# THE PHENOLIC COMPOUNDS CONTENT AND ANTIOXIDANT ACTIVITY OF SOME MEDICINAL PLANTS

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**Abstract**
*In the present study, the infusions from six medicinal plants have been evaluated in terms of their polyphenols content and antioxidant activity. The plants selected for this study are the following: elder (Sambucus nigra), chamomile (Chamomillae germanicae flos), sage (Salvia officinale), mistletoe (Viscum album), lemon balm (Melissa oficinalis), lavender (Lavandula angustifolia). The investigated samples show a wide variation in the values obtained both for the antioxidant capacity and for the content of total phenolic compounds. The values obtained for the content of phenolic compounds vary between 87.93 µg gallic acid equivalent/mL (mistletoe) and 984.33 µg gallic acid equivalent/mL (chamomile) and increase in the order of mistletoe < chamomile < lavender < sage <elder < lemon balm. All investigated samples show high values of antioxidant activity against the DPPH radical. The results indicate the highest antioxidant activity for lemon balm. The values vary between 1.05 and 14.63 µM Trolox equivalent /mL infusion and decrease in the order: lemon balm, sage, lavender, elder, chamomile, mistletoe. The high content of phenolic compounds of these medicinal plants can explain their use in the therapy of many disease.*

***Keywords:*** *antioxidant activity, medicinal plants, polyphenols*

## INTRODUCTION

Medicinal plants are used in the prevention and treatment of many diseases and ailments (Briskin, 2000, Hassan, 2012, Jamshidi-Kia et al., 2018, Sofowora et al., 2013). These beneficial effects are attributed to the rich composition in phytochemicals with antioxidant properties. It has been shown that free radicals and oxidative stress play a causational role in the pathology of many diseases and aging processes (Dröge 2002, Valko et al., 2006). Oxidative stress is a cellular status caused by the imbalance between the production of free radicals and the ability of the cell to scavenge these reactive molecules (Lushchak, 2014, Poprac et al., 2017). The cell protects itself from the devastating effects of free radicals and oxidative stress by developing a defensive antioxidant system (Espinosa-Diez et al., 2015, Halliwell 1996) that can be supplemented with natural exogenous antioxidants from plants. In traditional medicine, a widely used remedy is the infusion or decoction prepared from leaves, flowers and roots of medicinal plants (Briskin, 2000, Rafieian-Kopaei 2012). In the present study, the infusions from six popular medicinal plants have been evaluated in terms of their polyphenols content and antioxidant activity.

## MATERIALS AND METHODS

The plants selected for this study are the following: elder (*Sambucus nigra*), chamomile (*Chamomillae germanicae*), sage (*Salvia officinale*), mistletoe (*Viscum album*), lemon balm (*Melissa oficinalis*), lavender (*Lavandula angustifolia*). The dry plants were purchased from local market. Their antioxidant activities and total phenolic compounds content were determined in deionised water extracts as teas (2g:100mL, 5 min without additional heating.). The samples were filtered through Whatman No. 1 paper and the teas were analysed immediately.

The total phenolic content was determined colorimetric by using the Folin-Ciocalteu method (Soare et al., 2015). 2 mL Folin-Ciocalteu's phenol reagent (1:10) and 1.5 mL 7.5 % w/v $Na_2CO_3$ were added to 0.5 mL sample extract. The mixture was allowed to stand at room temperature in the dark for 60 min and then the absorbance was recorded at 765 nm. The total phenolic content (TPC) was calculated using a standard curve prepared using gallic acid and expressed as µg gallic acid equivalents (GAE)/mL tea.

