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Yields and quality indicators of selected hemp varieties (*Cannabis sativa* L.) grown in Serbia

Úroda a ukazovatele kvality vybraných odrôd konopy siatej (*Cannabis sativa* L.) pestovaných v Srbsku

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ABSTRACT

Interest in growing hemp (*Cannabis sativa* L.) is constantly increasing. Different varieties of hemp grown in Serbia (Institute of Field and Vegetable Crops, Bački Petrovac) were evaluated by plant height, stem yield, fibre content, fibre yield and seed yield. Carmagnola variety reached the highest height, Fedora 17 the lowest. Stem yields, which averaged 13.23±2.93 t/ha, had a high correlation relationship with plant height. The highest and demonstrably most significant fibre content was found in the variety Bialobrzeskia (30.77±0.78%). The fibre yield averaged 3.3±0.64 t/ha. The highest seed fertility was shown by the Novosadska variety. A significant difference (P≤0.05) between 2017 and 2018 in seed production was 51.28 kg/ha. The results of the experiment indicate the importance of the genotype x environment interaction in hemp production.

Keywords: *Cannabis sativa* L., hemp, yield, fibre, seed, genotype, environment

ABSTRAKT

Záujem o pestovanie konopy siatej sa neustále zvyšuje. Rôzne odrody konopy pestované v Srbsku (Ústav poľných a zeleninových plodín, Bački Petrovac) boli hodnotené podľa výšky rastlín, úrody stonky, obsahu vlákna, úrodnosti vlákna a úrody semena. Najvyššiu výšku dosiahla odroda Carmagnola, najnižšiu Fedora 17. Úrody stonky, ktoré boli v priemere 13,23±2,93 t/ha, s výškou rastlín mali vysoký korelačný vzťah. Najvyšší a preukázateľne najvýznamnejší obsah vlákna bol zistený v odrode Bialobrzeskia (30,77±0,78%). Úroda vlákna bola v priemere 3,3±0,64 t/ha. Najvyššiu úrodnosť semena vykazovala odroda Novosadska. Štatisticky preukazný rozdiel P≤0,05 medzi rokmi 2017 a 2018 v produkcii osiva bol 51,28 kg/ha. Výsledky experimentu poukazujú na dôležitosť interakcie genotyp x prostredie pri produkcii konopy siatej.

Kľúčové slová: konopa siata, *Cannabis sativa* L., úroda, vlákno, semeno, genotyp, prostredie

INTRODUCTION

Hemp (*Cannabis sativa* L.) is a naturally dioecious annual plant from the family *Cannabinaceae*. It is characterized by an upright stem that, depending on genetic makeup and pedoclimatic conditions, can reach up to 5 m (Farag and Kayser, 2017). Glandular trichomes secrete secondary metabolites - phytocannabinoids which protect hemp plants from pests (Andre et al., 2016). The typical aromatic smell of hemp comes from another important group of secondary metabolites terpenes, of which more than 120 have been recognized, consisting 1.283-2.141% of inflorescence dry weight and 0.125-0.278% of leaves dry weight (Jin et al., 2020). Due to the high content of bioactive compounds (García-Tejero et al., 2019) with certain medicinal properties (Ferber et al., 2020), terpenes are used in the cosmetics, food and perfume industry and are increasingly attracting attention (Lowe et al., 2021). The phytocannabinoid cannabidiol (CBD) has been confirmed to reduce pain and anxiety and promote relaxation (White, 2019). The combined action of secondary biomolecules could improve the treatment of inflammation, pain, and some mental illnesses (Anand et al., 2021). Popularization and expansion of the possibilities of using the nutritional, pharmaceutical and cosmetic potential of Cannabis are related to the low content of the psychoactive cannabinoid – tetrahydrocannabinol (THC) (Jin et al., 2020). During the 20th century, industrial hemp varieties with THC levels below 0.2% were developed in Europe to comply with current legislation (De Meijer, 1994).

