* and *Polygonum pensylvanicum* are extremely susceptible to nicosulfuron (Kapusta et al., 1994), many other species, including *Chenopodium album*, *Ambrosia artemisiifolia*, *Abutilon theophrasti*, *Xanthium strumarium* and *Solanum ptycanthum*, are not susceptible (Lueschen et al., 1992). Nicosulfuron applied at the seedling stage does not directly affect plant reproduction but it affects plant traits such as height,
fresh weight, leaf area and burs production in annual weeds like *X. strumarium* (Bozic et al., 2013).

*Helianthus annuus* L. (fam. Asteraceae/Compositae) is a species that occurs in many different forms: as "normal" crop plants, atypical plants known as “off-type” crop, wild sunflower, volunteer plants or weedy population of sunflower (Reagon & Snow, 2006). The origin of weedy sunflower populations may be different. Namely, seeds of wild forms may have been introduced unintentionally, and then wild sunflowers could have hybridized with sunflower crops during their seed production, making crop-wild hybrids. Furthermore, weedy sunflower could result from spontaneous evolution of volunteer populations, and volunteer population may hybridize with ornamental sunflowers grown in gardens (Muller et al., 2011). In Europe, sunflower volunteers are commonly present in the field (Ostrowski et al., 2010). Weedy populations of this species are present in France (Muller et al., 2009) and Spain (Poverene & Cantamutto, 2010), as well as in Hungary (Benécsné Bárdi et al., 2005) and Serbia (Saulić et al., 2013; Vrbničanin et al., 2014) and in Croatia, Romania, etc. Those populations are morphologically clearly different from the original volunteers (Muller et al., 2009). There is no information about weedy sunflower effects on crop productivity and yield loss have been studied by many researchers (Arslan et al., 2006; Karimmojeni et al., 2010; Zafar et al., 2010), but few studies have investigated effects of different factors on weed biological production (Conley et al., 2002; Vrbnicanin et al., 2006, 2017; Onć-Jovanović et al., 2011; Bozic et al., 2013, 2016). Generally, the influence of factors such as time of weed emergence, crop density or illumination on weed biomass and seed production have been studied frequently (Benvenuti et al., 1994; Conley et al., 2002; Teasdale & Cavigelli, 2010), while studies on the influence of herbicides on biological production have been rarer (Vrbnicanin et al., 2006; Bozic et al., 2013).

As weedy sunflower and *Sorghum halepense* are common dominant weeds in some Serbian maize fields, and nicosulfuron is known to be effective in controlling *S. halepense* (Baghestani et al., 2007; Nosratti et al., 2007), it is useful to know more about weedy sunflower response to that herbicide. To our knowledge, weedy sunflower response to herbicides in Europe has been rarely studied (Božić et al., 2011, 2019; Bozic, 2016). Bozic et al. (2016) found that the recommended rate (40 g a.i. ha⁻¹) of nicosulfuron reduced vegetative and generative production of this weed, and Božić et al. (2019) confirmed variability in weedy sunflower population responses to the same herbicide. As those studies showed that the recommended nicosulfuron rate was only able to reduce plant growth and different vegetative and generative parameters, rather than cause mortality of weedy sunflower, the objective of this study was to examine the influence of different rates of that herbicide on biological production of weedy sunflower under field conditions.

**MATERIALS AND METHODS**

Weedy sunflower seeds were collected in the autumn of 2005 from an area that had not been previously treated with any herbicides. The collected seeds were stored at room temperature in the laboratory.

A field experiment was conducted in Padinska Skela PKB Agroekonomik near Belgrade in 2008 and again in 2009. Seeds were sown in containers and transplanted to the field when the first pair of leaves developed.

| Month  | 2008 MMT (°C) | 2008 R (mm) | 2009 MMT (°C) | 2009 R (mm) |
|--------|---------------|-------------|---------------|-------------|
| April  | 12.1          | 37.60       | 13.8          | 17.00       |
| May    | 17.1          | 49.00       | 17.6          | 29.20       |
| June   | 20.9          | 39.60       | 18.9          | 98.20       |
| July   | 21.1          | 42.00       | 21.8          | 35.80       |
| August | 21.5          | 35.00       | 21.5          | 76.20       |
| September | 14.8          | 70.00       | 17.8          | 2.20        |
The plants were grown on alluvial black marsh soil in plots sized 5×4.2 m (21 m²). The experimental design was a completely randomized block with four replications. Distance between rows was 70 cm and inter-row spacing was 24 cm. Weather conditions in the experimental area in 2008 and 2009 are summarized in Table 1, while an overview of the trial timeline is shown in Table 2.

Nicosulfuron (Motivell, 40 g a.i. L⁻¹, SC, BASF, Germany) was applied when plants developed two pairs of leaves. A Neptune 15, Kwazar® knapsack sprayer with RS-MM 110°/04 nozzles was used for herbicide application. The amount of water was 300 L per hectare. The herbicide application rates were: 0, 10, 20, 40, 60 and 80 g a.i. ha⁻¹. Vegetative parameters (plant height, fresh weight and leaf area) were measured just before herbicide application and four times after it, i.e. 14, 30, 44 and 58 days after herbicide treatment (DAHT) in 2008 and 16, 33, 48 and 60 DAHT in 2009. After maturity, head (number of heads per plant, head weight, and head diameter) and seed (weight of seeds per head, weight of seeds per plant, and number of seeds per plant) production were recorded. All parameters were recorded on four randomly selected and harvested plants from each plot. Analysis of variance (ANOVA) was used to analyze and interpret the data.

