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## **Serbian Rutaceae species: comparison of flavonoid contents, coumarin compounds and radical scavenging activity**

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### **Abstract:**

**Pavlović, D., Zlatković, B., Živanović, S., Kitić, D., Golubović, T.: Serbian Rutaceae species: comparison of flavonoid contents, coumarin compounds and radical scavenging activity. *Biologica Nyssana*, 9 (1). September, 2018: 37-43.**

Family Rutaceae is presented in Serbian flora with 3 genera and 4 species: *Ruta graveolens* L., *Dictamnus albus* L., *Haplophyllum suaveolens* (DC.) G. Don and *H. boissierianum* Vis. & Pančić. Comparative study of the phytochemical variability and antioxidant activity of ethanolic and methanolic extracts, obtained from these species' areal parts, was performed. Total flavonoid content was determined spectrophotometrically and HPLC method was employed for quantification of coumarins. Fast antioxidant screening of samples was done in DPPH test. Flavonoid content of the tested samples varied from  $2.82 \pm 0.27$  to  $27.72 \pm 0.15$  mg of rutin equivalent (Ru)/g of extract. Psoralen, bergapten and xanthotoxin were detected and quantified only in *R. graveolens* extracts. All the tested samples exhibited notable potential in DPPH radicals scavenging assay. Yet, considerably lower activity was measured for *Haplophyllum* extracts. The present study brings forward the new data regarding the chemical profiles and antiradical activities of named Rutaceae species.

**Key words:** *Ruta*, *Dictamnus*, *Haplophyllum*, flavonoids, HPLC of coumarins, DPPH assay

### **Apstrakt:**

**Pavlović, D., Zlatković, B., Živanović, S., Kitić, D., Golubović, T.: Rutaceae vrste iz Srbije: poređenje sardžaja flavonoida, kumarina i sposobnosti uklanjanja slobodnih radikala. *Biologica Nyssana*, 9 (1). Septembar, 2018: 37-43.**

Familija Rutaceae u Srbiji broji 3 roda i 4 vrste: *Ruta graveolens* L., *Dictamnus albus* L., *Haplophyllum suaveolens* (DC.) G. Don i *H. boissierianum* Vis. & Pančić. Izvršeno je poređenje fitohemijske varijabilnosti i antioksidativne aktivnosti etanolnih i metanolnih ekstrakata herbi ovih biljnih vrsta. Ukupan sadržaj flavonoida je određen spektrofotometrijski dok je HPLC metod korišćen za kvantifikaciju kumarina. Brzi skining antioksidativne aktivnosti uzoraka je urađen DPPH testom. Sadržaj flavonoida je varirao od  $2.82 \pm 0.27$  do

27.72±0.15 mg rutin ekivalenata (Ru) po gramu ekstrakta. Psoralen, bergapten i ksantotoksin su detektovani i kvantifikovani samo u ekstraktima vrste *R. graveolens*. Svi testirani uzorci su ispoljili snažnu sposobnost uklanjanja slobodnih radikala u DPPH testu. Ipak, ova aktivnost je bila značajno niža kada su u pitanju ekstrakti *Haplophyllum* vrsta. Prikazana studija donosi nove podatke o hemijskom profilu i antiradikalnoj aktivnosti navedenih predstavnika familije Rutaceae.

**Ključne reči:** *Ruta*, *Dictamnus*, *Haplophyllum*, flavonoids, HPLC of coumarins, DPPH assay

## Introduction

Family Rutaceae comprises about 150 genera and 900 species distributed in warmer temperate and tropical regions (Jančić & Lakušić, 2017). The Rutaceae are herbaceous plants, shrubs or trees, predominantly glandular punctate, with commonly strong smell and frequent occurrence of spines as well as winged petioles. Depending on genus, the fruit morphology in Rutaceae is usually variable (Tutin et al., 1968; Jančić, 2002).

The family consists of six genera: *Citrus*, *Phellodendron* and *Ptelea* belong to subfamily *Aurantioideae* while *Ruta*, *Haplophyllum* and *Dictamnus* are members of subfamily *Rutinoideae*. *Rutinoideae* fruit is a capsule, usually 4 to 5-valved and seeds are with endosperm (Tutin et al., 1968). All three *Rutinoideae* genera are represented in Serbian flora (Diklić et al., 1973; Jančić & Lakušić, 2017).

