Jryza

ORYZA OIL & FAT CHEMICAL CO., LTD.

# WHITE JELLY FUNGUS EXTRACT

Sliming, Appetite Suppression, Fat Accumulation Inhibition, Fatty

Metabolism Enhancement, Constipation Relief, Moderated Intestinal

Function, Intestinal Flora Improvemament, Retaining Skin Moisture &

**Skin Beauty Effect** 

White Jelly Fungus Extract-P

(water-soluble powder, food-grade)

White Jelly Fungus Extract-PC

(water-soluble powder, cosmetic-grade)



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Sliming, Appetite suppression, Fat accumulation inhibition, Fatty metabolism enhancement, constipation relief, Moderated intestinal function, intestinal flora improvemament, retaining skin moisture & skin beauty effect

#### 1. INTRODUCTION

White jelly fungus (*Tremella fuciformis*) is an edible mushroom growing in broad leaf forests in Asian temperate zone including Japan. White jelly fungus constitutes a beautiful soft and transparent-white carpophore.

White jelly fungus is not only beautiful in its appearance, it also contains a number of nutrition components which are often deficient in females. It thus has been traditionally consumed as a beauty food item. It is said that the most famous ancient Chinese beauty, comcubine Yang of the Empire of the Tang Dynasty 1300 years ago, loved white jelly fungus as a beauty food item in royal cuisine. Today, upon established cultivation, white jelly fungus is a popular ingredient in home cooking. Especially as a desert in Chinese cuisine, white jelly fungus immersed in sweet syrup relishes uniquely and exotically, leaving behind an unforgettable luxurious tasting and texturing sensation.

Recently, investigations of ours and others provided convincing evidence for effects of white jelly fungus in suppressing fat accumulation and absorption, in constipation relief and in improving intestinal flora. In addition, white jelly fungus exhibits gastric mucosa protection and thus is used as a traditional Chinese medicine for prevention of gastric ulceration. Furthermore, effect of memory enhancement has been reported for white jelly fungus. Besides the beneficial physiological functionalities, the beautiful appearance of white jelly fungus has attracted scientists to understand the principle of beauty-formation.

Using our own special technique, Oryza Oil & Fat Chemical Co. Ltd. has succeeded in extracting polysaccharide from white jelly fungus, which we provide as White Jelly Fungus Extract. Composed of at least 80% total saccharides and 10-30% glucuronic acid, White Jelly Fungus Extract is a food item with a supple texture. It sucks water and swells, and gives rise to a large volume of highly

viscous transparent solution. No other polysaccharide can compete with white jelly fungus extract in high water-sucking capacity and in high viscosity in solution. Furthermore, viscosity of white jelly fungus extract is independent from pH change.

Growing body of evidence for the functionalities of whie jelly fungus extract ensures that our product White Jelly Fungus Extract will doubtlessly meet the needs of modern females in pursuit of health and beauty.



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# 2. Basic functionalities

2-1. Active sites of white jelly fungus extract



### 2-2. Chemical structure of white jelly fungus extract

White jelly fungus extract (WJF extract) is mainly composed of a 80~100 kD polysaccharide polymer, and small amount of protein and nucleic acid. The backbone structure of the polysaccharide is a long chain of repeating  $\alpha$ -1,3-mannoses (Fig. 2). Attached to mannoses are side chains composed of D-fucose, D-xylose and D-glucuronic acid. Glucuronic acid, the constituent sugar of hyaluronic acid, has a 17.6% content in WJF polysaccharide. The molar ratio of mannose, fucose, xylose and glucuronic acid is 9 : 1 : 4 : 3. WJF polysaccharide also contains a small amount of glucose, D-mannuronic acid and L-gluronic acid. The latter two are the constituent sugars of algin acid.



Fig 2. Structure of WJF Extract: repeating sugars

#### 2-3. Superior water-retaining capacity

WJF extract gives rise to a transparent and highly viscous solution which is very similar to that of hyaluronic acid. However, in contract to hyaluronic acid, the water-sucking and swelling capacity of the former is not significantly influenced by pH. In artificial intestinal fluid at pH7.4, both WJF extract and hyaluronic acid have similar water-retaining capacity (1.2 : 1).On the other hand, in acidic artificial gastric fluid at pH1.8, WJF extract holds 19-fold more water than hyaluronic acid (Fig. 3). In pH range of both acidic and neutral, WJF extract retains water of 450-fold of its own weight. Since swelling in stomach likely leads to satiety feeling, WJF extract is expected to suppress appetite. In addition, retaining large amount of water is beneficial for constipation relief.



