



## Nutraceutical and Food Preserving Importance of *Laetiporus sulphureus*<sup>#</sup>

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ARTICLE INFO	ABSTRACT
<p><sup>#</sup>This study was presented as an oral presentation at the 4th International Anatolian Agriculture, Food, Environment and Biology Congress (Afyonkarahisar, TARGID 2019)</p> <p><i>Review Article</i></p> <p>Received : 30/05/2019 Accepted : 25/07/2019</p> <p><b>Keywords:</b> Chicken of the Woods Food Preserving <i>Laetiporus sulphureus</i> Nutritional Therapeutic</p>	<p><i>Laetiporus sulphureus</i> (Bull.: Fr.) Murr. is popularly known as “sulphur polypore” or “chicken of the woods” due to its characteristic sulphur yellow coloured polypore’s and chicken-like taste and texture. This edible wild mushroom has been traditionally consumed as a source of nutrition and folk medicine in Asia and Europe for a long time. The numerous studies have shown that <i>L. sulphureus</i> nutritionally provides various key components such as carbohydrate, essential amino acids and fatty acids, vitamins, minerals, and fibre. Besides, the extracts prepared from fruiting bodies or mycelia of this mushroom have exhibited a number of medicinal properties such as immunomodulation, antitumor, anti-inflammatory, antioxidant, antimicrobial, and antihyperglycemic activities because of their biologically active components such as phenolics, triterpenes, and polysaccharides. <i>L. sulphureus</i> is also a suitable candidate to be used as a natural food preserving source.</p>

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## *Laetiporus sulphureus*'un Nutrasötik ve Gıda Koruyucu Olarak Önemi

MAKALE BİLGİSİ	ÖZ
<p><i>Derleme Makale</i></p> <p>Geliş : 30/05/2019 Kabul : 25/07/2019</p> <p><b>Anahtar Kelimeler:</b> Orman Tavuğu Gıda Koruma <i>Laetiporus sulphureus</i> Besinsel Tedavi Edici</p>	<p><i>Laetiporus sulphureus</i> (Bull.: Fr.) Murr., kükürt sarısı poliporları ve tavuk benzeri tadı ve dokusu nedeniyle “kükürt poliporu” veya “orman tavuğu” olarak bilinmektedir. Bu yenilebilir doğa mantarı, Asya ve Avrupa’da uzun zamandan beri hem besin kaynağı hem de halk hekimliğinde geleneksel tıpta kullanılmaktadır. Çalışma sonuçları <i>L. sulphureus</i>'un karbonhidrat, esansiyel amino asitler ve yağ asitleri, vitaminler, mineraller ve lif gibi temel besin öğelerini sağladığını göstermektedir. Ayrıca, mantar veya misellerinden hazırlanan ekstraktlar fenolikler, triterpenler, polisakaritler gibi biyolojik aktif bileşenler içerdiklerinden immünomodülasyon, antitümör, anti-enflamatuvar, antioksidan, antimikrobiyal ve antihiperlipidemik aktiviteler göstermektedir. <i>L. sulphureus</i>, doğal bir gıda koruyucu olarak kullanılmaya da uygundur.</p>

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## Introduction

*Laetiporus sulphureus* (Bull.:Fr.) Murrill., also called as Sulfur Tuft, Sulfur Shelf, Chicken-of-the-Woods, and Chicken Mushroom is a member of the class Basidiomycetes (Fomitopsidaceae, Polyporales) (Weber et al., 2004; Radic et al., 2009; Grienke et al., 2014). *Laetiporus* name is a combination of two words “*laeti*” (Lat.) and “*por*” (Lat.) and refers to a hymenial layer and the size of the specifically shaped fruiting bodies. The adjective “*sulphureus*” (Lat.) originates from the characteristic color of fruiting bodies (Sulkowska-Ziaja et al., 2018). It is a wood-rotting mushroom producing shelf-shaped, bracket-like fruiting bodies of pink-orange color, except for the fleshy margin, semicircular hats with a characteristic bright sulfuric-yellow color. It commonly grows in nature from late spring to autumn (Weber et al., 2004; Sulkowska-Ziaja et al., 2018). The remarkable yellowish or orange-colour of *L. sulphureus* contains non-isoprenoid polyene known as laetiporic acid A, B, C and 2-dehydro-3-deoxylaetiporic acid A as the main pigments of natural food colorants (Weber et al., 2004; Davoli et al., 2005). Moreover, the odor of *L. sulphureus* has been differently described over the years as being more or less pleasant, fungal, and strongly musky to strongly fungoid (Rapior et al., 2000; Wu et al., 2005) that can also be

