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# ASSESSMENT OF TOTAL PHENOLIC, TOTAL FLAVONOID, METAL CONTENTS AND ANTIOXIDANT ACTIVITIES OF *Trametes versicolor* AND *Laetiporus sulphureus*

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## ABSTRACT

In this study, total phenolic and flavonoid contents, biologically important antioxidant activities and precious metal contents of *Trametes versicolor* (L.) Lloyd and *Laetiporus sulphureus* (Bull.) Murrill fruiting bodies collected from Kastamonu province with their unique appearances and growing environments were determined. The content of phenolics in methanolic extracts expressed in gallic acid equivalents varied between 77.41 ±1.10 and 272,70 ±2.26 mg/g for *T. versicolor* and *L. sulphureus*, respectively. Also, the content of flavonoids in methanolic extracts expressed in quercetin equivalents varied between 13.82 ±0.21 and 44.29 ±0.54 mg QE/mg for *T. versicolor* and *L. sulphureus*, respectively. Antioxidant activity of mushroom extracts were expressed as percentage of DPPH radical inhibition and $IC_{50}$ values. Percentage of inhibition ranged from 15.83% to 61.03% with $IC_{50}$ value of 5.33 mg/mL for *L. sulphureus*, while 7.27% to 20.47% with $IC_{50}$ value of 18.10 mg/mL was obtained for *T. versicolor*. In addition, mushroom samples were examined for metal content by ICP-OES. While the most abundant precious metals of fruiting bodies were Ca (23.91 ±0.14), P (17.11 ±0.05), Mg (6.77 ±0.02) and Fe (3.84 ±0.02) as mg/kg DW for *T. versicolor*, they were P (24.52 ±0.09), Mg (4.59 ±0.01), Ca (0.49 ±0.01), and Fe (0.49 ±0.02) as mg/kg DW for *L. sulphureus*. The results showed that these two mushroom species rich in total phenolic contents can also be a very valuable source of P and Mg. While heavy metals Cu (123.93 ±0.30), Ni (180.99 ±0.64), Pb (54.62 ±0.58), and Cr (35.27 ±0.33) were found as µg/kg DW for *T. versicolor*, Cu (36.36 ±0.53), Ni (41.51 ±0.86), Pb (3.50 ±1.26), and Cr (8.23 ±0.55) were found as µg/kg DW for *L. sulphureus*.

**Key words:** phenolic content, total flavonoid, metal content, antioxidant activity, *Laetiporus sulphureus*, *Trametes versicolor*

## INTRODUCTION

Molecules with antioxidant properties that can be synthesized in the body take part in very important tasks such as scavenging free radicals against oxidative damage in cell metabolism, chelating prooxidant metal ions, and inhibiting prooxidative enzymes [Bakır et al. 2017]. Therefore, taking molecules with antioxidant properties from outside into the body is extremely important, especially for the immune system [Turkoğlu et al. 2007]. Antioxidant activities of mushrooms, which are also rich in proteins and carbohydrates, and low in fat and calories, are closely related to their polyphenolic and flavonoid contents [Pop

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et al. 2018, Stojanova et al. 2021]. Regarding their medicinal values, some wild mushrooms used as both food and medicine have an important place as a source of antioxidant compounds, which correlate well with their total phenolic and flavonoid contents [Turfan et al. 2018, Rašeta et al. 2020].

Although wild mushroom species have extensively been researched in the Northern Hemisphere, little has still been known about the phenolic contents and antioxidant properties of mushrooms in Turkiye, which has a great potential for edible wild mushrooms [Bakır et al. 2018a, Bozdoğan et al. 2018, Bulam et al. 2018]. In addition, the effect of chemical composition and metal content on the antioxidant properties of both edible wild and cultivated mushrooms was previously less studied in Turkiye [Bakır et al. 2017, Karadeniz et al. 2019, Şihoğlu Tepe 2021].

