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Research Article

OXIDATIVE STRESS INDEX AND ANTIOXIDANT CAPACITY OF *LEPISTA NUDA* COLLECTED FROM GAZİANTEP/TURKEY

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ABSTRACT

The objective of the present study was to determine antioxidant capacity and oxidative stress index of *Lepista nuda* (Bull.) Cooke mushrooms collected in Şahinbey (Gaziantep/Turkey) using novel research kits that could conduct high sensitive and reliable measurements with in vitro analysis methods. Its antioxidant activity was determined by DPPH method and TAS (total antioxidant status), TOS (total oxidant status) and OSI (oxidative stress index) values were determined using Rel Assay kits. Study findings determined that the TAS value was 3.102 mmol/L, the TOS value was 36.920 µmol/L and the OSI value was 1.190 for the mushroom. It was observed that the radical scavenging activity of DPPH increased in higher concentrations. It was concluded that *L. nuda* samples collected in regions with low TOS values could be used as better antioxidant sources.

Keywords: *Lepista nuda*, DPPH, antioxidant, oxidant, oxidative stress.

1. INTRODUCTION

Mushrooms are living organisms with cosmopolitan propagation [1]. Mushrooms now have a wide range of uses due to their nutritional and medical properties [2]. Mushrooms are consumed as a valuable nutrient since they contain low calories and low-fat content and high amounts of protein, fiber, vitamins and essential amino acids. Furthermore, the search for different tastes has increased the consumption of mushrooms [3,4]. Besides nutritional properties, mushrooms are also used for medical purposes. Mushrooms could produce various seconder metabolites including phenolic compounds, polyketides, terpenes and steroids [5,6]. These seconder metabolites that do not have nourishing properties are significant for their medical potential [7]. These seconder metabolites could have antibacterial, antifungal, antioxidant, antiviral, antitumor, antiallergenic, hypoglycemic, anti-inflammatory and immune system boosting action [8-11].

Lepista nuda (Bull.) Cooke is an edible mushroom, commonly seen in late summer-autumn and rarely spring, native to Europ e and North America. It grows in forests, parks and gardens which in nitrogen-rich soils [12,13]. In Turkey, many people consumes with relish by collecting this mushrooms. In the present study, total antioxidant status (TAS), total oxidant status (TOS),

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oxidative stress index (OSI) and DPPH free radical scavenging activity of *L. nuda* mushrooms collected in Şahinbey (Gaziantep/Turkey) were determined using "Rel Assay" brand commercial kits that contain highly reliable tests.

2. STUDIES

Laboratory Studies

Lepista nuda mushroom samples used in the study were collected in Şahinbey district (Gaziantep/Turkey). The morphological and ecological characteristics of the samples were recorded in the field conditions. Microstructural data were observed with a light microscope magnification, trade using Congo Red and 3% KOH solution. Identification of the samples was conducted according to Breitenbach and Kränzlin [12], Dähncke [14] and Roux [15]. After the collected mushroom samples were dried in the incubator at 40 °C. Dried mushrooms were pulverized with a mortar. 30 g pulverized material was weighed and placed into cartridges and the extraction was conducted with ethanol (EtoH) in a Soxhlet apparatus (Gerhardt EV 14). The extracts were condensed under pressure with a rotary evaporator (Heidolph Laborota 4000 Rotary Evaporator) and stored at 4 °C until they were tested.

Determination of TAS, TOS and OSI Values

Mushroom sample TAS and TOS values were determined using a Rel Assay brand commercial kit (Rel Assay Kit Diagnostics, Turkey). Trolox was used as the calibration standard for TAS, and the results are shown as mmol Trolox equiv./L. Hydrogen peroxide was used as the calibration standard for TOS, and the results are shown as μmol H_2O_2 equiv./L [16,17]. The TOS/(TASx10) formula was used to calculate OSI (arbitrary unit: AU), which indicates the percentage of oxidant compounds that are tolerated by antioxidant compounds [17].

Determination of Antioxidant Activity

When DPPH free radical scavenging activities of mushrooms were tested, stock solutions containing 1 mg/mL extract were prepared using Dimethyl Sulfoxide (DMSO). 50μL prepared fungal extract solution was added to 160μL 0.039% DPPH solution and allowed to incubate in the dark at ambient temperature for 30 minutes. Absorbance values were read at 517 nm after incubation [18]. Procedures for all concentrations were repeated separately. Caffeic acid and rosmarinic acid were also used as reference antioxidant compounds. Then DPPH free radical scavenging activities of fungal extracts were calculated with the formula below:

% DPPH scavenging = 100 x [(Abs Sample+DPPH)-(Abs Sample Blank)]/[(Abs DPPH)-(Abs Solvent)]