evaluated in food industry. Its fruiting bodies are very large in overlapping clusters of 5-50 cm, annual, spongy to leathery, up to 40 cm wide, with wet biomass over 40 kg and tubular hymenopores (Rapior et al., 2000; Luangharn et al., 2014b; Kovács and Vetter, 2015). They colonize in roots, butt, or heartwood of living tree trunks and decaying logs or dead trunks of deciduous species, and more rarely coniferous species (Radic et al., 2009; Luangharn et al., 2014b; Sulkowska-Ziaja et al., 2018). *L. sulphureus* is a cosmopolitan species and presents on all continents, except Antarctica. It is widely distributed in Asia, Europe and North America from tropical to subtropical zones. In certain parts of these continents, it is considered as a delicacy and can also be used as a substitute for chicken in a vegetarian diet due to its taste as meat of chicken, crab or lobster (Rapior et al., 2000; Ota et al., 2009; Petrović et al., 2013; Grienke et al., 2014). In Turkey, this species has been previously identified in different localities by some researchers (Pekşen and Karaca, 2003; Sesli, 2007; Sesli and Denchev, 2014). Fig. 1 represents photographs of the fruiting bodies of *L. sulphureus* naturally grown on the roots of rot *Prunus avium* trunk in a montane village of Keşap, Giresun, Turkey and found at the end of April.



Figure 1 Photos of rot *Prunus avium* trunk and fruiting body of *L. sulphureus* (Date: 23.04.2014)

This species is sold in the local markets and consumed in the local cuisine in Turkey as well as worldwide (Pekşen and Karaca, 2000; Pekşen et al., 2016; Mandić et al., 2018). Besides being a food, *L. sulphureus* fruit bodies have long been used in Asian folk medicine and thought to be capable of regulating the human body, improving health, and defending the body against illnesses (Ying et al., 1987; Zjawiony, 2004). Moreover, the fruiting bodies have been used for the treatment of pyretic diseases, coughs, gastric cancer, and rheumatism in Europe (Rios et al., 2012; Sulkowska-Ziaja et al., 2018). Burning of *L. sulphureus* is also used as mosquitoes and midges repellent (Ying et al., 1987). Lee et al. (2009) purified and characterized a thermostable extracellular xylanase having potential applications as bioconversion of lignocellulosic materials into fermentative products, improvement of digestibility of animal feedstock, and clarification of juices. Nutritional studies have recently confirmed that it is a sustainable food supply to growing population due to rich content of carbohydrate, protein, minerals, vitamins, polyunsaturated fatty acids, and fibre. In recent years, several primary and secondary biologically active components and extracts have been prepared from fruiting bodies or mycelia of *L.*