		5
	e	In artificial intestinal fluid (folds of it's own weight)
WJF extract	461	476
Hyaluronic acid*	24	393

 Table 1.
 Water-retaining capacity of WJF extract and hyaluronic acid

\*Sodium hyaluronate, food grade

#### [Method]

Artificial gastric fluid (pH 1.8) or intestinal fluids (pH 7.4) was added to 2 g of WJF extract or hyaluronic acid. After 30 min dissolving at room temperature and subsequent 2 hours of swelling at 37°C, the mixtures were passed through No. 2 filter paper for 10 minutes by spontaneous gravity fall. The remained polysaccharides on the filter paper were weighted for calculation of their water-retaining capacities.

### 2-4. Anti-oxidative effect

When added to cultured mouse corneocytes and fibroblasts, WJF extract exhibited a dose-dependent SOD-like activity (Fig. 4). Furthermore, feton reaction revealed a dose-dependent active oxygen scavenging effect (Fig. 5).



Fig 4. SOD-like activity of WJF extract



Concentration of WJF extract

Fig 5. Active oxygen scavenging effect of WJF extract

# 3. Sliming effect

### 3-1. Activating glycolytic enzymes (in vitro)

Kiho<sup>1)</sup> *et al.* reported that polysaccharide extract from *Tremella* sp. (TP), which belongs to the same species of white jelly fungus, significantly increased activities of glucokinase, hexokinase and glucose-6-phosphate dehydrogenase in normal and diabetic mice. Their results suggest that ingestion of TP stimulates glycolysis and thus may prevent fat accumulation.

Group		Dose	Enzyme activity (mean ± S.E, n=5)		E, n=5)
		(mg/kg)	0	3	6(h)
Glucol	kinase				
N:	Control		$10.8 \pm 1.8$	$9.1\pm1.9$	$11.2\pm2.1$
N:	TP	50		$14.1 \pm 2.6^{*}$	$15.4\pm2.0$
D:	Control		$12.5\pm0.8$	$12.3\pm1.5$	$12.4\pm1.6$
D:	TP	50		$18.0\pm1.1*$	$19.5\pm2.0^*$
Hexok	tinase				
N:	Control		$19.1\pm3.4$	$18.4\pm1.9$	$20.7\pm3.7$
N:	TP	50		$29.8\pm0.6*$	$28.1\pm2.0*$
D:	Control		$17.7\pm1.4$	$15.0\pm0.7$	$15.1\pm1.9$
D:	TP	50		$20.0\pm1.5*$	$21.4\pm1.6^*$
Glucose-6-phosphate (G-6-P) dehydrogenase					
N:	Control		$12.7\pm1.9$	$13.0 \pm 2.4$	$18.9\pm3.1$
N:	TP	50		$22.7 \pm 3.1*$	$27.1 \pm 5.8$
D:	Control		$15.7\pm0.9$	$16.6 \pm 0.1$	$16.0 \pm 2.3$
D:	TP	50		$23.6 \pm 1.7*$	$30.4 \pm 8.0*$

Table 2.	Effect of TP on activities of glycolytic enzymes
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\* Normal mouse (N) and streptozocin-Induced diabetic mouse (D)

\* Significant difference from the control at p < 0.05, by Student's *t*-test.

1) Kiho, T., Morimoto, H., Kobayashi, T., Usui, S., Ukai, S., Aizawa, K., Inakuma, T. Effect of a polysaccharide (TAP) from the fruiting bodies of *Tremella aurantia* on glucose metabolism in mouse liver. *Biol. Pharm. Bull.*, **64**, 417-419 (2000). Data in table 2 originate from this publication.

### 3-2. Suppressing fat accumulation (in vivo)

Mice were administered 0.5 ml of 1% solution of WJF extract daily for 30 days. Same amount of methylcellurose was administrated to control group mice. Body weight and fat pad increase were suppressed in the test group.



