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Ravlić, Marija; Baličević, Renata; Vinković, Željka; Brozović, Bojana; Sarajić, Ankica; Kranjac, David

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Alelopatski potencijal ruderalnih biljnih vrsta na rajčicu i salatu

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Fakultet agrobiotehničkih znanosti Osijek, Poljoprivredni institut Osijek

Faculty of Agrobiotechnical Sciences Osijek, Agricultural Institute Osijek

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THE ALLELOPATHIC POTENTIAL OF RUDERAL PLANT SPECIES ON TOMATO AND LETTUCE

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SUMMARY

The aim of the study was to evaluate the allelopathic potential of several ruderal plant species on the germination and initial growth of tomato and lettuce. The water extracts from dry aboveground biomass in a five-percent concentration were evaluated under the controlled laboratory conditions in a Petri dish bioassay. The research findings indicated that all species exhibited the allelopathic effects and were ranked by their average inhibitory potential, from the highest to the lowest, as follows: Melilotus albus, Portulaca oleracea, Verbascum phlomoides, Plantago lanceolata, Cichorium intybus, Rorippa austriaca, Vicia grandiflora, Eupatorium cannabinum, and Polygonum aviculare, respectively. A water extract of M. albus had the most significant negative impact, causing an average reduction of 43.9% in germination, 93% in the root length, and 89.4% in the shoot length. Conversely, a water extract of P. oleracea showed a notable stimulatory effect on both test species. The pH value of water extracts from the ruderal plant species ranged from 5.26 (P. aviculare) to 7.71 (V. grandiflora and M. albus) and did not influence their negative allelopathic potential.

Keywords: phytotoxicity, water extracts, inhibition, biological control, ruderal flora, crops

INTRODUCTION

In sustainable agricultural systems, the Integrated Weed Management (IWM) combines various preventative, cultural, mechanical, and biological weed-control methods, as well as the chemical ones, with a strong emphasis on a reduced use of synthetic herbicides. Nevertheless, the herbicides are still predominantly applied as a fast and efficient tool for weed control, and their overuse contributes to the environmental pollution, occurrence of weed-resistant populations, and a loss of biodiversity. Therefore, the incorporation of alternative and environmentally safe measures in weed management, such as allelopathy, is a necessity for modern agricultural production (Khamare et al., 2022; Kostina-Bednarz et al., 2023). Allelopathy, a biological phenomenon, represents direct or indirect, harmful or beneficial influence of one plant species on the germination, growth, and reproduction of the other through the production of chemical compounds—the allelochemicals (Rice, 1984).

The allelochemicals are mainly the secondary metabolites or their products, nonessential to the primary metabolism of plants, and include, among others, the different compounds, such as the phenols, flavonoids, coumarins, tannins, alkaloids, and terpenoids (Bachheti et al., 2020). The allelochemicals are present in all plant parts, from the roots to the seeds, and they are released into the environment through volatilization, leaching, decomposition, and via root exudation (Rice, 1984; Baličević et al., 2016). Various factors affect the allelopathic potential of particular species, such as a plant part, concentration of allelochemicals, and receptor species, as well as various environmental biotic and abiotic factors and stress conditions, which can enhance the production of allelopathic compounds (Serafimov et al., 2020.; Kostina-Bednarz et al., 2023; Žalac et al., 2022). In agricultural production, the allelopathic plants can be utilized as water extracts (natural herbicides), in crop rotation, as living or dead mulches, and cover crops, or their residues may be incorporated in the soil in order to suppress the weed germination and growth (Khamare et al., 2022).

Assist. Prof. Marija Ravlić (mravlic@fazos.hr), Prof. dr. Renata Baličević, Željka Vinković, univ. bacc. ing. agr, Assoc. Prof. Bojana Brozović, Assoc. Prof. Ankica Sarajlić, Assist. Prof. David Kranjac – Faculty of Agrobiotechnical Sciences Osijek, Josip Juraj Strossmayer University of Osijek, Vladimira Preloga 1, 31000 Osijek, Croatia

