SYSTEMATIC REVIEW
published: 30 May 2022
doi: 10.3389/fsufs.2022.882939

Systematic Review: Heat Treatments on Phenolic Content, Antioxidant Activity, and Sensory Quality of Malaysian Mushroom: Oyster (*Pleurotus* spp.) and Black Jelly (*Auricularia* spp.)

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Specialty section:
This article was submitted to Nutrition and Sustainable Diets, a section of the journal Frontiers in Sustainable Food Systems

Received: 24 February 2022
Accepted: 08 April 2022
Published: 30 May 2022

Citation:
Izham I, Avin F and Raseetha S (2022) Systematic Review: Heat Treatments on Phenolic Content, Antioxidant Activity, and Sensory Quality of Malaysian Mushroom: Oyster (Pleurotus spp.) and Black Jelly (Auricularia spp.).
Front. Sustain. Food Syst. 6:882939.
doi: 10.3389/fsufs.2022.882939

Pleurotus spp. and *Auricularia* spp. are popular species consumed by the Malaysian community. Recently, due to increased awareness, both mushrooms are also being consumed for their bioactive compounds, ergothioneine, and antioxidant properties and has been used since earlier ages as therapeutic remedies. The bioactive compounds such as phenol, flavonoid and ergothioneine found in both *Pleurotus* and *Auricularia* mushrooms were explored. Differences in heat treatments (microwave, hot air drying, and solar drying) and cooking methods may affect the content of bioactive compounds and their properties. Similarly, sensory acceptance by consumers may be affected too. Antioxidant properties using DPPH (1,1-diphenyl-2-picryl-hydrazyl) radical and FRAP (ferric reducing antioxidant power) assay of both raw and heat-treated mushrooms are included. Microwave drying retained color characteristics and bioactive compounds in both mushrooms. To add value to this review, a survey on the consumption pattern of *Pleurotus* and *Auricularia* species among Malaysians has been conducted online and concluded that *Pleurotus* species is the most considered species compared to *Auricularia* mushroom and almost half of the respondents were not aware that heat may deplete nutritional contents in mushroom despite agreeing both gave beneficial health in diets.

Keywords: black jelly mushroom, oyster mushroom, bioactive compound, heat, antioxidant

INTRODUCTION

Generally, *Pleurotus* spp. such as pearl oyster, gray oyster, pink oyster, and *Auricularia* spp., like jelly ear and black jelly mushrooms, are well-known among consumers. In addition to being low in calories, salt, fat, and cholesterol, *Pleurotus* spp. is also high in nutrients and vitamins, protein, carbohydrates, and fiber. *Auricularia* mushrooms are used in traditional Chinese medicine. Due to their bioactive ingredients, mushroom extracts have been recognized as antioxidants, anticarcinogenic, and anti-inflammatory by food and biochemical experts since 3500 BC. One of the mushrooms' bioactive compounds, ergothioneine, protects cells from oxidative stress caused by reactive nitrogen forms like nitric oxide. Furthermore, ergothioneine depletion in cells may cause DNA damage and protein and lipid oxidation (Paul and Snyder, 2010). Due to this reason, many food industries have used mushroom extracts in health care products and as added functional ingredients in food products. Heating treatments are used to extract or prepare foods for human consumption. Due to its thermolabile qualities, heating affects the residual nutrients and bioactive substances in food. Thermal processing is commonly used to extend food shelf life. As a result, heat-processed foods are regarded to be less healthful than fresh foods. Thermally processed foods, notably fruits and vegetables, have higher biological activity than non-thermally processed meals, according to recent studies (Kim et al., 2000; Dewanto et al., 2002).

In Malaysia, a few studies have shown that heat treatments on mushrooms affect phenolic chemicals, ergothioneine content, and antioxidant activity. This research benefits Malaysia's future food development, whereas nutritional imbalance has also occurred, particularly in poorer nations where animal protein is scarce and expensive, necessitating the development of an alternative protein source. Mushrooms are recognized as a vegetarian meat substitute since they are rich in nutrients.

SELECTION OF ARTICLES

The articles for this systematic review were chosen from two databases (Scopus and Google Scholar). The literature review took place between March 2021 and February 2022. The terms "heat treatment" OR "auricula" AND "pleurotus" OR "bioactive compound" were used in the search (Article title, Abstracts, Keywords). Additionally, the term "mushroom" was entered specifically in the [Search within results] section. A database search identified around 1,503 publications, and an extra 50 articles found in ScienceDirect, as displayed in **Supplementary Figure 1**.

LOCAL SURVEY ON OYSTER MUSHROOM AND BLACK JELLY MUSHROOM CONSUMPTION

An online local survey on oyster mushroom and black jelly mushroom was conducted. This survey contained two parts; Part A being a survey on Oyster mushroom consumption and Part B being a survey on Black jelly mushroom consumption. Both sections have to answer by respondents. This survey tested the respondents' general knowledge on oyster and black jelly mushrooms, including the benefits of both mushrooms, availability of both mushrooms on the market, mushroom price, and preferable dishes using the mushrooms. The type of questionnaire may vary, such as multiple-choice, open-ended, close-ended, Likert-scale, and dichotomous questions.

BACKGROUND OF RESPONDENTS

A local survey has been conducted in order to study the consumption of random Malaysians of Oyster mushroom and Black Jelly Mushroom. A total of 52 respondents have answered this survey, aged 21–60 years old. This survey has been answered by 88.5% (46) women and 11.5% (6) men, ages ranging mostly from 21 to 30 years old (61.5%), followed by 41 to 50 years old (21.2%) and 51 to 60 years old (11.5%).

OVERVIEW OF SELECTED MUSHROOM BIOLOGICAL ACTIVITIES AND CONSUMPTION PATTERN

Oyster and Black Jelly Mushroom: *Pleurotus* spp. and *Auricularia* spp.

Origin

Pleurotus species belong to a small group of higher fungi known as Basidiomycetes, which are categorized by fruit bodies with an eccentric stalk attached to the pileus, which opens up like an oyster shell during morphogenesis. *Pleurotus* is a genus of cultivated mushrooms with a wide range of species and there are over 30 species of *Pleurotus* mushroom, which have been found in wide parts of the world. The evolutionary relationship between *Pleurotus* species is still ambiguous, and several taxonomic issues remain unresolved. The earliest cultivation of oyster mushroom (*Pleurotus* species) has been recorded in Germany by Flank in 1917 (Adebayo and Oloke, 2017). *Pleurotus* species can grow in temperate and tropical zones but are more prevalent in the subtropical regions (Rajarathnam et al., 1987).

Wood ear, black jelly, tree ear, or ear fungus are local names for some *Auricularia* species. This mushroom species originated from China (Packialakshmi et al., 2017). Zhang et al. (2015) reported the cultivation of *Auricularia* mushroom started 1400 years ago in China. One *Auricularia* species it is believed existed 2000 years ago in China was *Auricularia heimuer,* it is reported in the Chinese medicinal book, "Shennong's Compendium of Materia Medica" (Sekara et al., 2015). In the Red Dragon country, a popular *Auricularia* mushroom is called "he i mù e'r" or in English "Juda's ear" and scientists may refer to this as *Auricularia auricula-judae. Auricularia auricula-judae, Auricularia fuscosuccinea,* and *Auricularia polytricha are* all from under the same genus called *Auricularia.* All saprophytic fungi with gelatinous, ear-to-shell-shaped fruiting bodies belong under the same genus.

Growth, Life Cycle, and Classification of Mushroom Species

Pleurotus ostreatus, Pleurotus sajor-caju, Pleurotus djamor and similar species can grow in habitats with a temperature of 15–31°C. In their natural habitat, *Pleurotus* spp. grow on broad-leaf hardwoods in the spring and fall, especially cottonwoods, oaks, alders, maples, aspens, ash, beech, birch, elm, willows, and poplars. *Pleurotus* spp. can thrive in a wide spectrum of natural lignocellulosic materials like cereal straws, all hardwoods, on wood by-products (sawdust, paper, pulp sludge), banana pseudostems, corn cobs, cotton waste, cotton boll locules, forest woods, sugarcane bagasse, coffee residues (coffee grounds, hulls, stalks, and leaves), banana fronds, cottonseed hulls, agave waste, soy pulp, and many more (Rajarathnam et al., 1987; Adebayo and Oloke, 2017). *Pleurotus* spp. undergoes two phases in their life cycle, which are vegetative growth and reproductive growth. Environmental factors that is suitable for *Pleurotus* spp.'s fruiting bodies formation is temperature of 10–21°C, 85–90% relative humidity, CO_2 concentration of below 1,000 ppm and light exposed to 1,000–1,500 lux. Many types of *Pleurotus* spp. have been cultivated by local farmers for domestic market (refer **Supplementary Table 1**). Additionally, some types of *Pleurotus* spp. are also being imported to meet the consumer demand. As such, very few edible oyster mushrooms were cultivated in Malaysia and some of them were even no longer seen in Malaysia any more (Samsudin and Abdullah, 2019). Those species are, *P. citrinopleatus*, *P. cyctidious, P. eryngii, P. flabellatus, P. pulmonarius*, and *P. tuber-regium* (Samsudin and Abdullah, 2019). **Supplementary Table 2** shows a list of *Pleurotus* spp. that are still available and being cultivated in Malaysia. Mat-Amin et al. (2014), stated in Malaysia, only abalone (*Pleurotus cystidiosus*), gray oyster (*Pleurotus sajor-caju*), white oyster (*Pleurotus florida*), and red oyster (*Pleurotus flabellatus)* are suitable to cultivate in Malaysia's climate and be successfully cultivated commercially. In general, oyster mushrooms are grown in the lowland parts of Malaysia.