#### [Method]

Four-week-old male mice (body weight:  $22 \pm 1$  g) were fasted for 12 hours before oral administration of either WJF extract or methylcellurose (control) once daily for 30 days. Body weight and fat pad around testis and kidney were measured at the end of the 30-day test period.

#### 3-3. Suppressing appetite (in vivo)

Kiho *et al.*<sup>2)</sup> reported TP addition to the drinking water at 0.5g/l decreased food intake for normal and non-insulin-dependent diabetic model mice. WJF extract is thus also expected to suppress appetite and be beneficial to calorie-controlling diet.

2) Kiho, T., Morimoto, Kochi, M., T., Usui, S., Hirano, K., Aizawa, K., Inakuma, T.,Effect of an acidic polysaccharide (TAP) from *Tremella aurantia* and its degredation product (TAP-H). *Biol. Pharm. Bull.*, **24**, 1400-1403 (2001).

#### 3-4. Suppressing fat absorption (*in vivo*)

WJF extract (50 and 200 mg/kg) was administrated to mice orally. An hour later, olive oil was given orally at 5 ml/kg later. Plasma triglyceride was measured 2, 4, and 6 hours after olive oil loading. A dose-dependent suppression of plasma triglyceride elevation was observed in the test group (Fig. 8). Moreover, plasma triglyceride elevation in 200 mg/kg WJF extract significantly

lowered at timepoints of 2ant 4 hours after olive oil intake. This result suggests a fat-absorption-suppressing effect of WJF extract.



**Fig 8.** WJF extract suppressed plasma triglyceride elevation in olive-oil-loaded mice  $(n = 6, \text{ mean} \pm \text{SD}, \text{ significant difference}^* : p < 0.05).$ 

#### [Method]

Six-week-old male mice were fasted for 18 hours. WJF extract was administrated at 50 or 200 mg/kg orally. Distilled water was given to the control group. An hour later, olive oil was given at 5 mL/kg. Blood was sampled 30 min before WJF extract administration and 2, 4 and 6 hours after oil-loading. Triglyceride was measured in plasma using the E-test.

# 4. Hypolipidemic effect

### 4-1. Lowering plasma triglyceride (in vivo)

Three-week administration of WJF extract in mice resulted in lowered plasma triglyceride, indicating hypolipidemic effect of this extract.

Group	Triglyceride (mg/dL)
Control	$216.0 \pm 12.4$
WJF extract (0.1 %)	$169.8\pm25.4$
WJF extract (0.2 %)	$167.1\pm15.4$
$(=5-6, mean \pm S.E)$	

Table 4. Effect of 3-week administration of WJF extract on plasma triglyceride

[Method]

Five-week-old male ddY mice were divided into 3 groups, with free access to either normal laboratory diet (CRF-1, powder chow), mixed diet containing 0.1% WJF extract and mixed diet containing 0.2% WJF extract, respectively. After 3 weeks, plasma triglyceride was measured.

#### 4-2. Lowering plasma cholesterol (in vivo)

Kiho *et al.* <sup>1)</sup> reported lower plasma cholesterol level in normal mice and in mice model of insulin-dependent diabetes mellitus administrated with 50 mg/kg *Tremella* sp. polysaccharide in comparison with mice in the control group.

Group	Dose	Relative pla	asma cholesterol level (	(mean $\pm$ S.E.)
(n=15)	(mg/kg)	0	3	6 (h)
Normal mice				
Control	0	100	$93.4\pm1.8$	$95.6\pm2.4$
TP	50	100	$74.6\pm4.7^*$	$91.7\pm2.7$
Diabetic mice				
Control	0	100	$94.7\pm2.8$	$93.3\pm4.8$
TP	50	100	$67.9\pm7.9^*$	$78.6\pm2.7*$

Table 5.	Effect of TP on lowering plasma cholesterol
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Each plasma cholesterol level at 0 h normal mice (n = 15) 124.6 ± 3.9 mg/dl, diabetic mice (n = 15) 141.5 ± 8.2 mg/dl] was set to 100.