*sulphureus* exhibiting immunomodulation, antitumor, anticoagulation, antioxidant, antibacterial, antifungal, insecticidal, anti-ulcer, insulin tropic, anti-HIV, and cytostatic activities (Grienke et al., 2014; Khatua et al., 2017; Sulkowska-Ziaja et al., 2018). Previous studies on this species have revealed many interesting metabolites that can be utilized in biotechnological studies about medicine and food industries on larger scale. These compounds include laetiporic acids, polysaccharides and alkali-soluble polysaccharides, fatty acids, amino acids, and  $\alpha$ -(1→3)-glucans (Luangharn et al., 2014a; Khatua et al., 2017). In some other studies, *L. sulphureus* was cultivated as fruiting bodies and hyphal mycelium on a larger scale in laboratory which may open way to commercial production (Agafonova et al., 2007; Pleszczyńska et al., 2013; Luangharn et al., 2014a; 2014b). In addition, food industry has started to search for novel natural substances to prolong the shelf life of food products. Regarding food-preserving properties of *L. sulphureus*, a number of successful antifungal studies against *Aspergillus flavus* in tomato paste kept at room temperature (25°C) for 15 days (Petrović et al., 2013) and chicken pate stored at +4°C for 21 days (Petrović et al.,

2014a) with the addition of *L. sulphureus* methanolic extract have been recently carried out. Thus, the species is considered as a natural resource of nourishment and food preservation as well as drug therapy and consequently it has increasingly become popular in scientific world.

However, gastrointestinal problems, occurrence of severe adverse effects including allergic reactions, vomiting and fever have been previously reported (Jordan, 1995; Watling, 1997). *L. sulphureus* consumption has also been reported to cause visual hallucinations and ataxia. Gas chromatographic analysis of the mushroom was negative for known hallucinogens. In that particular case, the toxic effects were attributed to a combination of factors, including the patient's age, the amount ingested, and the fact that it was eaten raw (Appleton et al., 1988). This review focuses on the nutrients and constituents of *L. sulphureus* and their biological activities as well as its food preserving property.

### Nutritional Value of *L. sulphureus*

The water content in *L. sulphureus* ranged from 66.67 to 72.69% (Palazzolo et al., 2012; Saha et al., 2014) and carbohydrate content from 64.90 to 74.47 g/100 g dw (Ayaz et al., 2011; Petrović et al., 2014a; Kovács and Vetter, 2015). Olennikov et al. (2008) detected the composition of carbohydrates as mannite, free carbohydrates, water-soluble polysaccharides (WSPS), base-soluble polysaccharides (BSPS), and chitin. Free sugars identified by HPLC as trehalose was the dominant sugar (4 g/100 g dw), followed by mannitol (3.54 g/100 g dw), while fructose was present in minor amount (0.46 g/100 g dw) (Petrović et al., 2014a). Turfan et al. (2018) detected total soluble carbohydrates, glucose, fructose, and sucrose as 266.82, 40.49, 6.31 and 0.32 mg/g, respectively. Crude protein contents of *L. sulphureus* varied considerably (10.61-21.00 g/100 g dw) (Ayaz et al., 2011; Petrović et al., 2014a; Saha et al., 2014; Kovács and Vetter, 2015). Agafonova et al. (2007) reported essential amino acids of arginine, histidine, isoleucine, leucine, lysine, methionine, threonine, and tryptophan as 0.47, 0.40, 0.11, 0.52, 0.28, 0.17, 0.20, 0.33% and 0.38, 0.58, 0.13, 0.30, 0.32, 0.18, 0.24, 0.79% for the fruiting bodies of *L. sulphureus* strains LS-BG-0804 and LS-UK-0704, respectively. Turfan et al. (2018) found total soluble protein and total free amino acid amounts of *L. sulphureus* as 83.27 and 3.63 mg/g, respectively. The fat content in *L. sulphureus* ranged from 2.96 to 4.50 g/100 g dw (Palazzolo et al., 2012; Luangharn et al., 2014a; Petrović et al., 2014a; Saha et al., 2014; Kovács and Vetter, 2015). Sinanoglou et al. (2015) determined that triglycerides were the most abundant in the neutral lipid fraction, whereas phosphatidylcholine in phospholipids. Total lipids were found to contain high degree of unsaturated fatty acids (UFA/SFA>3.4) and C18:2 $\omega$ -6, C18:1 $\omega$ -9, and C16:0 fatty acids dominated. In a study about the lipid composition of two strains, unsaturated fatty acids were in higher amounts and linoleic acid was dominant (47.50 and 48.19%, respectively) followed by oleic acid (19.33 and 19.43%, respectively) (Agafonova et al., 2007). But, according to Palazzolo et al. (2012), the dominant fatty acid was oleic acid (52.35%), followed by linoleic acid (20.62%). Petrović et al. (2014a) reported linoleic acid (63.27%) as