Genus *Ruta* L. consists of 5 species that grow in dry, usually rocky habitats. *Ruta graveolens* L. (*R. hortensis* Mill.) is a shrub-like herbaceous perennial aromatic plant that is native to Balkan Peninsula and Crimea, and rare wild growing species in SE Serbia. It is widely naturalized from gardens in S and SC Europe (Tutin et al., 1968; Diklić et al., 1973; Tasić et al., 2004). Fresh juice, whole plant and essential oil irritate skin and cause rash and pain (PDR, 2000). Underground parts of the plant are usually harvested at the beginning of flowering season (Stojković et al., 2007) and highly used in the traditional medicine in numerous countries to treat a variety of ailments, ranging from absence of menstruation to rheumatism and various mental conditions (PDR, 2000; Tucakov, 1997). Recent studies on rue herb and/or leaves confirmed, among other pharmacological activities, antimicrobial, abortifacient, anti-inflammatory, analgesic, antiandrogenic, antihyperlipidemic, antihyperglycemic, xantine oxidase inhibition, cytostatic and anticancer effects (Yang et al., 2006; Asgarpanah & Khoshkam, 2012; Pavlović et al., 2014). The plant contains active compounds like flavonoids, alkaloids, lignans, coumarin derivatives and essential oil (PDR, 2000). Herb and fresh leaves may be used as spice and flavoring agent, and they have been part of East Asian diets for many years as food and medicine (Yang et al., 2006).

*Dictamnus albus* L. (*D. fraxinella* Pers.) is also perennial plant with large and attractive flowers in terminal, bracteate inflorescences. Although several related taxa (*D. caucasicus*, *D. gymnostylis* and *D. hispanicus*) are maintained as separate species or subspecies, in Flora Europea as in Flora of SR Serbia *Dictamnus albus* is treated as a single, but polymorphic species. It is common in south and middle Europe and grows on sunny, dry, rocky slopes and bright forests (Tutin et al., 1968; Diklić et al., 1973). Plant parts of this species are rich in flavonoids, furanocoumarins, quinoline alkaloids, limonoids and essential oil (PDR, 2000; Michael et al., 2003). Its flowering branches are used as a folk remedy due to its stomachic, tonic, stimulant and antipyretic activities, but also as an emmenagogue, an effective uterine remedy and as an abortive agent. Phototoxic, mutagenic and embryotoxic effects have been documented (PDR, 2000; Tasić et al., 2004; Beis et al., 2005).

Genus *Haplophyllum* A. Juss. counts about 70 species of herbaceous perennial, sometimes woody below or semi-shrubs distributed from the Mediterranean region to eastern Siberia. They are closely related to *Ruta* (sometimes united with that genus), having simple to lobed leaves and small, yellow flowers with five, cupped petals in terminal cymose clusters. In Serbia are represent only *H. suaveolens* (DC.) G. Don (*H. ciliatum* Griseb., *H. biebersteinii* Spanc, *Ruta suaveolens* De Cand.) and *H. boissierianum* Vis. & Pančić (*H. albanicum* (Bald.) Bornm., *Ruta boissierianum* (Vis. et Panč.) K. Maly) (Tutin et al., 1968; Diklić et al., 1973).

*Haplophyllum suaveolens* is native to SE Europe: from Macedonia to east Ukraine (Diklić et al., 1973). Phytochemical study confirmed the presence of lignan and coumarin glycosides in aerial parts of that species (Ivanova et al., 2001).

*Haplophyllum boissierianum* is an endemic species of Balkan Peninsula. It grows at the territory of ex Yugoslavia and in Albania (Diklić et al., 1973). To the best of our knowledge, there are no data on its chemical composition or pharmacological effects.

In order to compare species of Rutaceae family growing wild in Serbia, we collected plant material of *R. graveolens*, *D. albus*, *H. suaveolens* and *H. boissierianum*. Thus, a comparative analysis of the variability of present flavonoids and coumarins

alongside antioxidant activity of the selected Rutaceae species was done.