Auricularia spp. mostly found in tropical, subtropical, and temperate regions, hence it is widely available worldwide. *Auricularia* spp. can be found scattered and in clusters on dead or dying tree branches, main trunk, decaying logs and others suitable (Packialakshmi et al., 2015). Also, this mushroom can adapt in monsoon season, hence it is producing large overlapping bunch due to its high humidity presence and the production of the basidiomes can be enormous. When growing conditions are favorable, the life cycle of *Auricularia* spp. begins with germination of haploid basidiospores and matures into short-lived haploid mycelia. The cycle ended with ballistospory, the mechanism by which mature basidiospores are expelled from the sterigmata (by removing surface tension). Finally, the wind disperses the spores after they fall below the cap. *Auricularia* spp. recorded found in Malaysia was *A. auricula judae, A. fuscosuccinea* and *A. polytricha* (refer **Supplementary Table 3** for vernacular name and common name for *Auricularia* spp.). "Cendawan telinga kera" is often found grow wildly on logs and tree trunks. Locals identified them as firmer texture and dark brown color. While "cendawan gelememeh" often identified as in bigger size, thinner and tougher than "cendawan telinga kera". If the locals spot the reddish-purple color *Auricularia* spp. and founded wet, then it should be "cendawan bibir". **Supplementary Figure 2** shows the picture of *Auricularia auricular-judae* and *Auricularia* polytricha. Mostly, *Auricularia* spp. recorded in Malaysia was found in Sarawak (Abdullah and Rusea, 2009).

Toxicity Studies on Mushrooms

Mushrooms are generally known as alternative remedies to medical drugs, either as a source of functional food or for drug development. Unfortunately, few studies have been reported to support the continued medicinal use of these remedies due to a lack of scientific research on their safety and efficiency. Acute and subacute toxicity testing on animals has been conducted to determine the toxicity assessment of ethanolic extract of the mushroom, according to Organization for Economic Cooperation and Development OECD guidelines (Deepalakshmi and Mirunalini, 2014). The acute toxicity test resulted in the maximal dosage level of 5,000 mg/kg body weight (Sprague Dawley rats) of orally administered *P. ostreatus* for 72 h; no mortality or major changes in the general behavior of rats were found. While, subacute toxicity resulted from 250 to 1,000 mg/kg doses of hepatic marker enzymes, none of the animals showed significant changes including in food and drink intake. In fact, no significant changes were recorded in vital organs like the liver, and this indicated that *P. ostreatus* had a high margin of safety due to the non-existence of toxicological effects recorded. Concern may arise from consumers as ergothioneine is usually consumed for its antioxidant properties, hence many studies investigated its mutagenic and toxicologic potential. In human and mammalian tissues, the concentration of ergothioneine was found to be 1–2 mM, implying that ergothioneine could act as a non-toxic antioxidant *in vivo* (Aruoma, 1999). While living organisms need trace amounts of certain elements such as iron, cobalt, copper, manganese, chrome, and zinc, excessive amounts of these elements are toxic to them (Erel, 2004, 2005). Mercury and arsenic are trace elements found in cultivated mushrooms, ranging from 0.01 to 0.17 mg/kg (Lelamurni and Razak, 2013). According to a report of the Joint Food and Agriculture Organization of the United Nations (FAO)/World Health Organization (WHO) Expert Committee on Food Additives, the provisional tolerable weekly intake (PTWI) of arsenic is 15 μg/kg body weight (equivalent to 2.1 μg/kg body weight per day), while PTWI for mercury is 4 μg/kg body weight. Mercury and arsenic levels were 10 and 170 g/kg dry weight, respectively, in the cultivated *A. polytricha*. The maximum PTWI for mercury for a 60 kg individual is 240 g, according to calculations. If a 60 kg individual ate 50 g of dried *A. polytricha* per day, their daily mercury intake would be 30 g, and their weekly mercury intake would be 210 g (Lelamurni and Razak, 2013). The highest PTWI for arsenic in a 60 kg human is 900 g. This person's daily arsenic consumption would be 510 g and weekly arsenic intake would be 3570 g if they consumed the same volume of *A. polytricha* on a daily basis (Lelamurni and Razak, 2013). This is even higher than the PTWI for arsenic. Therefore, a daily intake of 50 g dry weight of *A. polytricha* by a 60 kg individual may be considered potentially dangerous (Lelamurni and Razak, 2013). Based on these figures, the healthy daily intake of cultivated *A. polytricha* using the same fruiting substrate formulation as in this study must be less than 10 g dry weight (Lelamurni and Razak, 2013). *Auricularia polytricha* contains high amounts of a molecular toxin named auritoxin (Kim, 1993). Auritoxin had a polysaccharide content of 93.9% and a protein content of 6.8%. Mice were given median lethal doses of auritoxin of 56.4, 157.2, and 454.6 mg/kg. Convulsions occurred within 30 min of the injection, coma or sleeping occurred within an hour, and tremors, lacrimation, respiratory swelling, congestion, and death occurred within 24 h (Kim, 1993). The spleen was observed to be significantly swollen among the different organs of the mouse. The addition of auritoxin to human platelets prevented aggregation (Kim, 1993). The toxin inhibited the function of malic dehydrogenase *in vitro*. Other studies have shown consumption of 12 g *Auricularia auricula judae* in a day, does not give harmful effects on health, but helped abdominal obese women improve their blood LDL cholesterol, triglyceride, and bone density (Han et al., 2012).

Isolated Compounds and Antioxidant Properties of Oyster and Black Jelly Mushroom

Phenolics Compounds

Bioactive compounds in *Pleurotus* spp. such as phenolics, lipid, glycoproteins, peptides, and polysaccharides can be found in crude extracts, mycelia, and basidioma of *Pleurotus* spp (Corrêa et al., 2016). Patel et al. (2012) affirm that bioactive compounds in the *Pleurotus* mushroom has potential for therapeutic purposes in the future. Interestingly, bioactive compounds in the mycelium and fruiting bodies of oyster mushrooms were found to have immunostimulatory, anti-neoplastic, anti-diabetic, anti-atherosclerotic, anti-inflammatory, antibacterial, and anti-oxidative properties (Golak-Siwulska et al., 2018). Of the low molecular weight bioactive compounds, polyphenol is the most popular compound under research because of its anti-oxidative effects (Palacios et al., 2011; Muszyńska et al., 2013; Piska et al., 2017). Phenolics compounds have been known for their function of stabilizeing lipid oxidation, hence it is able to prevent oxidation damage and protect the human body (Gülçin et al., 2003; Ren et al., 2014). Phenolics and flavonoids have been found in *Pleurotus ostreatus* fruiting bodies and weighing 708 mg 100 g-1 D.M and 170 mg 100 g-1 D.M. Besides that, phenolics in mushrooms are reported to have anti-microbial activity and this may benefit further applications in the pharmacological industry as a natural source (Adebayo and Oloke, 2017). Researchclaims if ethanol solvents are used to extract phenolic content in *P. ostreatus*, the antioxidant activities would be 94.54%, higher than methanolic extract (Thillaimaharani et al., 2013). Phenolic compounds do not only exist in *P. ostreatus*, but are also found in, *P. djamor* (Acharya et al., 2017), *P. florida* (Thillaimaharani et al., 2013), and *P. gigantues* (synonyms: *Lentinus giganteus* and *Panus giganteus*) (Yun et al., 2020). Fruiting bodies of *P. giganteus* were found to have phenolic compounds (caffeic acid and cinnamic acid) and triterpenoids (Thillaimaharani et al., 2013). Triterpenoids were discovered to have a range of pharmacological functions, including anti-inflammatory, anti-diabetic and anti-cancer, and immune function control (Xu et al., 2018). Phan et al. (2014) emphasized the phenolics content in mushrooms had positively associated with antioxidant activity (free radical scavenging, ferric reducing power, and lipid peroxidation inhibition). Phenolic compounds in *P. djamor* also been recorded, which is 7.845 μg gallic acid/ mg dry extract (Acharya et al., 2017). The most common derivatives of phenolic compounds were found in *Pleurouts* spp. was benzoic acid and benzoic acid derivatives. In *P. djamor*'s extract, protocatechuic acid, gallic acid, vanillic acid and syringic acid from benzoic derivatives were found, while from cinnamic derivatives, caffeic acid and ferulic acid obtained (Alves et al., 2013). *Pleurotus florida* has been recorded to have high phenolic content which is 6.25 mg gallic acid/of dry extract extracted using ethanol solvents (Thillaimaharani et al., 2013). Interestingly, ethanol extract of *P. djamor* exhibited excellent antibacterial and antifungal activities against human bacterial pathogen and non-pathogens such as *E. coli* and *S. typhi* and pathogenic fungi such as *T. rubrum, E. floccosum*, and *M. gypseum*, compared to methanol extract and chloroform (Thillaimaharani et al., 2013).