RESULTS AND DISCUSSION

Bozic et al. (2016) reported that weedy sunflower could not be controlled with nicosulfuron satisfactorily, but this herbicide reduced different vegetative parameters of that species. The results of this research are consistent with other findings indicating that weedy sunflower is not susceptible to nicosulfuron. Namely, a doubled recommended rate (80 g a.i. ha⁻¹) caused no mortality of treated plants. Also, these results are in accordance with data confirming that many broadleaf weeds (Chenopodium album, Ambrosia artemisiifolia, Abutilon theophrasti, Xanthium strumarium and Solanum ptycanthum) could not be controlled satisfactorily with nicosulfuron (Lueschen et al., 1992; Dogan et al., 2005). But the results presented in Figure 1 and Table 2 show that different rates of nicosulfuron (0, 10, 20, 40, 60 and 80 g a.i. ha⁻¹) caused significant reductions in all vegetative parameters (plant height, fresh weight and leaf area) of treated weedy sunflower plants. Depending on the year and nicosulfuron rate applied, plant height declined by 29-56%, fresh weight by 63-85% and leaf area by 43-73%, compared to the untreated control samples. The level of reduction in listed vegetative parameters depends on the herbicide rate, time of assessment and year, but all of them decreased with increasing nicosulfuron rates. Statistically significant (p<0.05) or very significant (p<0.01) differences between treated and control plants were confirmed for all parameters. Depending on nicosulfuron rate, fresh weight of weedy sunflower declined by 52-91% (58 DAHT in 2008) and 56-92% (60 DAHT in 2009). Over the same post-application period, the decline in plant height was 19-65% (58 DAHT in 2008) and 23-66% (60 DAHT in 2009), while leaf area reduction was 50-90% (58 DAHT in 2008) and 56-91% (60 DAHT in 2009). Therefore, fresh weight and leaf area were more sensitive parameters (all applied herbicide rates reduced these parameters more than 50%) than plant height. This is not consistent with findings reported by Abbas et al. (2005) that plant height was the more sensitive parameter to some ALS-inhibiting herbicides than dry weight, but it is in accordance with the findings of Božić et al. (2019) who reported that plant height was the least sensitive parameter for estimating weedy sunflower response to nicosulfuron, compared to fresh weight and leaf area.

Table 2. Timeline and additional information about the experiment

| Year | 2008          | 2009          |
|------|---------------|---------------|
| Preceding crop | soybean | corn |
| Sowing date (in containers) | April 17 | April 17 |
| Date of transplantation | April 27 | April 25 |
| Date of herbicide application | May 13 | May 17 |
| Plant stage at herbicide application | 2 pairs of leaves | |
| Date of zero measurement | May 13 | May 17 |
| Date of first measurement | May 27 | June 02 |
| Date of second measurement | June 12 | June 19 |
| Date of third measurement | June 29 | July 04 |
| Date of fourth measurement | July 14 | July 16 |
| Date of harvest | September 27 | September 10 |
Figure 1. Effects of nicosulfuron on weedy sunflower under field conditions, determined based on: a) plant height; b) fresh weight; c) leaf area. Error bars represent ±SD.
Effects of herbicides (nicosulfuron in this case) to seed production of non-target weed species are important because seed production in a single growing season determines the potential weed problems in subsequent years. Knowledge of the preharvest herbicide application effects on seed production of weeds would be beneficial for developing long-term weed management strategies. Although post-emergence herbicides are generally applied at the seedling stage of weeds, herbicide application is closely related to plant fitness, which determines seed production. Thus, herbicides do not directly affect plant reproduction, but some of them affect the generative production of surviving weeds. For example, nicosulfuron significantly reduced the weight and number of burs of *X. strumarium* (Bozic et al., 2013), as well as the head and seed production of weedy sunflower (Bozic et al., 2016). Also, bentazone reduced the weight of burs of *X. strumarium* (Zhang et al., 1994). In the present study, the effects of nicosulfuron on some generative parameters (head weight, head diameter and weight of seeds per head) are not as notable as the effects on other generative (number of heads per plant, weight of seeds per plant, number of seeds per plant) and vegetative (plant height, fresh weight and leaf area) parameters. A significant effect of this herbicide was confirmed only for head weight, head diameter and weight of seeds per head (Table 4). Nicosulfuron reduced those parameters (Figure 2), which decreased as the rate of nicosulfuron increased. Similarly, Bozic et al. (2016) found that nicosulfuron reduced the number of heads per plant and number of seeds per plant in three weedy sunflower populations treated with the recommended rate of nicosulfuron (40 g a.i. ha\(^{-1}\)). The relationship between herbicide rate and head weight/diameter was not clear, and changes regarding these parameters were not in accordance with the changes caused by herbicide rates (Figure 2). The average weight of seeds per head is the only generative parameter which was not affected by nicosulfuron. The results indicate that plant growth and generative production is not completely correlative.

