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Eutectic point and freeze-drying curve of Tremella fuciformis containing sucrose

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Abstract

The eutectic point and drying curve of food multicomponent materials are the key to improve the efficiency of freeze-drying. The eutectic point, co-melting point, pre-freezing time, heating temperature and freeze-drying curve of Tremella fuciformis containing sucrose were investigated. The eutectic point and co-melting point temperature of materials were - 15.49 °C and - 8.35 °C respectively measured by differential scanning calorimetry (DSC). At the pre-freezing temperature of -25 °C for 3.5 h, the moisture of the sample was completely transformed into glassy ice. Different heating temperatures were set in sections during the drying process of multicomponent materials. The best heating temperature was 80°C for 3 hours, then 70 °C for 5 hours, then 60 °C for 12 hours, and finally 50 °C to constant dryness. Under the optimal operating conditions, Tremella fuciformis entered the desorption drying stage at the 8th hour, and the material could be dried to less than 5% moisture content at 25 h.

Keywords: *Tremella fuciformis*; freeze-drying; differential scanning calorimetry; drying curve.

Practical Application: Freeze-drying of *Tremella fuciformis* containing sucrose.

1 Introduction

Tremella fuciformis is a kind of edible fungus for medicine and food, containing polysaccharides, 18 kinds of amino acids and other nutrients. The main pharmacologically active ingredient of *T. fuciformis* is polysaccharides (Wu et al., 2019). This mushroom polysaccharide has various biological activities, including ameliorated ulcerative colitis, anti-oxidation and anti-tumor (Niu et al., 2021). In recent years, with the continuous acceleration of people's life rhythm, traditional white fungus dry products have been unable to meet people's needs for white fungus products. Instant Tremella Soup is a kind of Tremella product that is ready to eat after brewing. It can not only meet people's demand for Tremella products, but also meet the fast-paced life needs of modern consumers to reduce cooking time.

At present, people are conscious of the increasing demand for health products. In order to decrease the harm caused by preservatives, people continue to improve the drying methods of fresh food (Ratti, 2001). Many studies had shown (Zhong et al., 2017; Huang et al., 2010) that Tremella fuciformis is suitable for short-term storage, and long-term storage will inevitably breed microorganisms, and harmful metabolites are produced such as mycolic acid and sulfur dioxide. Research by Li Yahuan (Li et al., 2016) and others has shown that freeze-drying Tremella fuciformis has the best texture and nutritional content, and freeze-drying can reduce the accumulation of harmful metabolites.

Freeze-drying technology is a drying method for preparing high-quality instant *Tremella fuciformis* soup. The freeze-drying temperature was low, which can reduce the loss of heat-sensitive substances such as crude protein, polyphenols and vitamins when the edible fungus was dried (Li et al., 2019). Compared with other drying methods, the soluble substances in the material were prevented from moving outward due to the internal moisture gradient diffusion, causing nutrient loss; the resulting product has a sponge-like porous structure with good rehydration (Oikonomopoulou et al., 2011; Ma et al., 2017; Li, 2011). However, in order to prevent the melting of ice crystals in the material, the freeze-drying sublimation temperature should not be too high. More importantly, the thermal conductivity of the porous material under the vacuum was low, which leads to longer freeze-drying time (Berk, 2018). On the one hand, the equipment has to keep cooling, on the other hand, heat needs to been maintained, and vacuum state needs to be maintained. As a result, the operation cost of the equipment was higher, and the production cost was higher than other drying methods such as hot air drying and microwave drying (Law et al., 2014). On the other hand, in order to improve the palatability of instant *Tremella fuciformis* soup and meet consumer demand, manufacturers have added sucrose, wolfberry and other auxiliary materials to instant *Tremella fuciformis* soup, making it a multi-component solution before drying, with complex ingredients and one of multiple solutes. The crystallization situation was complicated. The part of the cooling treatment water freezes into crystal ice, and the crystalline part of other components forms a glass body, especially sucrose makes the material more difficult to freeze-dry, and the product more easily absorbs moisture.

