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Physicochemical Properties of *Chaga* (*Inonotus obliquus*) Mushroom Powder as Influenced by Drying Methods

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Abstract

The effects of drying methods on the physicochemical properties of *chaga* (*Inonotus obliquus*) mushroom powder were investigated. Scanning electron micrograph revealed that freeze drying produced smaller particle-sized samples which in turn resulted in higher porosity than did vacuum and hot-air drying. Samples prepared by freeze drying showed a significantly higher L^*-value as compared with those prepared by hot-air drying and vacuum drying ($p<0.05$). The lightness (L^*-value) significantly decreased with increasing relative humidity and storage temperature regardless of drying method ($p<0.05$). The yellowness (b^*-value) increased significantly with increasing relative humidity ($p<0.05$). Browning index was significantly lower in samples prepared by freeze drying ($p<0.05$) but not significantly different between samples dried by hot-air and vacuum drying. Freeze dried sample exhibited a significantly higher degree of rehydration than other samples ($p<0.05$) probably due to the small particle size. Water solubility of the freeze dried sample was higher than those of the other methods while swelling ratio of the same sample appeared to be lower than those of others. Freeze dried *chaga* mushroom powder contained significantly lower amount of total phenolics and total sugar as compared to other samples ($p<0.05$).

Key words: *chaga* mushroom, powder, physicochemical, quality, drying methods

INTRODUCTION

Mushrooms have received much interests due to their widespread applications in functional food as well as a source of physiologically beneficial medicines (1-3). They are good sources of thiamine, riboflavin and niacin and they contain all the essential amino acids. Mushrooms have also been used for thousands of years as some of the most effective, yet benign, of many plants that formed the pharmacopoeia of Oriental herbal medicines. *Inonotus obliquus* (*chaga*) mushroom is a black parasitic fungus that grows on birch, alder, beech and other hardwood trees in colder Northern climates between latitudes 45°N and 50°N (3,4). *Chaga* is a highly prized medicinal fungus that has been used in Siberian folk medicine as a cleansing and disinfecting substance, often used to treat stomach discomforts (5-7).

The fungus produces a thick mass on the trunks of trees sometimes measuring up to 40 cm thick and 1.5 meters in length. Folk medicine practitioners have traditionally removed these masses from the trees and prepared teas and other decoctions for the treatment of diseases ranging from stomach diseases, intestinal worms, liver and heart ailments and cancers with no observed side effects (8-17). *Chaga* is also rich in triterpenes, especially lanosterol-type triterpenes related to inotodiol. Other compounds isolated from *chaga* include betulin, polysaccharides, and soluble lignins.

More recent pharmacological studies using *chaga* in Poland, Russia, and the USA have shown anti-tumor activity related to the mammary glands and female sexual organs (9,10). The most active compound, inotodiol, has shown activity against influenza viruses A and B, and various cancer cells. Studies in Japan have also confirmed antiviral activity (inhibition of the protease enzyme of HIV-1) (18).

Chaga extract powder can be used to formulate various types of dietary supplement tablets and can be included in herbal tea blends. *Chaga* can also be blended with other medicinal mushroom extracts and powders with different species such as *shiitake* or *maitake*. These could be ideal powdered materials for formulating dietary supplements and natural cosmetics in improving immune system. Nevertheless, systematic studies on *chaga* mushroom drying are lacking in the literature and information on the physicochemical properties of *chaga* mushroom powder is very limited.

The goal of this study was to perform a systematic investigation of how different drying methods (*i.e.*,

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freeze drying (FD), vacuum drying (VD), and hot-air drying (AD)) influenced color, degree of rehydration, water solubility, swelling power, antioxidative properties represented by the amount of total phenolics and total sugars. These findings are of importance for further processing *chaga* mushroom to develop new types of functional foods.

