

## Planting density impact on weed infestation and the yield of *Miscanthus* grown on two soil types

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### ABSTRACT

The assessment of the weed infestation effect on biomass yield of *Miscanthus* × *giganteus* in the first year of its commercial yield was conducted on two types of soil with different productive ability – Luvic Chernozem and Calcic Gleysol. The formed mass of weeds was higher on Luvic Chernozem and the infestation had grown according to the stages of *Miscanthus* growth. The biomass of weeds depended on the planting density of *Miscanthus* as well as on the weather conditions during the studied years. Weed infestation of crops very significantly influenced the formation of aboveground biomass of *Miscanthus*, so that the yields in the first year of commercial harvesting in the control where the weeds were removed manually were significantly higher compared to the crops in which weeds were not removed. The obtained results showed that weeds significantly affect the initial growth and development of *Miscanthus* plants that are, in general, slow, especially in the year of the crop establishment. The study evaluates the impact of a manual method of suppression and weed infestation of crops on the commercial yield of *Miscanthus*.

**Keywords:** *Miscanthus*; C4 perennial grass; energy crop; biofuels; weed control

An interspecies cultivar *Miscanthus* × *giganteus*, as a C4 perennial rhizomatous grass species, has a high genetic yield potential as an energy crop for production of second biofuels generation. The biomass derived from *M. giganteus* plants is used in various industries, e.g. in furniture making, construction, petroleum and cellulose industries, as fuel (briquettes, oil, gas), in the production of biodegradable plastics, remediation of soils contaminated with heavy metals, for strengthening land areas at risk of erosion, and also as animal food (Stępień et al. 2014). In the year of establishment, the crop is sensitive to adverse climatic conditions (Lewandowski et al. 2000). In addition, Milovanović et al. (2011) indicate that weeds can also be a limiting factor in the year of plantation. According to Christian (1994) *Miscanthus* is a poor competitor to weeds in the period after planting because of its slow growth and a prolonged period of the crop establishment, especially on the soils

that have been fallow for many years. The study of Smeets et al. (2009) suggests that large row spacing after planting is also a suitable place for the development of weeds.

Weed control in the year of *Miscanthus* planting is the most important crop protection measure for its successful formation (Anderson et al. 2011). Regarding the fact that *Miscanthus* is often grown on marginal soils with prevailing herbal perennial weeds, the issue of chemical protection is more complex (Lesur-Dumolin et al. 2015). According to the current knowledge, weeds are a major problem in *Miscanthus* cultivation and also in the years until the plants tiller and cover the crop space. As Buhler et al. (1998) stated, the plantation of *Miscanthus* should be kept clean and without weeds in the second and third year of growing, so the plants could provide a commercial yield as soon as possible. Most authors recommended that use of

mechanical, cultural, and chemical weed management techniques are often necessary to facilitate crop establishment. Depending on planting method, mechanical interrow cultivation can also be done during the first growing season but generally would not be considered in subsequent years due to risk of injuring spreading rhizomes. No herbicides are currently labelled for use in *Miscanthus* grown for biomass, but herbicides used on field corn might be safe to *Miscanthus* (Anderson et al. 2010).

There is little information on weeds infestation and their impact on the yield of this crop, especially in Serbia, which gives the significance to this research as a base for developing successful weed management. The aim of this research was to investigate the weed infestation and its competitive impact on *Miscanthus* crop established on two types of soil and in two planting densities, with special emphasis on the realised yield in the first year of commercial cultivation.

## MATERIAL AND METHODS

The field trials were carried out in the period 2011–2014 at two sites in Serbia. At the site of the Institute for the Application of Nuclear Energy – INEP experimental field in Zemun, the soil type was Luvic Chernozem. At the second site in the Grabovac village near the thermal power station Nikola Tesla Obrenovac, the soil type was Calcic Gleysol.

Luvic Chernozem belongs to the first soil capability class, meaning it is a naturally fertile soil due to its optimal physical, chemical and biological properties, which results in high and stable crop yields. Opposite to this type of soil, Calcic Gleysol belongs to the third soil capability class, characterized by heavy mechanical composition and unfavourable water-air properties that are necessary to be improved in order to obtain the stable yields (Glamočlija et al. 2012).

