Differences in Polysaccharide Containment Obtained by Various Pulsed Light Irradiated Waste Mushroom Base of Yellow Flammulina Velutipes

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Abstract. The goal of this study was applied on the pulsed light of physical method, to promote the polysaccharide containment of mushroom base of Yellow Flammulina velutipes (Jinhua mushroom). Jinhua mushroom is a specific edible and medicinal mushroom in Taiwan that contains a variety of nutrients and bioactive components such as vitamin, carbohydrate, dietary fiber, protein, ergosterol, amino acid and umami constituents acknowledged to be beneficial for human health. Jinhua mushroom, whose appearance is also similar to white Flammulina velutipes but the functional ingredients are more than it. The polysaccharide of Jinhua mushroom base, which was irradiated the 0–80 pulses of pulsed light. Interestingly, the polysaccharide molecular weight was in the range of 3.56 to 4.30 x 10⁶ Da of base with pulsed light (0–80 pulses), along with increasing pulsed light irradiated pulses, the polysaccharides’ viscosity (cPs) was increased. Meanwhile, we determined the moisture adsorption ability and thermal decomposition of the Jinhua mushroom base’s polysaccharide, which could be applied to a design during cosmetic materials and biomedicals. Overall, this study is a complete, forward-looking and innovative research project in agricultural waste recycling.

1. Introduction

Yellow Flammulina velutipes (also call Jinhua mushroom) has become more popular to Taiwanese in recent years; its distinct texture and unique flavor are utterly different from white Flammulina velutipes. According to the literature, the biological activities are associated with the molecular weight distribution of polysaccharide. In addition, Pholiota nameko polysaccharide was purified by fractional precipitation, already demonstrated to have the antioxidant activity of polysaccharides [1].

This study was aim of the polysaccharides of Jinhua mushroom base by pulsed light irradiation of physical method. We applied physical treatment to irradiate the Jinhua mushroom base with pulsed light to study its polysaccharides content, to compare the base by different doses of pulsed light irradiated, and then contrasting the polysaccharide of ingredients, from which could be learned the reliable irradiation pulses of Jinhua mushroom base. In addition, the polysaccharides were obtained by hot water extracted of the base of Jinhua mushroom, which will irradiate at 0 (blank experimental control was unirradiated), 10, 30, 50, and 80 pulses by pulsed light [2].

Differential scanning calorimetry (DSC) was used to determine the energy changes of the moisture desorption and thermal decomposition of the polysaccharide of Jinhua mushroom base [3]. A novel
approach to discover the differences in polysaccharides obtained from different doses irradiation of the Jinhua mushroom base, that includes the viscosity characteristics, moisture desorption ability, and thermal decomposition properties. Moreover, we also compared the differences in structural functional groups after irradiation by infrared spectroscopy analysis. Therefore, this study can be applied to design during food processing, cosmetics processing, heat treatment, and storage conditions.

2. Materials and methods

2.1. Materials
Yellow Flammulina velutipes (strain with yellow colour) were donated by Da-Tian Farm Co. Ltd. (Changhua, Taiwan). The base (YB0) of ca. 4.0 cm in length is the waste part (see Figure. 1.).

![Figure 1. Photograph of sample treatment.](image)

2.2. Irradiation
Samples were subjected to irradiation using a Xenon RC-801 pulsed light system (Xenon Corp.) employing the RC-847 cabinet, lamp housing LH-840 and C-type 16″ linear lamp (190–700 nm). The apparatus generated three pulses per second at 169 J pulse⁻¹ [2]. An average dose was 11.50 kJ m⁻² pulse⁻¹ (Koyyalamudi et al. 2011). Each mushroom base was thinly scattered, placed in the middle of a stainless-steel plate and irradiated for 0, 10, 30, 50, and 80 pulses.

2.3. Jinhua mushroom base polysaccharides
The samples after irradiation were freeze-dried and ground into a fine powder (60 mesh) by using a mill (Retsch ultracentrifugal mill and sieving machine, Haan, Germany). Different doses of pulsed light irradiation of 0 (un-irradiated), 10, 30, 50, and 80 pulses, which are YB0, YB10, YB30, YB 50, and YB 80, respectively, were extracted with water in a 1: 30 (w/w) ratio at 121°C for 15 min. The mixture was cooled to room temperature and filtered by Whatman No. 4 filter paper. The combined filtrate was dialyzed by using a Cellu Sep T2 tubular membrane (MWCO: 6,000-8,000, Membrane Filtration Products, Inc., Seguin TX, USA) for 24 h. The retentate was concentrated to a small volume and then mixed with three volumes of 95 % ethanol [2,3]. The precipitate thus obtained was lyophilized and ground using a Retsch mill to obtain coarse powder of hot water extracted polysaccharides from 0, 10, 30, 50, and 80 pulse sample.