## Material and methods

All reagents and solvents used in this investigation were of analytical grade. The reference chemicals such as rutin and coumarins (psoralen, 5-methoxypsoralen, 7-methoxypsoralen, 8-methoxypsoralen and scopoletin) used for calibration curves and HPLC analyses were purchased from Sigma-Aldrich (St. Louis, USA) or Carl Roth (Karlsruhe, Germany).

Spectrophotometric measurements were performed using Evolution 60 Thermo scientific spectrophotometer (Fisher Scientific, UK) and Multiskan Ascent No354 (ThermoLabsystems, Finland) ELISA microplate reader. HPLC analysis was performed using the Agilent 1200 HPLC system (Agilent Technologies, Santa Clara, CA, USA) equipped with a binary pump and a diode array (DAD) detector.

## Plant material and plant extracts preparation

Plant material were collected from the species: *Ruta graveolens* in Sićevočka gorge, Sićevo village (southeast Serbia), *Dictamnus albus* and *Haplophyllum suaveolens* by Gabrovačka river in the city of Niš vicinity (south Serbia), and *Haplophyllum boissierianum* at Goč Mt. (central Serbia). The employed nomenclature is in accordance with Euro+Med PlantBase (<http://www2.bgbm.org/EuroPlusMed/>).

Aerial parts of these wild growing plants were collected in blossoming phase. *Ruta graveolens* was collected at the beginning and also at the end of flowering season.

Taxonomic identification and authentication were performed by Professor B. Zlatković (Faculty of Science and Mathematics, University of Niš).

Dry plant material was reduced to a fine powder and extracted with ethanol (70%, v/v) or methanol (80%, v/v) by maceration: drug-solvent ratio was 1:10 and extraction time was 5 days.

Ethanolic and methanolic extracts of *R. graveolens* (RE and RM: extracts obtained from plant material collected at the beginning of flowering season; RE2 and RM2: extracts obtained from plant material collected at the end of flowering season), *D. albus* (DE and DM), *H. suaveolens* (HsE and HsM) and *H. boissierianum* (HbE and HbM) were obtained after evaporation to the dryness under reduced pressure below 40 °C. Extraction yields were expressed in % of used dry plant material.

## Determination of total flavonoids

The contents of total flavonoids in the obtained extracts were determined spectrophotometrically at 430 nm using aluminium chloride (Lamaison and Carnat, 1990) and the results were expressed as mg rutin (Ru)/g of dry extract after quantification on the basis of a standard curve with rutin. Concentration span of rutin was 0.5-5 µg/ml and calibration curve equation:  $y = 0.0526x - 0.0278$  ( $R^2 = 0.987$ ). The total flavonoid assay was measured in triplicate.

## HPLC analysis of coumarins

For HPLC analysis of coumarins dry extracts were dissolved in methanol (5 mg/ml). Samples were gradually eluted with a two phase system, phase A = water and phase B = acetonitrile, flow rate of 1 ml/min, at 25 °C. Gradient profile was: 0-4 min 25%→30% B; 4-20 min 30%→40% B; 20-22 min 40%→100% B; 22-26 min 100% B; 27 min 30% B and 29 min 25% B. Purospher star RP-18C column (150 x 4.6 mm with 5 µm particle size, Merck) was used and injection volume was 10 µl.

Psoralen, 8-metoxypsoralen (syn. xanthotoxin, methoxsalen), 7-methoxypsoralen, 5-methoxypsoralen (syn. bergapten) and scopoletin were used as standards in concentration of 1 mg/ml (in ethyl acetate, chloroform, ethanol, chloroform and acetic acid, respectively). Chromatograms of extracts and standards were recorded under the same conditions. Detection was performed at 245 nm, 322 nm and 345 nm. Identification of components was carried out by comparing their UV spectra and peak retention times with UV spectra and peak retention times of standards (Mabry et al., 1970). Quantification was done on the basis of response factors of standards solutions at two pints (the ratio between a signal produced by standard and the quantity of standard which produced the signal). Response factors for psoralen, bergapten and xanthotoxin were 60.589, 35.088 and 59.125, respectively.