In view of the complexity of the multiple ingredients of instant *Tremella fuciformis* soup, the long drying time and the

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large blindness of process parameter control during the drying process, this study optimizes the freeze drying conditions of instant *Tremella fuciformis* soup, first using differential scanner calorietry (DSC) measured the eutectic point and eutectic temperature of the material (Kanari & Ozawa, 2003; Menczel et al., 2009; Harvey et al., 2018), and determined the pre-freezing temperature and other process parameters based on this. Then, based on this, the pre-freezing time, drying temperature and time and other process conditions were studied. Determine the best drying conditions, draw the freeze-drying curve, in order to reduce the freeze-drying time of instant *Tremella fuciformis* soup, improve drying efficiency, and reduce production costs (Ma et al., 2018).

2 Materials and methods

2.1 Materials

Fresh *Tremella fuciformis* was obtained from Youxi County, Fujian Province, China. Before the experiment, fresh *Tremella fuciformis* was stored in a refrigerator at a temperature of 5 ± 1 °C and a relative humidity of 90%.

2.2 Sample pretreatment

Fresh *Tremella fuciformis* removed pedicle and culture medium, washed with tap water and dried, cut into 1.0 cm ~ 1.5 cm × 1.0cm ~ 1.5 cm pieces of *Tremella fuciformis*. *Tremella fuciformis* add 1.5 times the weight of distilled water, add sucrose, boiled for 5 minutes, boiled process to continue to stir, to prevent tar. The boiled *Tremella fuciformis* was evenly spread on a stainless steel tray at a volume of 120g±1g per serving. The thickness of the sample was 10.0 ± 2.0mm. The eutectic point and eutectic point of some samples were determined by DSC, and the remaining samples were placed in a -25 °C refrigerator (DW-40L188, Haier Medical Laboratory Products Co., Ltd.) for 4 hours.The specific operation process is shown in Figure 1.

2.3 Freeze-drying

Samples frozen in a -25 °C refrigerator were quickly transferred to the pre-cooled lyophilizer (SCIENTZ-30ND freeze dryer, Ningbo Xinzhi Biotechnology Co., Ltd., China) . The cold trap temperature of lyophilizer was -50 °C ± 2 °C, and the vacuum degree was 10 Pa - 20 Pa. The operating parameters of the lyophilizer were set according to the test requirements.

2.4 Determination of eutectic point and co-melting point

A differential calorimetry scanner (DSC 200F3. German Netzsch Instrument Co., Ltd.) was used to detect the eutectic point and co-melting point of *Tremella fuciformis* soup material. Measurement parameters: under N_2 gas, cooling from 25 °C to -60 °C at a rate of 10 °C/min, record the eutectic point; under N_2 gas, heating from -60 °C to 25 °C at a rate of 5 °C/min, Record the co-melting point.

3 Results and discussion

3.1 Determination of eutectic point and co-melting point

The eutectic point temperature and the co-melting point temperature were the important basis for the freeze-drying process to set the pre-freezing temperature, sublimation temperature and cold trap temperature. Therefore, the determination of the