Antioxidant activity is evaluated as 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay by colorimetric method (Soare et al., 2016). 2 mL of 0.075 mM DPPH methanolic solution was mixed with 0.1 mL extract and vortexed thoroughly. The absorbance of the mixtures was measured at 517 nm after 20 minutes. A blank reagent was used to study stability of DPPH over the test time. The percentage of DPPH radical scavenging activity of extracts was evaluated according to the formula: % scavenging = $[A_0-(A_1–A_S)]/ A_0]100$, where $A_0$ is the absorbance of DPPH alone, $A_1$ is the absorbance of DPPH + extract and $A_S$ is the absorbance of the extract only. Trolox (T) was used as standards. The standard calibration curve was plotted as a function of the percentage of DPPH radical scavenging activity. The results were expressed as µM Trolox equivalents (TE) per mL tea. (µM TE/mL). The spectrophotometric measurements were performed with a Thermo Scientific Evolution 600 UV-Vis spectrophotometer with VISION PRO software. All determinations were performed in triplicate and all results were calculated as mean.

## RESULTS AND DISCUSSIONS

The values obtained for the content of phenolic compounds vary between 87.93 µg GAE/mL (mistletoe) and 984.33 µg GAE/mL (chamomile) and increase in the order: mistletoe < chamomile < lavender < sage < elderberry < lemon balm (figure 1). The high content of phenolic compounds of these medicinal plants can explain their use in the therapy of many ailments. The obtained results are similar to those reported by Katalinic et al., 2006 for sage and lavender and lower for chamomile and lemon balm. For mistletoe and elder, the results obtained in this study are higher than those reported by Katalinic et al., 2006.

In the specialized literature, there are numerous studies on the content of phenolic compounds and the antioxidant activity of medicinal plants.

*Lemon balm*: Phenolic compounds and antioxidant activity of leaves and flowers have been more studied in alcoholic extracts (Pereira et al., 2009, Mabrouki et al., 2018). Aqueous extract prepared in the form of infusion and freshly analysed, has an antioxidant activity expressed as IC50% = 32.9 µg/mL compared to the ethanolic and methanolic extract 28.2 µg/mL and 24.3 µg/mL, respectively. The content of phenolic compounds of these extracts is 389.65 ± 99.15 nmol GA/g plant (aqueous), 26.41 ± 0.09 nmol GA/g plant in ethanolic extract and 166.32 nmol GA/g plant in methanolic extract (Pereira et al., 2009).

In order to estimate the most efficient extraction method studies were carried out using acetone, hexane and ethanol as solvents (Mabrouki et al., 2018). It was reported that the ethanolic extract has the highest content of phenolic compounds (63.00 mg GAE/g dry ethanolic extract) and the highest antioxidant activity IC 59%. Spiridon et al., 2011 report for the content of phenolic compounds value of 54.90 mg GAE/g dry ethanolic extract and

EC50 = 87.28µg/mL (Spiridon et al., 2011). There is a great variation in the data presented in the literature because the biosynthesis of phenolic compounds and other compounds with antioxidant activity is dependent on environmental conditions, soil and agricultural technologies (Tusevski et al., 2014). Also, the extraction yield of these compounds depends on the solvent used and the working temperature.

*Elder:* Viapiana and Wesolowski qualitatively and quantitatively analyze the content of phenolic compounds and the antioxidant activity of the tea obtained by infusion of 24 commercial varieties of elder (Viapiana and Wesolowski, 2017). The content of phenolic compounds varies between 19.81 mg GAE/g dm and 23.90 mg GAE/g dm for fruits and between 15.23 mg GAE/g to 35.57 mg GAE/g dm for flowers. Similar results are obtained for plants from Jordan (20mg GAE/g dm) (Tawaha et al., 2007). Duymuş et al. report values of 67.15 mg GAE/g dm for fruit infusion (Duymus et al., 2014) and for flower infusion 498 mg CAE/L (Katalinic et al., 2006) and 42.67 g GAE/kg dw (Veljkovic et al., 2013). Regarding the spectrum of individual phenolic compounds, it is observed that flavonols are found in greater quantity than phenolic acids. The fruit infusion is rich in quercitin, rutin, kaempferol and gallic acid while the flower infusion is rich in myricetin, chlorogenic acid, p-coumaric acid (Viapiana and Wesolowski, 2017).