## Determination of radical scavenging capacity

The free radical scavenging activities of the extracts were determined using a stable 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical (Cuendet et al., 1997). Various concentrations of samples were mixed with methanol or ethanol solution of DPPH (0.05 mM), vigorously shaken and left in the dark at room temperature for 30 min. Inhibition of DPPH free radical in percent was calculated according to equation:

$$\% \text{ DPPH} = (\text{Ac} - \text{As} / \text{Ac}) \times 100$$

where Ac is the absorbance of the control reaction (containing ethanol or methanol instead of test solution), and As is the absorbance of the sample.

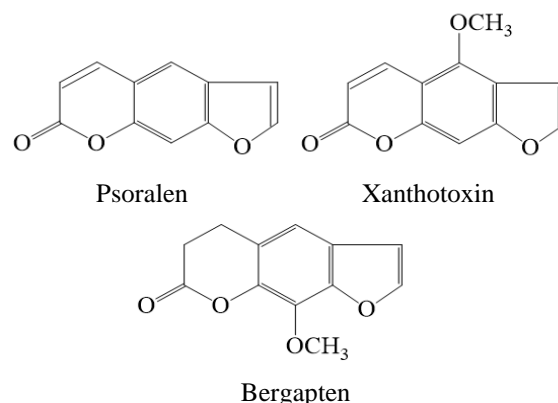
Regression curves were constructed and DPPH results calculated as the concentration of sample required for scavenging 50% of the free radical ( $IC_{50}$ ). Concentrations are expressed in mg/ml. Rutin was used as reference compound. All experiments were done in triplicates.

## Results and discussion

Plant material of *R. graveolens* collected at the beginning of the flowering season give the highest extraction yield. Extraction yields of ethanol extracts were generally higher than yields of methanol extracts. The only exception was *H. suaveolens* (Tab. 1).

Flavonoid contents and coumarin profiles of Rutaceae species growing wild in Serbia are quite diverse. Three coumarin compounds were identified in the investigated extracts (Tab. 1). All detected coumarins belong to furanocoumarin class. Psoralen, bergapten and xanthotoxin (Fig. 1) were present in all four tested *R. graveolens* extracts. Methanol extract of *R. graveolens* harvested at the end of flowering season had the highest furanocoumarin levels of all tested samples. Scopoletin and 7-metoxypsoralen were absent in all investigated samples. Thus, in extracts of *D. albus*, *H. suaveolens* and *H.*

*boissierianum*, none of the tested coumarins was detected.



**Fig. 1.** Structural formulas of psoralen, bergapten and xanthotoxin

Coumarins possess various biological and pharmacological activities (i.e. anticoagulant, estrogenic, vasodilator, hypothermic, antihelmintic, sedative, analgesic, anti-inflammatory and antiulcer activity). Coumarin derivatives are also considered as potential antibacterial agents and due to its wide range of structural modifications, they can serve as good molecular templates for new drugs (Veselinović et al., 2016). Furanocoumarins are found in many plants, including common plant foods consumed by humans, and are known to be absorbed into the

**Table 1.** Extraction yields, total flavonoids contents and contents of detected coumarins in tested Rutaceae species samples

Extract	Extraction yield (%)	Total flavonoids (mg rutin/g)	Psoralen (mg/g)	Bergapten (mg/g)	Xanthotoxin (mg/g)
RM	23.3	26.55±0.36	0.78	5.59	6.69
RE	27.3	27.72±0.15	0.90	5.93	6.52
RM2	19.5	23.58±0.16	1.11	8.99	9.24
RE2	24.0	26.77±0.27	0.90	5.62	6.45
DM	20.6	7.27±0.12	/	/	/
DE	21.4	2.82±0.27	/	/	/
HsM	14.9	13.95±0.12	/	/	/
HsE	11.3	15.88±0.05	/	/	/
HbM	16.1	16.50±0.25	/	/	/
HbE	17.7	15.82±0.32	/	/	/

RM – *Ruta graveolens* methanol extract (beginning of the flowering season)