At both experimental sites the field trials were carried out according to the divided plots system (split-plot) with three replications. The test culture was the interspecies cultivar *Miscanthus × giganteus*, for planting the rhizomes of 10–20 cm length were used, provided from a registered manufacturer. Manual planting was carried out in mid-April at a depth of 10 cm within the experimental plot size of 20 m<sup>2</sup> (5 × 4 m) with a row spacing of 1.5 m between each experimental plot.

The rhizomes were planted in two densities: two rhizomes per m<sup>2</sup> and three rhizomes per m<sup>2</sup>. In production technology the standard agricultural practice was applied (Dželetović et al. 2006). During the growing season in the years of research in control plots the weeds were manually removed. In the variant with weed infestation of crops (the second half of the experimental plot) sampling of weed species was carried out depending on the stages of *Miscanthus* growth (according to BBCH scale). The first sampling period was in 5-leaf stage, the second in the stage of intensive stem elongation (in 6 nodes detectable) and the third at beginning of tassel emergence. The samples were taken from the surface of one square meter, according to a random schedule, from each experimental plot. After the weeds picking, they were air-dried at room temperature and biomass of weeds was weighed.

Manual harvesting of *Miscanthus* stems was carried out at the end of winter. After additional drying in bundles at relatively stable moisture content (10–14%) the yield per whole elementary plots was measured and recalculated to the yield per hectare.

An analysis of the effect of planting density, type of soil and sampling period on the plantation infestation was determined by the three factorial analysis of variance, and for the subsequent comparison of significance the Duncan's test was used at a significance level  $\alpha = 0.05$ . Given that the effect of the second order interaction was significant, for further analysis, two factorial analysis of variance for every two factors were performed, with a fixed level of the third factor.

Both sites are located close to the city of Belgrade, so that the data on precipitation and temperatures from the Republic Hydrometeorological Institute of Serbia gathered from the weather station in Belgrade were used. In 2011, the total annual amount of precipitation was in relation to the multiannual average (1981–2014) decreased by 38.4%, while precipitation in growing season (April–October) decreased by 41.7%. The annual precipitation sum and precipitation during the *Miscanthus* growing season was less than the multiannual average; in 2012 it was 22.5% and 35.3%, respectively, and in 2013 it was 13.8% and 33.7%, respectively. The total annual amount of precipitation in 2014 of 1.095 mm and 927.3 mm in the vegetation period significantly exceeded the multiannual average.

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Average annual air temperature in the period of investigations was 0.6–1.5°C higher compared to the multiannual average. During the vegetation period of the first three years of research, the air temperatures were higher in the range from 1.1–2.5°C, while during the growing season in 2014 no difference of this parameter was registered.

## RESULTS AND DISCUSSION

The weed infestation of crops in the first year of research showed a dynamics of significant increase from the first to the third period of sampling (Table 1). Planting density at both sites significantly affected the biomass of weeds, which was, in general, higher in the crop with 2 rhizomes/m<sup>2</sup>. This is confirmed by the results of Borkowska and Molas (2010), finding that in the conditions of high planting density of *Miscanthus* (3 rhizomes/m<sup>2</sup>) compared to low (1 rhizomes/m<sup>2</sup>), the weeds formed significantly less mass. Effect of planting density was significantly manifested on Luvic Chernozem, while on Calcic Gleysol it was manifested only in the third period of sampling.

By analysing the biomass of weeds in relation to the soil type individually, it is evident that it was higher on Luvic Chernozem by 27.58% at two rhizomes density, and 46.93% at three rhizomes density.

Increased *Miscanthus* infestation on Luvic Chernozem can be explained by the fact that this type of soil is of higher natural fertility. Comparing the sampling periods, studied treatments of plant-

ing density and soil type, significant interactions were determined. Lesur-Dumolin et al. (2015) points out that *Miscanthus* is characterized by high to extreme weed infestation in the year of planting the rhizomes (from 35–92%).

Comparing the periods of sampling, studied treatments of planting density and soil types, interactions were also significant in the second year (Table 1). Planting density had an effect on Luvic Chernozem in the second and third period of sampling. In the second period (intensive stem elongation) greater biomass of weeds was registered at densities of 3 rhizomes/m<sup>2</sup>, while in the third period (beginning of tassel emergence), greater biomass was found at a density of 2 rhizomes/m<sup>2</sup>. On Calcic Gleysol planting density had an impact only in the second period of sampling, when the biomass of weeds was higher at 2 rhizomes planting density. The influence of soil type on the biomass of weeds at a planting density of 2 rhizomes/m<sup>2</sup> had expression only in the third period of sampling, when the average biomass of weeds was significantly higher on Luvic Chernozem soil type.