the most abundant fatty acid, followed by oleic acid (14.52%) and palmitic acid (11.68%). Bengu (2019) determined high amounts of linoleic (7.73%), stearic (28.55%), palmitic (28.60%), and oleic acids (33.94%) in *L. sulphureus*. Ericsson and Ivonne (2009) analysed the sterol composition of this mushroom and identified cerevisterol and ergosterol peroxide in addition to conventional sterols. Energy content of *L. sulphureus* was found as 341.06-375.62 kcal/100 g dw (Ayaz et al., 2011; Petrović et al., 2014a; Kovács and Vetter, 2015). Fibre content of *L. sulphureus* ranged from 4.12 to 6.43% dw (Luangharn et al., 2014a; Saha et al., 2014; Kovács and Vetter, 2015). Ash content in *L. sulphureus* was found to be low and varied from 4.00 to 9.03 g/100 g dw (Ayaz et al., 2011; Petrović et al., 2014a; Kovács and Vetter, 2015). The macro (Ca, P, Mg, Na, K, etc.) and micro elements (Fe, Zn, Mn, Cu, Si, Mo, B, etc.) of wild and cultivated *L. sulphureus* considerably differed due to habitat, geographical location and collection time of the mushroom samples, cultivation methods and analysis methods (Agafonova et al., 2007; Ayaz et al., 2011; Durkan et al., 2011; Palazzolo et al., 2012; Luangharn et al., 2014a; Saha et al., 2014; Kovács and Vetter, 2015; Turfan et al., 2018; Bengu, 2019). Petrović et al. (2014a) indicated the tocopherols in the order of  $\alpha$ -tocopherol >  $\gamma$ -tocopherol >  $\delta$ -tocopherol. Palazzolo et al. (2012) determined niacin (4.2 mg%), pantothenic acid (0.464 mg%), biotin (4.8  $\mu$ g%), B<sub>12</sub> (0.98  $\mu$ g%), and D<sub>3</sub> (0.75  $\mu$ g%). Malic acid, ascorbic acid, citric acid, tartaric acid, malonic acid, succinic acid, oxalic acid, fumaric acid, and quinic acid have been previously detected in both wild and cultivated strains of *L. sulphureus* (Olennikov et al., 2008; Ayaz et al., 2011; Petrović et al., 2014a).

### Bioactive Compounds of *L. sulphureus* and Their Pharmacological Activities

*L. sulphureus* contains N-methylated tyramine derivatives (Rapior et al., 2000), polysaccharides, lanostane triterpenoids, laetiporic acids, and other metabolites (Alquini et al., 2004; Weber et al., 2004; Davoli et al., 2005; Radic et al., 2009; Luangharn et al., 2014a; Khatua et al., 2017; Sulkowska-Ziaja et al., 2018) that are significantly correlated with various bioactivities.

The most important medically active primary metabolites of *L. sulphureus* comprise high-molecular weight compounds as polysaccharides, proteins, and polysaccharide-protein complexes. In addition, some pigments and nucleic acids have also been described to be biologically active (Grienke et al., 2014). *L. sulphureus* fruit bodies are a rich source of  $\alpha$ -(1,3)-D-glucans and their cell wall contains up to 88% (dw) of this glucan, whereas other mushrooms contain only 9-46% (Wiater et al., 2012). Antioxidant effects were reported for water-soluble and alkali-soluble polysaccharides extracted from *L. sulphureus* (Olennikov et al., 2009a; 2009b; Klaus et al., 2013). Olennikov et al. (2009a) isolated Laetiporan A as the major 56-kDa polysaccharide, that was found to be comprised of a  $\beta$ -1,3-glucan containing mannose, galactose, fucose, xylose, and rhamnose residues at position C-6. The pure polysaccharide prevented liver lesion in toxic hepatitis model by executing strong antioxidant effect on lipid peroxidation. Crude