Since extracts' antioxidation capacities are also related to their levels of phenolic and flavonoid material, there has been a lot of literature interest in bioactive compounds in *Auricularia* species. Wong et al. (2013) reported, *A. polytricha* from a local Malaysian food store had the strongest radical scavenging and metal chelating activities compared to mushrooms obtained locally—*H. tessulatus, P. eryngii, P. florida,* and *F. velutipes*. The author also reported the total phenolic content highest in *A. polytricha* (6.03 mg GAE/g dry matter) compared to *Pleurotus* spp.. Also, both *P. florida* and *A. polytricha* extracts reported had moderate antimicrobial activity. Total phenol contents has been reported in *A. auricula judae* (Packialakshmi et al., 2017), which are 10.25 mg of ferulic acid equivalent/ g and 8.94 g of catechol equivalent/g (Packialakshmi et al., 2015), both studies were used dried *Auricularia auricula-judae* originating from China. According to Bandara et al. (2019), inflammation is a natural response to a compromised immune system, and many Auricularia species, including *A. polytricha*, possess anti-inflammatory qualities due to the presence of phenolic compounds. Fresh fruit bodies of *Auricularia auricula-judae* grown in Malaysia were found in an earlier 2000s study to have 56.84 mg of gallic acid equivalents/g of extract (Kho et al., 2009).

Flavonoids

High flavonoid presence in *Pleurotus* spp. mushroom extracts may well account for the significant antioxidant activities recorded in the literature (Patel et al., 2012). Moreover, antimicrobial activity in flavonoids against *S. aureus* and *E. coli* has been reported (Elisashvili, 2012). Previously, Jaworska et al. (2015) reported that flavonoid content in *P. ostreatus* weighed 70 mg 100 g-1 dry matter. *P. djamor* of flavonoid acts as reducing agents, free radical scavengers, metal chelators, deactivators of singlet oxygen, good antioxidants, and have antimicrobial properties (Sudha et al., 2016). Phan (2015) reported the flavonoids in wild *P. giganteus* were 2.09% higher than in commercial strains. The studies conducted by Prabu and Kumuthakalavalli (2016) concluded that *P. florida* has significant antioxidant and antimicrobial properties due to its bioactive secondary metabolites and phenols (Adebayo and Oloke, 2017).

Flavonoids are the most abundant and extensively distributed class of plant phenolics, and they are extremely potent antioxidants. According to Ng and Rosman (2019), of the *Pleurotus* and *Auricularia* species studied, *P. sajor-caju* had the greatest flavonoid concentration, followed by *A. polytricha*. Furthermore, flavonoids, particularly catechin, quercetin, and chrysin, were the most abundant phenolic components in

Pleurotus spp. This is consistent with the current finding that *Pleurotus sajor-caju* (gray oyster) had the highest flavonoid content of the five mushrooms tested [*Agaricus bisporus* (white button), *Auricularia polytricha* (black jelly), *Flammulina velutipes* (enokitake), *Lentinula edodes* (shiitake), and *Pleurotus sajor-caju* (gray oyster)]. *P. sajor-caju,* which had the highest flavonoid content, also had the strongest anti-amylase and anti-aglucosidase activity, whereas *A. polytricha* had the least amount of activity. The TFC in *Auricularia* spp. was enhanced by steaming and pressure cooking. *L. polytricha* and *L. polytricha* edodes by 119 percent and 117 percent, respectively, although all cooked *P. sajor-caju* extracts showed a considerable loss of total flavonoid content. When compared to the WPC, cooking had a smaller effect on the total flavonoid content in mushroom extracts. *In vitro* digestion also resulted in the greatest increase in total flavonoid content (3,097%) and water phenolic content (281%) in pressure-cooked *A. polytricha* extract. Wong et al. (2013) reported the total flavonoid contents highest in *A. polytricha*, 6.95 mg QE/g dry matter compared to *Pleurotus* spp.. Total flavonoid contents has been reported in *A. auricula judae* (Packialakshmi et al., 2017) as 5.36 mg of quercetin equivalents/g, and 3.49 mg of rutin equivalents/g (Packialakshmi et al., 2015). Another study shows that cultivated *A. auricula* has a lower amount of total flavonoid content, which is only 0.86 mg of catechin equivalents/100 g, the mushroom tested was from a local market in Thailand (Srikram and Supapvanich, 2016).

Ergothioneine

In humans, a high concentration of ergothioneine exists in the bone marrow, liver, kidneys, and the lens of the eye/cornea. Even though ergothioneine is most commonly found in human tissues, its deficiency of it does not cause any harmful effect on human health, therefore it is not considered an essential dietary component for human consumption (Cheah and Halliwell, 2012). However, a thiol-containing amino acid, ergothioneine has been discovered to have an essential source in mushrooms, as well as its antioxidant properties. Another benefit of ergothioneine is that it protects cells from oxidative stress caused by reactive nitrogen forms such as nitric oxide. Dubost et al. (2007) found the highest ergothioneine content in oyster mushrooms compared to button, portabella, and shiitake mushrooms and other vegetables, such as spinach and broccoli. This shows oyster mushroom is a great source of ergothioneine. To the extent of readings, species recorded to have high ergothioneine were only reported in *P. ostreatus* (Ey et al., 2007), *P. cornucopiae, P. eryngii* (Duy Bao and Ohshima, 2013). Authors reported that the ergothioneine is mostly extracted from mushroom fruiting body and mushroom mycelia (Duy Bao and Ohshima, 2013). Ergothioneine in *Pleurotus* spp. is also functional to prevent and treat atherosclerosis via the reduction of oxidative stress (Abidin et al., 2017). Paul and Snyder (2010) reported in the review, that deficiency of ergothioneine in cells could lead to DNA damage and protein and lipid oxidation induced by oxidative stress.

Ergothioneine commonly exists in mushroom species such as in *Pleurotus* genus mushroom, *Boletus edulis, Agrocybe aegerita,* and many more (Lee et al., 2009; Chen et al., 2012; Kalaras et al., 2017). It is mainly found in the fruiting bodies and mycelia of the mushroom. This compound is also famous for its antioxidant properties and other beneficial content. Lo et al. (2012) reported the highest ergothioneine was in fruiting bodies and mycelia of *Volvariella volvacea* (537.27 mg/kg), *Boletus edulis* (258.03 mg/kg), *Pleurotus ferulae* (250.23 mg/kg), and *P. salmoneostramineus* (222.08 mg/kg). While, ergothioneine in *Auricularia* spp, such as *A. fuscosuccinea* and *A. polytricha,* was only 21.4 mg/kg and 1.4 mg/kg dry weight. In fact, the *Pleurotus* spp. has better content of ergothioneine than *Auricularia* spp., ranging from 26.2 to 250.2 mg/kg and 1.4 to 21.4 mg/kg dry weight. Although many studies reported ergothioneine popular content in fungi mushroom, there are some that were not detected in fruiting bodies and mycelia, for example *Inonotus obliquus* and *Termitomyces albuminosus* (Genghof, 1970; Lo et al., 2012).

DPPH (1,1-diphenyl-2-picryl-hydrazyl) Free Radical Assay

Lam and Okello (2015) in the study of the effect of different blanching methods on antioxidant properties of oyster mushrooms discovered that raw *P. ostreatus* gave 14.46 μmol TEAC/g of DPPH radical scavenging ability (extraction by methanol). Between commonly cultivated oyster species, the fruiting body of king trumpet or king oyster (91.84%) has higher DPPH percentages compared to the fruiting body of pink oyster (87.15%), white oyster (83.82%), Indian oyster (78.79%), and pearl oyster (61.98%) mushroom (Krakowska et al., 2020). While compared to *Auricularia* species, *Pleurotus* species has lower percentages of scavenging ability either by methanolic or aqueous extraction method (Oke and Aslim, 2011). In previous research, Corrêa et al. (2016) investigated and compared the hydrophilic and lipophilic compounds, as well as the antioxidant, anti-inflammatory, and antimicrobial activities of *P. ostreatoroseus* fruiting bodies and submerged culture mycelia (by ethanolic extraction). Five free sugars, four organic acids, four phenolic compounds, and two tocopherols were discovered in the bioactive formulations. In brain homogenates, the fruiting body-based formulation showed stronger reducing power, DPPH scavenging activity, also higher anti-inflammatory, and antibacterial effects than the mycelium-based preparation.