2.4. Jinhua mushroom base polysaccharides viscosity analysis
All samples were evaluated at room temperature, conducted by sine wave Vibro-viscometer A&D SV-10 (A&D Company, Limited, Tokyo, Japan), that was under constant frequency at 30 Hz and amplitude less than 1 mm [2].
2.5. Jinhua mushroom base polysaccharides molecular distribution analysis

The polysaccharide molecular weight distributions were applied on the high performance liquid chromatography (HPLC) with a Hitachi HPLC apparatus equipped with a 2490 refractive index (RI) detector. Polysep-GFC-P 4000 and Polysep-GFC-P 1000 (7.8 × 300 mm, Phenomenex) were employed after connection in series, and the temperature of the column was kept at 30 °C. The sample concentration was 1.0 mg mL⁻¹, and its injection volume was 10 μL. The eluent was ultrapure water, and the flow rate was 1.0 mL min⁻¹ [2,3].

2.6. Jinhua mushroom base polysaccharides infrared spectroscopy analysis

All sample was added into ca. 150 mg of dried KBr; the sample was fully mixed with KBr after grinding, and then put in a tablet machine pressed into a disc. Infrared spectrum were acquired with a Bruker Alpha FTIR spectrometer. Collections of spectra were in the transmission mode with an unpolarized light beam, at a resolution of 4 cm⁻¹ with eight scans, and a spectral range of 4000–400 cm⁻¹ [4].

2.7. Jinhua mushroom base polysaccharides DSC tests

DSC TA Q20-RCS90 (TA Instruments, USA) for conducting DSC analysis, the samples were sealed in 20 μL aluminum pans, and then the lid was pressed onto the crucible using the pressure of a heavy mechanical force and the seal tightened the crucible [2,3] Approximately 2.0 mg of the sample was used for received the thermal analysis experimental data. Non-isothermal tests of the heating rate selected for the programmed temperature ramp were 2, 4, 6, and 8 °C min⁻¹ for the range of temperature rise chosen from 30 to 300 °C for each thermal analysis.

3. Results and discussion

3.1. Polysaccharide viscosity and molecular distributions of different dose pulsed light irradiations

Pulsed light irradiation induced significant changes in the viscosity of the polysaccharide solution as shown in Table 1. As the irradiation dose increased from 0 to 80 pulses, the YB0–YB80 polysaccharide solution showed a significant increase in viscosity. The viscosity of the YB0–YB80 polysaccharide was from 1.77 to 1.86 cP (mPa·s), whereas the viscosity of the high viscosity 1.86 cP was irradiated at 80 pulses. The increase in the viscosity is attributed to the increase in degree of polymerization of polysaccharide [2]. In addition, the irradiation dose increased from 0 to 80 pulses, as the dose of 80 pulses, the polysaccharides had the greater molecular weight 4.30 x 10⁶ Da.

| Sample | Yield (%) | Viscosity (cPs) | Molecular weight (10⁶Da) |
|--------|-----------|----------------|-------------------------|
| YB0    | 5.47ᵃ     | 1.77ᵈ         | 3.56                    |
| YB10   | 4.49ᵉ     | 1.80ᶜ         | 3.93                    |
| YB30   | 4.63ᵈ     | 1.83ᵇ         | 2.95                    |
| YB50   | 4.75ᶜ     | 1.86ᵃ         | 3.84                    |
| YB80   | 5.14ᵇ     | 1.86ᵃ         | 4.30                    |

*Means with the same letter within a column are not significantly different (P>0.05).

3.2. Polysaccharides of different dose pulsed light irradiations by infrared spectroscopy analysis

Figure 2 displays the hydroxyl (–OH) groups of the stretching-vibration absorption peak wavenumbers, which were 4000–3850 cm⁻¹ and 3700–3100 cm⁻¹; of which the characteristic functional group of polysaccharide hydroxyl group clearly exhibited in the infrared spectrum. In addition, the polysaccharides of intermolecular hydrogen bonds 3650–3200 cm⁻¹ were particularly noticeable. For the O–H stretching-vibration and bending-vibration absorption peak the wavenumbers were 3000 cm⁻¹, 1550 cm⁻¹, 1400 cm⁻¹ and 980 cm⁻¹, respectively. The C–O stretching-vibration absorption peak
wavenumber was $1650 \text{ cm}^{-1}$, and C–O–H bending-vibration absorption peak wavenumber was $1580 \text{ cm}^{-1}$, respectively, which is also clearly shown in the spectrogram. From comparison of Table 1 and Figure 2, the results of polysaccharide molecular weight distributions and infrared analysis were consistent. We obtained the judgment, as Jinhua mushroom was irradiated by the pulsed light reaction, that it initially increases the molecular weight of the polysaccharide, along with continuous irradiation producing an effective resistance of subsided phenomenon; then continued increasing the dose pulsed light irradiation kept increasing the molecular weight of the whole polysaccharide.

**Figure 2.** Different doses of pulsed light irradiations infrared spectrogram.

**Figure 3.** Selected DSC tests of the YB0–YB80 polysaccharides at heating rate $4 \text{ °C min}^{-1}$ of the range of temperature rise chosen from $30–300 \text{ °C}$.