The plant species with high allelopathic potential include the crops such as rice (*Oryza sativa* L.), rye (*Secale cereale* L.), sunflower (*Helianthus annuus* L.), and sorghum (*Sorghum bicolor* (L.) Moench.), as well as the medicinal and aromatic plants (Dhima et al., 2009.; Khamare et al., 2022; Kostina-Bednarz et al., 2023). In a search for new bioactive molecules, the current research is also focused on the inhibitory potential of the edible or poisonous wild plants and weeds, particularly the invasive ones, with comprehensive studies conducted on a couple of dozen plants. For example, Mardani et al. (2016) studied an allelopathic potential of one hundred seventy-eight species, which included one hundred thirty-one medicinal and poisonous plants mainly belonging to the Asteraceae (twenty-three species), Fabaceae (nineteen species), and Poaceae (fourteen species) families. Using the sandwich method, Isin Ozkan et al. (2019) screened over seventy-nine wild medicinal species for their inhibitory potential on lettuce. In their study, Shinwari et al. (2013) evaluated thirty-eight invasive plant species and weeds using three different laboratory methods and identified the most noxious species among them. Due to their fast and simple procedure, laboratory screenings are often the first step in the evaluation of a large number of plant species (Wu et al., 2001) and simultaneously enable the assessment of allelopathic effect of water extracts, root exudates, and volatiles, as well as of a leaf litter and residues, both on an artificial medium and in the soil (Shinwari et al., 2013; Baličević et al., 2015; Mardani et al., 2016).

The aim of the research was to evaluate the allelopathic potential of water extracts prepared from a dry aboveground biomass of several ruderal weed species on seed germination and seedling growth of tomato (*Solanum lycopersicum* L.) and lettuce (*Lactuca sativa* L.).

MATERIAL AND METHODS

In the experiment, an allelopathic potential of the following plant species was assessed: Austrian yellowcress (*Rorippa austriaca* (Crantz) Besser, Brassicaceae), ribwort plantain (*Plantago lanceolata* L., Plantaginaceae), prostrate knotweed (*Polygonum aviculare* L., Polygonaceae), wild chicory (*Cichorium intybus* L., Cichoriaceae), hemp agrimony (*Eupatorium cannabinum* L., Asteraceae), large yellow vetch (*Vicia grandiflora* Scop., Fabaceae), woolly mullein (*Verbascum phlomoides* L., Scrophulariaceae), common purslane (*Portulaca oleracea* L., Portulacaceae), and white sweet clover (*Melilotus albus* Medik., Fabaceae). The aboveground biomass of plants was collected in a full flowering stage (phenological stage 6/65, Hess et al., 1997) from the ruderal sites in Osijek-Baranja County. A fresh biomass was air-dried for 48 h and subsequently oven-dried at 40 °C for 48 h. A dried biomass was ground into a fine powder and stored in a cool place until further use.

The preparation of water extracts followed a procedure specified by Norsworthy (2003), with some modifications. A 5 g amount of each plant material was mixed with 100 ml of distilled water and extracted for 24 h at room temperature. The mixtures were filtered through a muslin cloth to remove debris and subsequently through filter paper to obtain the water extracts in a 5% concentration. A sample of each prepared extract was taken for the pH value measurement.

The commercially purchased tomato (cv. *Cuor di Bue*) and lettuce (cv. *Majska kraljica*) seeds were used as the test species. The seeds were surface-sterilized with 1% NaOCl for 10 min and rinsed three times with distilled water prior to the experiment.

The effect of water extracts was evaluated under controlled laboratory conditions in a Petri dish bioassay. The experiment was set up as a completely randomized design with four replications. Each treatment in the experiment consisted of thirty seeds of the test species placed in the sterilized Petri dishes on a filter paper moistened with 4 ml of water extract. The distilled water was used in a control treatment. The seeds were incubated for fourteen (tomato) and six (lettuce) days at 22 ± 2 °C. A germination percentage was calculated, and the seedlings' root and shoot length was measured on the last day of the experiment. A germination percentage was calculated for each replication as G = (germinated seeds/total seeds) × 100.

All collected data were analyzed statistically by one-way analysis of variance (ANOVA), and the differences between the treatment means of the measured parameters for each test species were tested by the LSD test at $P < 0.05$.