A study conducted by Wong et al. (2013) showed that at different mushroom extract concentrations (from 4,000, 2,000, 1,000, 500, 200, 100, 50 μg/ml) *A. polytricha* has the highest DPPH scavenging activity, compared to *P. eryngii* and *P. florida*. *A. polytricha*'s high antioxidation capacity was demonstrated by its low EC50 values (which <50 mg dry matter/ml) (Wong et al., 2013). Additionally, EC50 values are shown, which represent the concentration of extract necessary to scavenge 50% of DPPH radicals (Wong et al., 2013). The *A. polytricha* were obtained in Kampar, Malaysia, at a local food store. In another study conducted by Kho et al. (2009), all extracts of *Auricularia auricula-judae* (freeze-dried, oven-dried, and mycelium) have a significant scavenging action on DPPH radicals, however, fresh fruit body extract could not be dissolved in methanol, which could be due to the high fiber content of fresh fruit bodies. As a result, the DPPH radical scavenging effect of this extract could not be evaluated. In fact, the butylated hydroxyanisole (BHA) was used for related concentrations as the positive control as it dissolved in methanol. From the findings, *A. auricula judae* values gave incomparable percentage of scavenging activity on DPPH free radicals. Differ from He et al. (2012), in the study *A. auricula* recorded the EC50 values of scavenging action on DPPH radicals was 1.62 mg/mL, vitamin C was used as standard antioxidant and positive control.

FRAP (Ferric Reducing Antioxidant Power) Assay

Kibar (2021) in the study conducted the FRAP analysis on the *P. ostreatus* mushroom before applying the different drying methods and cold storage treatments. From the FRAP analysis, it was found that the antioxidant activity of the FRAP assay was 68.53 (μmol g^{-1}TE). Contrary, from the findings of Lam and Okello (2015), the FRAP values of 11.21 μmol/g were found in raw *P. ostreatus*. Despite employing the same assays, comparing antioxidant activity to previously published data is difficult due to a lack of uniformity in techniques and data interpretation. The FRAP values of Kanagasabapathy et al. (2011) on ethanol aqueous extract of *P. sajor-caju* (26.29 μmol/g) and Zeng et al. (2012) (13 μmol/g extract) are likewise comparable. Mushroom antioxidant activity was shown to be substantially linked with total polyphenol content. The total polyphenol content and antioxidant assays (FRAP) were shown to have substantial positive relationships in this investigation. This shows that the differences in antioxidant capabilities observed in the methanol: water extracts were attributed mostly to the overall polyphenol concentration of the extracts. Despite this, some research has found no link between polyphenol concentration and antioxidant activity in certain vegetables (Ismail et al., 2004). *P. ostreatus*, in conclusion, is an excellent source of beta-glucan, dietary polyphenols, and antioxidants.

Since most of *Auricularia* spp. had an extensive amount of phenolic and valuable polysaccharide which exert antioxidant properties, many studies have been conducted locally (Kho et al., 2009; Mahfuzatunajla, 2012) and internationally (Packialakshmi et al., 2015, 2017; Srikram and Supapvanich, 2016) to examine their ability to scavenge free radicals by single electron transfer. In 2009, FRAP assay of grown local fresh fruit bodies of *Auricularia auricula judae* was found to have 56.84 μmol of $FeSO_4 \bullet 7H_20$ equivalents/g of extract (Kho et al., 2009). The author concludes, the significant positive correlation between FRAP value and total phenolic content indicates that phenolics contribute to *A. auricula-judae*'s antioxidant activity. These findings shed light on some of the uses of *Auricula auricula-judae* in folk medicine and add to the body of knowledge regarding the plant's medicinal properties. Mahfuzatunajla (2012) conducted a FRAP assay of locally grown mushrooms and discovered the highest antioxidant activity in *Auricularia auricula-judae* with the lowest IC_{50}, which is 52.7 μg/ mL. Total antioxidant capacity of the mushroom from Srikram and Supapvanich (2016) exhibited in the range of 0.86 mmol of Trolox equivalents/100 g of dry weight, among lowest from *L. squarrosulus, P. sajor-caju, V. vovacea*, and *L. polychrous*. *Auricula auricula*'s ferric reducing power was observed to be dependent on the concentration. At 4 mg/mL, *A. auricula* had a reducing power of 0.403 nm, the results indicate that *A. auricula* has a greater potential for hydrogen donation, implying that it possesses significant reducing power (Packialakshmi et al., 2017). Nattoh et al. (2016) examined reducing power assay on four different parts of the body of *Pleurotus djamor*, those being spawn mycelia phase, early young fruiting body, young fruiting body, and mature fruiting body. Results showed the ethanolic extract of young fruit body yield at 5 mg/ mL concentration had the highest (11.491) reducing power activity, higher than water extract of young fruit body of *P. djamor* (10.077). The bioactive compounds and both DPPH and FRAP assay of *Pleurotus* spp. and *Auricularia* spp. from studies can be simplified in this below **Supplementary Table 4**.

Heat Treatments on Isolated Compounds and Its Bioactivity of *Pleurotus* spp. and *Auricularia* spp.

Pre-treatments Before Drying

Pre-treatment methods are frequently used prior to various drying processes to avoid color changes, inhibit enzyme activity, reduce the number of microorganisms, enhance flavor retention, and improve the overall stability of mushrooms. Blanching, soaking in potassium metabisulphite (Brennan et al., 1999), and citric acid (Brennan et al., 2000) are all traditional pretreatment methods, but various means including whey fermentation, curd fermentation, and others have been reported. As for blanching, they are typically blanched at 95–98°C in water or antioxidative-containing aquatic solutions. This treatment might last from 20 s and 15 min. Blanching is the first procedure after washing and is mostly applied with chemical treatments such as sodium metabisulfite, citric acid, brine made from table salt, and sodium-calcium salt of versenic acid ($CaNa_2EDTA$). Blanching in only water and with added chemical treatments may give a different result for the mushroom. As an example, 15 min of water-only blanching mushrooms could reduce the darkening of the pilei induced by enzyme activity; due to the complete inactivation of peroxidase (Bernaś et al., 2006). Another study also found that blanching without chemical treatment could prevent the phenolic content losses (Mutukwa et al., 2019). A study conducted by Walde et al. (2006) on the effect of the drying rate of different pre-treatments reported soaking with fermented whey and blanching followed with curd can decrease the drying time of oyster mushrooms compared to control, only blanching, only whey, only curd, and blanching followed with whey treatment. Those blanching treatments involve a temperature of 90°C. The addition of citric acid and using a brine consisting of table salt and sodium-calcium salt of versenic acid could improve the color and texture of sterilized mushrooms while also increasing their microbiological resilience (Bernaś et al., 2006). According to Nöfer et al. (2018), prolonged drying time and high temperatures may degrade the mushroom aroma. According to Hassan and Medany (2014), pre-treatment of *P. ostreatus* and *P. eryngii* with 0.1% sodium metabisulfite at room temperature improved mushroom color and microbiological characteristics, while also retaining its nutrients. The highest rehydration ratio was found in the untreated mushrooms. Additionally, soaking mushrooms in 0.1% NaCl or 0.1% citric acid for 10 min at room temperature produced acceptable results, though they were not as good as those treated with sodium metabisulfite. Hassan and Medany (2014) found out blanching as a pre-treatment is ineffective for enhancing the quality of dried mushrooms. Not just them, Argyropoulos et al. (2011b) in the research concluded that blanching can cause undesired color and texture of mushrooms (hardens), while also inhibiting enzyme activity and reducing microbial populations.