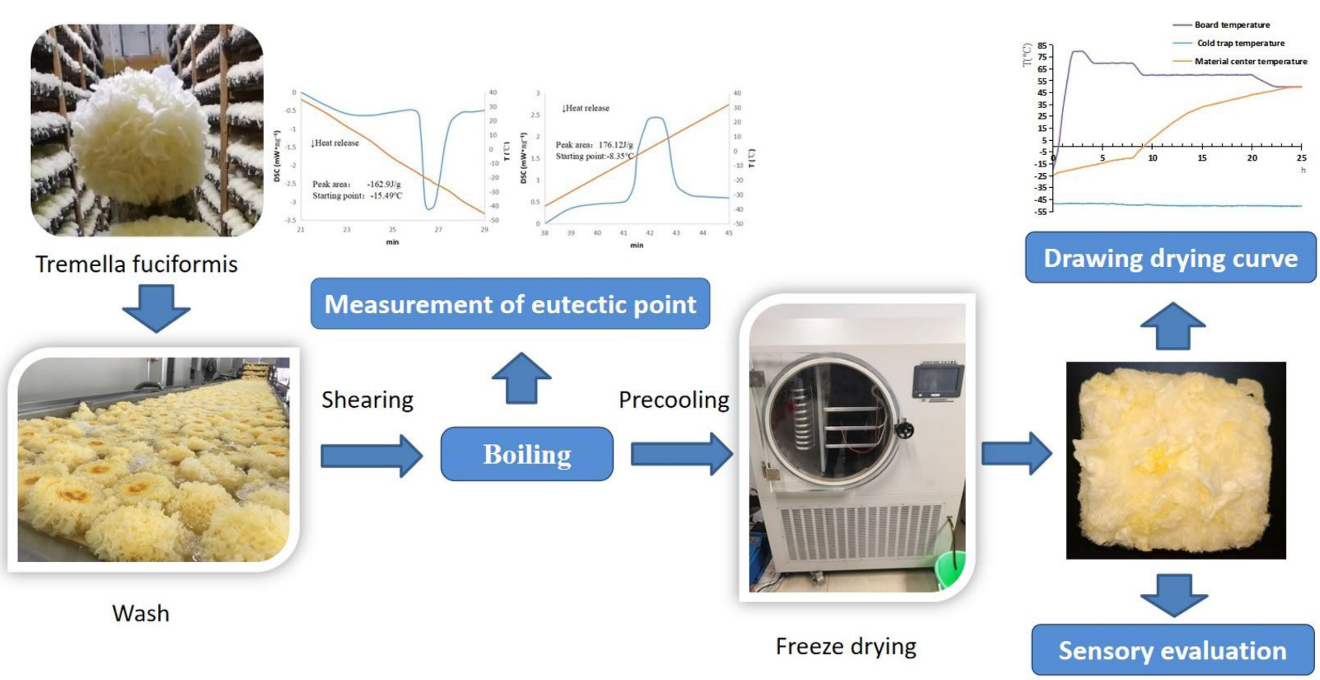

Figure 1. Schematic description about freeze-drying of *Tremella fuciformis*.

eutectic point temperature and the co-melting point temperature was of great significance.

The DSC method was used to determine the eutectic point and the eutectic point of the sucrose-containing *Tremella fuciformis* Soup material (Zhao et al., 2013). First, the temperature was lowered to -60 °C at a cooling rate of 10 °C/min, and then the temperature was increased to 25 °C at the same rate. In this process, two obvious One was the crystallization peak during cooling, and the other was the eutectic peak during heating. Through software analysis, the eutectic point temperature of the material was -15.49 °C and the Co-melting point temperature was -8.35 °C. It can be understood from Figure 2 that as the temperature continues to decrease, the free water in *Tremella fuciformis* soup continues to freeze, and the heat release of the material maintains a balance. There was an exothermic peak between 26 and 28 minutes, which drops sharply to -15.49 °C. The heat release curve changes the fastest. This was because *Tremella fuciformis* soup material needs to release a large amount of latent heat of phase change during the process of changing from liquid to solid. It also shows that the moisture in the material was frozen and solid. This temperature was the eutectic point of *Tremella fuciformis* soup. Figure 3 shows the change of heat absorption during the thawing process of *Tremella fuciformis* soup material. There was an obvious absorption peak between 41 min and 44 min, that was, at -8.35 °C, the DSC curve suddenly shows a peak change, indicating that the material was changing from solid to liquid. During the process, the latent heat of the phase change will be absorbed, and free-moving ions will appear. Therefore, the starting point, the Co-melting point was -8.35 °C.