RESULTS AND DISCUSSION

The water extracts from dry plant material of ruderal plant species manifested varying allelopathic effects on the germination of tomato seeds (Figure 1A). A statistically significant reduction in tomato germination was observed in most treatments, ranging from 13.8% to 84.5% if compared to the control. The most pronounced negative impact was noted in the treatments with *P. oleracea, V. phlomoides*, and *M. albus* water extracts. Similarly, *P. lanceolata* and *C. intybus* water extracts hindered tomato germination by more than 50%. Conversely, lettuce germination was significantly decreased only in the treatments with *V. grandiflora* and *P. oleracea* (Figure 1B).

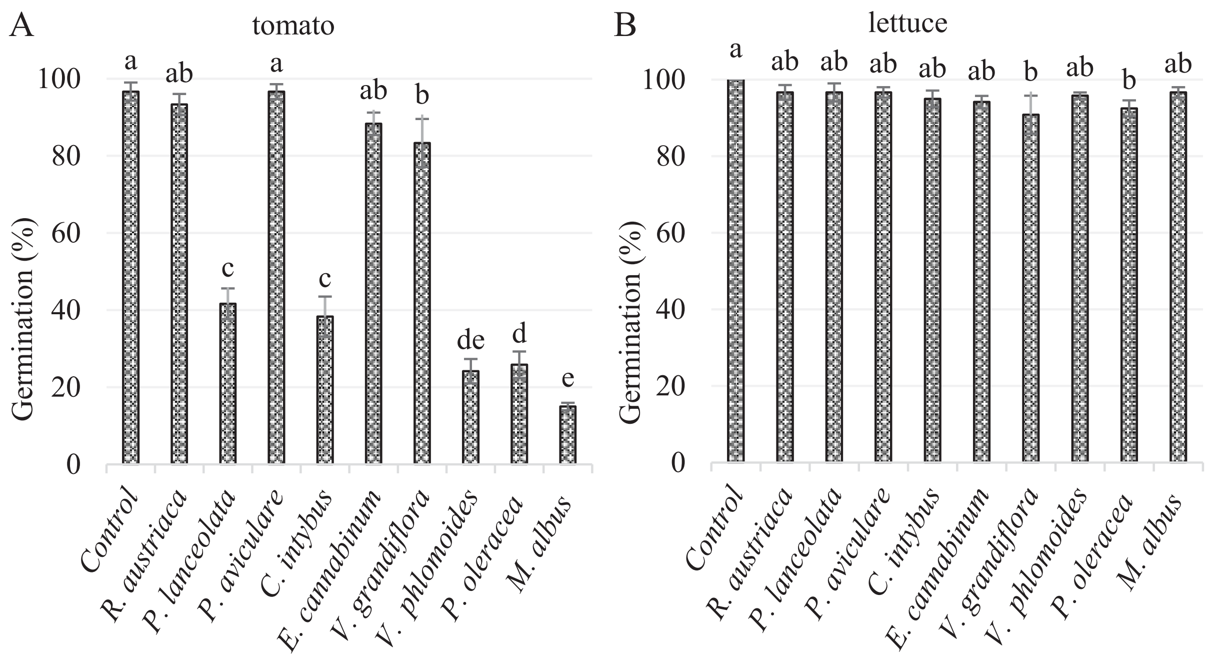

Figure 1. The allelopathic effect of water extracts from ruderal plant species on the germination of tomato (A) and lettuce (B)

Grafikon 1. Alelopatski utjecaj vodenih ekstrakata ruderalnih biljnih vrsta na klijavost rajčice (A) i salate (B)

A statistically significant reduction in the tomato seedlings' root length was observed in all treatments, except with the *P. aviculare* and *E. cannabinum* extracts (Figure 2A). The inhibition of tomato root length ranged from 37.5% with *V. grandiflora* to 94.5% and 95.1% with *P. oleracea* and *M. albus*, respectively. *P. aviculare*, on the other hand, stimulated the root length by 62.7% if compared to the control.