Around the world, industrial hemp has long been cultivated for its stems or grains and seeds (Zuardi et al., 2006), while inflorescences are harvested from high-cannabinoid varieties (Datwyler and Weiblen, 2006; Adesina et al., 2020). Hemp fibre is a raw material for the textile and construction industries (Small, 2017). In the focus of modern hemp breeding is the development of monoecious varieties suitable for dual use - production of both fibres and seeds, although dioecious varieties are better for fibre production (Berenji et al., 2013). Compared to other lignocellulosic plants, hemp has a higher biomass yield per growing area and cellulose content (Sipos et al., 2010). For the most part (93%), hemp fibre consists of cellulose, pectin polysaccharides, hemicellulose and lignins (Rehman et al., 2013). Interest in hemp - a sustainable industrial crop is growing and its fibre is an excellent substitute for cotton or synthetic materials in various applications (Schluttenhofer and Yuan, 2017). The end product of growing the crop (fibre, grain, or inflorescence) is based in the hemp plant habitus (Small, 2017).

The main objective of this study is that in the same agrotechnical conditions and the number of plants per unit area, when each plant has the same vegetation space, the yield potential of 6 hemp varieties grown in Europe is examined. The analysis refers to the 5 most important quantitative yield characteristics.

MATERIAL AND METHODS

Six varieties of hemp were cultivated: Fedora 17, Carmagnola, Tiborszallasi, Lovrin 110, Bialobrzeskia and Novosadska. The analyzed material includes two monoecious varieties, the French Fedora 17 and the Polish Bialobrzeskia. Monoecious varieties are generally intended for seed production, but are most often used as dual-purpose varieties. The dioecious varieties: Italian Carmagnola, Hungarian Tiborszallasi, Romanian Lovrin 110 and Serbian Novosadska originate from certain national hemp breeding programs, and are intended for the production of biomass (stalk) with a high fibre content (Kišgeci and Sikora, 2015).

Two-year trials (2017 and 2018) with six commercial varieties of hemp were based at the Institute of Field and Vegetable Crops Novi Sad at the site Bački Petrovac, Vojvodina province (Serbia). This area at 86 meters above sea level belongs to the continental climate with an average annual air temperature of 12 °C and an average temperature during the growing season of 18.1 °C. The annual sum of total sunshine is 2042 hours and the average relative humidity is 82.0%. The precipitation regime is of a Central European, specifically the Danube region, rainfall nature. The lowest amount of precipitation usually occurs in spring (March) and autumn (October), while the highest amount of atmospheric precipitation is in early summer (June). The soil is classicized as the calcareous chernozem soil on the loess terrace (Valentík, 2013). The basic climatological characteristics of the experiment are given in figure 1.

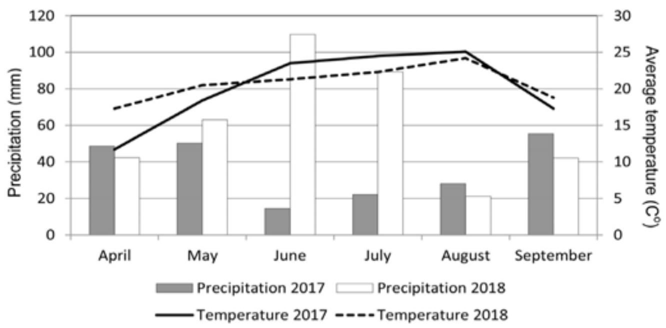

Figure 1. Sum of precipitation and average temperature for Bački Petrovac in 2017 and 2018

In order to obtain an identical number of plants on each plot, sowing was done with a pneumatic seed drill at a row spacing of 70 cm and between plants in a row spacing of 5 cm. Sowing was completed on April 16 in 2017 and April 26 in 2018 respectively, and the total area of each plot in both years was 300 m^2. After germination, when the plants were up to 5 cm high, manual thinning was performed to a final distance of 15 cm between the plants in a row in three repetitions per plot at 10 m^2. That means that in both years and on each plot was obtained 9.5 plants per m^2. Samples for morphological analysis and yield measurement were taken in three replicates (six randomly selected plants per replicate) in the physiological phase of full flowering. Samples of identical size to measure seed yield were taken at the time of seed physiological maturity. The method of Bredeman (1942) was used to determine the fibre content in the stem.