Reasonable determination of the pre-freezing temperature was very important in the freeze-drying process. If the final freezing temperature was set too high, the material cannot be completely frozen. It was easy to cause partial boiling and foaming during vacuum sublimation, so that the material cannot be guaranteed during drying. All the water in the material was directly sublimated and removed in the form of ice, so that part of the water in the material to be dried was vaporized in the liquid state, and shrinkage and loss of shape will occur during drying (Song, 2008). In addition, because the dissolved substances contained in the moisture in the unfrozen part cannot be precipitated in situ, but migrate to the surface with the internal moisture, causing the surface of the freeze-drying product to harden. If the freezing temperature was too low, it will not only extend the freeze-drying time, cause unnecessary waste of energy, but also cause greater damage to the material and reduce its quality. The eutectic point temperature was the temperature at which all the moisture in the material freezes into ice. In production research, in order to achieve complete freezing of the material, a temperature of 5-10 °C lower than the eutectic point was generally selected for freezing. If the pre-freezing temperature was too low, the freezing energy consumption were increased; if the temperature was too high, the material cannot be completely frozen, so it was determined that the final pre-freezing temperature of *Tremella fuciformis* soup material was -25 °C.

The Co-melting point temperature was the temperature at which the temperature of all materials frozen into ice was

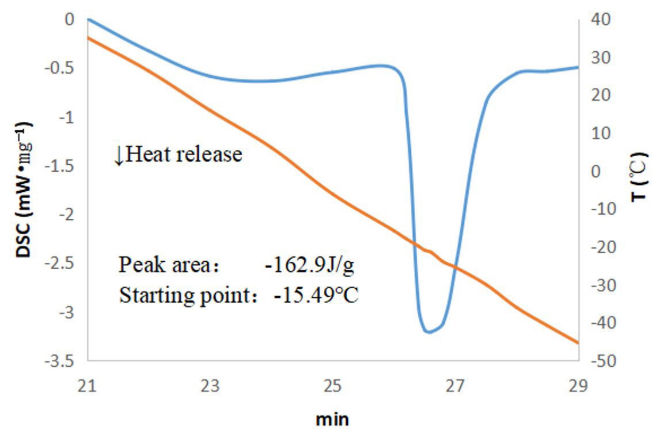

Figure 2. DSC cooling curve of sucrose-containing *Tremella fuciformis* soup material.

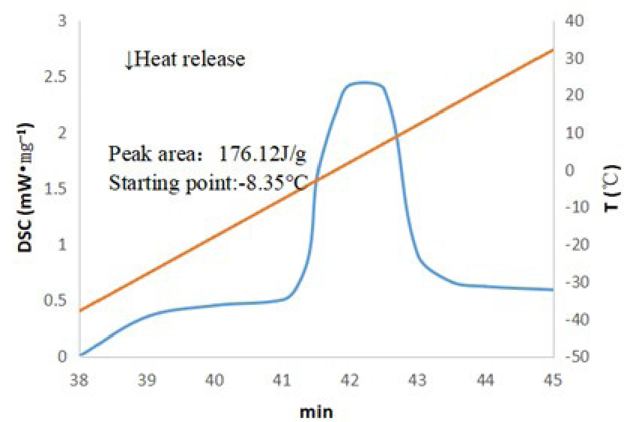

Figure 3. DSC heating curve of sucrose-containing *Tremella fuciformis* soup material.

raised to the moment when ice crystals begin to melt. In the freeze-drying sublimation stage, the temperature of the material cannot exceed the eutectic temperature. When the temperature of the material exceeds the eutectic temperature, the material will melt, and boiling under vacuum conditions will damage the structure of the product, and may also cause the loss of the material With a loose and porous structure, the relative density increases and the color becomes darker (Zhang et al., 2015). Therefore, in order to ensure the quality of dry products, in the sublimation drying stage to ensure that the temperature of the material does not exceed the Co-melting point , try to increase the temperature of the heating plate during the sublimation drying process to shorten the drying time.

3.2 Determination of pre-freezing time

Put the materials in the freeze-drying bin and equilibrate to the pre-freezing temperature. At this time, the materials were not completely frozen and cannot be directly dried by sublimation.