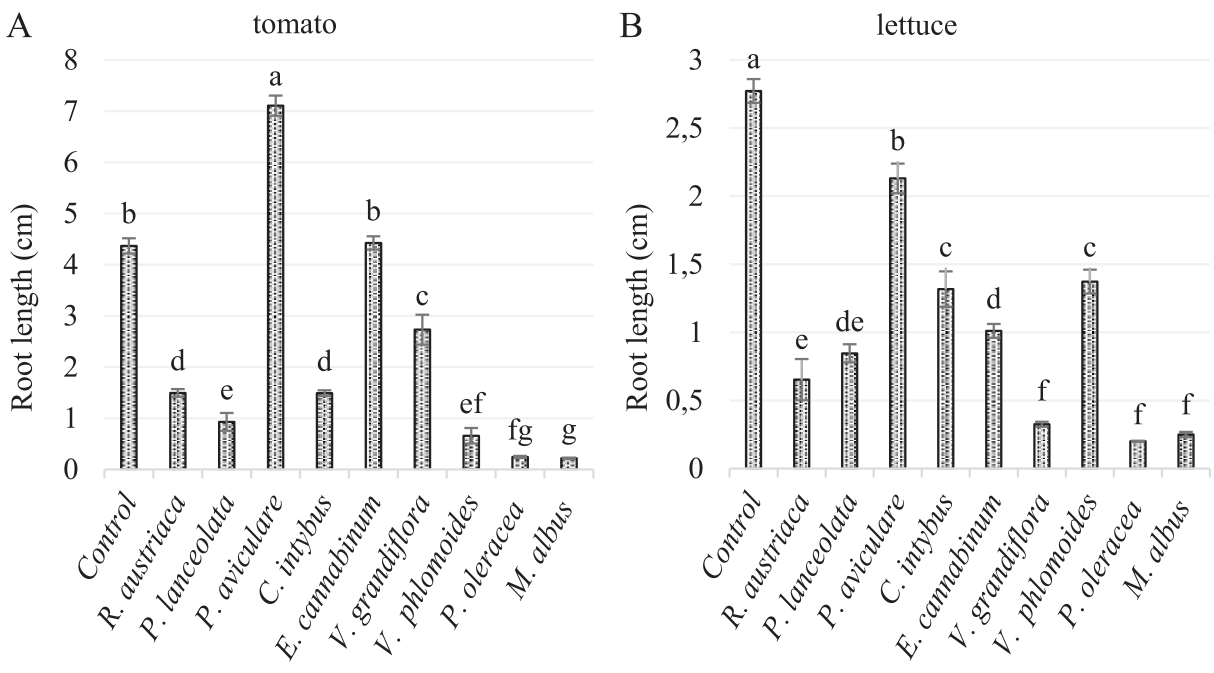

Figure 2. The allelopathic effect of water extracts from the ruderal plant species on the root length of tomato (A) and lettuce (B)

Grafikon 2. Alelopatski utjecaj vodenih ekstrakata ruderalnih biljnih vrsta na duljinu korijena rajčice (A) i salate (B)

In addition, the water extracts substantially impeded the root length of lettuce seedlings (Figure 2B). The most pronounced inhibitory effects were observed in the treatments with *P. oleracea*, *M. albus*, *V. grandiflora*, and the *R. austriaca* extracts, which reduced the root length by over 75% if compared to the control. Although statistically significant, the smallest negative impact was manifested by the *P. aviculare* water extract.

The shoot length of tomato seedlings was substantially decreased with all plant extracts besides *P. aviculare*, which stimulated the shoot growth by 87.9% if compared to the control (Figure 3A). While *E. cannabinum* and *V. grandiflora* reduced the shoot length by 39.9% and 42.1%, respectively, in all other treatments the inhibition was over 75% if compared to the control. Similarly, *P. aviculare* also stimulated the lettuce shoot length, while *P. lanceolata* and *C. intybus* had no effect thereupon (Figure 3B). A significantly negative effect was recorded for other extracts, with *M. albus* and *P. oleracea* demonstrating the greatest inhibition potential and reducing the shoot length by 82.6% and 68.4%, respectively.