The data obtained were statistically evaluated using multifactor analysis of variance (ANOVA) and correlation analysis in STATISTICA CZ version 10.0 (TIBCO Software Inc., Palo Alto, California, USA, 2010). Differences were assessed using Fisher's LSD test. The statistical significance of the differences was assessed at the level of 95% probability and the statistically high significance at the level of 99% significance. Correlation analysis was used to determine the relationships of the obtained data. The data are presented as an average ± standard deviation.

RESULTS AND DISCUSSION

The height of the plant ranged from 1.77 to 2.75 m on an average of 2.21±0.37 m (Table 1). The lowest growth was found to be the Fedora 17 variety (1.77±0.05 m), while the highest growth was found in Carmagnola (2.75±0.17 m). Although the wide geographical distribution confirms the distinct adaptability of hemp, the results of the survey confirm the superiority in the stem height of the dioecious compared to monoecious varieties. This confirms the result of Bennett et al. (2006) or Cosentino et al. (2012) according to which monoecious varieties are characterized by both lower stem height and thinner basal stem diameter than dioecious. Plant height is a quantitative trait that is positively correlated with stem yield (Sikora et al., 2011) and therefore has a significant impact on production economics. The year of cultivation did not have a demonstrable effect on the height of the plant, which implies a pronounced genetic determination of this trait. The height of the hemp plant is also affected by genetics through photosensitivity. Varieties that do not require light reduction to less than 14 hours for the transition from the vegetative to the generative phase are classified among the northern group and their height at the time of physiological maturity reaches about 1.5 m. This habitus provides a mechanized harvest using a cereal combine without any problems, but due to the low yield of the stalks this kind of varieties are used exclusively for seed production (Amaducci et al., 2008). In addition, in some hemp breeding programs, multi-purpose varieties with a medium-height stem have been created (Adesina et al., 2020). In addition to the genetic basis, another part of the factors with a significant impact on the height of the hemp plant relates to environmental conditions. These factors are related to the microclimatic conditions in which individual plants develop, and primarily relate to the crop density, or the size of the vegetation space that individual plants dispose. According to Amaducci et al. (2002) a very dense crop provides limited essential resources to plants (light) and due to competition in dense growth there is a reduction in average plant height.

The average stem yield in the experiment ranged from 8.84±0.24 t/ha as recorded for Fedora 17 to 16.45±0.73 t/ha for Carmagnola (Table 1). Due to the pronounced positive correlation between plant height and stem yield (Sikora et al., 2011), dioecious varieties selected for biomass yield (especially Carmagnola and Novosadska) achieved significantly higher stem yields compared to monoecious varieties. Data on maximum hemp stem yields vary depending on the region, variety and applied technology. While Finnan and Styles (2013) state that hemp produces more than 10 t/ha of biomass per year, Struik et al. (2000) talk about up to 25 t/ha of total dry aboveground mass, of which 20 t covers dry stem. Small (2017) estimated fresh and air-dry weight of a hectare of fiber hemp, relative to living hydrated plant. According to him, the total yield of fresh biomass reaches 40 t/ha (100%), of which 30% (12 t) are green leaves and 70% (28 t) are green stem. In the air-dry mass, the leaves make up 12.5% (5 t) and the air-dry stem 26.3% or 10.5 t, of the total yield of fresh biomass. These results cover a very wide range of production regions as well as hemp types. In our experiments, in comparison with the mentioned averages, significantly better yields were achieved in both monitored years, thanks primarily to favorable agroecological conditions in the region of southern and southeastern Europe and the choice of varieties included in the experiment and using appropriate agricultural techniques. Contemporary hemp production is based on the interaction of genotype x environment x management (GxExM) and a responsible approach to these factors is the basis of success (Amaducci et al., 2015; Cosentino et al., 2013).