MATERIALS AND METHODS

Materials and powder preparation

Fresh *chaga* mushrooms were obtained from the Korean Ginseng Corp. (Imported from Baikal Herb Ltd.; harvested in March 2006) and stored at room temperature before use. Three drying treatments were used on the samples as follows: 1. Hot-air drying: samples were dried at 50°C using a hot-air drying oven (DMC-122SP, Daeil Engr. Co., Korea) to a final moisture content of approximately 4~6%, moisture-free basis (MFB). 2. Vacuum drying: drying at 50°C and vacuum pressure of 0.1 MPa in a vacuum dry oven (VOS-301SD, Tokyo Rikakikai Co., Japan) to the final moisture content as stated earlier. 3. Freeze drying: samples were frozen at -45°C for 24 hr in a deep freezer (VLT 1450-3-D-14, Thermo Electron Corp., Asheville, NC, USA) prior to freeze drying using a freeze-dryer (PPU-1100, Tokyo Rikakikai Co., Japan) at a vacuum pressure of 8.5 Pa.

Dehydrated mushrooms were cut into approximately $3\times3\times3$ cm pieces and milled using an analytical mill (M20, IKA, Staufen, Germany) with different particle size sieves (D-55743, FRITSCH, Idar-Oberstein, Germany) to yield particle sizes of 150~250 μm. The powder was placed in a desiccator prior to measurements. For color and browning index measurements, samples were further stored in a standard static system of thermally stabilized desiccators at 20°C, 35°C, and 50°C. Binary saturated salt solutions made using laboratory grade and at least 99.5% pure LiCl, NaBr, and KNO_3 were used to control the water activity (a_w) at 0.11, 0.58, and 0.93, respectively.

Scanning electron microscopy (SEM)

The structures of freeze dried, vacuum dried, and hot-air dried samples were examined using a low vacuum scanning electron microscope (Hitachi S-4300, 5.0 kV, Hitachi Ltd., Tokyo, Japan) operated at 5.0 kV, WD 15 mm using the high vacuum mode. The samples were fixed on the SEM stub, which were subsequently coated with lead/platinum in order to provide a reflective surface for the electron beam. The coated samples were subsequently viewed under the microscope.

Color and browning index

Color of mushroom powder was evaluated using a colorimeter (model CR-200, Minolta Co., Osaka, Japan) calibrated with a white standard (Y=94.2, x=0.3131, y=0.3201), and reported as CIE L^*- (lightness), a^*- (redness), and b^*- (yellowness) values. Measurements were made in triplicate and mean values were compared.

One gram of *chaga* mushroom powder was diluted with 40 mL of distilled water. Ten mL of 10% tricholoacetic acid solution was added to the mixture and left to stand at room temperature for 2 hr. Absorbance was determined spectrophotometrically at 425 nm after filtering through a filter paper (Whatman No. 2).

Degree of rehydration, water solubility, and swelling power

Degree of rehydration was determined using the modified method of Medcalf and Gilles (19). Mushroom powder was dispersed in distilled water (1 g to 20 mL distilled water) with shaking for 1 hr at 120 rpm in a 20°C water bath. Samples were then centrifuged at $3,000\times g$ for 20 min. The centrifuge tubes were placed up side down for 1 min and the supernatant decanted. The degree of rehydration was calculated as follows:

$$\text{Degree of rehydration (\%)} = \frac{\text{Weight of rehydrated sample} - \text{Initial sample weight}}{\text{Initial sample weight}} \times 100$$

Water solubility and swelling power were determined following the modified methods of Dubois et al. (20) and Leach et al. (21). In the capped centrifuge tube, 0.1 g of sample was suspended in 10 mL distilled water. The samples were heated at 60°C for 30 min with stirring at 120 rpm. The heated sample was immediately cooled rapidly in an ice water bath for 3 min and centrifuged at $424.84\times g$ at 4°C for 30 min. The supernatant decanted, samples were dried for 3 hr at 105°C. Percentage water solubility and swelling power were determined using the following formulas:

$$\text{Water solubility (\%)} = \frac{\text{Dry weight}}{\text{Sample weight}} \times 100$$

$$\text{Swelling power (g/g)} = \frac{\text{Weight of swollen sample}}{\text{Sample weight} \times (100 - \text{\% Solubility})} \times 100$$

Total phenolics

The phenolic contents of *chaga* mushroom powder were measured by a modified Folin-Ciocalteu colorimetric method (22). Each sample (10 g) was extracted with 50 mL of 80% methanol for 1 hr, and then the

methanolic extract was added to a tube containing deionized water (final volume 100 mL). The mixture was filtered through a Whatman filter paper 2. Nine mL of the diluted extract was mixed with 1 mL of Folin-Ciocalteu reagent. After 3 min, 1 mL of 40% sodium carbonate was added to the mixture and left to stand at room temperature for 1 hr. Absorbance was determined spectrophotometrically at 725 nm. Tannic acid was used as a standard and determination of total phenolics was carried out in triplicate.