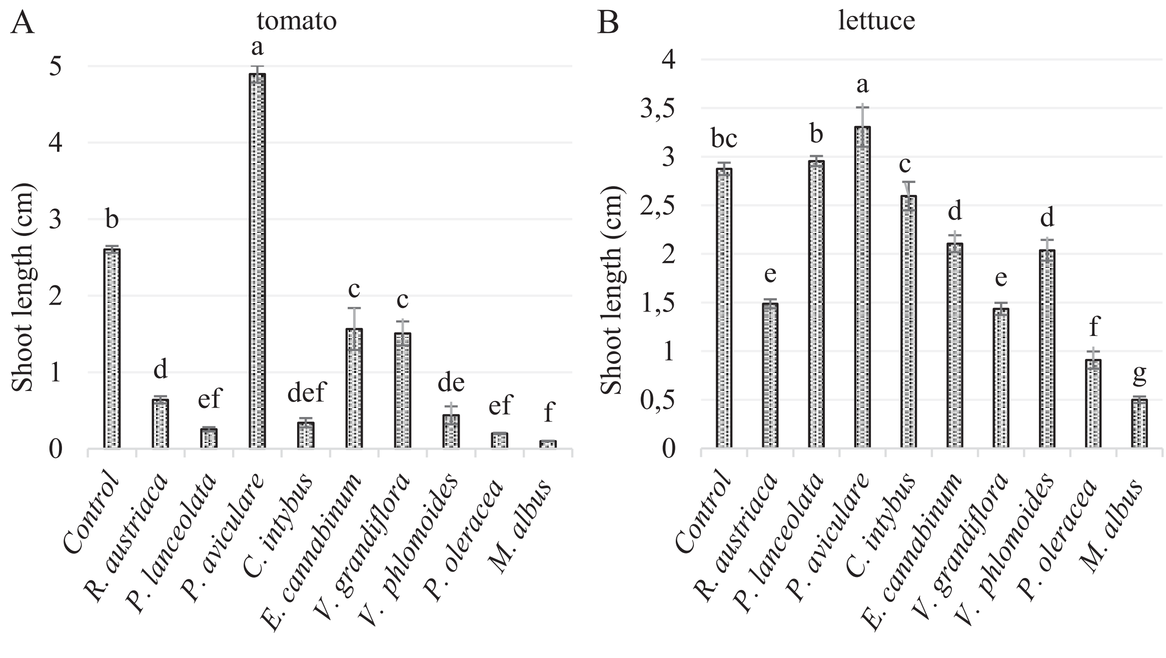

Figure 3. The allelopathic effect of water extracts from the ruderal plant species on the shoot length of tomato (A) and lettuce (B)

Grafikon 3. Alelopatski utjecaj vodenih ekstrakata ruderalnih biljnih vrsta na duljinu izdanka rajčice (A) i salate (B)

The experiment results proved that the investigated ruderal plant species manifested various allelopathic effects, which depended on the test species and on a measured parameter. According to their average inhibitory potential, the ruderal plant species can be ranked from the highest to the lowest, as follows: *M. albus*, *P. oleracea*, *V. phlomoides*, *P. lanceolata*, *C. intybus*, *R. austriaca*, *V. grandiflora*, *E. cannabinum*, and *P. aviculare*. A severe reduction in the seed germination of tomato, and seedlings' growth of both test species was detected in a treatment with *M. albus* extract. These findings are in accordance with a study conducted by Mardani et al. (2016), who showed that both *M. albus* and the yellow sweet clover (*Melilotus officinalis* (L.) Pall.) are highly allelopathic since their leaves suppressed the root length of lettuce by over 90%. Similarly, Shinwari et al. (2013) screened an allelopathic effect of thirty-eight weed species and concluded that *M. officinalis* and *M. albus* ranked as first and as second in their inhibitory potential with their leaf litter, volatiles, and root exudates, reducing the lettuce root length up to 98%, 39%, and 81%, respectively. *M. albus* is an annual or biennial species, most often a weed of ruderal habitats, used as a valuable honey plant, as well as a medicinal plant due to its high coumarin content (Knežević, 2006; Wu et al., 2021). According to Stefanović et al. (2015), a phytochemical analysis of *M. albus* extracts revealed the presence of phenolic compounds, flavonoids, and tannins, with their contents differing depending on the extract type (ethanol, acetone, or ethyl acetate). In our study, the *P. oleracea* extracts averagely reduced the germination, root, and shoot length of seedlings by 40.4%, 93.6% and 80.4%, respectively. The high concentrations of *P. oleracea* seed extracts proved to be inhibitory toward lettuce, while the very low concentrations stimulated the endive (*Cichorium endivia* L.) germination and seedlings' growth (Shehata, 2014). Furthermore, in a cogermination experiment, the *P. oleracea* seeds decreased the germination of common bean (*Phaseolus vulgaris* L.) and onion (*Allium cepa* L.) seeds, as well as the root and shoot length of the broad bean (*Vicia faba* L.), according to Rashidi et al. (2021).

