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Effect of Technical Variables on the Total Phenolic and Antioxidant Activity in Cooking of Canned White Lingzhi (*Ganoderma Lucidum)* Fruit

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Abstract.

Lingzhi (*Ganoderma lucidum)* has high nutritional values with different beneficial effects on human health. The production of fresh *Ganoderma lucidum* is increasing in recent time but the further processing step is still limited. Fruit boby of *Ganoderma luvitcidum* has a short shelf-life owing to its high rate of respiration and perishibility. In order to increase the added value of *Ganoderma lucidum*, a processing of canned white lingzhi (*Ganoderma lucidum)* in respect of maintaining the total phenolic and antioxidant activity in maximum level was investigated. By examining on different variables of the cooking time, temperature and salt concentration; the highest phenolic and antioxidant activity inside the canned white lingzhi (*Ganoderma lucidum)* could be achieved by cooking within 4 minutes, at 90^{o}C in the presence of 5 g/L of salt. Canned white lingzhi may be consumed as functional food.

Keywords: *Ganoderma lucidum, Lingzhi, cooking, phenolic, antioxidant,*

I. INTRODUCTION

Ganoderma species are among those fungi that can thrive under hot and humid conditions. *Ganoderma lucidum* is among fve species of *Ganoderma* mushroom collected from different sites in Vietnamese National Parks.[1] *Ganoderma lucidum*, or Lingzhi is a white-rot fungus of division basidiomycetes. It composed of water, carbohydrate, crude fat, crude fibre and crude protein. Triterpenoids, steroids, glycoproteins, phenols and polysaccharides, these components are helpful in maintaining good health and meeting nutritional requirements. *G. lucidum* has been reported to possess therapeutic effects such as antitumor, anti-oxidant, cholesterol lowering, detoxification, blood pressure lowering, antimicrobial, antiacetylcholinesterase, immune-modulating, blood glucose lowering, anti-allergic effects and physical fitness.[2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14]

Artificial cultivation of *G. lucidum* in polypropylene bags with variety of cheap and readily available substrates such as grass, sawdust, malt extract, and wheat bran was implemented.[15, 16, 17, 18] Malt extract medium, temperature range of 25-35˚C and acidic pH (5.0) were conducive for the mycelial growth of *G. lucidum*.[19] Salicylic acid (SA) increased the biosynthesis of triterpenoid in *G. lucidum* fruit bodies by spraying this chemical during the fruiting.[20] Commercial *G. lucidum* products are available in various forms, such as powders, dietary supplements, alcoholic beverage and tea.[21] There were few studies mentioned to the processing and preserving of *Ganoderma lucidum.* An evaluation of alcoholic beverages based on *Ganoderma lucidum* was conducted.[22] Alcohol concentration (40%) with very agreeable sensory characteristics and possible pharmacological activity could be obtained. The effect of drying on crude ganoderic acids and watersoluble polysaccharides content in *Ganoderma lucidum* was evaluated.[23] The nutritional analysisand mineral content of two varieties of mushroom species include oyster (*Pleurotus ostreatus*) and Reshii (*Ganoderma lucidum*) was evaluated.[16] Exopolysaccharide produced from *Ganoderma lucidum* in a repeated-batch fermentation.[24] A research investigated on the preservation of *Ganoderma lucidum* fresh fruit body. [25]

Ganoderma lucidum fresh fruit body is easily decayed or rotten owing to respiration. In order to increase the added value of *Ganoderma lucidum*, we investigated on processing of canned white lingzhi (*Ganoderma lucidum)* in respect of maintaining the total phenolic and antioxidant activity in maximum level. By examining on different variables of the cooking time, temperature and salt concentration; the highest phenolic and antioxidant activity inside the canned white lingzhi (*Ganoderma lucidum)* could be achieved.