\* Normal mouse (N) and streptozocin-Induced diabetic mouse (D)

\* Significant difference from the control at p < 0.05, by Student's *t*-test.

### 4-3. Enhancing bile acid secretion (in vivo)

Kiho *et al.*<sup>1)</sup> reported that adding *Tremella* sp. polysaccharide to drink water at 0.5g/l significantly increased amount of bile acid in feces of mice. Cholesterol level was also lowered in feces. These results suggest that the effect of TP in lowing plasma cholesterol is likely via enhancing bile acid secretion.

Table 6.	Effect of TP on bile acid and cholesterol in feces		
Group	Total bile acid	Total cholesterol	
(n=4 - 5)	(mmol/g feces)	(mg/g feces)	
Control	$96 \pm 5.6$	$4.73\pm0.14$	
TP	$135 \pm 4.3*$	$4.11 \pm 0.13^{*}$	

Feces were gathered for one week after 8 weeks of the test. Each value represents the mean  $\pm$  S.E of 4 - 5 mice. Significantly different from the control : \* *p*<0.05.

### 4-4. Hypolipidemic effect (human trial)

The healthy benefits of WJF extract was further investigated in 6 healthy male volunteers (our employees) aged between 27 and 55. Daily 50 mg of WJF extract was intaken for 3 weeks. Blood parameters were examined before and after the test period. As summarized in table 7, decreases were found in LDL-cholesterol, Triglyceride (TG) and free fatty acids. In contrast, HDL-cholesterol was increased, suggesting an improving effect for dyslipidemia. Fig. 9 illustrates high TG values exceeding 150 mg/dL (the key diagnostic criteria for metabolic syndromes) in two test subjects were drastically lowered after 3-week intake of WJF extract. In one of them, a decrease of TG from around 300 mg/dL to 150 mg/dL (near the normal range) was reached. In the other subject, 170 mg/dL TG was lowered to the normal range. In addition, LDL-cholesterol moved toward the median value of 110 mg/dL after intake of WJF extract, and in two test subjects was decreased to the normal range after intake of WJF extract. These results demonstrate that WJF extract lowers plasma TG and LDL-cholesterol especially effectively in individuals with dyslipidemia, and thus is beneficial for metabolic syndromes.

Table 7. Blood parameters before and after WJF extract intake in human

Item	unit	Before intake	After intake
LDL-cholesterol	mg/dL	$131\pm37.8$	$123\pm31.7$
HDL-cholesterol	mg/dL	$61.3\pm26.9$	$66.3\pm28.5$
Total cholesterol	mg/dL	$206\pm36.4$	$204\pm27.9$
Triglyceride (TG)	mg/dL	$121\pm97.8$	$82.7\pm43.3$
Free fatty acids	mEq/L	$0.52\pm0.32$	$0.51\pm0.22$

Mean  $\pm$  S.E of 5 test subjects

* reference values				
Item	unit	Standard value		
LDL-cholesterol	mg/dL	70~139		
HDL-cholesterol	mg/dL	40~77		
Total cholesterol	mg/dL	130~219		
Triglyceride (TG)	mg/dL	30~149		
Free fatty acids	mEq/L	0.13~0.77		



Fig 9. Blood parameters before and after WJF extract intake in human

# 5. Intestinal regulation & flora improvement

### 5-1. Promoting Lactobacillus bulgaricus growth (in vitro)

WJF extract-P or oligofructose was added to *Lactobacillus bulgaricus* cultured in MRS broth at 1.25%. Growth of *Lactobacillus bulgaricus* was promoted in the medium complemented with WJF extract.



Fig 10. WJF extract promoted growth of Lactobacillus bulgaricus

#### 5-2. Promoting Bifidobacterium bifidum growth (in vitro)

WJF extract or oligofructose was added to *Bifidobacterium bifidum* cultured in MRS broth at 1.25%. Growth of *Bifidobacterium bifidum* was promoted in the medium complemented with WJF extract.



