

VARIABILITY OF THE NUMBER AND WEIGHT OF 1000 SEEDS OF WEEDS PRESENT IN ALFALFA NATURAL SEEDS FROM DIFFERENT LOCATIONS IN SERBIA

VARIJABILNOST BROJA I MASE 1000 SEMENA KOROVA PRISUTNOG U NATURALNOM SEMENU LUCERKE SA RAZLIČITIH LOKALITETA U SRBIJI

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ABSTRACT

Alfalfa is the most significant perennial and multi-crop forage legume in Serbia. Seed production has numerous problems, which causes high variability in seed yield. One of the problems in this production is the weed seeds that are in the natural alfalfa seed after harvesting. Minimum purity must be greater than 95 %, up to 2 % of other species are allowed, and up to 0.5 % of weeds, but no quarantine weeds such as dodder (*Cuscuta*.sp). Weed seeds were detected in natural alfalfa seeds: *Sinapis arvensis* L., *Amaranthus retroflexus* L., *Rumex obtusifolius* L., *Cuscuta campestris* Yunk., *Rumex acetosella* L., *Plantago lanceolata* L., *Plantago major* L., *Rumex crispus* L., *Galium aparine* L., *Convolvus arvensis* L. as expected, the variability of the number of weed seeds detected from the seed lots over three years was high: CV% = 14.78 for wild sorghum – *S. halepense* (L.) Pers. In the 2022, to CV% = 65.47 for ribwort plantain – *Plantago lanceolata* L. in the 2020. For the weight of 1000 seeds of the examined weeds, the variability was determined from CV% = 5.869 for the ribwort plantain – *Plantago lanceolata* L. (year 2020), to CV% = 34.41 for yellow dock – *Rumex crispus* L. (year 2022).

Key words: Number of weed seeds, natural alfalfa seeds, weight of 1000 weed seeds.

REZIME

Lucerka je naznačajnija višegodišnja i višeočkosna krmna leguminoza u Srbiji. Proizvodnja semena je sa brojnim problemima što uzrokuje visoku varijabilnost za prinos. Jedan od problema u ovoj proizvodnji čine i semena korova koja se nakon ubiranja nalaze u naturalnom semenu lucerke. Na drugoj strani za stavljanje semena u promet minimalna čistoća semena mora da bude viša od 95 %. Dozvoljeno je do 2 % drugih vrsta i do 0.5 % korova, ali u semenu lucerke ne smeju da budu prisutni karantinski korovi kao što su: vilina kosica (*Cuscuta* spp.). U ovim istraživanjima su predstavljeni rezultati varijabilnosti broja semena i mase 1000 semena detektovanih korova u naturalnom semenu pet partija semena lucerke sa različitih lokaliteta u Srbiji, tokom tri godine. U naturalnom semenu lucerke je detektovano seme sledećih korova: *Sinapis arvensis* L., *Amaranthus retroflexus* L., *Rumex obtusifolius* L., *Cuscuta campestris* Yunk., *Rumex acetosella* L., *Plantago lanceolata* L., *Plantago major* L., *Rumex crispus* L., *Galium aparine* L., *Convolvus arvensis* L. Varijabilnost broja semena detektovanih korova bila je visoka za divlji sirak- *Sorghum halepense* (L.) Pers. (CV% = 14.78 u 2022.), do CV% = 65.47 za uskolisnu bokvicu - *Plantago lanceolata* L. u 2020. godini. i za masu 1000 semena od CV% = 5.86 *Plantago lanceolata* L. (2020.), do CV% = 34.41 za *Rumex crispus* L. (2022.).

Ključne reči: Broj semena korova, naturalno seme lucerke, masa 1000 semena korova.