At planting density of 3 rhizomes/m<sup>2</sup>, the type of soil had an impact on all three sampling periods, and weed infestation in the crop was significantly higher on Luvic Chernozem. Regarding the sampling periods, dynamics of weed infestation in crops showed a significant increase from the first to the third period on both soil types. By denser planting of *Miscanthus* (3 rhizomes/m<sup>2</sup>) the weed infestation of the crop was lower because the plants formed a larger number of secondary stems

Table 1. Influence of planting density, soil type and sampling period on dry weight of weeds in the first, second and third year of monitoring (g/m<sup>2</sup>; means ± standard deviation)

Plant density	Sampling period	Year 1		Year 2		Year 3	
		Luvic Chernozem	Calcic Gleysol	Luvic Chernozem	Calcic Gleysol	Luvic Chernozem	Calcic Gleysol
2 rhizomes/m <sup>2</sup>	I	223.0 ± 38.6 <sup>AaY</sup>	186.8 ± 46.4 <sup>AaX</sup>	272.5 ± 15.3 <sup>AaX</sup>	261.8 ± 28.4 <sup>AaX</sup>	386.0 ± 4.4 <sup>BaX</sup>	248.7 ± 142.7 <sup>AaX</sup>
	II	307.0 ± 22.5 <sup>BbX</sup>	250.8 ± 2.2 <sup>AbX</sup>	278.0 ± 15.4 <sup>AaX</sup>	275.6 ± 47.2 <sup>AaY</sup>	616.0 ± 14.2 <sup>BbY</sup>	424.2 ± 68.6 <sup>AbX</sup>
	III	494.0 ± 35.5 <sup>BcY</sup>	365.0 ± 20.7 <sup>AcY</sup>	679.5 ± 50.6 <sup>BbY</sup>	383.5 ± 50.4 <sup>AbX</sup>	572.5 ± 8.8 <sup>BbX</sup>	372.2 ± 87.2 <sup>AbX</sup>
3 rhizomes/m <sup>2</sup>	I	155.0 ± 11.5 <sup>AaX</sup>	164.5 ± 45.6 <sup>AaX</sup>	291.0 ± 8.3 <sup>BaX</sup>	200.8 ± 25.1 <sup>AaX</sup>	1027.0 ± 40.6 <sup>BcY</sup>	200.5 ± 70.2 <sup>AaX</sup>
	II	423.0 ± 12.8 <sup>BbY</sup>	209.8 ± 32.4 <sup>AaX</sup>	378.0 ± 9.6 <sup>BbY</sup>	206.9 ± 23.7 <sup>AaX</sup>	433.0 ± 26.1 <sup>AaX</sup>	367.4 ± 63.2 <sup>AbX</sup>
	III	379.0 ± 9.6 <sup>BbX</sup>	277.0 ± 15.2 <sup>AbX</sup>	394.5 ± 26.3 <sup>BbX</sup>	331.4 ± 74.2 <sup>AbX</sup>	648.0 ± 7.9 <sup>BbX</sup>	355.5 ± 38.9 <sup>AbX</sup>

To highlight the significance of differences between the type of soil the capital letters were used, between the period of sampling – lowercase was used, and between the density of rhizomes planting X and Y letters; the values highlighted with the same letters do not differ at the significance level of  $P \leq 0.05$

and with their mass covered better the space between the rows. Weed infestation of crops in the second year was higher for about 13% as a result of favourable water regime. Lesur-Dumolin et al. (2015) points out that a large weed infestation in the second year of cultivation is the result of infestation in the year of *Miscanthus* plantation.

Effect of planting density, soil conditions and sampling period on weed biomass was also significant in the third year of growing *Miscanthus* (Table 1). An analysis of weed infestation by soil types at planting densities of 2 rhizomes/m<sup>2</sup> in all the three sampling periods showed that infestation was 50.55% higher on Luvic Chernozem. In crop with 3 rhizomes/m<sup>2</sup> infestation was also higher on Luvic Chernozem but the significant variations were recorded only in the first and third sampling period. Planting density significantly affected the biomass of weeds on Luvic Chernozem. In the first sampling period the mass of weeds was significantly greater at higher planting density (3 rhizomes/m<sup>2</sup>), while in the second sampling period the mass of weeds was greater at a lower planting density of *Miscanthus* (2 rhizomes/m<sup>2</sup>). This can be explained by the intense increase of *Miscanthus* in this growing season. Planting density had no influence on the mass of weeds in *Miscanthus* crops on Calcic Gleysol. Weed infestation was also in the third year higher on Luvic Chernozem, which was certainly caused by the fact that this type of soil had better production characteristics.