Solar Drying

Natural sun-drying is still widely utilized in many cultures due to its low-investment, simple operation, and energy-savings. Direct solar drying includes spreading out the mushroom to be dried in a thin layer on large outside free open surfaces and leaving it until it reaches the necessary moisture content. During the day, the mushroom must be flipped over to allow stored moisture to escape. The drying surface is usually concrete with polyethylene nets, although sensitive food is placed on perforated trays. Obviously, the drying rate is low. The crops must be left outdoors for 10–30 days, depending on their type and the local weather (Belessiotis and Delyannis, 2011). Moreover, the open sun drying procedure can also cause significant loss and quality degradation due to pollution, insufficient drying, microbial development, and other factors. Therefore, technology has been developed, such as solar drying, to alleviate these limitations, in which the mushrooms are maintained in a UV irradiation chamber equipped with UVB lamps for drying. One study reported that there is a benefit of exposing mushrooms to a UVB light, which is the greater generation of vitamin D2, especially when it is sliced and followed by freeze-drying (Nölle et al., 2017). The study was conducted by exposing the samples on both sides and irradiating for 20 min at room temperature until a dose of 1.5 J/cm^2 (2.54 mW/cm^2) was reached independently of slicing or drying (Nölle et al., 2017). Solar dryers are rather basic equipment. They range from relatively rudimentary ones used in small, desert, or remote communities to more complex industrial systems. Forced convection cabinet solar dryer type is recommended as it assists in keeping constant conditions inside the drying chamber, such as temperature and air velocity, increases the drying rate, and decreases the moisture rate (Bhavsar and Patel, 2021). Moreover, the efficiency of natural solar dryer and forced convection solar dryers was determined to be 19.27 and 35.60%, respectively. Reyes et al. (2013) in the study, dehydrated mushrooms using a hybrid solar dryer that had a three-square-meter solar panel and electrical resistances. As to the results, solar energy succeed in conserving between 3.5 and 12.5% of total energy. Mishra et al. (2016) studied the antioxidant properties and mineral composition of oyster mushrooms in relation to the different drying processes used. Solar dried oyster mushrooms were discovered to be extremely rich in a variety of antioxidants and minerals, compared to sun-dried oyster mushrooms.

Hot Air Drying

The simplest and most extensively used type of drier is hot air drying. This method is frequently used to dry wild mushroom species such as *Craterellus cornucopioides*, *Boletus edulis*, *Morchella* spp., *Cantharellus cibarius*, and *Lentinula edodes*. Typically, mushrooms are placed on a tray and pushed into a cabinet or tunnel dryer for hot air drying. When heated to a temperature of 50–70°C (Argyropoulos et al., 2011a) and combined with air convection, the hot wind evaporates the moisture contained within the mushroom, fulfilling the aim of drying. High temperatures and rapid air convection are favorable for mushroom drying in terms of drying speed alone. However, if the water on the surface of the mushroom flows too quickly, the internal water does not have enough time to reach the surface, and the surface might become overheated. As this process develops, the moisture content of the mushroom falls, the drying rate consumes more energy, and dramatically slows, making drying more difficult. Inadequate hot air drying of mushrooms can result in distinctive color changes and structural deformations (Kotwaliwale et al., 2007). Inah et al. (2021) studied the effect of oven-hot air drying on the sensory attributes of the *A. auricula judae*. The results showed the optimal process variables for treating mushrooms are 110 minutes of hot air drying with a temperature of 75°C. The greatest desirability index 0.648. (64.8%) was achieved.

Microwave-Vacuum Drying Technique

In the initial days of drying, hot-air drying is undeniably practical. As days go by, the dehydration became impractical in the food industry, as hot-air-dried mushrooms will lose color and shape and the process takes substantially longer and consumes significantly more energy. Microwave drying has been used to solve these issues, either alone or in combination with hot air drying. Microwave drying is faster than conventional drying due to less heat loss, volumetric heating, easy automatic control, and lower equipment footprint. Microwave drying may considerably reduce drying time and improve product quality. Microwave-assisted or accelerated drying technologies have been extensively studied (Zhang et al., 2006). Microwave-assisted drying is ideal for heat-sensitive items like mushrooms. Microwave energy has the ability to increase product quality, but the inappropriate application might cause irreversible changes in dried vegetable quality. Also, for mass transfer control, the microwave energy is pulsed to enhance drying efficiency because continuous heating does not accelerate moisture removal. Microwave drying mushrooms at reduced power density with warm air has increased moisture diffusivity, rehydration, and flavor retention. According to Askari et al. (2009), unlike other agricultural goods, microwave treatment can degrade the quality of mushrooms. Therefore, vacuum and microwave drying have been combined to increase dried mushroom quality. Also, due to high initial and ongoing expenses, microwave-vacuum processing has lately been utilized in conjunction with hot-air drying. It can be used after pre-drying with hot air to remove moisture when the drying rate slows. The material is dry on the surface and damp in the center. Microwave energy can selectively heat the interior of products, causing a high steam pressure. The dry surface acts as a barrier, preventing rapid pressure release, resulting in puffed produce. Vacuum, on the other hand, allows rapid mass transport at low temperatures, speeding up the final drying cycle. When working with temperature sensitive materials like mushrooms, combining drying procedures may be required to produce high-quality dried products. Das et al. also discovered a method to rapidly dry mushrooms using microwaves and convective hot air, achieving commercial quality in only 72 min (Das and Arora, 2018). Argyropoulos et al. discovered that combining hot air and microwave vacuum drying produced good quality mushrooms with less color change, soft texture, and a high rehydration rate. Combined drying can make mushroom structure airy and crunchy, allowing for innovative snack development (Argyropoulos et al., 2011a).

Effects of heat treatments on isolated compounds and antioxidant activities of *Pleurotus* spp. and *Auricularia* spp.

Analysis of Phenolic Compounds Content

Singhal et al. (2020) explain in their study that the phenolic content increased during the first hour of drying using a tray/cabinet drier and then declined with time. Also, two phytochemical kinetic models were used. Since the capacity of drying equipment to preserve nutrients at low cost is still being studied, a kinetics analysis of nutritional characteristics is necessary. This helps understand how food molecules degrade throughout processing (Sarpong et al., 2018). For free radical scavenging activity (percent inhibition) and phenolic content degradation, a first-order kinetic model could be predicted better than a second-order model using statistical parameters. Additionally, the D value of phytochemicals is being examined. The figures indicated the time (hr.) required to reduce the phytochemical content by 90% at each temperature (60, 65, and 70°C). Ascorbic acid, which has the greatest D values, degraded the fastest during heat treatment, followed by total flavonoids and total phenolics. The total phenolic content of dried *Pleurotus sajor-caju* samples was assessed by Mishra et al. Sun dried sample had 31.4 mg GAE/g dry weight, which was greater than hot air dried sample (28.7 mg GAE/g dry weight) (Mishra et al., 2016). Phenolic molecules could be utilized to assess antioxidant capability. The antioxidant activity of mushrooms is linked to their phenolic component concentration. A study conducted by Mutukwa (2014) concluded that among the chemical pre-treatments, vinegar and potassium bisulfite produced the highest total phenolic chemicals. The drying process had no effect on total phenolic content. The interaction of the blanching method with chemical pre-treatment on total phenolic compound content was significant. The vinegar and potassium bisulfite treated samples with no blanching had 8.31 mg/g GAE and 8.4 mg/g GAE of phenolic, respectively (Mutukwa, 2014). Water blanched samples with a chemical preparation had smaller total phenolic content than water blanched samples with no chemical pre-treatment. Unblanched samples had greater total phenolic content than blanched samples among those that received a chemical pre-treatment. This could be explained by the fact that potassium bisulfite is a reducing agent, and so the increased total phenolic content reported in potassium bisulfite-treated samples could be a result of this molecule. Polyphenols were found in white vinegar, but no flavonoids, therefore the vinegar treatment may have enhanced the total phenolic content of the sample (Lopez et al., 2005). Furthermore, leaching may be the cause of the decreased total phenolic compounds in the blanched samples. Blanching may have facilitated the loss of phenolic chemicals. This may include the release of bound phenolic compounds and/or disruption of phenolic compound and cell wall structure, facilitating phenolic compound leaching out after blanching.