In terms of mineral composition, while the most abundant macro elements existed in wild growing and cultivated mushrooms were stated as potassium, phosphorus, sulphur, magnesium, calcium, and sodium, the most common micro elements were reported as iron, zinc, aluminium, copper, and manganese with a trace element of selenium having antioxidant activity. Furthermore, the accumulation of various heavy metals such as aluminium, arsenic, cadmium, chromium, iron, lead, manganese, mercury, nickel, selenium, and zinc in edible wild and cultivated and medicinal mushrooms has previously been investigated worldwide as well as in Turkiye. Both metal and heavy metal accumulations in wild mushrooms change depending on environmental conditions and intrinsic factors of mushrooms, and analytical methods studied [Bakır et al. 2017, Turfan et al. 2018, Kalač 2019, Bulam et al. 2019a, b, Keskin et al. 2021].

In this study, it was aimed to find an answer to the question of whether medicinal *Trametes versicolor* (L.) Lloyd and edible *Laetiporus sulphureus* (Bull.) Murill grown in Turkiye could be evaluated as a medicinal and functional food with their antioxidant properties, and phenolic and flavonoid contents. For this purpose, the total phenolic compounds and flavonoids were determined by spectrophotometric methods, the antioxidant activities of these two mushrooms were investigated by DPPH radical scavenging method, and the precious metals for human health and heavy metal contents were elucidated by the ICP-OES method.

## MATERIAL AND METHODS

**Reagents used.** The reagents and chemicals used in the experimental preparations and analysis processes were purchased from Merck KGaA (Darmstadt, Germany) and included methanol for the extraction study and determination of total flavonoids, DPPH (1,1-diphenyl-2-picrylhydrazyl) for the antioxidant activity assays, Folin-Ciocalteu's phenol reagent, gallic acid (3,4,5-Trihydroxybenzoic acid), and carbonic acid disodium salt ($Na_2CO_3$, anhydrous, powder) for the determination of total phenolic content, aluminium (III) chloride ($AlCl_3$) and Quercetin, ≥95% (HPLC), solid (3,3′,4′,5,6-Pentahydroxyflavone, 2-(3,4-Dihydroxyphenyl)-3,5,7-trihydroxy-4H-1-benzopyran-4-one) for the determination of total flavonoids, and nitric acid, ACS reagent, ≥90.0% ($HNO_3$) and hydrogen peroxide solution, 30% (w/w) in $H_2O$, containing stabilizer ($H_2O_2$) for preliminary preparation of metal content analysis.

In this study, deionized water (18.2 MX/cm) obtained from Milli-Q system (Human Power I Plus, Korea) was used to prepare all aqueous solutions. Absorbances were measured using a 1 cm thick pair of identical quadruple baths using a SHIMADZU UVM-1240 UV-VIS spectrophotometer (manufactured by Shimadzu Corp., Kyoto, Japan).

**Collection of mushrooms.** Fresh mushrooms (*T. versicolor* and *L. sulphureus*) were collected from the districts of Inebolu, and Devrekani in Kastamonu province of Turkiye. Mushroom species have been identified according to classical herbarium taxonomy methods [Phillips and Reid 2006]. The collected mushrooms were divided into 100 g portions. The samples were then dried in chamber dryer (NUVE KD 400, Turkiye) at 30°C for 48 h. Dried mushrooms were homogenously pulverized for usage in the analyzes.

**Preparation of mushroom extracts.** Mushroom extracts were prepared according to a standard protocol with minor modifications [Bakır et al. 2018 a, b]. The prepared mushroom material (1 g) was transferred to dark-colored bottles and then dissolved in 10 mL of 75% methanol solution. The mixture was allowed to stand at 25°C for 5 h. The resulting homogenate was centrifuged at 5000 rpm for 10 min (at 18°C). The supernatant from this process was again centrifuged at 7500 rpm for 10 min (at 4°C) [Lee et al. 2004]. The

final supernatant was removed (100 mg/mL) and used for the measurements of total phenolic and flavonoid contents and for DPPH analysis.