Figure 11. WJF extract promoted growth of Bifidobacterium bifidum

### 5-3. Improving intestinal flora (human trial)

Six healthy male were obliged to intake 50 mg WJF extract daily for 3 weeks. Bacteria in feces was measured before and after the intake period at a  $10^2$ -fold dilution. As illustrated in Fig 12, *Clostridium perfringens*, which is the pathogenicity for gas gangrene, hemorrhagic enteritis, enterotoxin-intoxication and food intoxication, was reduced. Reduced counts were also found for 4 other intestinal bacteria. In contrast, beneficial bacteria *Bifidobacterium* spp. and *Lactobacillus* spp. increased. Especially, counts of *Bifidobacterium* bifidum was close to the standard value of  $10^{11}$  CFU/g after the intake. Similarly, *Escherichia coli* count was also improved to near the standard of  $10^{10} \sim 10^{11}$  CFU/g. Increase in lactose-degradating bacteria suggested the prebiotic effect of WJF extract.



Fig 12. Intestinal bacteria before and after 3-week intake of WJF extract

# 6. Constipation relief





WJF extract can hydrate stool and promote healthy bowel movement, leading to constipation relief. Because of it's high molecular weight (>1000kD), extremely high water-retaining capacity, stability in acidic gastric fluid and effect in improving intestinal flora.

### 6-2. Constipation relief (human trial)

Sixteen constipation patients were divided into 3 groups, each given 0.1, 0.2 or 0.3% WJF extract solution (100 mL), respectively, twice a day for 7 days. Constipation relief was reported by 2 out of 5 patients in the 0.1% group, 5 out of 6 in the 0.2% group and by all 5 patients in the 0.3% group.



Based on information provided by the test subject after the 7 days test period.

#### Fig 13. Constipation relief effect of WJF extract

# 7. Enhancing brain function

#### 7-1. Neurite outgrowth of brain neurons (in vitro)

According to Kim *et al*<sup>3</sup>, neurite outgrowth of PC12h cells was promoted by hot-water extract of white jelly fungus (TF).



**Fig 14.** Neurite outgrowth promoted by hot-water extract of *Tremella fuciformis*. The neurite length is expressed as the mean  $\pm$  S.E.M. of the individual cell (n = 150) in each group. Separate measures of one-way ANOVA of the swimming time among the groups were followed by Scheffe test. ## p < 0.01, ### p < 0.001 vs. control (vehicle).

#### [Method]

Hot-water extract of *Tremella fuciformis* was added to DMEM/Hams F (12, 1:1) medium at various ratio for culturing PC12h cells. After 72 hours incubation at 37°C and 5% CO<sub>2</sub>, the cells were fixed with gluthalaldehyde and the neurite outgrowth was measured under microscope.

3) Kim. J.H., Ha.H.C., Lee.M.S., Kang.J.I., Kim.H.S., Lee.S.Y., Pyun.K.H., Shim.I. Effect of *Tremella fuciformis* on the neurite outgrowth of PC12h cells and the improvement of memory in rats. *Biol. Pharm. Bull.*, **30**, 708-714 (2007).

### 7-2. Enhancing memory (in vivo)

According to Kim *et al.*<sup>3)</sup>, oral daily administration of TF for 14 days significantly reversed scopolamine-induced learning and memory deficits in rats. When tested in Morris water maze, the performance of mice treated with scopolamine and administrated with TF was nearly that of rats untreated with scopolamine. This result suggests that TF improves memory and learning ability.



**Fig 15.** TF improved memory of rats treated with scopolamine. Mean values of the four trials per day are shown. Repeated measures of ANOVA for swimming time among the groups were followed by the LSD test. \* p < 0.05, \*\* p < 0.01 vs. sham; # p < 0.05 vs. control.

[Method]

Rats were assigned to 4 groups each with 7-10 rats: the sham group, the scopolamine-treated group with TF-administration at 100 mg/kg for 14 days, and the scopolamine-treated group with TF-administrated at 400mg/kg for 14 days. Learning ability and memory of rats in all the 4 groups were evaluated with the Morris water maze test.

# 8. Suppressing plasma glucose elevation

### 8-1. Suppressing plasma glucose elevation in diabetic mice (in vivo)

Kiho *et al.*,<sup>1,2)</sup> reported that administration of 0.05% TP in drink water for 10 weeks suppressed plasma glucose-increase in diabetic mice by approximately 30%. Similarly, plasma insulin was decreased by approximately 30% in these mice.