The investigation showed that different processing processes did influence *A. auricula-judae* total phenolic content (Kho et al., 2009). Oven-dried *A. auricula-judae* fruit bodies had 2.75 times more total phenolics than fresh fruit bodies. The amounts of total phenolic contents in freeze-dried tomatoes increased 2.6% to 5.9% compared to the fresh ones, and the total phenolic contents in oven-dried tomatoes increased 13% to 29% compared to the fresh ones. This may be due to the drying process releasing phenolic chemicals from the matrix. Processing may speed up the release of bound phenolic compounds by breaking down cellular components. The high temperature of the oven-drying procedure may denature oxidative and hydrolytic enzymes that could damage phenolic compounds in *A. auricula-judae*. This may have increased the antioxidant capacity of *A. auricula-judae* by reducing phenolic acid loss. The freeze-dried *A. auricula-judae* fruit bodies had 3.54 times the total phenolic content of the oven-dried fruit bodies. This may be related to greater overall phenolic extraction efficiency. During freeze-drying, ice crystals form within the fruit bodies' matrix. Ice crystals can cause more cell rupturing, allowing for improved solvent access and extraction. The cooking process reduced the water-soluble phenolic content (WPC) of all mushroom extracts by 29% to 85% (Ng and Rosman, 2019). With the loss of WPC in all cooked mushroom extracts, especially *F. velutipes* and *L. edodes*, came a loss of FRAP values, indicating WPC contributes to their antioxidant activity. This trend was absent in *A. bisporus, A. polytricha,* and *P. sajor-caju* (Ng and Rosman, 2019), perhaps due to the existence of heat-resistant antioxidant substances other than phenolics such as b-glucan and amino acid, ergothioneine (Soler-Rivas et al., 2009) and heat-stable component (superoxidase dismutase and quinone oxidoreductase). After conducting *in vitro* digestion (simulates human gastrointestinal digestion to assess phenolic content digesting stability on the reducing power, and radical scavenging capabilities in digested mushroom products), Ng and Rosman (2019) found the pressure-cooked *A. polytricha* extract had the largest increase in WPC (281%). In the digested microwaved samples, WPC increased significantly for *A. polytricha* (156%) and *L. edodes* (116%) but decreased significantly for *A. bisporus* (-48%) and *P. sajor-caju* (-60%).

Analysis of Flavonoid Content

Singhal et al. (2020) studied the effect of heat on flavonoids in mushrooms (microwave blanched for 30 s, then osmotic dehydration in 15% sodium chloride solution for 10 min). The study concluded that as temperature and time increased, the flavonoid content decreased. Flavonoids lose much more at 70°C than at 60°C. In apricots, Madrau et al. (2009) found that 75°C results in a greater loss of flavonoids than 55°C. The bigger the enthalpy change, the more heat-sensitive the chemical (Karaaslan et al., 2014). Hence, ascorbic acid and flavonoids are more heat-sensitive than total phenolics. Furthermore, positive enthalpy change values indicate that the breakdown of flavonoids and ascorbic acid constituted an endothermic reaction. A similar result was reported by Sarpong et al. (2018). The total flavonoids in oyster mushroom with different blanching methods (control, water, and steam) and different chemical treatments (no chemical, with potassium bisulfite, lemon juice, and vinegar) was assessed by Mutukwa (2014). The water-blanched samples had the highest total flavonoids, while the un-blanched samples had the lowest. The drying method had no impact on total flavonoid content. Total flavonoid content was affected by the blanching procedure with chemical pre-treatment. Blanching and soaking in different pre-treatments increased overall flavonoid concentration. The unblanched samples showed the lowest total flavonoid concentration, except for those treated with potassium bisulfite. While the blanched samples exhibited higher flavonoid concentration than the unblanched samples. Water blanching yielded more total flavonoids than steam blanching. Heat may increase flavonoid availability, resulting in increased flavonoid content in blanched samples. A 15-min heat treatment at 100°C increased free flavonoids while decreasing bound flavonoids, according to Choi et al. (2006). These alterations may be due to heat disrupting the cell wall and leading to the release of bound flavonoids. Given that blanching mushrooms and soaking them in chemicals both cause nutrient leaching, these findings suggest that polyphenols in the flavonoid class are less susceptible. Blanching and chemical pre-treatments reduce polyphenol oxidase activity. Therefore, combining both treatments should help maintain polyphenols, which contribute to greater total flavonoids. It is also possible that the chemical pre-treatments affected the total flavonoid content directly. This may have aided in the total flavonoid concentration detected in lemon juice treated samples (Gattuso et al., 2007). The amount of flavonoids in *Pleurotus ostreatus* dried fruit extracts resulted in the following order: microwave, freeze-drying, hot air drying, and sun-drying (Piskov et al., 2020). Despite the expexctations of Piskov et al. that low-oxygen and low-temperature freeze-drying would limit flavonoid losses, the microwave drying sample extracts had the highest flavonoid content. Toor and Savage (2006) explained that this result could be because the permeability of microwave radiation damages the cellular components of tissue, hence increasing the accessibility of flavonoids for extraction. Microwave drying also preserves more flavonoids than hot air drying due to its rapid heating. Sun-dried samples had the lowest flavonoid levels, this is most likely related to drying time and consequently oxidation processes, as shown by Oprica et al. (2019) in their studies of dehydrogenation in plants. True retention of minerals, vitamins, and bioactive compounds in mushrooms has been studied by Lee et al. (2019) while undergoing steaming, blanching, microwaving, boiling, and roasting. Alpha-glucan shows higher values in all cooking methods except for raw, while total flavonoid shows higher values in the microwave and roasting method other than that the values decreased. Also, Ng and Rosman (2019) claimed the flavonoid content was improved by 119% and 117% in *A. polytricha* after steaming and pressure cooking, respectively, while there was a considerable loss of flavonoid content in all cooked *P. sajor-caju* extracts. When compared to the phenolic content, cooking had a smaller effect on the flavonoid in mushroom extracts. Total flavonoid content (3,097%) of pressure-cooked *A. polytricha* extract increased the most after *in vitro* digestion.

Determination of Ergothioneine Content

There was a statistically significant difference in the amount of ergothioneine present in no blanching, water blanching, and steam blanching of oyster mushrooms (Mutukwa, 2014). The highest concentrations of ergothioneine were found in the unblanched samples, followed by the steam blanched samples, and the lowest concentrations were found in the water blanched samples. This loss was most likely caused by leaching. Nguyen et al. (2012) steamed, boiled, and microwaved *Flammulina velutipes* mushrooms for 2–5 min and discovered that all of these treatments reduced the amount of ergothioneine in the mushrooms, with the boiled mushrooms showing the greatest reduction in this component. According to their findings, heat degradation was not considered, and the reduction was attributable to leaching instead. Increased ergothioneine yield was seen as the microwave-convective drying temperature was raised up to 70°C, after which the yield began to decline progressively as the drying temperature was raised even higher (Bhattacharya et al., 2013). Microwave-convective heating enables the temperature of the product to rise dramatically, resulting in a flux that aids in the quick evaporation of water vapor from the product. True retention of minerals, vitamins, and bioactive compounds in mushrooms has been studied by Lee et al. (2019) while undergoing steaming, blanching, microwaving, boiling, and roasting. The ergothioneine concentration of mushrooms was significantly reduced by all of the cooking methods. Boiling induced the greatest loss of ergothioneine in the shiitake mushrooms when compared to the other cooking procedures. Lee et al. (2019) came to the conclusion that microwaving and roasting were preferable to other cooking methods in terms of retaining functional compounds such as ergothioneine, polyphenols, and flavonoids while boiling resulted in the greatest reduction in the amount of functional compounds present in mushrooms.