RE – *Ruta graveolens* ethanol extract (beginning of the flowering season)

R2M – *Ruta graveolens* methanol extract (end of the flowering season)

R2E – *Ruta graveolens* ethanol extract (end of the flowering season)

DM – *Dictamnus albus* methanol extract

DE – *Dictamnus albus* ethanol extract

HsM – *Haplophyllum suaveolens* methanol extract

HsE – *Haplophyllum suaveolens* ethanol extract

HbM – *Haplophyllum boissierianum* methanol extract

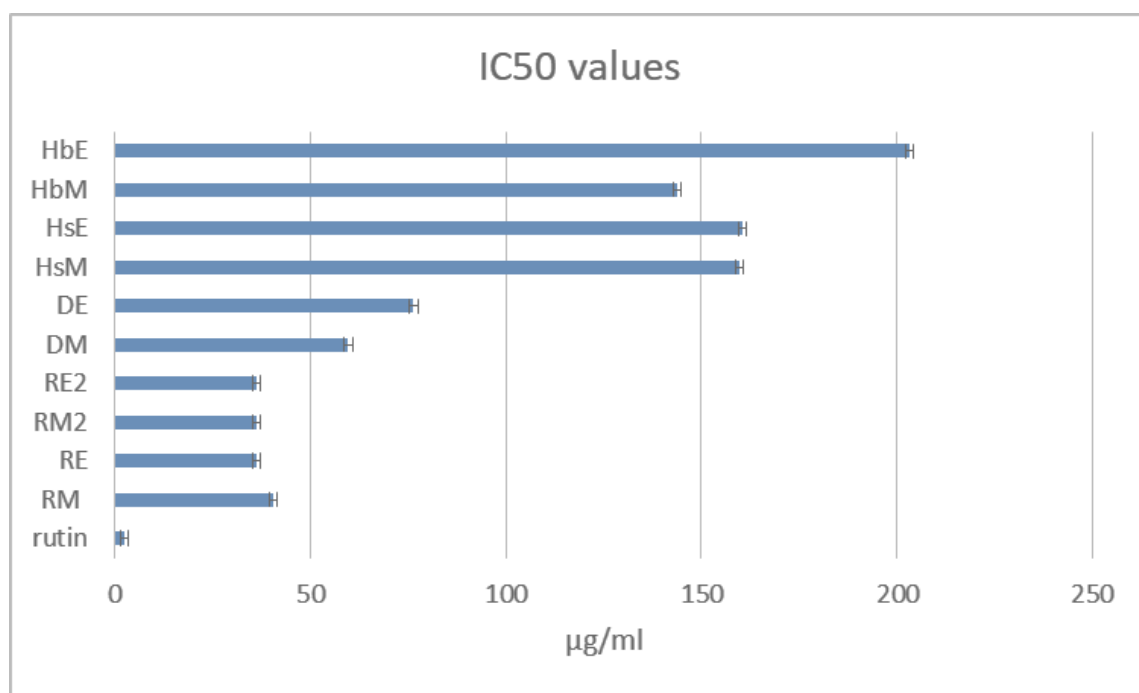
HbE – *Haplophyllum boissierianum* ethanol extract

bloodstream rapidly following consumption. They exhibit strong photoactivity and are capable of intercalating with DNA in the presence of UV light that lead to phytophotodermatitis but also genetic mutations and carcinogenicity (Melougha et al., 2018). Psoralens (psoralen and its derivatives such as bergapten and xanthotoxin, **Fig. 1**) are part of PUVA therapy: photochemotherapy with psoralens (as the photosensitizing agents) and ultraviolet light type A (long-wave ultraviolet A or UV-A, 320-380 nm) radiation as the treatment for psoriasis, eczema and some other dermatologic conditions (Melougha et al., 2018). High cytotoxic response after simultaneous exposure of melanoma cells to psoralens and UVA radiation *in vitro* suggests the usefulness of PUVA therapy to treat melanoma *in vivo* (Wrześniok et al., 2017). Considering amounts of furanocoumarins quantified in the tested extracts (**Tab. 1**), *R. graveolens* could be treated as a valuable natural source of these compounds. Xanthotoxin is the most abundant coumarin compound in the tested *R. graveolens* samples.