**Total phenolic content analysis.** The total phenolic contents of methanolic extracts were determined by using the methods reported by Slinkard and Singleton [1977] and Chandler and Dodds [1983] which included Folin-Ciocalteu reagent and gallic acid as standard. For the application of this method, 4.5 mL of deionized water and 0.1 mL of Folin-Ciocalteu reagent were added. After 3 min, 0.3 mL $Na_2CO_3$ (2%) solution and 0.1 mL extract solution were added and vigorously shaken. After a 2 h waiting period, the absorption was measured at 760 nm. The concentrations of the phenolic compounds were calculated according to the following equation, obtained from the standard gallic acid graph:

$$\text{Absorption} =$$

$$= 0.553 \text{ Gallic Acid } (\mu g) + 0.060, R^2 = 0.946$$

**Total flavonoid content analysis.** Total flavonoid content was determined by using the method reported by Arvouet-Grand et al. [1994]. For this analysis, 3 mL of methanol (2%) of aluminium trichloride ($AlCl_3$) solution and 3 mL of mushroom extract were mixed. After 15 min, the absorbance was read against blank sample at 415 nm. Total flavonoid compound concentrations were calculated according to the following equation obtained from the standard quercetin graph:

$$\text{Absorption} =$$

$$= 0.014 \text{ Quercetin } (\mu g) + 0.043, R^2 = 0.945$$

**Antioxidant activity analysis.** In this study, DPPH (1,1-diphenyl-2-picryl-hydrazyl) radical scavenging method was used for the antioxidant activity determination of mushroom samples. For this aim, methanolic extracts of mushroom samples were prepared and their ability to capture free DPPH radicals was determined using spectrophotometric method. According to this method, the maximum absorbance of the purple-colored solution of the DPPH radical is measured at 515 nm and the progress of the reaction can be monitored with decreasing color change due to the antioxidant content of the mushroom samples [Bozdogan et al. 2018].

In this study, DPPH solution at concentration of $3.08 \times 10^{-5}$ M was used as control solution. The absorbance change in the solution was measured at four different concentrations for each added mushroom extract at 1.66–6.66 mg/mL. The percent radical scavenging activity was calculated by the following formula: % inhibition = $[(A_0 - A_1) / A_0] \times 100$, where $A_0$ is the absorbance in the presence of control absorbance and $A_1$ samples. The amount of antioxidant required to reduce the initial DPPH concentration by 50% is referred to as the antiradical activity and is termed $IC_{50}$ (mg / mL) [Frankel and Meyer 2000]. The $IC_{50}$ value was determined from the graphic slope by the formula "y = mx + c", from the plot obtained for the standard Trolox and mushroom extracts [Mukherjee et al. 2011].

**Metal content analysis.** For the analysis of metal contents, dried mushroom samples were homogenized and stored in polyethylene bottles. Mushroom samples were prepared by applying a different method than the extract method used in the total phenolic and flavonoid contents and antioxidant activity measurements mentioned earlier. For this purpose, 0.5 g sample was extracted with 7 mL of $HNO_3$ (65%) and 1 mL of $H_2O_2$ (30%) for 30 min in a microwave extraction system to obtain mushroom extracts, and finally diluted to 50 mL with deionized water. The microwave system was carried out with temperature increased to 200°C within 15 min and kept constant for 15 min [Yamaç et al. 2007]. Then, the mushroom extracts were analyzed in SpectroBlue ICP-OES for metal and heavy metal (Co, Cu, Cd, Pb, Ni, Cr, Na, Ca, Al, Fe, Zn, Ba, P, Mg, As, Mn, B) determinations.

**Statistical analysis.** The significant relationship between the antioxidant concentration of mushroom and total phenolic and flavonoid contents was calculated by using descriptive statistical analysis with SPSS software for Windows version 13.0 (SPSS Inc., Chicago, IL, USA).

