

# Grifola Frondosa

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*Grifola frondosa* (*G. frondosa*) is a Basidiomycetes fungus that belongs to the family of Grifolaceae and the order of Polyporales.

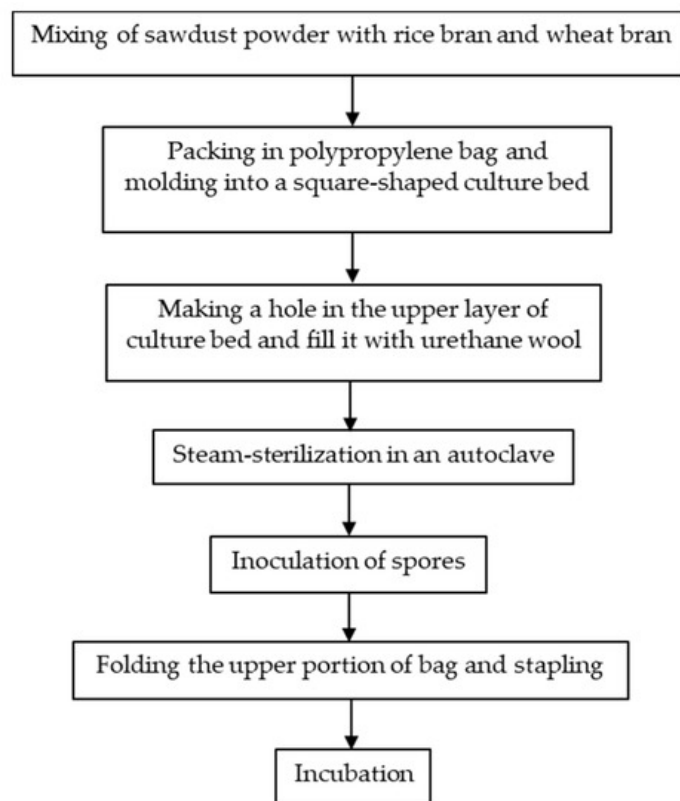
Keywords: *Grifola frondosa* ; maitake mushroom ; polysaccharide

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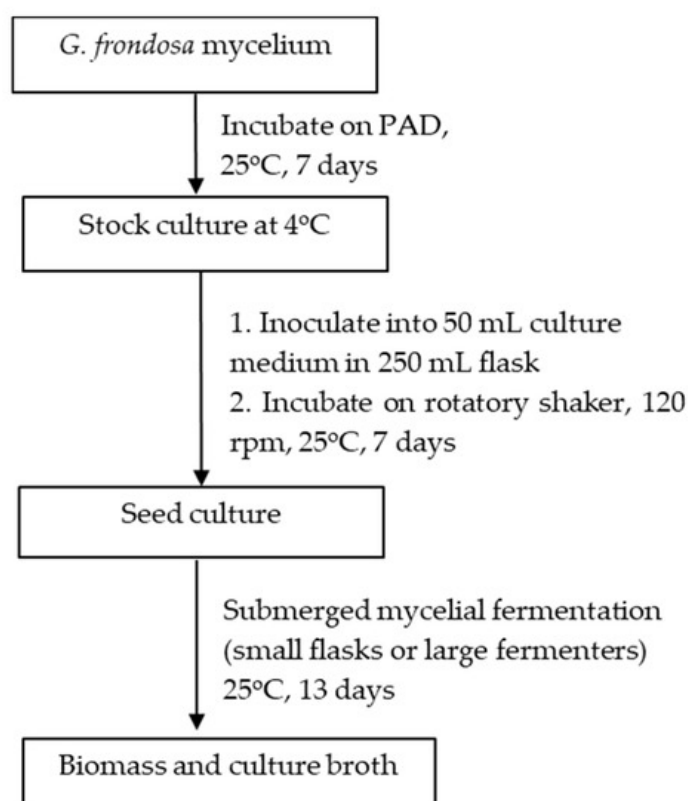
## 1. Introduction

In Japan *Grifola frondosa* (*G. frondosa*) edible fruiting body is known as maitake. In Japanese, *mai* means dance and *take* means mushroom. *G. frondosa* is known as “hui-shu-hua” (grey tree flower) in Chinese, possibly due to its appearance. *G. frondosa* grows around the stumps of broadleaf trees or trunks and is edible when young. The environment of the northeastern part of Japan is suitable for the growth of *G. frondosa*. The temperate forests in eastern North America, Europe and Asia are also ideal for its growth. Meanwhile, it is a common mushroom in the United States and Canada, known as sheep’s head, king of mushrooms, hen-of-the-woods, and cloud mushroom <sup>[1]</sup>.