Total sugars

To analyze the total sugars in the sample, 1 g of *chaga* mushroom powder was mixed with 10 mL of distilled water. Total sugars were determined using a phenol-sulfuric method (20). All measurements were done in triplicate.

Statistical analysis

All experimental data for each treatment were analyzed by ANOVA, and Duncan's multiple range tests (α=0.05) were performed to determine any significant difference among various treatments using SAS.

RESULTS AND DISCUSSION

Scanning electron micrograph (SEM)

Completely different surface structures of freeze-dried, vacuum dried, and hot-air dried materials were observed by SEM, as shown in Fig. 1. There were differences in size and density of the particles among the samples. Freeze dried samples had more orderly, densely, and highly porous structure, while vacuum dried and hot-air dried samples were less porous and more spacious and a somewhat broken structure was noticed especially in hot-air dried *chaga* mushroom powder. Uniform and fine structure of freeze dried *chaga* mushroom powder could be due to removing the majority of water content by sublimation. The highly porous structure of the freeze dried sample was also seen in garlic powder (23) and ginger powder (24).

Color

Table 1 illustrates the color differences in the *chaga* mushroom powders. Since the enzymes that caused the quality degradation were destroyed during processing *via* high temperature of 50°C or freezing, the non-enzymatic browning was considered a major cause of color changes of *chaga* mushroom powder. The lightness (L^*-value) decreased significantly with increasing relative humidity and storage temperature regardless of drying method ($p<0.05$). The reduction of lightness was greater at higher drying temperatures for all drying methods. In addition, hot-air dried sample showed the lowest L^*-values for all temperature conditions. Temperature changes from 20 to 50°C appears to lead more decrease in L^*-values than did changes in relative humidity from 11 to 93%, regardless of drying method. Hot-air dried *chaga* mushroom powder had lower L^*-values than those of freeze dried and vacuum dried samples for all conditions.

The redness (a^*-values) did not show the clear trend with temperature and relative humidity variation, however a^*-values of vacuum dried sample was slightly higher than those of freeze dried and hot-air dried sample. However, the yellowness (b^*-value) increased significantly with increasing relative humidity ($p<0.05$). Temperature did not have a major effect of b^*-values. b^*-values of vacuum dried sample were also slightly higher than those of freeze dried and hot-air dried sample.

Browning index

Browning reaction in food during storage is heavily temperature and moisture dependent. The effects of relative humidity and storage temperature, and drying methods on the browning index of *chaga* mushroom powder are shown in Table 2. The freeze dried sample had the highest value of 0.221, vacuum dried and hot-air dried sample showed 0.183 and 0.161, respectively. Browning

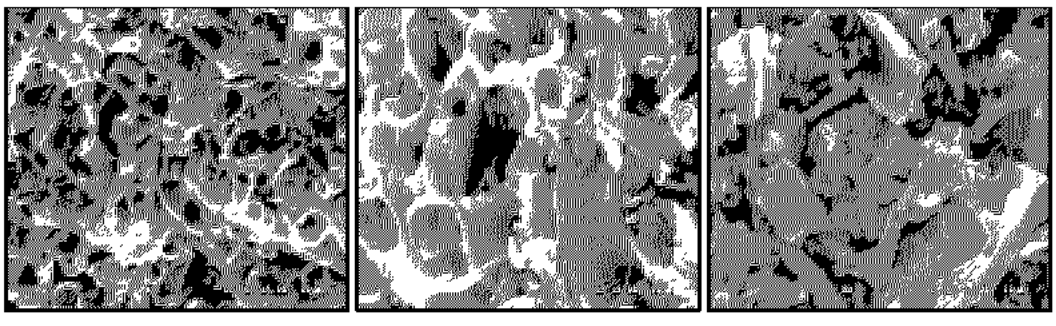

Fig. 1. Scanning electron microscope photographs (5.0 kV ×5000) of *chaga* mushroom powder prepared by different drying methods.