Since TP significantly lowered glycogen level in liver of both normal and diabetic mice, the mechanism of the hypoglycemic activity of TP is supported to be not the conversion of glucose into glycogen, but rather the acceleration of glucose metabolism.

### 8-2. Reducing fasting hyperglycemia (human trial)

Six healthy male volunteers aged between 27 and 55 were administrated with 50 mg WJF extract daily for 3 weeks. Decrease of fasting plasma glucose level was observed (Fig. 6). Especially in two subjects with plasma glucose level exceeding the normal range of 60-109 mg/dL, significant improvement was reached after the intake of WJF extract. This result demonstrated that this extract improved hyperglycemia.



Fig 16. WJF extract reduced plasma glucose level in human

# 9. Protecting gastric mucosa

### 9-1. protecting gastric mucosa (in vivo)

Experiment using rats has revealed effect of WJF extract solution in suppressing acute gastric inflammation.

Table 8.	Effect of WJF extract in suppressing acute gastric inflammation ( $N = 10$		
	Group Hyperemic and		
		diffuse hyperemic area (mm <sup>2</sup> )	
	Control	$5.41 \pm 1.53$	
	WJF extract	$3.23 \pm 1.49^{p < 0.05}$	

#### [Method]

Nine-week-old male rats were given 3.3 ml of 0.8 % WJF extract solution orally twice a day for 10 days. After 24-hour fasting, 3.3 ml of 0.8% WJF extract solution was given again, followed by administration of 1 ml absolute ethanol an hour later to induce acute gastric inflammation. After another hour, stomach was sampled and hyperemic and diffuse hyperemic areas were measured.

### 9-1. Improving gastric inflammation (human trial)

Ten patients of mild gastric inflammation were administrated with 200 ml of 1% WJF extract solution twice a day (morning and evening) for 7 days. Improvement of the gastric inflammation was observed.



Day after beging of intake

# 10. Stability of White Jelly Fungus Extract

#### 10-1. Thermostability of solution at various pH

White Jelly Fungus Extract-P was dissolved in water at pH 2, 4, 7, and 9. After heating at 121°C for 1 hour, content of glucuronic acid was determined and, found to be almost constant over the pH range tested.



Thermostability of glucuronic acid at various pH

#### 10-2. Stability of the viscosity of the solution at various pH

Viscosity of 0.5 % White Jelly Fungus Extract-P solution was measured in term of continuous shearing velocity ( $0\sim200 \text{ l/s}$ ) at 25°C using a B-type viscometer, and found to be stable at pH range of 2-12. Thirty-day storage at room temperature did not decrease, viscosity of the solution remained the same. White Jelly Fungus Extract-P solution is thus stable over a broad pH range.



Stability of solution viscosity at various pH

### 10-3. Collagen Co-precipitation with Polyphenols

Mixing of polyphenol-rich solution with collagen solution likely makes precipitation of polyphenols and collagen. This phenomenon has prevented polyphenol-rich materials to be prescribed with collagen in beauty-oriented beverages. To solve the problem, we conducted experiments using water-soluble polyphenol-rich materials selected from our products. It was found WJF Extract-P was effective to prevent collagen precipitation with the materials listed in Table 1, when WJF Extract-P was added to polyphenol solutions prior to mixing with collagen solution.

Addition to that, Kiwi Seed Extract-WSP and Yuzu Seed Extract-WSP solutions were found not to make precipitation or cloudiness when mixed with collagen solution.

No precipitation or cloudiness observed in	Kiwi Seed Extract-WSP
	Yuzu Seed Extract-WSP
WJF Extract-P successfully avoided precipitation in	Resveratrol-WSP0.5
	Purple Rice Extract-P
	Walnut Polyphenol-WSP10
	Evening Primrose Extract-WSPS
WJF Extract-P failed to avoid precipitation in	Litchi Seed Extract-WSP

 Table 9.
 Polyphenol-rich materials examined and status after mixing with collagen solution

#### 10-4. Stability during long-term storage

White Jelly Fungus Extract-P, -PC was stored at 25°C for 2 years. Content of polysaccharide and glucuronic acid was slightly decreased, which however was still upper the specification. White Jelly Fungus Extract-P, -PC is thus stable during long-term storage.