Japan was one of the countries that first started the artificial cultivation of *G. frondosa* in the mid-1980s. There are in general three methods for the artificial cultivation of the *G. frondosa* fruiting body, they are bottle culture, bag culture and outdoor bed culture. Bag culture is the most popular cultivation method in Japan <sup>[2]</sup> because of its advantages such as the low cost of plastic bags, small space requirements and easily-controlled indoor environment. Bag culture can achieve higher yields of mature *G. frondosa* mushrooms than bottle culture and requires a shorter cultivation time than outdoor bed cultures. As shown in [Figure 1](#) <sup>[2]</sup>, the major steps of bag cultivation include substrate preparation, substrate sterilization, mycelium inoculation and incubation. In addition to the fruiting body, there is also an increasing demand for *G. frondosa*’s mycelium and its bioactive metabolites. Solid-state fermentation (SSF) <sup>[3]</sup> and submerged fermentation <sup>[4]</sup> are two common methods of mycelium cultivation. A common substrate for SSF is sawdust supplemented with rice bran or wheat bran <sup>[5]</sup>. Submerged or liquid fermentation is usually more efficient, providing a higher mycelial productivity in a shorter time, requiring smaller plant space and allowing for more effective product quality control <sup>[6]</sup>. A typical submerged fermentation process is presented in [Figure 2](#).



**Figure 1.** A typical bag culture procedure for the *G. frondosa* fruiting body.



**Figure 2.** Submerged culture fermentation of the *G. frondosa* mycelium adapted from [4].

*G. frondosa* is edible and is regarded as a healthy food because it is a good source of protein, carbohydrates, dietary fiber [7][8][9][10][11][12][13], vitamin D<sub>2</sub> (ergocalciferol) [13][14][15] and minerals (K, P, Na, Ca, Mg) [7][9][12][15][16], with low fat content and caloric value [15]. *G. frondosa* is delicious, with a sweet and umami taste, which is mainly attributed to its high trehalose, glutamic and aspartic amino acid and 5'-nucleotide content [10][11][13][17]. Due to its delicious and special taste, *G. frondosa* is not only used as a food ingredient, but also as a food-flavoring substance in dried powder form. Apart from its high nutraceutical value, *G. frondosa* is reported to possess a wide range of pharmacological effects. *G. frondosa* was first discovered to have antitumor activity in the 1980s from hot water extracts of the *G. frondosa* fruiting body [16][17]. The major bioactive components were found to be  $\beta$ -glucans [17][18][19][20]. The D-fraction, a  $\beta$ -glucan complex with about 30% protein, was first discovered by Nanba's group in the late 1980s [21]. Since then, the D-fraction has been widely studied

and gradually developed into commercially available complementary medicines and healthcare products. In addition to the D-fraction, there are many other bioactive polysaccharide fractions that are obtained from *G. frondosa*, such as the MD-fraction [22], X-fraction [23], Grifolan [24], MZ-fraction [25] and MT- $\alpha$ -glucan [26]. The different polysaccharide fractions isolated from *G. frondosa* possess various bioactive effects such as immunomodulation [24], antitumor [25], antiviral [27], antidiabetic [26] and anti-inflammation [28]. In recent years, an increasing number of studies have attributed or linked the health and therapeutic effects of *G. frondosa* polysaccharides to their capacity for modifying gut microbiota, microorganisms that play an important role in human health and diseases. In particular, gut microbiota play a role in maintaining immune homeostasis, which may have a connection to the antitumor effects of polysaccharides [29]. The regulation of gut microbiota composition by *G. frondosa* polysaccharides has also been suggested to contribute to the treatment of metabolic disorders such as non-alcoholic fatty liver disease (NAFLD) [30] and diabetes [31], indicating their potential for preventing or treating hyperglycemia and hyperlipidemia. Apart from polysaccharides, other molecular fractions isolated from *G. frondosa* fruiting bodies or mycelial biomass have shown promising medicinal values as well. For instance, the protein components of *G. frondosa*, including glycoprotein, have shown anti-tumor [32], immune-enhancing [33], anti-diabetic, anti-hypertensive, anti-hyperlipidemic [34] and anti-viral effects [35]. Moreover, other small biomolecules in *G. frondosa* have been found to possess health benefits such as anti-inflammation [36], hypoglycemia [37], antitumor [38] and antioxidation [39].

## 2. Chemical and Nutritional Compositions

### 2.1. Proximate Composition

Generally, proximate composition is determined by the methods suggested by the Association of Official Analytical Chemists (AOAC). The total carbohydrate content can be calculated by subtracting the percentages of ash, crude fat and protein [7][40]. For the determination of crude protein, the nitrogen conversion factor is 4.38 instead of the usual 6.25, due to the large amount of chitin that is usually contained within the fungus, a component that may interfere with the correct calculation of the result of total nitrogen [41].