Increased weed infestation of crops in the third year of growing *Miscanthus* within the set combinations was probably caused by the high amounts of precipitation during the summer months and relatively low crops tillering. In the first and second

studied years, the water regime was not favourable for the intensive growth of *Miscanthus* plants. Similar results were obtained by Price et al. (2004). They point out that insufficient water supply can slow the growth and development of crops, and a seasonal yield differences are mainly the result of the water stress. Semere and Slater (2007) recorded higher weed infestation of the crop in the third year of cultivation (96%) compared to the second year (48%), despite the use of herbicides, which was explained by the fact that *Miscanthus* has a weak initial growth, and from the aspect of agronomic practices, large row space planting leaves plenty of space for the growth of weeds.

The first two years were unfavourable for the growth of *Miscanthus*, especially in the crops without weed suppression. The yields in the first two years are almost negligible and with no commercial value. Most authors point out that in conditions of temperate continental climate in the cultivation of *Miscanthus*, this plant provides a commercial yield from the third year (Christian et al. 2008, Angelini et al. 2009). Following these findings, the average yield of *Miscanthus* biomass on Luvic Chernozem was 13.13 t/ha, and on Calcic Gleysol it was 11.92 t/ha (Table 2).

Weeds in the crop decreased the biomass yield of *Miscanthus*. On Luvic Chernozem weed infestation was generally higher than on Calcic Gleysol. However, the difference in biomass yield compared to the control was statistically very significant at both sites. On Calcic Gleysol the difference in yield amount between the control and the weedy crops was lower than on Luvic Chernozem. Field trials in Illinois (Anderson et al. 2010) showed a greater than 40% reduction in dry weight and number of shoots per plant for non-weeded check compared

Table 2. Average yield of *Miscanthus* biomass (t/ha) in the third year

Planting density	Treatment	Luvic Chernozem	Calcic Gleysol
2 rhizomes/m <sup>2</sup>	weed infestation	0.37 ± 0.1 <sup>AaX</sup>	0.78 ± 0.14 <sup>AaX</sup>
	control	18.60 ± 7.08 <sup>BbY</sup>	10.33 ± 1.26 <sup>AbX</sup>
3 rhizomes/m <sup>2</sup>	weed infestation	0.51 ± 0.19 <sup>AaX</sup>	0.86 ± 0.16 <sup>AaX</sup>
	control	7.66 ± 0.58 <sup>AbX</sup>	13.50 ± 1.32 <sup>BbX</sup>
Total	weed infestation	0.44 ± 0.16	0.82 ± 0.14
	control	13.13 ± 7.49	11.92 ± 2.08

To highlight the significance of differences between the type of soil the capital letters were used, between the treatments – lowercase was used, and between the density of rhizomes planting X and Y letters; the values highlighted with the same letters do not differ on the significance level of  $P \leq 0.05$

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to weeded control. Also, Haines et al. (2015) and Lesur-Dumolin et al. (2015) point out that the low yields of *Miscanthus* biomass are associated with high weed infestation of crops that prevents good tillering in the year of plantation. Planting density influenced higher biomass yield only on the control at Zemun site (Luvic Chernozem), while the type of soil significantly affected only the control in which the weeds were manually suppressed.

In conclusion, the formed biomass of weeds was higher on Luvic Chernozem and the infestation had grown according to the stages of *Miscanthus* growth. Weeds in the crop decreased the yield of *Miscanthus* biomass. At both sites, increased weed infestation was recorded at lower plant densities. On Luvic Chernozem weed infestation was generally higher than on Calcic Gleysol. Planting density had higher influence on biomass yield only on the control variant at Zemun site (Luvic Chernozem), while the type of soil significantly affected only the control in which the weeds were manually suppressed. Since there are no adequate herbicides registered for the suppression of grass weeds in a *Miscanthus* crop, most authors recommend the use of mechanical protection measures (interrow cultivation, hilling) and the use of total herbicides before planting or in early spring, after the harvest and before the growth of *Miscanthus* plants.

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