INTRODUCTION

Alfalfa is characterized by high seed yield variability (Ahmad et al 2020; Bolaños-Aguilar et al. 2002) and in Serbia (Pajcin et. al., 2020; Stanisavljević et. al., 2012). Weeds in the alfalfa seed crop also contribute to this (Janjić et al., 2005), with the most dangerous quarantine weed – field dodder (*Cuscuta* sp.), (Sarić-Krsmanović et. al., 2020). However, regardless of these and other numerous problems in achieving high and stable yields (Pajić and Marković, 2016), the production of alfalfa seeds is characterized by relatively low investments per unit area (499.8 € ha⁻¹, coverage margin 807 € ha⁻¹, cost - effectiveness coefficient 2.47, profit rate 59.56%), which makes this production economical. An important contribution to the economy of alfalfa seed production is efficient seed processing that will allow as little seed loss as possible (Đokić et. al., 2021). For the purpose of such processing, for each lot of seeds, it is necessary to analyze natural alfalfa seeds for the presence of weed seeds and other impurities and based on their number and size, the shape of the seed coat, etc. choose the appropriate finishing system, and the correct setting of the machines (Đokić

et. al., 2023). During the seed processing, natural seeds pass through a system of machines that separate impurities such as dry stalks, weeds and broken seeds (Uhlarek et. al., 2018).

To choose which machines will be used during seed processing, it is necessary to know which weeds and which impurities are present in natural alfalfa seeds. At the same time, weed seed characteristics such as the weight of 1000 seeds, as well as the numerical share of individual weed seeds in the natural alfalfa seed that will be processed.

According to the regulations on the quality of seeds of the Republic of Serbia (Gazette of SFRY, 1987; 47/87) and ISTA rules (ISTA; 2021), declared alfalfa and red clover seeds in the sample must not contain any seeds of *Cuscuta* sp.

The aim of these tests was to determine which weeds were present, what their weight of 1000 seeds was and how many of them were present in the sample, from five localities – seed lots of natural seeds - during three years.

MATERIAL AND METHODS

For testing, natural alfalfa seeds were taken from five localities – seed lots, Zrenjanin, II) Novo Miloševo, Negotin,

Banatsko Karađorđevo, Srpska Crnja. In all localities, seed production was from the second cut.

Alfalfa crops were sown in 2019, so seed was sampled from the second, the third, and the fourth year of seed production.

During all years, from all seed lots, 4 (repetitions) x 50 g weight of the average sample were taken and weed seeds were separated, the number and weight of 1000 seeds were determined, in accordance with the regulation on seed quality ("Official Gazette of SFRY, 1987; 47 /87; and Annex, 34/2013) which is harmonized with ISTA rules (ISTA, 2021).

For all tested traits the variability was determined through the coefficient of variation (CV %) for each seed lot over three years. For the statistical analyses, a statistical program Minitab Inc., version 16.1.0, free version was used.

RESULTS AND DISCUSSION

As expected, a high variability was found for the number of weed seeds: in 2020 from CV% = 34.64 (red sorrel - *Rumex acetosella* L.) to CV% = 65.47 (ribwort plantain - *Plantago lanceolata* L.); in 2021 from CV% = 33.32 (red sorrel - *Rumex acetosella* L.) to CV% = 48.58 (wild sorghum - *Sorghum halepense* L. Pers.); in 2022 from CV% = 14.78 (wild sorghum - *Sorghum halepense* L. Pers.) to CV% = 60.93 (curly dock - *Rumex crispus* L.). There is an evident increase in the number of weed seeds in the tests conducted on the seeds from 2022, the lowest number of weed seeds was in the alfalfa seed crop from 2021, and the lowest from the 2020 sample. It is common in agronomic practice that with the aging of alfalfa, the crop thins out (Stanisavljević et al., 2012) and in those circumstances, weed control, especially broad-leaved weeds, is much less effective (Konstantinović and Meseldžija, 2005).

Therefore, it is inevitable that part of the weed seeds after harvesting will be in the unprocessed-natural alfalfa seed (Širbanović et al., 2014), and the method of harvesting is also very important for the reduction of weed seeds (Radenović, 2000).

During the examination of samples from five locations-lots from 2020, weed seeds were detected and variability was determined for the of 1000 seeds: from CV% = 5.869 ribwort plantain - *Plantago lanceolata* L. to CV% = 13.40 (wild sorghum - *Sorghum halepense* L. Pers.); (Table 1).