## RESULTS AND DISCUSSION

The results of total phenolic and flavonoid contents of two basidiomycetes species are presented in Table 1. The total phenolics were found in gallic acid equivalents (GAE) as 77.41 and 272.70 mg/g for *T. versicolor* and *L. sulphureus*, respectively. Pop et al. [2018] investigated total polyphenol content of

*T. versicolor* harvested from Romania and the concentration of total polyphenol content was 15.40, 46.22 and 8.18 mg GAE/100g fresh weight (FW) for methanolic, water, and acetone extracts, respectively. Their result was almost one-fifth of the one identified in this study for methanolic extract of *T. versicolor*. In another study, the total phenol content of polysaccharide extracts, after dialysis and reprecipitation, of *T. versicolor* was found to be 1.8 g/100 g dry weight (DW) [Kozarski et al. 2012]. On the other hand, Klaus et al. [2013] found lower total phenolic contents as 0.3 and 0.7 g/100 g DW for hot water and hot alkaline polysaccharide extracts of *L. sulphureus*, respectively. Sułkowska-Ziaja et al. [2012] reported total contents of phenolic compounds as 10.40 mg GAE/g DW for *L. sulphureus*. In addition, the content of flavonoids in methanolic extracts expressed in quercetin equivalents (QE) varied between 13.82 and 44.29 mg/mg for *T. versicolor* and *L. sulphureus* in present study, respectively. Rašeta et al. [2020] detected the most total phenol and total flavonoid contents of *T. versicolor* as 163.5 mg GAE/g DW and 37.8 mg QE/g DW in ethanol extracts, respectively. However, total flavonoid content could not be found in methanolic extracts of *T. versicolor* by Pop et al. [2018]. While Stojanova et al. [2021] found total phenol content in aqueous and ethanolic extracts of *T. versicolor* as 12.88 and 10.31%, respectively, they detected total flavonoid content in the same mushroom extracts as 5.95 and 7.52%, respectively. In another study, Turfan et al. [2018] determined total phenolics and total flavonoids of *L. sulphureus* as 28.68 and 12.81 mg/g, respectively. Furthermore, the antioxidant activity of mushrooms has usually closely been related with their total phenolic and flavonoid contents [Olennikov et al. 2011, Klaus et al. 2013, Petrović et al. 2014, Pop et al. 2018, Turfan et al. 2018, Rašeta et al. 2020, Stojanova et al. 2021, Şihoğlu Tepe 2021].

DPPH free radical scavenging activity expressed as $IC_{50}$ (mg extract/ mL) values are presented in Table 1. $IC_{50}$ values are very useful in comparing the antioxidant activities of mushroom's extracts with a negative correlation. The results of Table 1 show that *L. sulphureus* has higher values in terms of total phenolics and flavonoids than *T. versicolor* and has a better effect in terms of radical scavenging activity parallel to these values. While Pop et al. [2018] determined antioxidant activity of DPPH assay for methanolic extract of *T. versicolor* as 0.213 mM Trolox equivalents (TE)/ 100 g FW, Rašeta et al. [2020] found the most antioxidant activity of DPPH assay for water extract of the same species as 11.9 $IC_{50}$, µg/mL. Kozarski et al. [2012] determined $EC_{50}$ value of *T. versicolor* polysaccharide extracts as 0.23 mg extract/mL regarding scavenging ability on DPPH radicals. Stojanova et al. [2021] found $IC_{50}$ values of aqueous and ethanolic extracts of *T. versicolor* in terms of DPPH as 0.04 and 0.27 mg/mL, respectively. In another study, Orhan and Üstün [2011] reported total phenolic contents for *L. sulphureus* and *T. versicolor* as 9.78 and 9.58 mg/g extract, respectively, and inhibition percentages against DPPH radical as 3.88% and 2.97%, respectively. Sevindik et al. [2018] detected DPPH free radical scavenging percentage of *L. sulphureus* as 66.33, 56.44, 47.72 and 28.6% in the concentrations of 100, 75, 50 and 25%, respectively. These results confirmed that *L. sulphureus* had higher total phenolic and flavonoid contents and antioxidant activity values than *T. versicolor*, as it was found in present study.

It was found that inhibition values of both mushrooms' methanolic extracts increased with concentration. Inhibition values in the concentrations of 1.66–

**Table 1.** Total phenolic and total flavonoid contents, $IC_{50}$ values, and concentration equations of *T. versicolor* and *L. sulphureus* calculated by the DPPH method at different concentrations*

| | Total phenolic mg GAE/g | Total flavonoid mg QE/mg | $IC_{50}$ mg/mL | Concentration equations | $R^2$ |
|---|---|---|---|---|---|
| *T. versicolor* | 77.41 ±1.10 | 13.82 ±0.21 | 18.10 ±0.10 | y = 2.571x + 3.477 | 0.984 |
| *L. sulphureus* | 272.70 ±2.26 | 44.29 ±0.54 | 5.33 ±0.05 | y = 8.951x + 2.272 | 0.992 |

*Data are expressed as mean value ± standard deviation (SD) of three parallel measurements.