Analysis of Heat on Antioxidant Activities

Piskov et al. (2020) studied the effects of different drying methods (microwave, hot air, and solar drying) on the maximum antioxidant properties (ferric reducing ability, reducing power activity, antiradical scavenging activity, total antioxidant capacity, the activity of inhibiting lipid oxidation) and ACE-inhibiting activity. From the study, electrochemical analysis revealed that hot air-dried oyster mushrooms possessed a high overall antioxidant capacity. The remaining mushroom extracts were accepted in the following order: freeze-drying > sun-drying > microwave drying. Contrary to expectations, microwave drying extracts with the highest antioxidant activity measured using spectrophotometric methods (Reducing Power Activity, ABTS, and FRAP assays) had the lowest total antioxidant activity evaluated using the electrochemical approach. Study shows *P. sajor-caju* treated with sun dry has high FRAP values than solar dry and hot air treatments, while DPPH assay has shown higher values in solar dry compared to sun dry and hot air (Mishra et al., 2016). Lam and Okello (2015) observed FRAP antioxidant activity of blanched treatment of *P. ostreatus* is reduced compared to raw/fresh mushroom. Tan et al. (2015) examined the DPPH free radical scavenging assay of mushrooms cooked in boiling, microwave, and pressure cooking compared to the uncooked sample. The antioxidant activity of *Pleurotus pulmonarius* increased by 151% when boiling for 5 min compared to uncooked samples (16.56 ± 0.18%). The antioxidant activity of *Pleurotus citrinopileatus, P. flabellatus*, and *P. floridanus* was not affected by boiling. *Pleurotus cystidiotus* and *P. eryngii* had reduced antioxidant activity (relative percentage 12 and 77%) than raw samples. Lee et al. (2007) found that hot water extracts of *P. citrinopileatus* (heated at reflux for 1 h) scavenged 20.7–2.3 % at 20 mg/ml. After 5 min of boiling, *P. citrinopileatus* had 18.8% scavenging ability at 5 mg/ml. To compare the DPPH free radical scavenging activity of boiled (100°C, 1 h) *Agaricus bisporus* (button mushroom) to uncooked (15.6–65.8%), Jagadish et al. (2009) found that the boiled (100°C, 1 h) *Agaricus bisporus* decreased by 8.3–53.1%. The increased boiling time may have reduced the scavenging ability of bioactive chemicals in mushrooms. The antioxidant activity of *P. floridanus, P. flabellatus,* and *P. pulmonarius* (0.46; 4.60 and 6.72, respectively) increased significantly after microwave heating (1.45, 6.50, and 14.43 correspondingly). However, *P. citrinopileatus, P. cystidiosus,* and *P. eryngii* demonstrated lower antioxidant activity than the uncooked sample, with relative percentages of 44, 65, and 70%. Kettawan et al. (2011) reported that the texture, color, and form of each mushroom variety may alter antioxidant activity. *P. cystidiosus* and *P. eryngii* had thick, meaty, solid, and hard features where the microwave heat may not have entered the mushroom tissue or broken the cell wall to free the antioxidant compounds. However, Hayat et al. (2009) observed that microwave treatment could speed up the release of phenolic chemicals from orange peels by thermally destroying the cell wall and subcellular compartments. Microwave cooking retained more phenolic acids in Boletus mushrooms than pressure cooking, steaming, boiling, or frying, according to Sun et al. (2014). When compared to all other processed and uncooked mushrooms, *P. floridanus* had the highest scavenging ability (>200%). The increase in antioxidant activity may be attributed to the fiber complexes being released during pressure cooking (low moisture level and high temperature). The DPPH radical scavenging activities of *P. flabellatus* and *P. pulmonarius* increased by 10.01 and 10.02, respectively. *P. citrinopileatus* and *P. eryngii* demonstrated 50 and 67% decreased scavenging ability by pressure cooking compared to raw samples. Choi et al. (2006) found that increasing heating time and temperature will greatly influence *Lentinula edodes*' DPPH radical scavenging activity. *P. pulmonarius, P. floridanus,* and *P. flabellatus* have thinner and softer tissue than *P. eryngii* and *P. cystidiosus*. Thus, high-pressure cooking may liberate phenolic chemicals more easily from mushroom tissues. Thermal reactions may also produce greater antioxidants. Furthermore, it was discovered that cooking ordinary beans (*Phaseolus vulgaris*) for longer periods of time increased their free radical scavenging activity (Rocha-Guzmán et al., 2007). An antioxidant assay, FRAP, and DPPH method showed, that the steamed *A. polytricha* mushroom showed the highest antioxidant activities compared to boiling, microwaving, and stir-frying the mushrooms (Shamaruddin et al., 2021). The antioxidant activity of cooked *Auricularia polytricha* has been investigated by Ng and Rosman (2019). It is found out that boiling *A. polytricha* would be the best option for cooking to achieve best antioxidant values, while for *P. sajor caju* preferably by pressure cooking. Interestingly, cooked *A. polytricha* and *P. sajor caju* showed decreasing value in water soluble phenolic content, however that doesn't affect the FRAP values, in fact the FRAP values of boiled and steamed *A. polytricha* and *P. sajor caju* was increased. This can be explained by the presence of non-phenol molecules such as b-glucan and amino acid, ergothioneine, which are heat-resistant anti-oxidants (Soler-Rivas et al., 2009). Moreover, steaming and pressure cooking are believed to improve the total flavonoid content in *A. polytricha* (not in *P. sajor caju*), therefore the FRAP values increased. The study conducted using autoclave and the temperature arrangement mimicked the domestic cooking styles, which are 95°C (steam), 100°C (boiling), and 121°C (pressure cooking). Kho et al. (2009) in their study found that because of the high fiber content in fresh fruit bodies of *A. auricula-judae* obtained from a local farm at Tanjung Sepat, Selangor, Malaysia, the extract of fresh fruit bodies could not be dissolved in methanol. As a result, the DPPH radical scavenging effect of this extract could not be calculated. Therefore, the EC50 value of BHA, was employed as a positive control, and the results was 12.05 μg/mL. The extracts of freeze-dried fruit bodies of *A. auricula-judae* had the lowest EC50 value of 2.02 mg/mL. BHA is a pure or concentrated synthetic phenolic antioxidant. It can scavenge reactive oxygen species like DPPH free radicals by giving them labile hydrogen and leaving behind an oxidized phenolic ion that is stabilized by the resonance of the benzene ring. This could explain why BHA had a stronger free radical scavenging activity than *A. auricula-judae* extracts. It was found that 1 mg/mL air-dried and oven-dried ear mushrooms grown in Taiwan with an EC50 value of 91 μg/mL showed a 100 percent scavenging effect (Mau et al., 2001). It is possible that the variation in species and growing environment accounts for the significantly high free radical scavenging potency of ear mushrooms grown in Malaysia. When compared to the EC50 value (2.02 mg/mL) exhibited by freeze-dried fruit bodies of *Auricula judae*, fresh fruit bodies of *Phenillus linteus*, a wild fungus, had a low EC50 value of 22.1 g/mL (Song et al., 2003). The scavenging capabilities of freeze-dried Grifola and Morchella mycelia with EC50 values of 4 to 5 mg/mL, on the other hand, were lower than those of *A. auricula-judae* (Mau et al., 2004). At a concentration of 4 mg/mL, methanol extracts of *A. auricula-judae* freeze-dried and oven-dried fruit bodies had DPPH radical scavenging effects of 94.77 and 74.04%, respectively, in the current study. When compared to the processed (freeze-dried and oven-dried) *A. auricula-judae*, processed tomatoes are thought to have only marginally higher DPPH radical scavenging activity. It is possible that the microwave, pulsed electric field, and ultrasonic treatments alter the antioxidant activity of Auricularia polysaccharides, but not their main chemical structures. The microwave-derived Auricularia polysaccharides had reduced molecular weight, higher glucose content, and improved antioxidant activities, indicating that molecular weight may be a key factor determining antioxidant activities (Miao et al., 2020).

Consumer Preferences on Heat-Treated Oyster and Black Jelly Mushroom: *Pleurotus* spp. and *Auricularia* spp.

Sensory Analysis of Pleurotus spp. and Auricularia spp.

Studies conducted by Aishah and Rosli (2013) stated that the taste and features of *Pleurotus sajor-caju* remain the same when dried. Çelen et al. (2010) stated at 50°C heat treatment of mushrooms risked their taste and protein. The texture, color, or shape of each mushroom variety can influence the reduction of antioxidant activities. *Pleurotus cystidiosus* and *Pleurotus eryngii* have thick, meaty, strong, and firm characteristics, suggesting that the microwave heat did not reach the mushroom tissue or disrupt the cell wall, allowing the antioxidant compounds to be freed. Mutukwa (2014) in his experiment on the effect of drying treatment on the sensory quality of oyster mushrooms, found that oyster mushrooms in the oven method gave lower yellow and browning color ratings than solar-dried oyster mushrooms in terms of appearance. White colors decreased with an increase in the intensity of yellow and brown. Maillard browning may be the product of the higher brown and yellow color ratings of solar drying. Overall acceptability was still recorded as highest for oven and sundried oyster mushrooms. L*, a*, and b* values after heat treatments for oyster and black jelly mushroom were recorded in **Supplementary Table 5**.

Hot-air drying at 40, 50, 60, 70, and 80°C has been applied to *Auricularia auricula-judae,* results show as temperature increases to the 70°C the appearance is denoted as good in shape and resilience (Choi et al., 2014). Resilience reduction and low moisture content of tissue were observed at 80°C. In terms of chromaticity, as the temperature increases, the *L, *a, and *b values also increase, while the hardness may vary for each temperature. One study was conducted by Chen et al. (2020) on the effect of different drying temperatures on the color of the *Auricularia cornea,* the milky-white mushroom, and the light yellow ventral surface. A major index for selecting the drying technique is the changes in the color of edible fungi during drying. The color of the dry fruiting bodies and the rehydrated fruiting bodies influences sales price significantly. The selection of an appropriate drying temperature is therefore especially important. Results show as the temperature of drier increases from 35, 45, 55, and 65°C, the L* (lightness and darkness), a* (red and green), and b* (yellow and blue) values of the dry fruiting bodies showed random changes. At 35 and 65°C the ΔE values of the mushroom are lower than the other temperature, the color is exactly as that of the mushrooms in sun-drying treatments (Chen et al., 2020). While, at 65°C the *L and *a values of the fruiting body of *A. cornea* indicates the highest brightness and lowest redness which can be said the mushroom was in good color. When compared to *A. bisporus* dried thoroughly with conventional hot air, combining hot-air and microwave-vacuum drying resulted in a dried product of greater quality (Argyropoulos et al., 2011a). Furthermore, using the combined process, dried mushrooms with a puffed structure and crispy texture were created, which might be deemed relevant attributes for making a snack-type product. A study was conducted by Shamaruddin et al. (2021) on the effect of sensory quality of the different domestic cooking of black jelly mushroom. The authors concluded that it is normal that *Auricularia* spp. regains a darker color when exposed to heat, especially during boiling due to the Maillard reaction. Moreover, the firmness of the black jelly mushroom getting decreases when exposed to high temperature compared to raw mushroom. This is because of as high temperature penetrated the mushroom, cells will shrink and reduce intercellular space which later causes tighter hyphae organization within tissue (Zivanovic and Buescher, 2004). Also, the heterogeneous structure of black jelly mushroom will gave varying results during texture analysis. Heating indeed alters mushroom tissue, causing membranes to denature and permeability to rise, resulting in water loss and softening of the cells, hereto changing the structure. Because the mushroom cell wall is made up of glucan and chitin and does not provide the same support as a plant cell wall, the effect of cooking treatment is more visible in mushrooms than in vegetables. In addition, hydrolysis in macrostructures of mushroom cells resulting in additional tissue relaxation. Overall, between heat treatments there is no significant difference in terms of overall acceptability such in appearances, texture, color, flavor. Only that chemical treatments were found the least for overall acceptability.