extracellular polysaccharides produced from a submerged mycelial culture of *L. sulphureus* provoked a hypoglycaemia effect in streptozotocin-induced diabetic rats, indicating that these substances could be useful in diabetes mellitus treatment (Hwang and Yun, 2010). Bioactivities related to bioactive proteins include antitumor, antiviral, antimicrobial, antioxidative, and immunomodulatory properties (Kang et al., 1982; Xu et al., 2011). Mushroom proteins can be structurally categorized as classical proteins/peptides (including enzymes), or lectins, i.e. carbohydrate-binding proteins. Kanska et al. (1994) characterized lectins from *L. sulphureus* in detail. A protein-polysaccharide fraction from fruit bodies of *L. sulphureus* consisting of 84% polysaccharide and 5% protein exerted antitumor activity against sarcoma 180 in mice (Kang et al., 1982). Laetiporic acids, i.e. non-carotenoid polyene pigments, identified in *L. sulphureus* fruiting bodies have shown well-known antioxidant properties and their high stability may also render them attractive as food dye (Weber et al., 2004; Davoli et al., 2005).

About 75% of bioactive secondary metabolites are composed of triterpenoids (acids, esters and lactones, alcohols, ethers and peroxides, aldehydes and ketones, glycosidic triterpenes, miscellaneous triterpenes) with different structures, whereas other secondary metabolite classes are produced to a lesser extent. The second largest group of secondary metabolites (~14%) is composed of organic acids. *L. sulphureus* also contains other compounds belonging to different chemical classes, i.e. benzofurans, flavonoids, coumarins, and N-containing compounds (Grienke et al., 2014). In general, volatile components of *L. sulphureus* can be categorized as (i) fatty acids and methyl-branched carboxylic acids, (ii) C8 compounds and benzoic volatiles, and (iii) volatile amines (Rapior et al., 2000; Wu et al., 2005; Petrović et al., 2013). More than 40 major volatiles were identified by (HR) GC-MS and gas chromatography-olfactometry (GC-O) methods. However, because of difficulties in assaying such compounds, no biological activity has been linked to these volatile components (Grienke et al., 2014). The primarily EtOH and MeOH extracts of *L. sulphureus* were evaluated for cytotoxic, antioxidant and antimicrobial effects (Keller et al., 2002; Karaman et al., 2010, 2009; Ozen et al., 2011; Turfan et al., 2018). Orhan and Üstün (2011) evaluated the acetylcholinesterase (AChE) inhibitory activities of *L. sulphureus* and possible use of its fruiting bodies in the treatment of Alzheimer's disease. Six lanostane-type triterpene acids from *L. sulphureus* were assayed in an MTT assay for their apoptotic potential against HL-60 cells (human myeloid leukaemia cells) and acetylated triterpenes showed more potent effects than non-acetylated derivatives (León et al., 2004). Lear et al. (2009) identified (7)-laetirobin benzofuran from *L. sulphureus* as a cytostatic compound with rapid cell entry. Ergosterol peroxide having potential cytotoxic activity was isolated from *L. sulphureus* (Krzyczkowski et al., 2009). Fan et al. (2014) isolated three mycophenolic acid derivatives from *L. sulphureus*. Among them, 6-((2E, 6E)-3, 7, 11-trimethyldodeca-2, 6, 10-trienyl)-5, 7-dihydroxy-4-methylphtanlan-1-one exhibited moderate cytotoxicity against HL-60, SMMC-7721, A-549 and MCF-7 cells with IC<sub>50</sub> values of 39.1, 31.1, 27.4, and 35.7 µmol/L,