As shown in Table 1, *G. frondosa* is made up of around 83–96% moisture and 4–17% dry matter in its fresh fruiting body [7][8][9][10][11][12][13] and mycelium [11][17][42], indicating the watery texture of *G. frondosa*. Carbohydrates and protein are the major constituents contributing to the dry weight of *G. frondosa*, taking up around 70–80% and 13–21%, respectively, of the fruiting body. Based on the average values of component percentage, it could be found that the mycelium of *G. frondosa* has a similar moisture content, a lower content of carbohydrate and crude ash and a higher content of crude fat and protein, compared with the fruiting body of *G. frondosa*.

**Table 1.** Proximate composition of *G. frondosa*'s fruiting body and mycelium.

Components <sup>1</sup> (%)	Fruiting Body								Mycelium			
	[8] *	[9] #	[12] #	[10] 2,#	[13] #	[11] 2,#	[7] 2,#	Average	[43]	[44] 2	[11] 2	Average
Moisture	83.1	89.1	90.9	86.1	90.4	95.6	95.2	90.1 ± 4.5	84.8	96.7	92.3	91.3 ± 6.0
Dry matter <sup>3</sup>	16.9	10.9	9.1	13.9	9.6	4.4	4.8	9.9 ± 4.5	15.2	3.3	7.7	8.7 ± 6.0
Carbohydrate <sup>4</sup>	70.4	74.9	72.3	68.8	71.8	66.3	70.3	70.7 ± 2.7	66.3	45.0	60.4	57.2 ± 11.0
Crude ash	6.5	4.8	6.6	7.0	7.1	6.2	4.9	6.1 ± 0.9	6.4	4.0	4.7	5.0 ± 1.3
Crude fat	4.5	1.5	3.3	3.1	2.4	6.5	5.6	3.8 ± 1.8	4.2	24.7	6.5	11.8 ± 11.2
Crude protein	18.6	18.9	17.8	21.1	18.8	21.0	19.2	19.3 ± 1.3	23.1	26.4	28.4	26.0 ± 2.7

<sup>1</sup> Moisture and dry matter were based on fresh weight; others were presented based on dry weight. <sup>2</sup> For easy comparison, only the mean value is used. <sup>3</sup> The dry matter was represented by  $(1 - \text{moisture}/\text{total}) \times 100\%$ . <sup>4</sup> Amount of carbohydrate was calculated by subtracting crude ash, crude fat and crude protein. \* Fruiting body was grown naturally. # Fruiting body was grown artificially.

According to the research findings of Kurasawa and coworkers [8], the composition of the *G. frondosa* fruiting body resembles that of normal cultivated mushrooms. It is worth mentioning that the crude fat content of the *G. frondosa* fruiting body is generally lower than the average crude fat content in cultivated mushrooms (4.3%), and the amounts of protein and carbohydrates are slightly higher than the average of other mushrooms (17.2% and 70.3%), indicating the excellent nutritional values of *G. frondosa*.

## 2.2. Soluble Sugar Content

The content of soluble sugar within *G. frondosa* is mostly determined by the method described in the research work of Ajlouni and coworkers [42]. As shown in Table 2, the total sugar content in *G. frondosa* is higher in the fruiting body (90–190 mg/g) than in the mycelium (70–90 mg/g). The content of total sugar in the fruiting body of *G. frondosa* is also superior to some edible mushrooms such as *Lactarius glaucescens* and *Craterellus odoratus* [45], which may be one of the reasons for the good taste of *G. frondosa*. Table 2 also shows variations in both total soluble sugar content and individual sugar content among different *G. frondosa* samples, which may be attributed to factors such as cultivation period and cultivation environment. Trehalose, a disaccharide that comprises two molecules of glucose, is the major sugar component of both the fruiting body and mycelia of *G. frondosa* [9][10][11][43][44]. Compared with the amount in the mycelium (40–60 mg/g), the fruiting body contains more trehalose than the mycelium, around 50–160 mg/g in dry weight. In addition to trehalose, the fruiting body also contains glucose and mannitol, whereas the mycelium has glucose and mannitol, together with arabinol and fructose.

**Table 2.** Soluble sugar content of *G. frondosa* fruiting body and mycelium in dry weight.

Component	Fruiting Body (mg/g Dry wt.)			Mycelium (mg/g Dry wt.)		
	[9] #	[10] 1,#	[11] 1,#	[43]	[44] 1	[11] 1
Arabinose	n.d. <sup>2</sup>	n.d.	n.d.	n.d.	n.d.	5.37
Arabitol	n.d.	n.d.	n.d.	n.d.	12.65	2.01
Fructose	n.d.	n.d.	n.d.	1.00	n.d.	2.99
Glucose	59.30	14.02	2.42	8.00	19.72	2.18
Lactose	n.d.	n.d.	n.d.	n.d.	n.d.	0.93
Mannitol	7.20	9.36	1.00	n.d.	9.92	2.30
Mannose	n.d.	n.d.	n.d.	n.d.	n.d.	1.92
Ribose	n.d.	n.d.	8.34	n.d.	n.d.	4.04
Trehalose	45.80	161.83	99.94	65.00	41.60	65.32
Total	112.30	185.21	111.7	74.00	83.89	87.06

<sup>1</sup> For easy comparison, only the mean value is used. <sup>2</sup> Not determined or not detected. # Fruiting body was grown artificially.