It is also evident that far more broadleaf weed seeds were detected, which is expected considering that broadleaf weeds in the alfalfa seed crop are much more difficult to control compared to narrow leaf weeds. This is in accordance with the research of Konstantinović et al., (2007) that in perennial alfalfa crops, there are seasonal changes in the weed community, and that during the first year, different types of typical field weeds are present, and that as the crop ages, the presence of ruderal weeds increases weeds. According to the same author, perennial weed species of economic importance are: *Cirsium arvense* (L) Scop., *Sorghum halepense* (L) Pers., *Convolvulus arvensis* L., then quarantine species from the genus *Cuscuta* as well as species from the fam. *Asteraceae*. For their suppression, he recommends integral agritechnical measures with the herbicides.

Weeds from the genus *Cuscuta* spp. is the most difficult to control because it lives on many plants (*Sarić-Krsmanović et al., 2022*), and is especially dangerous in alfalfa (*Sarić-Krsmanović, et al., 2020, 2015*). In addition, seeds in the soil can be dormant for many years, and are transmitted by domestic and wild animals. If the seed of the dodder (*Cuscuta* sp.) is present in the natural alfalfa seed, it is very difficult to remove it, considering that it is similar to the alfalfa seed (size and shape). For these reasons, processing alfalfa seeds is very expensive because of removing weed seeds, especially quarantine weeds (Đokić et al., 2021, 2023).

On the samples from 2021, for the weight of 1000 seeds, the variability was determined from CV%= 9.052 (curly dock - *Rumex crispus* L.) to CV% = 19.27 (wild sorghum - *Sorghum halepense* L. Pers.); (Table 2).

Table 1. Number and weight of 1000 weed seeds in lots of natural alfalfa seeds in 2020

| Natural alfalfa seeds | | Weed seeds in 50 g of natural alfalfa seeds | | |
|--|-----------------------------|--|----------------------|--------------------------|
| Variety | The origin of natural seeds | Common and Latin name of the weed | Number of weed seeds | Weight of 1000 seeds (g) |
| Banat | Zrenjanin | Curly dock - <i>Rumex crispus</i> L. | 3 | 1.513 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 2 | 0.356 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 2 | 2.178 |
| | | Broadleaf plantain - <i>Plantago major</i> L. | 2 | 4.742 |
| Banat | Novo Miloševo | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 4 | 3.432 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 1 | 1.956 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 1 | 0.418 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 2 | 1.098 |
| Zaječarska 83 | Negotin | Field bindweed - <i>Convolvulus arvensis</i> L. | 3 | 7.165 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 7 | 2.989 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 4 | 1.212 |
| | | Curly dock - <i>Rumex crispus</i> L. | 6 | 1.325 |
| | | Cockspur grass - <i>Panicum crus-galli</i> L. | 6 | 1.396 |
| Banat | Banatsko Karađorđevo | Field bindweed - <i>Convolvulus arvensis</i> L. | 3 | 7.145 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 7 | 2.978 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 4 | 1.278 |
| | | Cockspur grass - <i>Panicum crus-galli</i> L. | 6 | 1.336 |
| Kruševačka 28 | Srpska Crnja | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 2 | 3.919 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 4 | 1.989 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 2 | 0.345 |
| | | Curly dock - <i>Rumex crispus</i> L. | 5 | 1.611 |
| Variability expressed through the coefficient of variation (CV%) for the number of seeds and the weight of 1000 weed seeds | | Curly dock - <i>Rumex crispus</i> L. | 45.83 | 9.801 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 34.64 | 10.55 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 65.47 | 5.869 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 48.99 | 13.40 |

For separating weeds with a specific seed structure, *Rumex* sp., a roll machine can be used that has canvases over which the seed material passes. Seeds with a smooth cotyledon pass, while weed seeds with a specific cotyledon stick to the canvas and are separated from the alfalfa seeds.

In 2022, the variability of the weight of 1000 seeds, was determined from CV% = 10.68 (ribwort plantain -*Plantago lanceolata* L.) to CV% = 34.41 (Curly dock - *Rumex crispus* L.); (Table 3).

In addition to the number of weed seeds, for the successful separation of weed seeds during processing, the type of seed, i.e. its weight, the shape and structure of the seedcoat, as well as the correct selection and adjustment of seed processing machines are very important (Đokić et al., 2021; Đokić et al., 2023).