respectively. He et al. (2015) obtained seven new drimane-type sesquiterpenoids, sulphureuines B-H, together with four known compounds from cultures of mushroom *L. sulphureus* and tested for their cytotoxicities against five human cancer cell lines. As a result, compound 10 showed potent cytotoxic activity against HL-60, SMMC-721, A-549, SW-480, with IC<sub>50</sub> values of 37.5, 14.8, 15.6, and 36.1 µM, respectively. Keskin et al. (2017) determined that 10% cytotoxicity was observed at 0.25 mg/ml dosage used in MCF-7 cell line. At all dosages used, viability rates were increased in L929 Fibroblast cell lines. In addition, apoptotic and necrotic effects were observed in the samples.

Turfan et al. (2018) found total phenolics and total flavonoids of *L. sulphureus* as 28.68 and 12.81 mg/g, respectively. In the study of Turkoglu et al. (2007), the fraction exhibited dose dependent DPPH radical scavenging ability as it inhibited 14, 26, 55, and 86% at the concentrations of 100, 200, 400 and 800 µg/ml, respectively. While in β-carotene/linoleic acid system, the ethanolic extract showed 57.4% of inhibition at 80 µg/ml concentration that increased to 82.2% at 160 µg/ml concentration. Klaus et al. (2013) found that, the hot alkali extract of *L. sulphureus* was the most effective in the DPPH radical scavenging, reducing power, and ferrous ion-chelating ability. Very strong correlation was also observed between α-glucan content (17.3 g/100 g dw) and the EC<sub>50</sub> values of these antioxidant activity assays as 0.5, 4.0 and 1.5 mg/ml, respectively. In another study, the extracts of *L. sulphureus* fruiting bodies and mycelia had 0.68 and 4.92 mg/ml EC<sub>50</sub> values of DPPH radical scavenging activity, respectively. In the same study, total phenolics contents of *L. sulphureus* fruiting bodies and mycelia extracts were 12.14 and 6.85 mg GAE/g, respectively (Prasad et al., 2015). Popa et al. (2016) reported that the highest total phenols amount (283.9 mg GAE/100g) was found in fruiting bodies extract among the dried fruiting bodies, dried mycelia broth, and mycelia-free broth submerged cultures. In the pharmacognostic standardization study of Acharya et al. (2016), the fraction showed potent activity in superoxide radical scavenging, DPPH radical scavenging, chelating ability of ferrous ion, reducing power, and total antioxidant assays where EC<sub>50</sub> values ranged from 0.11 to 1.38 mg/ml concentration and the fraction was composed of different bioactive compounds such as phenolics, flavonoids, carotenoids, and ascorbic acid. Beauvericin, an antimicrobial secondary metabolite including a cyclodepsipeptide, has been detected in *L. sulphureus*. Moreover, it has also possessed insecticidal and nematocidal activity (Zjawiony, 2004). In another study, results showed that the ethanol extract had a narrow antibacterial spectrum against Gram (-) bacteria, though strongly inhibited growth of Gram (+) bacteria examined. However, the fraction did not show any activity against *Klebsiella pneumoniae* at the evaluated concentrations (Turkoglu et al. 2007). In a study, results indicated that hexane extracts possessed better antifungal and slightly better antibacterial activity compared to chloroform extracts though both were less active than the commercial antimicrobial agents (Sinanoglou et al., 2015). Zhang et al. (2018) determined that antibacterial activity of ethanol, petroleum ether, and ethyl acetate extracts from *L. sulphureus* was better than antifungal activity. Gasecka et