Figure 1. White lingzhi (*Ganoderma lucidum)* fresh fruit

II. MATERIAL AND METHODS

2.1 Material

We cultivated *Ganoderma lucidum* on sawdust as solid-state medium to get fruit body. They were cultivated following VietGAP to ensure food safety. Fresh white lingzhi (*Ganoderma lucidum)* fruit body had short shelf life, with a high rate of respiration. It consumed oxygen and released carbon dioxide. This respiration also created

weight loss. It's necessary to process fruit body as soon as possible. After harvesting, they were conveyed to laboratory within 8 hours for experiments. Fruits were washed thoroughly under turbulent washing to remove dirt, dust and adhered unwanted material. Perasan (peroxyacetic acid) solution was also used for washing as primary treatment.

2.2 Methods

2.2.1 Sample preparation

Each sample of white lingzhi (50 g) was placed on a 1-L beaker with 300 mL of salt solution (0, 1, 3, 5, and 7 g/L) and cooked in hot water bath using different temperature (80, 90, and 100^{o}C) and time (2,4, and 6 minutes). Five grams (5 g) of the cooked lingzhi was macerated, mixed, and stirred with 20 mL methanol then placed on a vial. Afterwards, the fresh and cooked lingzhi-methanolic extracts were analyzed for total phenolic content and DPPH scavenging activity.

2.2.2 Total phenolic content determination

Total phenolic content was measured using the Folin–Ciocalteu method with some modifcations. The absorbance was measured using a UV–vis spectrophotometer at 710 nm against a reagent blank. The total phenolic content was expressed as milligrams of catechin equivalents per 100 gram of dry sample weight (mg of CE/100 g) using the calibration curve of (±)-catechin.[38]

2.2.3 Antioxidant (DPPH) activity

The total free radical scavenging capacity of the lingzhi - methanolic extract was estimated by the DPPH.[26] One (1) mL of the extract was adjusted to 5 mL volume with the addition of distilled water. Freshly prepared, 1 mL DPPH solution (0.1 mM in absolute methanol) was mixed with the extract. The reaction mixture was shaken well and held for 30 minutes at room temperature, and the absorbance of the resulting solution measured at 517 nm against a reagent blank. The radical scavenging activity was measured as a decrease in the absorbance of DPPH, and expressed as percent radical quenching compared to that without the extracts.

2.3 Statistical analysis

The experiments were run in triplicate with three different lots of samples. Data were subjected to analysis of variance (ANOVA) and mean comparison was carried out using Duncan's multiple range test (DMRT). Statistical analysis was performed by the Statgraphics Centurion XVIII.

III. Results & Discussion

3.1 **Efffect of cooking time and temperature on the total phenolic and antioxidant activity of canned white lingzhi (*Ganoderma lucidum)* fruit**

Phenolic compounds are one of the major sources of antioxidant in vegetables, so as in lingzhi (*Ganoderma lucidum).* Antioxidant is responsible for protecting the body from the damage caused by free radicals. One of the functions of antioxidant is to scavenge the unpaired electron of the free radical. The antioxidant activity of phenolics is mainly due to their redox properties, which allow
them to act as reducing agents, hydrogen donors, and singlet oxygen quenchers.[27] From table 1, the cooking time and temperature for canned white lingzhi (*Ganoderma lucidum*) fruit should be 90^{o}C, 4 minutes to maintaining the highest content of total phenolic and antioxidant activity.

This study showed that cooking temperature and time had an effect in the levels of phenolics and antioxidant activity in canned white lingzhi (*Ganoderma lucidum)* fruit. In general, the phenolic content of the lingzhi fruit (*Ganoderma lucidum)* decreased as the temperature was increased. Boiling vegetables for ≤5 minutes would be better for preserving or enhancing the total phenolic content.[7] When subjected to high temperatures, phenolics are decomposed or undergo oxidative condition which may reduce its amount that may be extracted from the sample.[27] Increasing temperature, more bound phenolics are released due to cellular breakdown together with the oxidative and hydrolytic enzymes. In respect of % DPPH scavenging activity of canned white lingzhi (*Ganoderma lucidum*) fruit, it decreased with the increasing of cooking temperature and time. A research found that the cooking process can be beneficial since it softens the vegetable tissues, consequently leads to better extraction of bioactive compounds.[28] One study found out that pressure cooking signifcantly improves the scavenging abilities of three species of mushroom. The increased in antioxidant activity at high temperature could also indicate that water insoluble and heat resistant compounds might be released from the rigid tissues or have been developed in mushrooms during cooking.[29] One research assessed the effects of three different cooking methods (boiling, microwave and pressure cooking) on the antioxidant activities of six different types of oyster mushrooms (*Pleurotus eryngii*, *P citrinopileatus*, *P. cystidiosus P. flabellatus*, *P. floridanus* and *P. pulmonarius*). Pressure cooking improved the scavenging abilities of *P. floridanus* (>200 %), *P. flabellatus* (117.6 %), and *P. pulmonarius* (49.1 %) compared to the uncooked samples. On the other hand, the microwaved *Pleurotus eryngii* showed 17 % higher in the reducing power (TEAC) value when compared to the uncooked sample.[30]