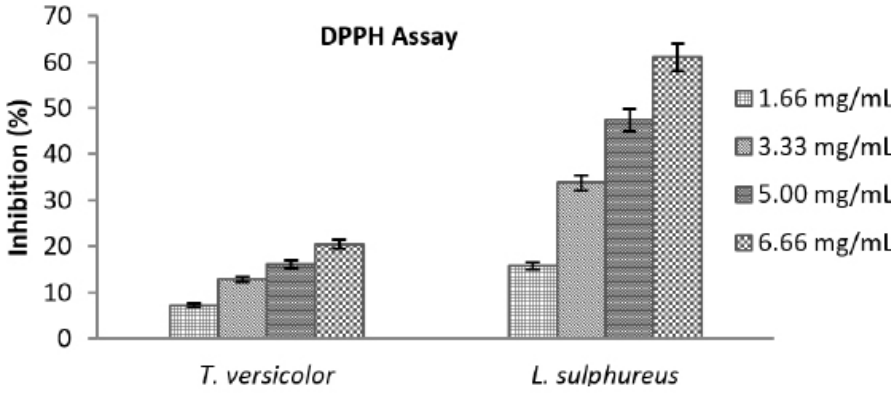


**Fig. 1.** Free radical-scavenging capacities of the extract measured in different concentrations (measured by DPPH assay). The calculated results are given as mean ±SEM (standard error of the mean)

6.66 mg/mL were 7.3%, 12.9%, 16.1%, 20.5% for *T. versicolor*, respectively, and 15.8%, 33.8%, 47.4%, 61.0% for *L. sulphureus*, respectively (Fig. 1).

Accordingly, the % inhibition values demonstrated that at each concentration *L. sulphureus* exhibited a more effective inhibition against the DPPH radical in present study. Stojanova et al. [2021] determined antioxidant capacity in terms of capture of DPPH radicals in aqueous and ethanolic extracts of *T. versicolor* as 38.04–80.66% and 39.95–80.17%, respectively. On the other hand, Olennikov et al. [2011] detected total phenolic contents and DPPH antiradical activities of ten *L. sulphureus* ethanolic extracts as 1.44–5.34 mg/g and 98.16–904.04 $IC_{50}$, µg/mL, respectively. In the same study, they also found the DPPH antioxidant activity of phenolic compounds extracted from *L. sulphureus* as 0.98–24.88 $IC_{50}$, µg/mL. Both Stojanova et al. [2021] and Olennikov et al. [2011] reported that the high free radical scavenging activity and total antioxidant activity might be due to the presence of phenolic and flavonoid type compounds.

A relative correlation was obtained between the inhibition percentages calculated after the measurements with the DPPH method and the concentration and showed good significance at ($p < 0.01$, $n = 4$) values. Pearson's correlation coefficients (r) of 0.995 and 0.997 between concentration and percent inhibition were obtained for *T. versicolor* and *L. sulphureus*, respectively. The 95% confidence interval of the difference test for the concentration-inhibition pairs was at a level of significance $p < 0,05$ for *T. versicolor* and *L. sulphureus*. The standard deviation value (SD) for concentration-inhibition comparisons was found at 2.152. Statistically significant correlation coefficients between total phenolic and total flavonoid contents, which are directly responsible for the change in antioxidant properties, and $IC_{50}$ values, which are important for the comparison of antioxidant activity, were found to be significant ($p < 0.01$, $n = 2$) for both mushrooms ($r = –1.00$). Therefore, the negative correlation found confirmed the inverse relationship between $IC_{50}$ and total phenolic and total flavonoid contents. Consequently, a statistically significant ($p < 0.01$) correlation coefficient ($r = 1.00$) was found between the total phenolic contents and the total flavonoid contents for all mushrooms.