Fibre content in the two-year trial ranged from 21.33±0.97 to 30.77±0.78% with the average of 25.32±3.50% (Table 2). The varieties Bialobrzeskia and Novosadska contained the highest proportion of fibre. There is a low correlation when comparing fibre content and plant height (-0.28) or stem yield (-0.37) (data not shown). In the statistical evaluation of fibre content for the varieties Carmagnola (21.33±0.97%), Tiborszallasi (23.17±1.44%) and Lovrin 110 (23.38±0.28%), there was no significant difference in fibre content. A demonstrable difference of the variety was found in the variety Bialobrzeskia (30.77±0.78%). Although the two analyzed years differ in terms of the amount of available moisture (Figure 1) climatic conditions in 2017 and 2018 had no effect on the fibre content. Significant differences in fibre content between cultivars do not coincide with differences in stem height and yield and do not follow the division into monoecious and dioecious varieties in this regard. Considering that the monoecious variety Bialobrzeskia had a lower plant height and a lower total stem yield compared to the average of the trial, the high fibre content speaks in favor of significant selection

Table 1. Plant height and stem yield of selected hemp varieties

Variety	Plant height [m]			Stem yield [t/ha]		
	2017	2018	Average	2017	2018	Average
Bialobrzeskia	1.87±0.11	2.03±0.07	1.95^{ab}±0.09	11±0.73	10±1.18	10.47^{a}±1.27
Carmagnola	2.63±0.09	2.20±0.10	2.75^{e}±0.17	16±0.95	17±0.69	16.45^{c}±0.73
Fedora 17	1.67±0.07	1.88±0.12	1.77^{a}±0.05	8±0.54	9±0.33	8.84^{a}±0.24
Lovrin 110	2.03±0.93	2.09±0.51	2.06^{bc}±0.21	14±0.64	14±0.14	14.33^{b}±0.38
Novosadska	2.41±0.34	2.51±0.69	2.46^{d}±0.20	14±0.75	16±1.10	15.23^{bc}±0.19
Tiborszallasi	2.30±0.52	2.20±0.18	2.25^{cd}±0.23	13±1.20	15±0.93	14.09^{b}±1.75

Different superscript letters indicate values that differ significantly from each other in the same columns of the table by according to Fisher's test at $P \leq 0.05$

for this trait (Burczyk et al., 2005). If we consider that Thygesen et al. (2005) assume that hemp grown for fibre in the total mass of stem contains 20-40% fibre and 60-80% of woody core, the results of the experiment indicate the biological space for defining the genetics and technology to improve this quantitative trait.

The fibre yield averaged 3.3±0.64 t/ha, with extreme values (maximum and minimum) for Novosadska (4.24±0.24 t/ha) and Fedora 17 (2.24±0.14 t/ha). Correlation analysis revealed that fibre yield and fibre content interact. The coefficient of this relationship was low 0.37 (data not shown). The monoecious variety Fedora 17 proved to be the least suitable for biomass production in specific conditions, primarily due to low stem yield (associated with relatively low height) and average fibre content (25.22±1.47%). The best result was given by the variety Novosadska, which was selected in specific agro-climatic conditions of southern and southeastern Europe, for which it shows exceptional ecological adaptability. No significant differences in fibre yield were observed in other dioecious cultivars, which is in accordance with the claim of Berenji et al. (2013) that the highest superior fibre quality and yields are achieved with dioecious hemp of longer vegetation period. In addition to the choice of genetics (variety) adequate for a particular agro-ecological region, the yield and quality of fibre are also affected by appropriate agro-technical measures performed at the appropriate time and in a quality manner. Deng et al. (2019) states that to achieve fibre yield over 2200 kg/ha requires 330-370,000 plants per hectare with the application of 250-270 kg/ha of nitrogen, 85-95 kg/ha of phosphorus and 210-240 kg/ha of potassium. The results of our experiments indicate a higher potential for fibre yield in European varieties, although it should be noted that Deng et al. (2019) in their research used only one variety grown in central China. Thanks to the very high fibre content in the stem, the Polish monoecious variety Bialobrzeskia achieved a yield at the level of dioecious varieties. Other large-scale studies conducted with more extensive material at several sites over several years have confirmed that the cultivars adapted to the specific environment and proper agrotechnical practice are responsible for obtaining a high yield of quality hemp fibre (Struik et al., 2000; Tang et al., 2016).