The samples will freeze until the temperature was maintained for 1 to 2 hours. Therefore, the pre-freezing time was determined by measuring the freezing curve of the material. The freezing curve of 2.5cm thick *Tremella fuciformis* soup material when the pre-freezing temperature was -25 °C was shown in Figure 4. The temperature of *Tremella fuciformis* soup material entering the warehouse was 32 °C, and the temperature was cooled to -2 °C in 35 minutes, and the average rate was 0.97 °C/min. Between 35 and 55 minutes, the temperature drops slowly and was maintained at about -2 °C, because when the material changes from liquid to solid, more phase change heat was absorbed, so the temperature drops slowly (Daraoui et al., 2010). When the material completes the phase change heat stage, the cooling rate increases. At 100 minutes, the temperature drops to -18 °C, with an average rate of 0.36 °C/min. After 100 min, the temperature drop rate becomes slower. Because the temperature difference between the material temperature and the air medium becomes smaller, the temperature drop rate becomes smaller accordingly. The temperature drop rate was maintained at -23 °C for 150 min, and the material can be used for about 0.5 h. It reached the pre-freezing temperature, so this study determined that the freezing time of *Tremella fuciformis* soup at the pre-freezing temperature of -25 °C was 3.5 h, at which time *Tremella fuciformis* soup was completely frozen.

3.3 The influence of the heating plate temperature on the material temperature and product quality in the sublimation analysis process

Put the samples of *Tremella fuciformis* Soup pre-frozen to -25 °C on the freeze-drying shelf. When vacuum air was pumped from the freeze-drying bin to 20 Pa, set the shelf heating temperature to 30 °C, 40 °C, 50 °C, and 60 °C. Perform sublimation analysis tests at 70 °C and 80 °C to analyze the changes in the material temperature during the sublimation and analysis process under different shelf heating temperatures. The results were shown in Figure 5. The test results show that at the six temperatures set in the test, as the shelf heating temperature increases, the time required for sample sublimation analysis shows a downward trend. The sublimation stage of the sample heated at 30°C takes the longest time, which was 12h, and the sublimation stage of the sample heated at 80 °C takes the shortest time of 7.5 hours; it showed that increasing the shelf heating temperature in the sublimation analysis stage can effectively shorten the freeze-drying processing time. However, it was found in the experiment that when the shelf heating temperature was 70 °C and 80 °C, bubbling occurred on the surface of part of *Tremella fuciformis*. This may be due to the rapid heating of the sublimation stage and the sublimation stage has not yet ended and directly entered the analysis stage. In addition, the test found that the higher the temperature, the partial color of the product deepened, and the brightness and whiteness decreased. This may be because the temperature was too high and the product has part of the Maillard reaction.

After sensory evaluation, *Tremella fuciformis* soup prepared at 30 °C, 40 °C, 50 °C and 60 °C has a thick soup shape within 5 minutes of brewing, with stretched ears, moderately soft and hard texture, and good quality; The products prepared at the heating temperature of 70 °C and 80 °C have relatively low viscosity after brewing, and the ears were relatively hard.

In conclusion, the higher the heating plate temperature, the higher the drying rate of *Tremella fuciformis* soup material, but too high temperature will affect the product quality. From the analysis of the material center temperature during the drying process, *Tremella fuciformis* soup material can be heated in stages. In the sublimation stage, as long as the material center temperature was not higher than the Co-melting point temperature, increase the temperature as much as possible. In the analytical drying stage, the shelf heating temperature was controlled at 50 °C ~ 60 °C was appropriated, too high temperature will affect the color of the product.

3.4 The effect of heating plate temperature control program on freeze-drying time and quality of instant Tremella fuciformis soup

Comprehensive consideration of freeze-drying efficiency and freeze-drying product quality, set up three heating plate temperature control procedures to investigate the impact on

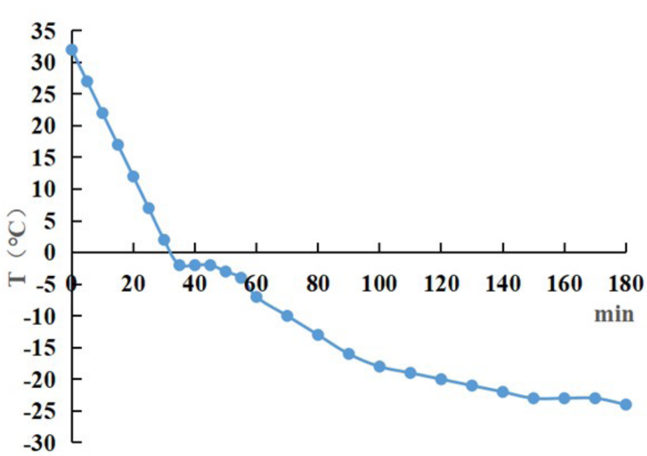

Figure 4. The freezing curve of *Tremella fuciformis* soup at a refrigeration temperature of -25 °C.