Flavonoids represent one of the most important groups of phenolic natural products and, like coumarins, they are also biosynthesized from L-phenylalanine. These metabolites are found widely distributed in Rutaceae in seeds, fruits, flowers and leaves (Middleton, 1984). In our previous research we concluded that rue leaves and herb should be harvested at the beginning of blossoming stage and that wild growing plants present richer source of

secondary metabolites in comparison to cultured rue (Pavlović et al., 2014). Considerable amounts of flavonoids, expressed in mg of rutin per g of dry extract, were found in investigated samples. The highest flavonoid levels were detected in extracts of *R. graveolens* followed by *H. boiserianum*, *H. suaveolens* and *D. albus* extracts. Flavonoids are represented in the range from  $2.82 \pm 0.27$  mg/g (DE) to  $27.72 \pm 0.15$  mg/g (RE) (**Tab. 1**). Beside numerous other proven activities, flavonoids and other polyphenols have been reported to be responsible for the antioxidant activities of botanical extracts. The antioxidant activity of these substances is due to the ability of reducing free radical formation and to scavenge free radicals (Pavlović et al., 2011). According to some authors, total flavonoids content could be treated also as an antioxidant assay alongside total phenolic content determination (Mihajilov-Krstev et al., 2015).

Preliminary screening of antioxidant activity was done in DPPH test. According to this radical scavenging assay, *R. graveolens* extracts possess notable and quite similar anti-radical activity with  $IC_{50}$  range from  $36.36 \pm 1.20$   $\mu$ g/ml (RM2) to  $40.65 \pm 2.53$   $\mu$ g/ml (RM). Considerably lower activity was measured for extracts obtained from two tested *Haplophyllum* species (the lowest for HbE:  $203.26 \pm 4.74$   $\mu$ g/ml), while both *D. albus* extracts showed intermediate activities (DM:  $59.80 \pm 1.53$   $\mu$ g/ml and DE:  $76.48 \pm 2.30$   $\mu$ g/ml). As expected, rutin exhibited much stronger antiradical potential with



**Fig. 2.** Radical scavenging activity  $IC_{50} \pm SEM$  in  $\mu$ g/ml of the tested samples and rutin as reference compound

IC<sub>50</sub> value 2.68±0.25 µg/ml. IC<sub>50</sub> values of the tested samples and a reference compound rutin are presented in **Fig. 2**. Since the measurement of antioxidant activity of a plant extract that is a complex mixture, cannot be evaluated satisfactorily by a single antioxidant test, several test procedures must be engaged in order to fully comprehend properties of the examined extracts (Pavlović et al., 2013).

## Conclusions

Generally, the present study brings forward the new data regarding the chemical composition of selected Rutaceae species alongside its preliminary antioxidant activity. Further phytochemical and pharmacological evaluations are necessary to bring more light to potential applications of these species.