In terms of mineral composition, seventeen metals (Na, Ca, Al, Fe, Zn, Ba, P, Mg, As, Mn, B, Co, Cu, Cd, Pb, Ni, and Cr) were determined for two mushroom species. Element concentrations are presented in Table 2. According to the results, the most abundant elements were Ca, P, Mg, and Fe, respectively, for *T. versicolor*. These were followed by Na and Al and the other minor ones were Ni, Cu, Pb, and Cr. For *L. sulphureus*, the most abundant elements were P, Mg, and Na, respectively and other minor elements followed a similar order as if they were *T. versicolor.* The correlation coefficients between two mushroom species in this study were found ($r = 0.891$) for 17 metals and it was significant at ($p < 0.01$). Significance

**Table 2.** Levels of elemental contents for *T. versicolor* and *L. sulphureus**

| Element | Element contents in mushroom species (mg/kg DW) | |
|---|---|---|
| | *T. versicolor* | *L. sulphureus* |
| Na | 2.09 ±0.006 | 1.88 ±0.01 |
| Ca | 23.91 ±0.14 | 0.49 ±0.01 |
| Al | 1.45 ±0.01 | 0.89 ±0.01 |
| Fe | 3.84 ±0.02 | 0.49 ±0.02 |
| Zn | 0.29 ±0.001 | 0.21 ±0.001 |
| Ba | 0.06 ±0.001 | 0.07 ±0.09 |
| P | 17.11 ±0.05 | 24.52 ±0.09 |
| Mg | 6.77 ±0.02 | 4.59 ±0.01 |
| As | 0.01 ±0.001 | 0.01 ±0.001 |
| Mn | 0.18 ±0.001 | 0.03 ±0.001 |
| B | 0.06 ±0.001 | 0.04 ±0.002 |
| Element | Element contents in mushroom species (µg/kg DW) | |
| | *T. versicolor* | *L. sulphureus* |
| Co | 1.52 ±0.04 | 1.11 ±0.06 |
| Cu | 123.93 ±0.30 | 36.36 ±0.53 |
| Cd | 2.58 ±0.06 | 2.88 ±0.06 |
| Pb | 54.62 ±0.58 | 3.50 ±1.26 |
| Ni | 180.99 ±0.64 | 41.51 ±0.86 |
| Cr | 35.27 ±0.33 | 8.23 ±0.55 |

*Data are expressed as mean value ± standard deviation (SD)

values of the results of the intermetallic One-Sample Test were found to be $p < 0.05$ for both mushrooms. In the literature, there have been very few studies showing the change in antioxidant properties with the metal contents of mushrooms [Bakır et al. 2017, Karadeniz et al. 2019].

All samples were analyzed by an ICP-OES to obtain the concentration of metals and heavy metals. In this study, while maximum and minimum metal contents of mushrooms were found as mg/kg DW for Na (1.88–2.09), Mg (4.59–6.77), P (17.11–24.52), Ca (0.49–23.91), and Fe (0.49–3.84), respectively, the maximum and minimum heavy metal contents of mushrooms were determined as µg/kg DW for Cr (8.23–35.27), Ni (41.51–180.99), and Cu (36.36–123.93), respectively. Kalač [2019] previously reported the lowest and highest metal contents of both edible wild and cultivated mushrooms for Na, Mg, P, Ca, and Fe as 50–750, <500–1500, 2500–10.000, 50–750 and 50–300 mg/kg DW, respectively. In this study, Na, Mg, P, Ca, and Fe contents of two mushrooms were detected lower than general content in the literature. Although *T. versicolor* has been used for traditional and modern medicine applications for a long time, information about the element contents of *T. versicolor* fruiting bodies is very rare in the literature. Therefore, present study is important in terms of determination of mineral composition of *T. versicolor* carpophores. In a study, macro elements of *T. versicolor* were determined in mg/kg for Na, Ca, and Mg as 214.6, 161.62 and 133.54 by Akgul et al. [2017], respectively. Keskin et al. [2021] found K, Ca, and Mg as 1461, 622.7 and 381 mg/kg DW for *T. versicolor*, respectively. Ayaz et al. [2011] reported K, Ca, Mg, and Na for *L. sulphureus* as 18.500, 4200, 2100 and 285 mg/kg DW, respectively. In another study, Tur-