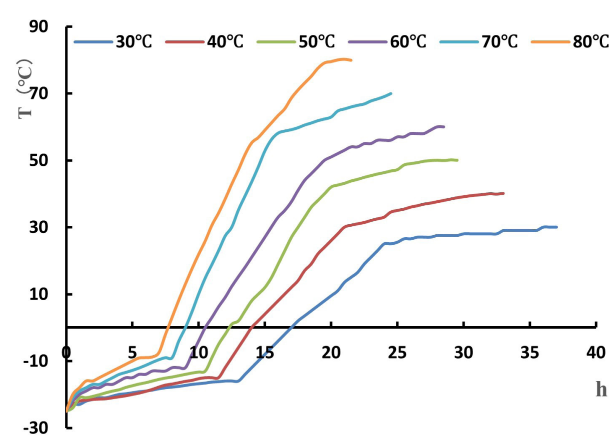

Figure 5. The influence of heating temperature on the material temperature of *Tremella fuciformis* soup in the sublimation analysis process.

Table 1. The influence of the heating plate temperature control program on the freeze-drying time and the quality of instant *Tremella fuciformis* soup.

Temperature control program	Drying time (h)	Moisture content	Rehydration ratio	Product sensory evaluation
A	22.47±0.63[a]	3.71±0.21[a]	10.13±0.29[b]	Light yellow, with a small amount of bubbling on the surface, the soup has a certain consistency, and the texture of the ears was hard
B	23.45±0.57[ab]	3.87±0.13[a]	11.38±0.45[a]	It was off-white and has a good shape. The soup was smooth and sticky when tasted; the ears were stretched and the texture was moderately soft and hard. However, the product was easy to absorb moisture due to the high difference between the product and the room temperature when the product was out of the warehouse.
C	24.56±0.51[b]	2.96±0.23[b]	11.55±0.36[a]	Off-white, the soup has a smooth and sticky feeling when tasting; the ears were stretched, and the texture was moderately soft and hard

Note: Different letters indicate significant differences in the data of each group ($p<0.05$). The temperature control program is A: 80 °C 6h → 70 °C 5h → 60 °C to constant dry; B: 80 °C 3h → 70 °C 5h → 60 °C to constant dry; C: 80 °C 3 h → 70 °C 5 h → 60 °C 12 h → 50 °C to constant dryness.

freeze-drying time and product quality (Pang et al., 2017). It can be seen from Table 1 that the moisture content of the samples prepared by the three temperature control programs was less than 5%, and the drying time was arranged in order as A < B < C, but the heating plate was set to A temperature control program (80 °C 6 h → 70 °C 5 h → 60 °C to constant drying), the temperature was too fast, the product has a small amount of bubbling, and the color was yellow, the rehydration ratio was the same as B (80 °C 3 h → 70 °C 5 h → 60 °C to constant drying) and C. There were obvious differences in temperature control procedures, and the latter two have better product quality. On the other hand, the temperature of the B program product was 60 °C when it was out of the warehouse, which was too high from room temperature. The product was easy to absorb moisture and was not easy to operate in the large-scale production process. Therefore, the C temperature control program was selected as the appropriate setting temperature for the heating plate. That is, 80 °C 3 h → 70 °C 5 h → 60 °C 12 h → 50 °C to constant dryness.