Weed seeds are classified up to the Genus level, in some even up to the Species level according to Heinisch (1954). The genus and species of the weed seeds is very important for the seed processing process. This determines the mass, shape, size, structure of the seed coat, on the basis of which it is determined which machines will be the most efficient for separating weed seeds from natural alfalfa seeds.

During the examined period, variability of the number of weed seeds was varied from CV% = 14.78 (wild sorghum - *Sorghum halepense* (L.) Pers.) in 2022 (Table 3), to CV% = 65.47 (ribwort plantain - *Plantago lanceolata* L.) in 2020 (Table 1). Also, variability of the 1000 seeds weight was determined from CV% = 5.869 (ribwort plantain - *Plantago lanceolata* L. in 2020- Table 1) to CV% = 34.41 (curly dock - *Rumex crispus* L. in 2022 - Table 3). Generally, the number of the weed seeds in the samples had higher variability than the weight of 1000 weed seeds (Tables 1-3).

CONCLUSION

On samples of natural alfalfa seeds from five localities during a three-year study, the presence of seeds of various weeds was determined, which differed in the number of seeds present in the sample, and the weight of 1000 seeds of weeds. A high variability of the weed seed number was determined during 2020 from CV% = 34.64 (red sorrel - *Rumex acetosella* L.), to 65.47 (ribwort plantain - *Plantago lanceolata* L.). During 2021, variability was determined from CV%=33.32 (red sorrel - *Rumex acetosella* L.) to CV% = 48.58 (wild sorghum - *Sorghum halepense* L. Pers.). On samples from 2022 variability varied from CV% = 14.78 (wild sorghum - *Sorghum halepense* L. Pers.) to CV% = 60.09 (curly dock - *Rumex crispus* L.). A slightly lower variability was determined for the weight of 1000 seeds: from CV% = 5.869 (ribwort plantain - *Plantago lanceolata* L.) to CV% = 13.40 (wild sorghum - *Sorghum halepense* L. Pers.) on weed seeds from 2020; from CV% = 9.052 (curly dock - *Rumex crispus* L.) to CV% = 19.27 (wild sorghum - *Sorghum halepense* L. Pers.) on seeds from 2021; and from CV% = 10.68 (ribwort plantain - *Plantago lanceolata* L.) to CV% = 34.41 (curly dock - *Rumex crispus* L.) on seeds from 2022. The obtained results indicate all the complexity for determining the optimal finishing system to be applied for cleaning weed seeds from natural alfalfa seeds.

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Table 2. Number and weight of 1000 weed seeds in lots of natural alfalfa seeds in 2021.

| Natural alfalfa seeds | | Weed seeds in 50 g of natural alfalfa seeds | | |
|---|-----------------------------|--|----------------------|--------------------------|
| Variety | The origin of natural seeds | Common and Latin name of the weed | Number of weed seeds | Weight of 1000 seeds (g) |
| Banat | Zrenjanin | Curly dock - <i>Rumex crispus</i> L. | 6 | 1.398 |
| | | Field dodder - <i>Cuscuta campestris</i> Yunk. | 4 | 1.345 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 8 | 4.121 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 5 | 2.074 |
| | | Broadleaf plantain - <i>Plantago major</i> L. | 1 | 5.112 |
| Banat | Novo Miloševo | Cleavers - <i>Galium aparine</i> L. | 2 | 3.245 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 2 | 1.098 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 3 | 0.389 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 4 | 3.189 |
| Zaječarska 83 | Negotin | Field dodder - <i>Cuscuta campestris</i> Yunk. | 8 | 1.178 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 2 | 0.298 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 3 | 2.475 |
| | | Redroot pigweed - <i>Amaranthus retroflexus</i> L. | 3 | 0.495 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 2 | 1.321 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 3 | 1.702 |
| Banat | Banatsko Karađorđevo | Redroot pigweed - <i>Amaranthus retroflexus</i> L. | 3 | 0.478 |
| | | Field bindweed - <i>Convolvus arvensis</i> L. | 4 | 6.089 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 10 | 3.747 |
| | | Curly dock - <i>Rumex crispus</i> L. | 3 | 1.658 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 2 | 1.009 |
| Kruševačka 28 | Srpska Crnja | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 5 | 3.978 |
| | | Field dodder - <i>Cuscuta campestris</i> Yunk. | 9 | 1.314 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 7 | 1.956 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 4 | 1.223 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 4 | 0.382 |
| | | Curly dock - <i>Rumex crispus</i> L. | 9 | 1.458 |
| Variability expressed through the coefficient of variation (CV%) for the number of seeds and the of 1000 weed seeds | | Curly dock - <i>Rumex crispus</i> L. | 35.25 | 9.052 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 33.32 | 14.21 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 40.01 | 9.952 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 48.58 | 19.27 |