### Table 3. A one-way ANOVA (F-values) showing nicosulfuron effects on variables of vegetative production (height, fresh weight, leaf area) of weedy sunflower

| Year | Variable | DAHT 0 | 14/16 DAHT | 30/33 DAHT | 44/48 DAHT | 58/60 DAHT |
|------|----------|--------|------------|------------|------------|------------|
| 2008 | Height   | 0.077190 NS | 209.9668* | 327.1072* | 247.5211* | 288.5828* |
|      | Fresh weight | 0.031643 NS | 237.5402* | 334.5761* | 580.0327* | 773.4443* |
|      | Leaf area  | 0.017044 NS | 118.2339* | 128.1340* | 678.2579* | 682.7121* |
| 2009 | Height   | 0.047211 NS | 119.5478* | 456.2376* | 153.1121* | 153.5548* |
|      | Fresh weight | 0.064331 NS | 199.3650* | 421.3351* | 472.3026* | 556.2187* |
|      | Leaf area  | 0.014470NS | 217.3328* | 211.2043* | 512.2095* | 354.7219* |

NS (p>0.05) - no significant differences; *(P<0.01) - very significant differences

### Table 4. A one-way ANOVA (F-values) showing the effects of nicosulfuron on generative production (number of heads per plant, head weight, head diameter, weight of seeds per head, weight of seeds per plant, number of seeds per plant) of weedy sunflower

| Year | Variable | Weedy sunflower maturity |
|------|----------|--------------------------|
|      | number of heads per plant | 16.38704* |
|      | head weight | 0.021543 NS |
|      | head diameter | 0.042054 NS |
|      | weight of seeds per head | 3.103589 NS |
|      | weight of seeds per plant | 45.23876* |
|      | number of seeds per plant | 71.43657* |
| 2008 | number of heads per plant | 39.77504* |
|      | head weight | 0.013479 NS |
|      | head diameter | 0.092078 NS |
|      | weight of seeds per head | 1.23549NS |
|      | weight of seeds per plant | 63.38726* |
|      | number of seeds per plant | 56.75634* |

NS (p>0.05) - no significant differences; *(p<0.01) - very significant differences
Differences in weedy sunflower response to nicosulfuron were evident between the years, although not so prominent, which probably resulted from different environmental conditions. Namely, the period between sowing and sampling dates in 2008 was characterized by somewhat higher rainfall than the corresponding period of 2009. This is coinciding with previous studies which confirmed that environmental conditions significantly affected the activity and effects of post-emergence herbicides (Božić et al., 2012, 2015b; Božić et al., 2019).

Figure 2. Effects of nicosulfuron on biological production of weedy sunflower under field conditions, determined based on: a) number of heads per plant; b) head weight; c) head diameter; d) weight of seeds per head; e) weight of seeds per plant; f) number of seeds per plant. Error bars represent ±SD.
In conclusion, nicosulfuron was found to have a significant impact on weedy sunflower vegetative production (plant height, fresh weight and leaf area) and on some generative parameters (number of heads per plant, weight of seeds per plant and number of seeds per plant).

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Uticaj nikosulfurona na biološku produkciju hibridne forme korovskog suncokreta (*Helianthus annuus*)

**REZIME**

Efekat nikosulfurona na biološku produkciju hibridne forme korovskog suncokreta (*Helianthus annuus*) ispitivan je u poljskim ogledima. Biljke ove korovske vrste su posejane na parcelicama veličine 5x4,2 m, sa rastojanjem biljaka u redu od 24 cm i razmakom između redova od 70 cm. Biljke u fazi dva para razvijenih listova su tretirane nikosulfuronom, koji je primenjen u sledećim količinama: 0, 10, 20, 40, 60 i 80 g a.s. ha⁻¹. Vegetativni parametri (visina biljaka, sveža masa i površina listova) su mereni pet puta tokom vegetacije, što uključuje prvo merenje neposredno pre primene herbicida i četiri merenja u intervalima od oko dve nedelje. Produkcija glavica i semena (broj glavica po biljci, masa glavice, prečnik glavice, masa semena po glavici, masa semena po biljci, broj semena po biljci) je određena nakon sazrevanja semena. Primena nikosulfurona je prouzrokovala redukciju rasta hibridne forme korovskog suncokreta u poređenju sa kontrolom. Sve količine ovog herbicida su redukovale vegetativne (visina biljaka, sveža masa i površina listova) i neke generativne (broj glavica po biljci, masa semena po biljci, broj semena po biljci) parametre, koji su se smanjivali sa povećanjem količine nikosulfurona. Efekat na ostale generativne parametre (masa glavice, prečnik glavice, masa semena po glavici) nije bio uporediv sa efektom na ostale parametre.

**Ključne reči:** nikosulfuron; generativni parametri; vegetativni parametri; hibridna forma korovskog suncokreta