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## References

- Asgarpanah J., Khoshkam R. 2012: Phytochemical and pharmacological properties of *Ruta graveolens* L. *Journal of Medicinal Plants Research* 6(23): 3942-3949.
- Beis R., Öztürk Y., Aydin S., Güreş F. 2005: Pharmacological and toxicological effects of gas plant (*Dictamnus albus* L.). *Turkish Journal of Pharmaceutical Sciences* 2 (3): 111-124.
- Cuendet M., Hostettmann K., Potterat O. 1997: Iridoid glucosides with free radical scavenging properties from *Fagraea blumei*. *Helvetica Chimica Acta* 80: 1144-1152.
- Diklić N., 1973: Rutaceae In: Josifović M, Stjepanović L., Janković M.M., Gajić M., Kojić M., Diklić N. (eds.), *Flora SR Srbije* V. Srpska akademija nauka i umetnosti, Beograd.
- Ivanova A, Mikhova B, Stambolijska T, Kostova I. 2001: Lignan and coumarin glycosides from *Haplophyllum suaveolens*. *Zeitschrift für Naturforschung C*, 56(5-6): 329-33.
- Jančić R., Lakušić B. 2017: *Botanika farmaceutika*. Lento, Beograd.
- Lamaison J.L., Carnat A. 1990: Teneurs en principaux flavonoids des fleurs de *Crataegus monogyna* Jacq et de *Crataegus laevigata* (Poir) D. C.) en fonction de la vegetation. *Pharmaceutica Acta Helvetica* 65, 315-320.
- Mabry T.J., Markham K. R., Thomas M. B. 1970: *The Systematic Identification of Flavonoids*. Springer-Verlag, Berlin.
- Melougha M.M., Chob E., Chuna O.K. 2018: Furocoumarins: A review of biochemical activities, dietary sources and intake, and potential health risks. *Food and Chemical Toxicology* 113: 99–107.
- Michael J.P. 2003: Quinoline, quinazoline and acridone alkaloids. *Natural Products Reports* 20(5): 476-93.
- Middleton E.J. 1984: The flavonoids. *Trends in Pharmacological Science*, 8:335-338.
- Mihajilov-Krstev T., Zlatković B., Ilić M., Stankov-Jovanović V., Mitić V. 2015: Antimicrobial and antioxidant potential of wild growing *Silene baccifera* (L.) Roth. (Caryophyllaceae) fruits juice. *Biologica Nyssana* 6 (2): 55-58.
- Pavlović D.R., Branković S., Kovačević N., Kitić D., Veljković S. 2011: Comparative Study of spasmolytic properties, antioxidant activity and phenolic content of *Arbutus unedo* from Montenegro and Greece. *Phytotherapy Research* 25(5): 749-54.
- Pavlović D.R., Tasić-Kostov M., Marčetić M., Lakušić B., Kitić D., Savić S., Kovačević N. 2013: Evaluation of *in vivo* effects on surfactant-irritated human skin, antioxidant properties and phenolic composition of five Ericaceae species extracts. *Rivista Italiana Delle Sostanze Grasse* 90 (4): 255-264.
- Pavlović D.R., Vukelić M., Najman S., Kostić M., Zlatković B., Mihajilov-Krstev T., Kitić D. 2014: Assessment of polyphenol content, *in vitro* antioxidant, antimicrobial and toxic potentials of wild growing and cultured rue. *Journal of Applied Botany and Food Quality* 87: 175 – 181.
- PDR, 2000: *PDR for Herbal Remedies*. 2nd Edition, Thomson PDR, Medical Economics Company, Montvale, New Jersey.
- Stojković D., Jančić R., Lakušić B., Slavkovska V. 2007: *Samonikle lekovite biljke Stare planine priručnik za održivo sakupljanje*. Svetska unija za zaštitu prirode Zavod za zaštitu prirode Srbije, Beograd.
- Tasić S., Šavikin Fodulović K., Menković N. 2004: *Vodič kroz svet lekovitog bilja*. Valjevac, Valjevo.
- Tucakov J., 1997: *Lečenje biljem*. VII Edition, Rad, Beograd.
- Tutin T.G., 1968: Rutaceae In: Tutin T.G., Heywood V.H., Burges N.A., Moore D.A., Valentine D.H., Walters S.M., Webb D.A. (eds.), *Flora Europaea* 2. University Press, Cambridge.
- Veselinović J.B., Matejić J.S., Veselinović A.M., Sokolović D. 2016: Coumarin structure as a lead

- scaffold for antibacterial agents - molecular docking study. *Biologica Nyssana* 7(2): 167-170.
- Wrześniok D., Beberok A., Rok J., Delijewski M., Hechmann A., Oprzonek M., Rzepka Z., Bacler-Żbikowska B., Buszman E. 2017: UVA radiation augments cytotoxic activity of psoralens in melanoma cells. *International Journal of Radiation Biology* 93(7): 734-739.
- Yang R.-Y., Tsou S.C.S., Lee T.-C., Wu W.-J., Hanson P.M., Kuo G., Engle L.M., Lai P.-Y. 2006: Distribution of 127 edible plant species for antioxidant activities by two assays. *Journal of Science and Food Agriculture* 86: 2395-2403.