Table 1. Efffect of cooking time and temperature on the total phenolic and antioxidant activity of canned white lingzhi (*Ganoderma lucidum)* fruit

Cooking time and temperature	80^{o}C, 6 minutes	90^{o}C, 4 minutes	100^{o}C, 2 minutes
Total phenolic (mg CE/ 100g)	7.45±0.03^{c}	9.78±0.01^{a}	8.59±0.02^{b}
Antioxidant activity (% DPPH scavenging activity)	74.19±0.02^{c}	80.03±0.01^{a}	77.29±0.03^{b}

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant (α = 5%).

Table 2. Efffect of salt concentration on the total phenolic and antioxidant activity of canned white lingzhi (*Ganoderma lucidum)* fruit

Salt concentration (g/L)	0	1	3	5	7
Total phenolic (mg CE/ 100g)	9.78±0.01^{d}	10.33±0.02^{c}	11.21±0.03^{b}	13.41±0.02^{a}	13.50±0.02^{a}
Antioxidant activity (% DPPH scavenging activity)	80.03±0.01^{d}	82.19±0.03^{c}	83.47±0.02^{b}	85.14±0.02^{a}	85.20±0.02^{a}

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant (α = 5%).

3.2 Effect of salt concentration on the total phenolic and antioxidant activity of canned white lingzhi (*Ganoderma lucidum)* fruit

In order to obtain the longest shelf life for a product, osmotic treatment by salt is typically employed in association with other methods of food preservation.[31] Salt is the second most-used food additive, after sugar, in the food processing industry. Salt has diverse functions, ranging from simple flavoring or flavor enhancement to providing complex functional properties in various food systems. Salt is usually added to canned vegetables during processing primarily for flavoring since thermal processing provides the preservative function.[32] Osmotic treatment is a method that is often used to maintain the quality and stability of dry food products.[33] It helps to minimize the thermal damage on color, flavor, and texture, and prevents enzymatic browning of dried products.[34]

From table 2, the optimal salt concentration for cooking the canned white lingzhi (*Ganoderma lucidum*) fruit should be 5 g/L to maintaining the highest content of total phenolic and antioxidant activity. This result was similar to finding by one research when they found that sodium chloride 5 % was adequate for mushroom preservation during 90 days.[35] According to one study, the mushrooms preserved in 1.5 % brine were characterized for a week at room temperature.[36] The effects of different levels of osmotic pretreatments prior to drying and different drying methods on nutritional quality of dried mushroom slices was evaluated.[37] As salt concentration increased from 0 to 5 and 10%, the protein content reduced from 26.78 to 25.99 and 24. 95%, the fat reduced from 2.42 to 2.19 and 1.94, and fiber from 12.82 to 9.41 and 9.01%, respectively.[38, 39, 40]

IV. CONCLUSION

Ganoderma lucidum (Reishi) is a popular mushroom which has been used as medicine because of its promoting effects on health and life expectancy. *G. lucidum* contains tremendous compounds with a high grade of biological activity, which increase the immunity and show antitumour, antimicrobial, anti-inflammatory, antioxidant and acetylcholinesterase inhibitory activity. Fresh *Ganoderma lucidum* can't be stored for a long duration. In order to accelerate its added value for human consumption, we have successfully investigated different aspects in processing of canned white lingzhi (*Ganoderma lucidum*) fruit body.

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