Contents at the production and after storage for 2 years

## 11. Safety profile of White Jelly Fungus Extract

### 11-1. Acute toxicity text (LD<sub>50</sub>)

White Jelly Fungus Extract-P was administrated orally to 10 male and female mice at 1000 mg/kg for 14 days. Neither death nor toxic symptoms were observed. Body weight was decreased in male mice. However, no abnormality was found in blood parameters. The oral administration LD<sub>50</sub> value for White Jelly Fungus Extract-P was thus above 1000 mg/kg for both male and female mice.

#### 11-2. Subacute toxicity test

White Jelly Fungus Extract-P (227 mg/kg) was administrated to 10 male and female mice orally for 30 days. Neither death nor toxic symptoms were observed.

#### 11-3. Genotoxicity Test

1) Mutagenicity Test (Ames test)

Ames test was conducted for White Jelly Fungus Extract-P using *S. typhimurium* TA98 and TA100. At concentration of  $19.5 \sim 5000 \,\mu\text{g/plate}$ , no mutagenicity was found.

2) Sperm Teratogenicity Test

White Jelly Fungus Extract-P (1.25 g/kg) was administrated to 15 male mice for 5 days. Thirty days late sperms were examined under microscope. No teratogenicity was found.

#### 11-4. Human Intake test

Fifteen mg White Jelly Fungus Extract-P (no diluent) was administrated to 6 male volunteers daily for 3 weeks. No abnormality was observed in any of the following blood parameters: total bilirubin, total protein, albumin, AST, ALT,  $\gamma$ -GTP, LDL-cholesterol, total cholesterol, triglyceride, phospholipid, free fatty acid, HDL-cholesterol, sodium, calcium, serum iron, TIBC, UIBC, urea nitrogen, urea acid and glycemia.

#### 11-5. Residual Agricultural Chemicals

White Jelly Fungus Extract-P was examined for 447 agricultural chemical residues, according to the food hygiene regulation and pesticide legislation. All items were below the detection limits.

Test trustee: Masis Co., LTD Date of issue of the report: April 19, 2007 Contract No. : 11280

# 12. Nutrition facts of White Jelly Fungus Extract-P

Component	Result	Note	Method
Protein	1.0 g/100 g	1	Combustion
Lipid	0.1 g/100 g		Acid fat dissolution
Ash	5.6 g/100 g		Direct ashing
Carbohydrate	0.0 g/100 g	2	
Energy	175 kcal/100 g	3	Modified out-watering
Dietary fiber	84.9 g/100 g		Enzymatic-weight method
Sodium	2300 mg/100 g		Atomic absorption spectrometry
Sodium chloride equivalent	5.8 g/100 g		Sodium equivalent value

Note 1) Nitrogen, protein conversion factor : 6.25

Note 2) 100 - (moisture + protein + fat + ash + dietary fiber)

Note 3) Factors for calculating the energy value : protein 4; lipid 9; sugar 4; dietary fiber 2

Test trustee: SRL, Co., LTD

Date of issue of report: October 15, 2007

Trustee No.: 133653

# 13. Recommended Dosage

Based on various experiments and safety data, we recommend  $50 \sim 200 \text{ mg}$  as the daily dose of White Jelly Fungus Extract-P.

# 14. Application of White Jelly Fungus Extract

	Functionality	Dosage form
Food	sliming, suppressing hyperlipidemia, improving intestinal flora, enhancing brain function, suppressing plasma glucose elevation, protecting gastric mucosa	Beverage (refreshing drinks, lactobacillus beverage, etc.), hard and soft capsules, tablets, candies, hewing gum, gum, cookies, wafer, jelly, <i>etc</i> .
Cosmetic	moisturizing	Lotion, pack, bodygel

Solubility for various solvents at room temperature

Product	Water	Hydrous ethanol	Glycerin	Dietary oil
White Jelly Fungus	Soluble	Incoluble	Incolubio	Incolubio
Extract-P, -PC	Soluble	Insoluble	Insoluble	Insoluble

\* Viscosity of White Jelly Fungus Extract-P, -PC solution increases in volume and viscosity as increasing concentration. For preparation of solution, stir the powder quickly into water and stir until there is no lumps. Due to this feature, preparation of solution containing more than 1% White Jelly Fungus Extract-P, -PC is technically difficult.