fan et al. [2018] detected Na, Mg, P, and Ca as 8.00, 16.86, 1524.50 and 18.78 mg/kg for *L. sulphureus*, respectively. Mineral bioaccumulation in mushrooms was generally dependent on the genetic factors, mushroom species/strains and their metabolisms, their habitats and growing conditions, environmental factors, geographical variations, collection time and maturity stages, preservation methods and cooking treatments, storage conditions after collecting/harvesting, part of the mushroom analyzed and analytical methods, cultivation techniques but also strongly influenced by the chemical composition of the substrates/soil [Turfan et al. 2018, Bulam et al. 2019a, b, Kalač 2019, Karadeniz et al. 2019, Bulam et al. 2021].

Heavy metals in both edible wild and cultivated mushrooms were stated for Cr, Ni, and Cu as 0.5–10, <1–15 and 20–100 mg/kg DW, respectively, by Kalač [2019]. In present study, Cr, Ni, and Cu contents of two mushrooms were determined lower than general content in the literature. In a study, heavy metal contents of *T. versicolor* were found in mg/kg for Fe, Zn, and Cu as 154.34, 15.68 and 8.94 by Akgül et al. [2017], respectively. Keskin et al. [2021] determined Fe, Zn, Cu, Cd, Hg, and As as 73.9, 11.4, 6.80, 0.19, 14.58 and 0.68 mg/kg DW for *T. versicolor*, respectively. Fe, Zn, Cu, and Cd were detected as 28.6, 38.6, 2.8 and 0.33 mg/kg DW for *L. sulphureus* by Ayaz et al. [2011], respectively. However, Co, Ni, and Pb metals were not detected in *L. sulphureus* in the same study. Sevindik et al. [2018] reported Fe, Zn, Cu, and Pb contents of *L. sulphureus* as 138.44, 47.42, 1.90 and 1.73 mg/kg FW, respectively. In another study, Turfan et al. [2018] determined Cr, Cu, Ni, and Fe contents of *L. sulphureus* as 4.03, 14.35, 9.54 and 80.62 mg/kg, respectively. The toxic heavy metal (As, Co, Cu, Cd, Pb, Ni, and Cr) concentrations of both *T. versicolor* and *L. sulphureus* in present study were lower than those of the essential elements and acceptable for human consumption in terms of nutritional and toxic levels stated by Ayaz et al. [2011] and Kalač [2019]. It was also stated that toxic metal bioaccumulation in fruiting bodies of mushrooms was affected by environmental factors such as the amounts of organic matter, the metal content, and pH of the soil and culinary treatments and intrinsic factors regarding the taxonomic position, mycelium age, and developmental stage of the mushroom [Bakır et al. 2017, Kalač 2019, Keskin et al. 2021].

## CONCLUSIONS

DPPH radical scavenging activity was determined from methanolic extracts of *T. versicolor* and *L. sulphureus* and evaluated by comparing with total phenolic and flavonoid contents. In conclusion, *L. sulphureus* showed better scavenging activity and reducing power than *T. versicolor* in terms of both total phenolic and flavonoid contents and inhibition values. It was shown that the antioxidant activity of two mushrooms had a very strong positive correlation with their total phenolic and flavonoid contents. In addition, metal contents of these mushrooms, which could be utilized as natural antioxidant sources due to their bioactive compounds and mineral contents, were experimentally investigated. A strong positive correlation was also found between two mushroom species and their 17 metal contents. It was thought that both *T. versicolor* and *L. sulphureus*, their extracts, and bioactive compounds, which were also rich in P and Mg, could be used in traditional and complementary alternative medicine and drug, dietary supplement, and nutraceutical formulations. Furthermore, they could be of potential interest as novel natural preservatives/replacers to produce and package functional foods as well as human healthcare, anti-aging products, and cosmetics in near future.

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