## 2.3. Free Amino Acid Content

The content of free amino acids in *G. frondosa* was quantitatively measured by the method described in the research work of Mau and coworkers using HPLC [10][44]. Results regarding the free amino acid content of *G. frondosa* are exhibited in Table 3. The total free amino acid content in the fruiting body of *G. frondosa* is around 15–60 mg/g in dry weight, which is higher than that in many other edible mushrooms, such as *Dictyophora indusiata* and *Tricholoma giganteum* [10]. The mycelium of *G. frondosa* contains a relatively higher total free amino acid content in comparison with the fruiting body. There is also a great variety of amino acids in *G. frondosa*. There are around eighteen kinds of free amino acids, including essential amino acids such as L-histidine and L-methionine, in both the fruiting body and the mycelium of *G. frondosa*, indicating that *G. frondosa* is an excellent source of amino acids.

**Table 3.** Free amino acid assay of *G. frondosa*'s fruiting body and mycelium in dry weight.

Component (mg/g Dry wt.)	Fruiting Body			Mycelium		
	[13] 1,#	[11] 1,#	[10] 1,#	[44]	[11] 1	
	In Sawdust	In Log				
L-Alanine	2.15	3.13	5.22	2.77	3.26	14.59
L-Arginine	3.02	3.21	1.66	0.64	0.97	12.39
L-Aspartic acid	1.61	1.25	1.88	0.42	2.75	19.40
L-Glutamic acid	8.01	9.10	12.62	0.67	3.76	2.10
GABA	n.d. <sup>2</sup>	n.d.	0.28	n.d.	n.d.	17.09
Glycine	1.53	1.53	2.46	0.57	1.93	7.81
L-Histidine <sup>3</sup>	1.53	0.94	19.50	0.59	4.10	n.d.
L-Isoleucine <sup>3</sup>	0.12	0.12	0.56	0.33	2.80	6.67
L-Leucine <sup>3</sup>	0.05	0.09	0.27	0.35	4.92	6.39
L-Lysine <sup>3</sup>	1.56	1.28	5.70	1.11	0.22	23.49
L-Methionine <sup>3</sup>	n.d.	n.d.	4.50	1.40	0.67	n.d.
L-Phenylalanine <sup>3</sup>	0.26	0.28	2.71	0.80	1.66	9.98
L-Serine	2.91	2.82	2.01	0.97	2.73	10.74
L-Threonine <sup>3</sup>	1.43	1.44	n.d.	4.40	8.23	10.85
L-Tryptophan <sup>3</sup>	n.d.	n.d.	n.d.	0.27	n.d.	12.01
L-Tyrosine	1.77	0.73	1.53	n.d.	2.15	17.99
L-Valine <sup>3</sup>	0.96	0.91	0.39	0.60	4.13	9.41
Total	29.26	29.38	61.29	15.9	44.28	180.91

<sup>1</sup> For easy comparison, only the mean value is presented. <sup>2</sup> Not determined or not detected. <sup>3</sup> Essential amino acid. <sup>#</sup> Fruiting body was grown artificially.

However, large variations exist in the amount of each amino acid in *G. frondosa*. For instance, Mau et al. (2001) and Tsai et al. (2006) found that threonine was the major free amino acid in *G. frondosa* [10][44], whereas Huang and coworkers found that histidine and glutamic acid were the major free amino acids in the fruiting body (>12 mg/g) and lysine, aspartic acid and tyrosine were the major free amino acids in the mycelium (>17 mg/g) [11]. Huang and coworkers also obtained a larger amount of free amino acid from *G. frondosa* than other groups in both the fruiting body and the mycelium, a result that might be due to the different sources of the fruiting body and the preparation methods of the mycelium by the different research groups. The choice in cultivation substrate was also found to affect the variety and amount of amino acids in *G. frondosa*. As shown in Table 3, *G. frondosa* fruiting bodies cultivated in sawdust and log substrates have different amounts of each amino acid [13], although the total amino acid content was similar. Moreover, GABA (γ-aminobutyric acid), a biologically active compound which is related to the therapeutic effect of *G. frondosa*, is mainly detected in the mycelium but not in the fruiting body [11] (Table 3).

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