Trends in Oyster and Black Jelly Mushroom Consumption

Based on the 52 respondents (refer to **Supplementary Figure 3**), most of them had eaten oyster mushroom (82.7%), while for others it depended on the dish cooked before consumption, and only a few among the 52 respondents did not ever consume oyster mushrooms (3.8%). This finding corresponds with the previous study which found out that the gray oyster mushroom was the most preferable among consumers in Malaysia, and one of the most valuable crops that will be commercially farmed by the Malaysian government (Mohd Tarmizi et al., 2013). The 82.7% mostly included white oyster mushroom and gray oyster mushroom in their meals, since most of them claimed oyster mushroom is delicious and has been influenced by their family. Soup (78%) and frying (68%) are the most favored cooking styles for mushroom dishes, respondents claimed such styles give the most flavorful taste to the mushrooms and suit the texture of the mushrooms. Mohd Tarmizi et al. (2013) found mushrooms had also been included in Malay cuisine as floss (serunding), in curry dishes, and as satay.

Generally, the questions asked in the Black jelly section were similar to the questions asked in the oyster mushroom section. However, contrary to our expectation, the actual respondents that had black jelly mushrooms or wood ear mushroom (40.3%) were much lower than and not even close to the amount who had oyster mushrooms. Almost half of the respondents voted they never ate black jelly mushroom and wood ear mushroom

(46.2%) (refer **Supplementary Figure 4**). The Department of Agriculture of Malaysia listed that among 17 types of mushroom, the cultivation of black jelly mushroom was only 1.17% compared to oyster mushroom at 90.89% (Rosmiza et al., 2016). This shows demand for black jelly mushroom is still very low, hence more local farmers go for oyster mushrooms instead. Interestingly, respondents who had eaten black jelly mushroom because of its supposed benefits afterwards considered it delicious, while family influences were the lowest contributor to self-consumption.

Availability of the Oyster Mushroom and Black Jelly Mushroom in the Local Community

According to the respondents, most of them had never seen pink oyster mushroom, hence they had never tried it. This is understandable since pink oyster mushroom is almost never seen in the local market (**Supplementary Figure 3**). According torespondents, only white oyster and gray oyster mushroom are commonly found in the supermarket. Moreover, the high number of people consuming oyster mushroom may because of the moderate price offered at the supermarket, neither expensive nor cheap but still affordable for regular consumption. Since most supermarkets in Malaysia mixed imported vegetables and local vegetables, respondents had no idea the origin of the mushroom (54%), while 46% of respondents claimed the mushroom was locally farmed.

In terms of the mushroom availability, black jelly mushroom was easier to find (89.3%) in the market compared to wood ear mushroom (14.3%), according to respondents. Moreover, respondents mentioned that besides the market, online platforms were also among the best options to get black jelly mushrooms. However, the price available in supermarkets was considered somewhat expensive-to-average for the 28 respondents who consume black jelly mushroom. It can be seen, even though the price is considered a bit expensive, people still consume black jelly mushroom, therefore it can be said price is not the sole influence the consumer for vegetable intake. One study showed that during fruit season, the prices were lower than other fruits, and they were sold in abundance even by the roadside (FAMA, 2010), yet those same findings showed that consumption of low-cost seasonal fruits did not rise. Thus, the price was not the sole determinant of seasonal fruit consumption choices. Similarly, another study comparing consumers from several Asian countries found that price is not a significant influence on Malaysian food choices (Prescott et al., 2002).

Nutritional Value of Oyster Mushroom and Black Jelly Mushroom

On the nutritional side of oyster mushrooms, 54% of the 50 respondents who consume oyster mushroom and 2 respondents who did not agreed that the style of cooking may decrease the nutrients, while another 42% disagree with that. It can be concluded that the knowledge of depletion of nutrients by higher temperature is not familiar enough. This can be seen in the results survey, even though washing has no relation to the depletion of nutrients, 8% of respondents still thought washing caused nutrients to decrease in mushrooms. However, 84% of respondents agreed depletion of mushroom nutrients could be caused by high temperature during cooking, style of cooking, and UV radiation; the least expected to cause low nutrients in mushrooms was storage in a refrigerator or freezer (see **Supplementary Figure 3**). Benefits of oyster mushroom also were asked in this survey, some respondents agreed the oyster mushroom is of high nutritional and health-promoting value (35%), good antioxidant properties from bioactive compounds (phenolic content) (22%), and is a good source of protein (21%).

In terms of nutrients of black jelly mushroom, respondents agreed this mushroom contained high nutritional and pharmacological value and good antioxidant properties from bioactive compounds (phenolic content). Furthermore, decrease in nutritional value may be caused by high temperature during cooking, style of cooking, UV radiation, and storage in refrigerator or freezer, according to respondents.

Intake of Oyster Mushroom and Black Jelly Mushroom Consumption

On the other hand, the amount of oyster mushroom intake was mostly not conscious by the respondents who consume this mushroom (54%), and only 46% of them were aware of their intake of this mushroom, therefore it can be seen that both types of respondent probably never thought of food poisoning as really being caused by oyster mushroom (56%) or had actually known about this mushroom causing poisoning (44%). Since only gray and white oyster mushroom are consumed by people in any great quantities, it still considered safe, as most literature recorded no toxicity or poisoning cases reported from oyster species (Deepalakshmi and Mirunalini, 2014; Sharma and Gautam, 2015). Interestingly, respondents who never consume black jelly mushroom (18 people), were conscious about the toxicity of those mushrooms, rather than respondents who had eaten this mushroom (11 people).

CONCLUSION AND RECOMMENDATION

It can be concluded that microwave drying have a lot of benefits as heat treatment to contained bioactive compound and the quality of oyster and black jelly mushroom, followed by freeze-drying and oven drying. Blanching mushrooms only retained flavonoids only but no other valuable components, therefore instead of blanching, using pre-chemical treatments alone could enhance the extraction of phenol in the mushroom, especially potassium bisulfite and vinegar. Ergothioneine yielded greater after microwave treatments. Phenol and flavonoid were not the only compounds that can be seen as indicators of high antioxidant values, this review analysis found antioxidant properties may be enhanced by other valuable compounds such as beta-glucans. Therefore, the microwave is seen to have a better chance to retain these compounds. Apart from that, boiling the mushroom has become a good option for domestic cooking to retain bioactive compounds, even if not as much as microwave treatment for industry purposes, it can be practical for domestic use in retaining healthy nutrition in cooked meals. Based on the observation conducted, many still have not realized that temperature and style of cooking could impact the nutritional values of these mushrooms, hence it is a huge loss if these beneficial contents cannot be retained in meals. This analysis did not compare the bioavailability of bioactive chemicals released from mushroom extracts and absorbed through the digestive system. More research using an *in vivo* model is needed to offer a more extensive nutritional bioavailability profile across diverse mushroom species. Knowledge about how heat treatments and *in vitro* digestion influence bioavailability in mushrooms would be beneficial to both consumers and food manufacturers for future production and invention.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/**Supplementary Material**, further inquiries can be directed to the corresponding author/s.

AUTHOR CONTRIBUTIONS

II conducted the research as well as the writing of the article. SR gave input on drafting the review article. FA gave constructive comments to improve the article. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supports by the Grant Scheme: 600-RMC/GPK 5/3 (229/2020) from Universiti Teknologi MARA.

ACKNOWLEDGMENTS

The authors would like to thank the Grant Scheme: 600-RMC/GPK 5/3 (229/2020) from Universiti Teknologi MARA. The authors would like to thank the facilities and technical assistance from lab staff from Universiti Teknologi MARA.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsufs.2022.882939/full#supplementary-material

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Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.