3. CONCLUSION

TAS, TOS and OSI Values

L. nuda TAS (mmol/L), TOS (μmol/L) and OSI values were determined with Rel Assay kits. Study findings demonstrated that *L. nuda* TAS value was 3.102 mmol/L and the TOS value was 36.920 μmol/L. The OSI value was calculated as 1.190. In studies conducted about the oxidative stress on mushrooms, it was determined that the TAS value of *Tricholoma terreum* (Schaeff.) P. Kumm was 0.38, the TOS value was 16.76 and the OSI value was 4.41 [19]. TAS value of *Fomitopsis pinicola* (Sw.) P. Karst was 1.57, the TOS value was 2.03 and the OSI value was 0.13 [20]. TAS value of *Geastrum pectinatum* Pers was 1.278, the TOS value was 13.858 and the OSI value was 1.084 [21]. Furthermore, the TAS value of *Pleurotus eryngii* (DC.) Quél. was 1.93, the TAS value of *Auricularia polytricha* (Mont.) Sacc. was 0.93 and the TAS value of *Macrolepiota procera* (Scop.) Singer was 2.823 in various studies conducted previously [22-24]. The TAS and TOS results for *L. nuda* were higher than those of the previous studies conducted by different researchers wit h different mushroom species. Higher TAS levels indicate that the mushroom has

a higher antioxidant capacity. However, a higher TOS value is indicative of higher levels of oxidant compounds produced by environmental and metabolic factors in the mushroom. OSI value indicates the percentage tolerance of the oxidant compounds formed by the environmental and physical effects by the organism using the antioxidant compounds available internally. *L. nuda* OSI value was lower than that of the *T. terreum*. This finding demonstrated that *L. nuda* could produce more antioxidant molecules that could suppress oxidant compounds when compared to *T. terreum*. In another study, it was reported that the *Omphalotus olearius* (DC.) Singer mushrooms collected from different regions had similar TAS values, but different TOS values due to regional differences [11]. High levels of *L. nuda* TAS value suggested that this mushroom can be used as a natural antioxidant source, but when TOS and OSI values are taken into consideration, it is recommended to collect this mushroom at cleaner and safer regions.

DPPH Free Radical Scavenging Activity

DPPH free radical scavenging activity was determined using mushroom EtoH extracts, and the % inhibition values are presented in Table 1.

It was reported that mushroom samples obtained from different countries such as Brazil, China, Korea, Spain, India, Portugal, Taiwan and Turkey were good antioxidant sources in studies conducted worldwide [25]. Murcia et al. [26], identified the antioxidant capacity of seven different mushrooms including *L. nuda* mushrooms collected in Spain using the linoleic acid assay method in the study they conducted. Mercan et al. [27], determined the antioxidant capacity of *L. nuda* mushroom collected in Denizli province using DPPH and Carotene/Linoleic acid systems method in their study. Elmastaş et al. [28], investigated the antioxidant activities of seven fungi species that they collected in the Black Sea Region using the DPPH and Chelating effect on ferrous ions methods in their study. As a result of this study, it was determined that *L. nuda* mushroom, which was one of the seven different mushroom species investigated, had antioxidant capacity. Barros et al. [29], identified the antioxidant capacities of seven mushroom species that included *L. nuda* and collected in Portugal using the DPPH method in 2006. In the present study, it was determined that *L. nuda* mushroom ethanol extract increased DPPH free radical scavenging activity as the extract concentration increased. In our study, it was identified that *L. nuda* had a potent antioxidant capacity despite the low activity it exhibited when compared to caffeic and rosmarinic acid used as standard.

Table 1. DPPH Free Radical Scavenging Activity Percentage

Concentration (μg/mL)	Cafeic acid (%)	Rosmarinic acid (%)	*L. nuda* (%)
25	8.62±0.91	6.03±0.15	2.79±4.21
50	21.34±0.66	47.72±0.76	12.83±2.99
75	38.39±0.66	56.44±1.98	33.78±3.25
100	54.47±0.05	66.33±1.01	50.20±5.27

*The results represent % inhibition values. Values are presented as mean ± SD; n = 3

In the present study, the antioxidant capacity of the *L. nuda* mushroom samples collected in Gaziantep - Turkey were determined using reliable and sensitive analysis methods that were not used in previous studies. It was determined that although the mushroom had antioxidant capacity, it was suggested that the samples collected from this region should be used with care due to the detected high TOS values. Furthermore, oxidant compounds that can be produced by environmental and physiological factors can vary from one region to the other. Thus, considering the oxidative stress status of the mushroom, it was suggested that the fungus should be collected from more adequate and cleaner regions for consumption.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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