**Table 2.** DSC analysis of YB0–YB80 polysaccharides with heating rate 2, 4, 6, and $8 \text{ °C min}^{-1}$ of the range of temperature rise chosen from $30–300 \text{ °C}$.

| Sample | Heating rate | desor$T_o$ | desor$T_p$ | desor$\Delta H$ | decompp$T_o$ | decompp$T_p$ | decompp$\Delta H$ |
|--------|--------------|------------|------------|----------------|--------------|--------------|-----------------|
| YB0    | 2            | 81.80      | 127.54     | 238.20         | 227.44       | 237.70       | 72.69           |
|        | 4            | 93.45      | 140.81     | 87.83          | 230.37       | 242.91       | 66.75           |
|        | 6            | 91.13      | 130.29     | 247.70         | 240.80       | 251.77       | 118.20          |
|        | 8            | 87.93      | 161.39     | 228.20         | 225.38       | 234.47       | 87.98           |
| YB10   | 2            | 99.38      | 158.63     | 212.90         | 211.01       | 226.03       | 85.77           |
|        | 4            | 149.20     | 156.56     | 125.30         | 226.36       | 241.12       | 75.27           |
|        | 6            | 121.71     | 164.12     | 184.20         | 218.85       | 227.17       | 75.00           |
|        | 8            | 91.62      | 138.15     | 214.80         | 243.23       | 254.47       | 102.1           |
| YB30   | 2            | 91.93      | 133.89     | 214.80         | 223.78       | 233.95       | 71.01           |
|        | 4            | 94.18      | 159.45     | 193.70         | 227.00       | 240.31       | 74.40           |
|        | 6            | 104.48     | 151.31     | 283.60         | 232.39       | 245.69       | 101.70          |
|        | 8            | 103.37     | 151.52     | 202.30         | 239.99       | 251.92       | 101.90          |
| YB50   | 2            | 87.10      | 129.68     | 189.00         | 228.64       | 238.31       | 79.09           |
|        | 4            | 95.66      | 146.37     | 182.80         | 227.08       | 241.42       | 79.66           |
|        | 6            | 115.81     | 169.83     | 185.80         | 224.86       | 244.70       | 74.71           |
|        | 8            | 98.87      | 147.18     | 175.10         | 237.06       | 252.82       | 115.20          |
| YB80   | 2            | 84.19      | 131.38     | 226.10         | 228.40       | 239.80       | 98.66           |
|        | 4            | 100.03     | 152.96     | 162.90         | 230.87       | 243.84       | 87.40           |
|        | 6            | 105.44     | 146.67     | 181.50         | 238.46       | 250.72       | 117.60          |
|        | 8            | 93.65      | 150.27     | 207.40         | 242.53       | 255.83       | 108.20          |
3.3. Polysaccharides of different dose pulsed light irradiations by DSC thermal analysis

From Table 2, the initial conversion (onset) temperatures of moisture desorption and thermal decomposition were ca. 93 and 230 °C (selected YB0 at heating rate 4 °C min⁻¹), respectively. The thermal decomposition behavior was by heat treatment, that also obtained the Jinhua mushroom base polysaccharide’s peak maximum temperature of ca. 240 °C (see Figure 3). Furthermore, Table 2 shows YB0–YB80 polysaccharides of Jinhua mushroom base have superior absorbent properties, as the YB0–YB80 desorption of moisture was heated more than 140 °C to reach equilibrium. It is essential for the polysaccharides of Jinhua mushroom involving the hydroxyl group (–OH) and the ether group (–O–) that they have good moisture absorbance and high sensitivity to ambient humidity [2]. Among all polysaccharide samples, the YB80 have the greater molecular weight 4.30 x 10⁶ Da and viscosity 1.86 cP. The results of DSC thermal analysis proved again that polysaccharides of Jinhua mushroom base could increase the molecular weight by pulsed light irradiation. As also, the Jinhua mushroom base with different doses of pulsed light of 0, 10, 30, 50, and 80 pulses, along with increasing the temperature of moisture desorption to reach equilibrium, as YB80 illustrated its equilibrium temperature more than 152 °C. Obviously, after pulsed light irradiation, the polysaccharides’ structure was changed in this study.

4. Conclusion

Comparisons of the different doses of pulsed light of 0–80 pulses showed that the Jinhua mushroom, acquired and demonstrated after pulsed light, the polysaccharides’ structure was changed. Overall, the result of viscosity, molecular distribution, infrared spectroscopy, and DSC thermal analysis of different dose pulsed light irradiations was demonstrated; the Jinhua mushroom was irradiated by the pulsed light reaction. In the initial period it could be increased the molecular weight of the polysaccharide, during continuous irradiations producing an inhibition response, but continued increasing the dose pulsed light irradiation, which promoted the molecular weight for whole polysaccharide. This study is a forward-looking and innovative research project of the improved functional ingredients of Jinhua mushroom in food processing and heat treatment. In the future, our goal is to enhance the processing technology for health foods of edible and medicinal mushrooms to find improved processing technology and energy efficiency, a less polluting method and improved functional ingredients technology in the food, medicines and food safety areas.

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5. References

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