The extracts of *V. grandiflora* significantly reduced the seed germination and growth of lettuce seedlings. A leaf litter of several other species of this genus, such as bird vetch (*V. cracca* L.), *V. truncatula* Fischer ex Bieb., *V. nissoliana* L., and *V. abbreviata* Fisch., suppressed the root growth of lettuce up to 50% (Mardani et al., 2016). The species belonging to the Asteraceae family, such as giant goldenrod (*Solidago gigantea* Ait.), annual fleabane (*Erigeron annus* L.), cockleburs (*Xanthium* sp.), and thistles (*Cirsium* sp.), demonstrated a strong allelopathic potential in several studies (Chon et al., 2003.; Baličević et al., 2015; Baličević et al., 2016). In our study, however, *E. cannabinum*, belonging to this plant family, was among the species with a lower allelopathic effect. Novak et al. (2018) also stated that both plants—namely, the common cocklebur (*Xanthium strumarium* L.) and the common ragweed (*Ambrosia artemisiifolia* L.)—exhibited a lower inhibitory effect when compared to the tree of heaven (*Ailanthus altissima* (Mill.) Swingle, Simaroubaceae) and jimsonweed (*Datura stramonium* L., Solanaceae).

Only the *P. aviculare* extract exhibited positive effects and promoted the root and shoot length of tomato, as well as the shoot length of lettuce. In our previous study, the stem and leaf extracts of *P. aviculare* decreased the germination and growth of lettuce, except for a shoot length in a treatment with a leaf extract, which was 26.1% higher if compared to the control (Baličević et al., 2016). Tsytsiura and Sampietro (2024) also recorded a reduction of germination up to 98.3 % and of the root and shoot length up to 65.7% and up to 61.1% of the oilseed radish (*Raphanus sativus* L. var. *oleiformis* Pers.) with the *P. aviculare* extracts. The differences in the results among the studies can be attributed to the different plant parts, extract concentrations, and/or the test species used in the studies.

A higher inhibitory effect of the extracts on the seedlings' root length was observed if compared to shoot length and seed germination. This is consistent with the results of Novak et al. (2018), who screened an inhibitory potential of eight invasive ruderal and segetal species and detected no significant effect on the germination of sunflower and oat (*Avena sativa* L.). A more pronounced effect on the root length of seedlings when compared to the shoot length could be due to the direct contact of roots with the extracts on a filter paper (Baličević et al., 2016).

The test species differed in their sensitivity to the allelopathic potential of water extracts. For example, *P. lanceolata* reduced the tomato germination and shoot length by 59.9% and 90.2%, respectively, but had no significant effect on lettuce. On average, tomato exhibited greater sensitivity to the applied extracts if compared to lettuce. The differences in sensitivity between the test species and even between the genotypes of the same species were reported in other studies (Norsworthy, 2003; Shehata, 2014; Novak et al., 2018; Serafimov et al., 2020; Rashidi et al., 2021). Baličević et al. (2015) detected that the crops and weed species responded differently to the *S. canadensis* extracts, with barley (*Hordeum vulgare* L.) and redroot pigweed (*Amaranthus retroflexus* L.) being the most sensitive. A seed size of the test species may influence its susceptibility to an allelopathic potential, with the seeds of a smaller size being more sensitive (Köhler et al., 2018).