Sage: In sage preparations, among the phenolic acids, the majority constituent is caffeic acid (Lu and Foo, 2002).

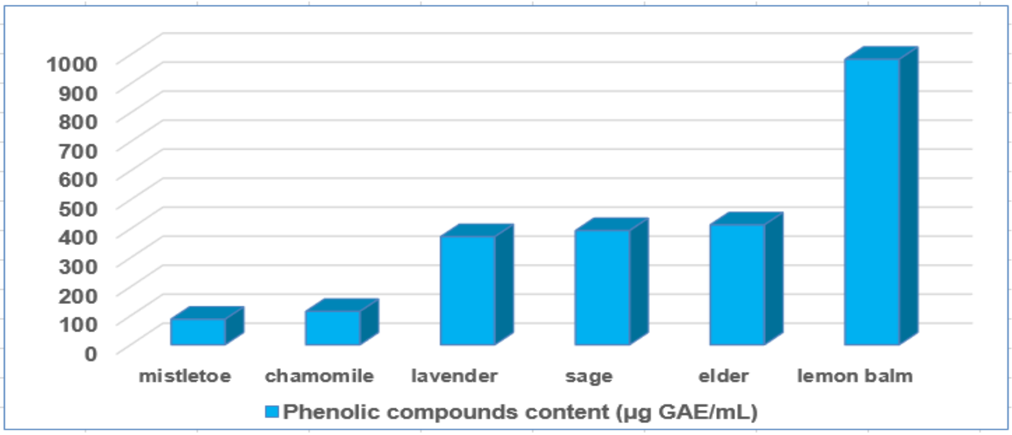


Figure 1. Phenolic compounds content in studied medicinal plants

Among its derivatives, there is a large amount of rosmarinic acid (a dimer of caffeic acid) which has been reported to be one of the main ones responsible for the antioxidant activity and beneficial effects of sage preparations. Among the phenolic acids, there are also ferulic, isoferulic and chlorogenic acids. Salvia also has a wide spectrum of flavonoids, these being present as flavones (apigenin, luteolin), flavonols (kaempferol and quercetin) and their glycosides. Anthocyanins are present in sage flowers. A study on the distribution of the spectrum of anthocyanins in 10 species of Sage shows that the red and pink flowers contain pelargonidin, the blue ones delphinidin and the purple ones derived from cyanidin (Haque et al., 1981).

In a paper that studies the influence of the harvest period and drying method of *Salvia officinalis* L. leaves on the content of phenols, TPC values between 8,9-11,6 mgGAE/g dw for the infusion and 11,6-17,1 mg GAE/g dw for the methanol-acetone extract are reported (Francik et al., 2020).

*Chamomile:* For chamomile infusion and decoction the scientific literature reports a content of phenolic compounds of 86.09 mg GAE/100 mL respectively 72.17 mg GAE/100 mL for the decoction for 15 minutes. The infusion was made from 2 g of the plant with 60 mL of boiling water and left for 15 minutes (Fotakis et al., 2016). The results obtained are higher than the results obtained in this study.

*Lavender*: The main phenolic compounds present in the methanol extract of lavender are rosmarinic and chlorogenic acid and the flavonone glycosides apigenin and luteolin (Celep et al., 2018). HPLC analysis showed the presence of rosmarinic acid, ferulic acid glucoside, caffeic acid, morin, coumarin and herniarin (Dobros et al., 2022).

*Mistletoe*: It has been determined that the host tree has an important influence on the content of phenolic compounds in mistletoe. All species of mistletoe have a high content of procatechinic acid, p-hydroxybenzoic acid, caffeic acid, ferulic acid and sinapic acid. They are found free, esterified or glycosylated (Nazaruk and Orlikowski, 2015). The mistletoe grown on *Sorbus aucuparia* has a high content of salicylic acid, while the mistletoe grown on apple has a high content of rosmarinic acid (Vicas et al., 2011).