Table 3. Number and weight of 1000 weed seeds in lots of natural alfalfa seeds in 2022

| Natural alfalfa seeds | | Weed seeds in 50 g of natural alfalfa seeds | | |
|--|-----------------------------|--|----------------------|-----------------------|
| Variety | The origin of natural seeds | Common and Latin name of the weed | Number of weed seeds | 1000 seeds weight (g) |
| Banat | Zrenjanin | Charlock mustard - <i>Sinapis arvensis</i> L. | 8 | 0.915 |
| | | Redroot pigweed - <i>Amaranthus retroflexus</i> L. | 6 | 0.278 |
| | | Bitter dock - <i>Rumex obtusifolius</i> L. | 12 | 1.145 |
| | | Field dodder - <i>Cuscuta campestris</i> Yunk. | 11 | 1.332 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 7 | 0.259 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 5 | 2.398 |
| | | Broadleaf plantain - <i>Plantago major</i> L. | 14 | 5.156 |
| Banat | Novo Miloševo | Curly dock - <i>Rumex crispus</i> L. | 2 | 1.256 |
| | | Cleavers - <i>Galium aparine</i> L. | 2 | 3.232 |
| | | Field bindweed - <i>Convolvus arvensis</i> L. | 7 | 6.089 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 11 | 1.099 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 13 | 0.315 |
| Zaječarska 83 | Negotin | Divlji sirak - <i>Sorghum halepense</i> (L.) Pers. | 9 | 3.178 |
| | | Field dodder - <i>Cuscuta campestris</i> Yunk. | 6 | 1.199 |
| | | Field bindweed - <i>Convolvus arvensis</i> L. | 9 | 6.012 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 9 | 2.087 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 9 | 0.256 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 7 | 2.442 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 8 | 1.413 |
| Banat | Banatsko Karadorđevo | Redroot pigweed - <i>Amaranthus retroflexus</i> L. | 5 | 0.448 |
| | | Redroot pigweed - <i>Amaranthus retroflexus</i> L. | 7 | 0.445 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 7 | 0.335 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 2 | 1.912 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 12 | 3.745 |
| | | Curly dock - <i>Rumex crispus</i> L. | 7 | 1.672 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 4 | 1.035 |
| Kruševačka 28 | Srpska Crnja | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 10 | 3.946 |
| | | Curly dock - <i>Rumex crispus</i> L. | 9 | 1.615 |
| | | Redroot pigweed - <i>Amaranthus retroflexus</i> L. | 9 | 0.456 |
| | | Field dodder - <i>Cuscuta campestris</i> Yunk. | 9 | 1.475 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 7 | 1.941 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 5 | 1.223 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 9 | 0.389 |
| Variability expressed through the coefficient of variation (CV%) for the number of seeds and the weight of 1000 weed seeds | | Curly dock - <i>Rumex crispus</i> L. | 60.09 | 34.41 |
| | | Red sorrel - <i>Rumex acetosella</i> L. | 27.22 | 18.84 |
| | | Ribwort plantain - <i>Plantago lanceolata</i> L. | 51.93 | 10.68 |
| | | Charlock mustard - <i>Sinapis arvensis</i> L. | 36.74 | 20.37 |
| | | Wild sorghum - <i>Sorghum halepense</i> (L.) Pers. | 14.78 | 10.99 |
| | | Field dodder - <i>Cuscuta campestris</i> Yunk. | 29.04 | 13.69 |

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