Table 1. Effects of drying methods, storage temperature, and relative humidity on color and browning index of dried *chaga* mushroom powder

Drying method	Temperature (°C)	Relative humidity (%)	L^*	a^*	b^*	Browning index (O.D. at 420 nm)
Freeze drying		Control	43.84^{a}	8.13^{e}	14.49^{g}	0.221^{g}
	20	11	42.90^{b}	7.95^{ef}	15.48^{e}	0.232^{f}
		58	42.79^{c}	8.26^{d}	17.54^{d}	0.250^{e}
		93	42.76^{c}	8.64^{b}	18.08^{c}	0.275^{c}
	35	11	40.47^{f}	7.83^{f}	14.44^{g}	0.264^{d}
		58	41.42^{d}	8.13^{e}	15.42^{f}	0.271^{c}
		93	39.27^{g}	8.76^{a}	18.44^{b}	0.281^{b}
	50	11	40.92^{e}	8.21^{d}	15.68^{e}	0.282^{b}
		58	41.02^{e}	8.18^{e}	13.57^{h}	0.285^{b}
		93	37.79^{g}	8.32^{c}	20.08^{a}	0.295^{a}
Vacuum drying		Control	43.11^{a}	8.43^{d}	16.89^{d}	0.183^{g}
	20	11	42.59^{b}	7.90^{e}	15.90^{f}	0.268^{f}
		58	42.59^{b}	8.84^{b}	18.23^{b}	0.281^{e}
		93	41.32^{d}	8.88^{a}	18.14^{c}	0.288^{e}
	35	11	41.05^{e}	8.49^{d}	16.58^{e}	0.350^{d}
		58	41.26^{d}	8.46^{d}	16.95^{cd}	0.374^{c}
		93	40.17^{e}	8.59^{bc}	16.89^{d}	0.398^{b}
	50	11	41.59^{bc}	8.59^{bc}	14.75^{g}	0.355^{d}
		58	41.46^{c}	8.58^{bc}	16.18^{f}	0.439^{ab}
		93	38.65^{f}	8.56^{c}	19.89^{a}	0.450^{a}
Hot-air drying		Control	42.41^{a}	7.94^{d}	15.04^{c}	0.161^{h}
	20	11	41.56^{b}	8.06^{c}	15.93^{bc}	0.219^{g}
		58	41.89^{ab}	8.31^{b}	16.39^{b}	0.243^{f}
		93	39.73^{cd}	8.87^{a}	17.47^{a}	0.263^{e}
	35	11	40.03^{c}	7.65^{f}	13.84^{f}	0.380^{d}
		58	40.28^{c}	7.94^{d}	14.26^{d}	0.401^{c}
		93	37.22^{e}	8.01^{c}	15.04^{c}	0.476^{b}
	50	11	39.71^{d}	7.73^{e}	12.28^{g}	0.385^{d}
		58	39.74^{cd}	6.99^{g}	14.01^{e}	0.412^{c}
		93	36.09^{f}	7.97^{d}	15.04^{c}	0.480^{a}

$^{a-h}$Means with different letters in the same column within the drying method are significantly different according to Duncan's multiple range test ($p<0.05$).

Table 2. Effects of drying methods on degree of rehydration, water solubility, and swelling powder of dried *chaga* mushroom powder

Drying method	Degree of rehydration (%)	Water solubility (%)	Swelling power (g/g)
Freeze drying	382.39±16.16^{a}	14.53±3.19^{a}	5.61±1.29^{b}
Vacuum drying	353.24±3.00^{b}	8.33±0.20^{b}	7.99±0.79^{a}
Hot-air drying	340.10±17.25^{c}	8.20±2.04^{b}	8.02±2.45^{a}

$^{a-c}$Means with different letters in the column are significantly different according to Duncan's multiple range test ($p<0.05$).

index also increased with increasing relative humidity and storage temperature. Browning indices were are significantly different among the three drying methods. Vacuum drying resulted in a higher browning index than did freeze drying and hot-air drying at higher 50°C and lower drying temperatures 20°C, except at 50°C and 93% relative humidity, at which hot-air dried sample showed the highest browning index of 0.48. However, hot-air dried sample showed a higher browning index value at the medium temperature of 35°C than those of freeze and vacuum dried samples in all relative humidity.