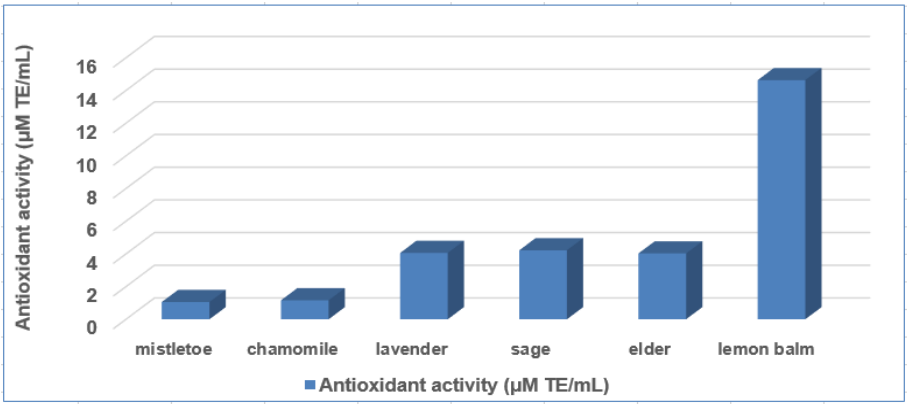


Figure 2. Antioxidant activity in studied medicinal plants

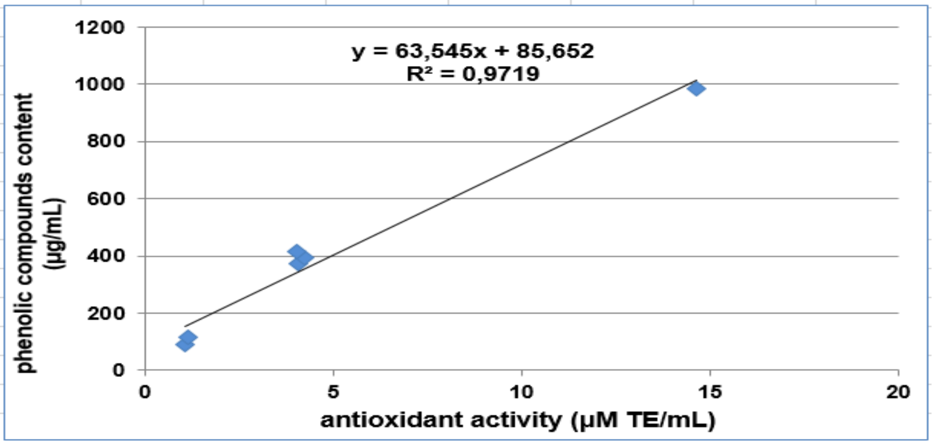


Figure 3. Correlation between antioxidant and phenolic compounds content

The highest content in total phenolic compounds and flavonoids was found in mistletoe grown on *Fraxinus excelsior*. The flavonoids isolated from the methanolic extract contain chalcones and flavanones, all found in glycosylated form and having methoxy groups in the molecule.

*Antioxidant activity:* All the medicinal plants studied show high values of the antioxidant activity against the DPPH radical (figure 2).

The results indicate the highest antioxidant activity for lemon balm (14.63 μM TE/mL). The values vary between 1.05 and 14.63 μM TE/mL and decrease in the order: lemon balm, sage, lavender, elder, chamomile, mistletoe.

Positive correlations (r=0.97) were observed between the antioxidant activity determined with DPPH and the content of phenolic compounds (figure 3), which indicates that in these samples the antioxidant activity is due to the content of phenolic compounds.

## CONCLUSIONS

All the medicinal plants studied showed significant antioxidant activities and high content of phenolic compounds. The results obtained show that the lemon balm (*Melissa oficinalis*) plant presented the highest values for the investigated biochemical indices, which recommends these teas as a source of phenols and antioxidants.

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