al. (2018) reported ergosterol content of *L. sulphureus* having antioxidant activity as 0.540 mg/g dm. Petrović et al. (2014b) prepared aqueous, ethanol, and methanol extracts obtained after application of classical and ultrasound-assisted extraction methods. All fractions presented high potentials to inhibit growth of microorganisms while extracts obtained by ultrasound technique were proved to be more potent than that of classical extract. In some cases, the activity of methanolic and ethanolic formulations prepared by ultrasound technique exceeded that of commercial antibiotics. Besides antioxidant, antimicrobial, cytotoxic, and anticancer studies, some researches on other medicinal attributes of both *L. sulphureus* derived extracts and individual compounds, such as anti-HIV (Mlinaric et al., 2005), anti-malaria, anti-ulcer, hepatoprotective (Sun et al., 2014), anti-inflammation (Saba et al., 2015), immunomodulatory (Wang et al., 2019), hypolipidemic, anti-diabetic, and anti-thrombin activities have been previously reported (Radic et al., 2009; Grienke et al., 2014; Khatua et al., 2017; Sulowska-Ziaja et al., 2018).

### Food Preserving Studies

The growth of microorganisms and free-radical production are the two basic reasons of food spoilage, so the natural sources of antioxidant and antimicrobial compounds should be considered important. Being a mushroom that can be easily found, recognized in nature and easily cultured, *L. sulphureus* is a suitable material for the researches. Therefore, there have been some studies in the literature conducted on the use of *L. sulphureus* as a natural food preserving agent that could bring functional preservations due to its determined antimicrobial and antioxidant properties. In a study realized by Petrović et al. (2013), antimicrobial activity of methanol, acetone, and dichloromethane extracts from *L. sulphureus* in combination with and without 0.01 M food additive, potassium disulfite (E224) have been *in vitro* investigated against eight bacterial and eight fungal species, and methanol extract of *L. sulphureus* showing the best inhibitory activity with MIC value of 20 mg/ml was also studied for *in situ* control of *Aspergillus flavus* in tomato paste usually considered as semi-finished food. *A. flavus* has been able to produce dangerous mycotoxin of aflatoxin in foods (Kalantari et al., 2012), so it was important to assess the antifungal activity of this mushroom's extract. *In situ* results demonstrated complete inhibition of *A. flavus* growth in tomato paste after 15 days of the treatment at 25 °C with the concentrations of 15 mg/ml of *L. sulphureus* methanol extract. In another study, Petrović et al. (2014a) determined the nutritional value, bioactive compounds, *in vitro* antioxidant, antitumor, and antimicrobial properties of *L. sulphureus*. Moreover, a suitable model system with chicken pate was developed to test the *in situ* antifungal preserving properties of *L. sulphureus* methanolic extract against *A. flavus*. The inhibition of *A. flavus* growth in chicken pate was dose dependent and higher concentrations of *L. sulphureus* extract (3.56-5.49 mg/ml) completely inhibited mycelial growth after 21 days storage at 4 °C. The prevention of mould development is highly recommendable, from the slaughtering of animals to the packaging of the final

product (Mizakova et al., 2002). Because of consumers' awareness, food products preserved with natural additives are becoming more popular (Hooley and Patel, 2005). When choosing a natural antioxidant for the purposes of prolonging the shelf life of food, the impact on the sensory and taste qualities should also be taken into account. *L. sulphureus* with proven antimicrobial and antioxidant properties can functionally preserve meat products, affect their taste in a positive way, and make them acceptable to the customer (Petrović et al., 2014a).

### Conclusion

*L. sulphureus* has revealed presence of essential nutrients and can be used in low-calorie diets. Various studies have also provided evidences that *L. sulphureus* contains several bioactive components with outstanding properties to treat different types of disorders. However, most of the researches based on *in vitro* studies and few *in vivo* experimentations were only performed. Therefore, in future research, clinical studies are essential to confirm the safety, bioavailability and the effects of these mushroom derived compounds. In addition, new collaborated biotechnological studies must be conducted to be able to use *L. sulphureus* as health-promoting functional food as well as natural food additive prolonging shelf life of the foods and food safe colorant and odor compound in food industry.

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