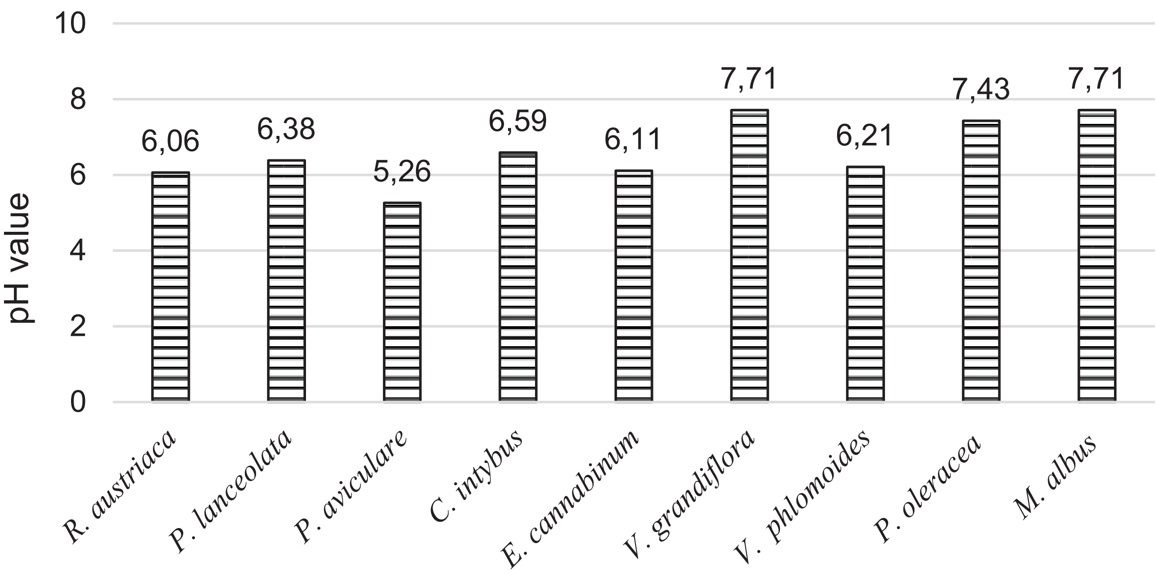

Figure 4. The pH values of water extracts from ruderal plants species

Grafikon 4. Vrijednosti pH vodenih ekstrakata ruderalnih biljnih vrsta

The pH value of a majority of extracts ranged between 6 and 7, while the lowest was recorded for *P. aviculare* (5.26) and the highest (7.71) for *V. grandiflora* and *M. albus* (Figure 4). Considering that the extracts of the same or of similar pH values had a different effect on the test species—that is, the pH of the extracts with the highest inhibitory potential was different and ranged from 6.21 to 7.71— it can be concluded that the seed germination and seedlings' growth were not affected by the pH values. Qasem (2002) also reported that the extracts of the same pH values reduced germination by 100% and 14.1%, respectively, while the extracts with the highest inhibitory potential had different pH values of 4.17 and 7.17, respectively.

CONCLUSION

The water extracts from dry biomass of the tested species manifested a different effect on tomato and lettuce seedlings; however, all extracts reduced at least one of the measured parameters. The test species differed in their susceptibility to the water extracts, with tomatoes being more sensitive when compared to lettuce. On average, the greatest inhibitory potential was recorded for *M. albus*, *P. oleracea*, and *V. phlomoides*, while *P. aviculare* manifested stimulatory effects. The aforementioned ruderal plants represent promising species for further studies.

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ALELOPATSKI POTENCIJAL RUDERALNIH BILJNIH VRSTA NA RAJČICU I SALATU

SAŽETAK

Cilj istraživanja bio je procijeniti alelopatski potencijal nekoliko ruderalnih biljnih vrsta na klijanje i početni rast rajčice i salate. Vodeni ekstrakti suhe nadzemne biomase u koncentraciji od 5 % testirani su u kontroliranim laboratorijskim uvjetima u Petrijevim zdjelicama. Rezultati istraživanja pokazali su da su sve vrste imale alelopatski učinak, te su poredane prema prosječnome inhibitornom potencijalu, od najvišega do najnižega, kako slijedi: Melilotus albus, Portulaca oleracea, Verbascum phlomoides, Plantago lanceolata, Cichorium intybus, Rorippa austriaca, Vicia grandiflora, Eupatorium cannabinum i Polygonum aviculare. Najznačajniji negativan utjecaj imao je vodeni ekstrakt vrste M. albus, koji je u prosjeku smanjio klijavost za 43,9 %, duljinu korijena za 93 % i duljinu izdanka za 89,4 %. Nasuprot tome, vodeni ekstrakt vrste P. oleracea imao je značajan stimulirajući učinak na obje ispitivane vrste. Vrijednost pH vodenih ekstrakata ruderalnih biljnih vrsta kretala se od 5,26 (P. aviculare) do 7.71 (V. grandiflora i M. albus) i nije utjecao na njihov negativan alelopatski potencijal.

Ključne riječi: fitotoksičnost, vodeni ekstrakti, inhibicija, biološka kontrola, ruderalna flora, usjevi

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