A higher degree of non-enzymatic browning occurring during hot-air drying and vacuum drying might be due to both Maillard reaction and ascorbic acid oxidation. An abrupt increase in browning index at 35°C and 58% relative humidity was clear in vacuum and hot-air dried

samples, which are in good agreement with the findings of Labuza et al. (25). Ko et al. (26) also showed similar results with *shiitake* mushrooms and Kim et al. (27) with red pepper.

Degree of rehydration, solubility, and swelling power

Degree of rehydration, water solubility, and swelling power of *chaga* mushroom powder dried by different methods differed significantly ($p<0.05$) (Table 2). Rehydration is not the reverse of drying (or dehydration). During the process of drying, textural changes, solute migration, and volatile losses occur and they are irreversible. Especially, heat reduces the degree of hydration of starch and the elasticity of cell walls and coagulates proteins to reduce their water-holding capacity. In other words, the rate and extent of rehydration may be used as an indicator of food quality, those foods that are dried under optimum conditions, such as freeze drying, suffer less damage and rehydrate more rapidly and completely than poorly dried foods. Structural and geometrical differences resulting from different drying methods probably accounted to some extent for the differences in water sorption properties of the freeze-dried and spray dried materials.

The freeze dried sample showed the highest degree of rehydration and water solubility, but showed the lowest swelling power. Those effects were due to the compact and the highly porous structures as visualized by SEM, presenting a high water accessibility. Similar results were reported with garlic powder (23) and ginkgo nut powder (28). In the case of vacuum drying and hot-air drying, during the process of drying, the samples undergo severe temperature treatment resulted in a lower degree of rehydration. Kim (29) also reported the rehydration rate of freeze dried *lycium chinense* Miller was 3 times higher than that of hot-air dried sample.

Vacuum-dried and hot-air dried *chaga* mushroom powder exhibited a higher swelling power than that of the freeze dried sample. Lorenz and Hinze (30) explained that molecular interactions became weaker during hot temperature treatment causing the swelling power to increase. Kim et al. (28) showed a similar effect of drying temperature with ginkgo nut powder.

Total phenolics and total sugars

Phenolic compounds in plant foods include a wide range of compounds and a broad spectrum of functional activities. These compounds have been considered important in plant foods because of their flavor and color, especially enzymatic browning, currently there is substantial interest in their potential health benefits, antioxidant activity, and antimicrobial effects.

Table 3. Effects of drying methods on total phenol and total sugar contents of dried *chaga* mushroom powder

Drying method	Total phenolics (mg/mL)	Total sugar (mg/mL)
Freeze drying	474 ± 4^{c}	14.48 ± 0.05^{b}
Vacuum drying	495 ± 12^{b}	14.96 ± 0.02^{b}
Hot-air drying	556 ± 33^{a}	15.82 ± 0.01^{a}

[a-c]Means with different letters in the column are significantly different according to Duncan's multiple range test ($p<0.05$).

The amount of total phenolics in processed *chaga* mushroom powders with different drying methods are shown in Table 3. The hot-air dried sample contained the highest total phenolics as compared to other drying methods. A similar result was shown with hot-air dried tomatoes (31). It is probably due to the liberation of phenolic compounds from the matrix during the process.

Haard and Chism (32) wrote that plants normally contain high contents of phenolics, as the metabolic intermediates, which usually accumulate in the vacuoles. Food processing might accelerate binding of phenolic compounds, and oxidative and hydrolytic enzymes releasing from the breakdown of cellular constituents. Even though these enzymes would destroy the phenolic compounds, high temperature hot-air drying processing would deactivate these enzymes and avoid the loss of phenolic acids and; therefore, lead to the increase of total phenolics. Cha et al. (33) also reported that polyphenolic compound contents of *cudrania tricuspidata* were increased with increasing bleaching temperature. The amounts of total sugar in processed *chaga* mushroom powders dried by different methods are shown in Table 3. Total sugar content of hot-air dried sample was significantly higher than those dried by other methods.

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