Seed yield gradually increased in the evaluation of individual varieties in the order: Tiborszallasi, Carmagnola, Lovrin 110, Bialobrzeskia, Fedora 17, Novosadska. The average was found at 758.47±27.11 kg/ha. The minimum yield on average two years (734±18.71 kg/ha) was achieved by the variety Tiborszallasi and the maximum Novosadska (778.67±6.59 kg/ha). Seed yields differed significantly between the two analyzed years for this locality. The yield of seeds in 2017 was 732.93±13,99 kg/ha and in 2018 the harvest increased by 51.08 kg/ha. The reason for this difference is environmental conditions (Pejić et al., 2018), i.e. more precipitation during the period

Table 2. Fibre content, fibre yield and seed yield of selected hemp varieties

Variety	Fibre content [%]			Fibre yield [t/ha]			Seed yield [kg/ha]		
	2017	2018	Averege	2017	2018	Averege	2017	2018	Averege
Bialobrzeskia	34±0.20	28±0.45	30.77^{a}±0.78	3.52±0.27	2.90±0.07	3.21^{a}±0.33	745.10±15.25	765.24±7.16	755.17^{bc} ±4.67
Carmagnola	19±0.50	24±0.34	21.33^{c}±0.97	3.99±0.14	3.01±0.35	3.50^{a}±0.41	729.00±36.12	777.00±29.60	753.00^{ab}±53.25
Fedora 17	25±1.15	26±0.98	25.22^{bc}±1.47	2.04±0.65	2.44±0.18	2.24^{b}±0.14	731.00±9.18	821.00±24.14	776.00^{cd}±10.73
Lovrin 110	13±0.45	24±1.62	23.38^{c}±0.28	3.31±0.46	3.41±0.50	3.36^{a}±0.26	737.00±42.31	771.00±7.90	754.00^{ab}±23.34
Novosadska	29±0.35	27±0.12	28.02^{ab}±0.86	4.14±0.14	4.34±0.24	4.24^{c}±0.24	747.00±12.35	810.34±28.71	778.67^{d}±6.59
Tiborszallasi	23±0.26	23±0.79	23.17^{c}±1.44	3.05±0.10	3.45±0.87	3.25^{a}±0.24	708.5±20.23	759.5±15.19	734.00^{a}±18.71

Different superscript letters indicate values that differ significantly from each other in the same columns of the table by according to Fisher's test at $P \leq 0.05$

of intensive growth (July and August), which created moisture reserves that plants used at the time of grain filling. The results showed that the monoecious variety Bialobrzeskia with seed yield at the level of the average of the experiments in these conditions can be used for dual purposes (fibre and seed) and the monoecious variety Fedora 17 exclusively for seed production. The best results were shown by the Novosadska variety, which originates from the breeding program located in the region where the experiments were performed. This confirmed the importance of considering genetics (choice of varieties), environmental conditions in which production is carried out and application of technology in accordance with set production goal. The importance of the adaptability of the cultivated material is also indicated by the results of the examination of the native population in comparison with the French assortment in the conditions of Iran (Abdollahi et al. 2020). These studies have shown that locally adapted local populations achieve significantly higher seed yields compared to introduced varieties, with a potential of over 3000 kg/ha that exceeds the material tested in our experiments, when the seed yield does not exceed 800 kg/ha.

CONCLUSIONS

The representative choice of hemp assortment analyzed for the most important yield components indicates the existence of variability for these traits. Detected variability, with the inclusion of foreign (non-European origin) germplasm, can be the basis for defining a hemp breeding program designed to improve production. The results showed that when planning hemp production based on a defined purpose, the choice of variety and sexual profile (monoecious or dioecious) should be taken into account. For the conditions of southern and southeastern Europe, for the purpose of biomass production (stem and fibre), dicotyledonous varieties proved to be better, while monoecious varieties are more appropriate for seed production. The second factor of successful production refers to the consideration of environmental microclimatic conditions and adaptation of agricultural techniques in order to provide the best conditions for plant development. In that sense, further expand the research in terms of other agronomic research, including the level of fertilization, canopy adjustment should continue and the use of irrigation.

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