3.5 Freeze drying curve drawing

Pre-freeze *Tremella fuciformis* soup material in the refrigerator at -25 °C for 3.5h and place it on the freeze-drying shelf. When the freeze-drying bin pumps vacuum to 20 Pa ± 5Pa, the shelf heating temperature of the freeze dryer was set to 80 °C 3 h → 70 °C 5 h → 60 °C 12 h → 50 °C to constant drying, record the change of the material center temperature and the temperature of the heating shelf during the freeze-drying process, and draw the freeze-drying curve, as shown in Figure 6.

It can be seen from the freeze-drying curve in Figure 6 that the temperature of the material in the early stage of freeze-drying was relatively low, but as the sublimation progresses, the temperature of the material gradually rises. When the sublimation drying progresses to 7.5 h, the center temperature of the material reaches the Co-melting point temperature. At this time, the sublimation drying stage basically ends; after 8h, the material enters the analytical drying process, and the center temperature of the material continues to rise, when the drying progresses to 23.5 h , The center temperature of the material was already close to the temperature of the heating shelf, and the entire freeze-drying process ends when the time was extended to about 25 h. The freeze-drying time was 30 h at a heating temperature of 50 °C. This freeze-drying temperature control program

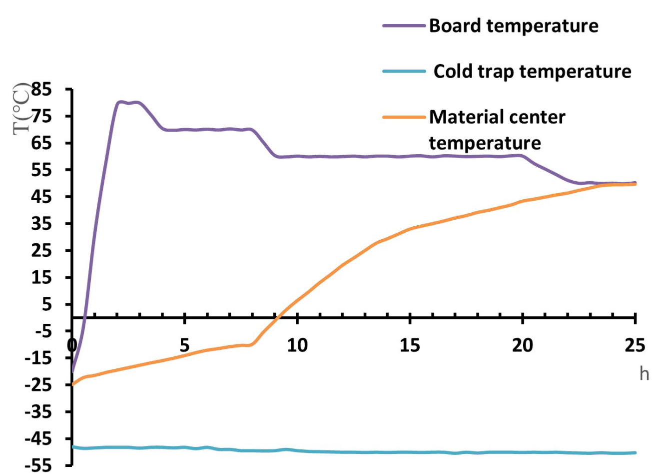

Figure 6. Freeze-drying curve of *Tremella fuciformis* soup.

reduces the operating time by 20% and reduces the production cost(Schössler et al., 2012).

3.6 Sensory evaluation

The instant *Tremella fuciformis* soup developed by this project did not add thickeners. It was evaluated by a sensory evaluation team composed of 10 R&D personnel and consumers. The product was beige. After adding hot water above 90 °C, Within 3 minutes, the product soup was thick and thick, the ears were stretched, and the texture was moderately soft and hard.

4 Conclusions

1. Using DSC, the eutectic point temperature of *Tremella fuciformis* soup material after adding sucrose and other auxiliary materials is -15.49 °C, and the Co-melting point temperature is -8.35 °C. On this basis, it is determined that the pre-freezing temperature is -25 °C, and it is clear that the pre-freezing temperature of *Tremella fuciformis* soup is maintained at -25 °C for 3.5 hours, which can ensure that the 2.5 cm thick material can be totally frozen.

2. The higher the heating plate temperature, the higher the drying rate of *Tremella fuciformis* soup material, but too high temperature will affect the product quality. In the analysis and drying stage, the heating temperature of the shelf should be controlled at 50 °C ~ 60 °C. Excessive temperatures will affect the color of the product. Considering the freeze-drying efficiency and the quality of freeze-drying products, select 80 °C 3 h → 70 °C 5 h → 60 °C 12 h → 50 °C to constantly dry as the heating plate temperature control program.

3. The freeze-drying curve of *Tremella fuciformis* soup shows that the temperature of the center of the material continues to rise, and analytical drying is entered after 8 h, and the entire freeze-drying process is completed in about 25 h. The freeze-drying time is 30 h at a heating temperature of 50 °C. This freeze-drying temperature control program reduces the operating time by 20% and reduces the production cost.

4. The product is off-white. After adding hot water above 90 °C to brew, the product soup will be thick and thick within 3 minutes, the ears will be stretched, and the texture will be moderately soft and hard.

Acknowledgements

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