# 15. Packaging

White Jelly Fungus Extract-P (water-soluble powder, food grade)

5 kg Interior packaging: aluminum-coated plastic bag Exterior packaging: cardboard box

White Jelly Fungus Extract-PC (water-soluble powder, cosmetic grade)

5 kg Interior packaging: aluminum-coated plastic bag Exterior packaging: cardboard box

# 16. Storage

Avoid high temperature and humidity. Keep tightly sealed. Store in cool, dry and dark place.

# 17. Expression

#### <food>

White Jelly Fungus Extract-P

Example : White Jelly Fungus Extract, White Jelly Fungus Polysaccharide

#### <cosmetic>

White Jelly Fungus Extract-PC

INCI name: Tremella Fuciformis Polysaccharide

\* Please refer to your local standard and regulations.

# **PRODUCT STANDARD**

# PRODUCT NAME : WHITE JELLY FUNGUS EXTRACT-P (FOOD)

This product is powder extracted from *Tremella fuciformis* Berk. It guarantees a minimum of 80.0 % total saccharides and 10.0 - 30.0 % glucuronic acid.

<u>Appearance</u>	White or pale yellow powder with slight unique aroma.	
Total Saccharides	Min. 80.0 %	(Sodium Hyaluronate (2) Method 2.
		The Japanese Standards of Quasi-Drug
		Ingredients.)
Glucuronic Acid	10.0 - 30.0 %	(Orcinol - Sulfuric Acid Saccharides Assay)
Loss on Drying	Max. 10.0 %	(Analysis for HygienicChemists,
		1g, 105 °C, 2 hr)
Purity Test		
(1)Heavy Metals (as Pb)	Max. 20 ppm	(Sodium Sulfide Colorimetric Method)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 2 ppm	(Standard Methods of Analysis in Food
		Safety Regulation, The Third Method,
		Apparatus B)
<b>Standard Plate Counts</b>	Max. $1 \times 10^3$ cfu/g	(Analysis for Hygienic Chemists)
Moulds and Yeasts	Max. $1 \times 10^2$ cfu/g	(Analysis for Hygienic Chemists)
<u>Coliforms</u>	Negative	(Analysis for Hygienic Chemists)
<b>Composition</b>	Ingredient	Content
	White Jelly Fungus Extract 100 %	

# **PRODUCT STANDARD**

# PRODUCT NAME : WHITE JELLY FUNGUS EXTRACT-P (COSMETIC)

This product is powder extracted from *Tremella fuciformis* Berk. It guarantees a minimum of 80.0 % total saccharides and 10.0 - 30.0 % glucuronic acid.

<u>Appearance</u>	White or pale yellow powder with slight unique aroma.	
<b>Total Saccharides</b>	Min. 80.0 %	(Sodium Hyaluronate (2) Method 2.
		The Japanese Standards of Quasi-Drug
		Ingredients.)
Glucuronic Acid	10.0 - 30.0 %	(Orcinol - Sulfuric Acid Saccharides Assay)
Loss on Drying	Max. 10.0 %	(Analysis for HygienicChemists,
		1g, 105 °C, 2 hr)
Purity Test		
(1)Heavy Metals (as Pb)	Max. 20 ppm	(Sodium Sulfide Colorimetric Method)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 2 ppm	(Standard Methods of Analysis in Food
		Safety Regulation, The Third Method,
		Apparatus B)
Standard Plate Counts	Max. $1 \times 10^2$ cfu/g	(Analysis for Hygienic Chemists)
Moulds and Yeasts	Max. $1 \times 10^2$ cfu/g	(Analysis for Hygienic Chemists)
<u>Coliforms</u>	Negative	(Analysis for Hygienic Chemists)
<b>Composition</b>	Ingredient	Content
	Tremella Fuciformi	s Polysaccharide 100 %

**ORYZA OIL & FAT CHEMICAL CO., LTD.,** striving for development of new functional additives and material for your health and beauty.

From product planning to OEM – For any additional information and assistance, please contact:

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\* This cataloque was created based on research findings. For expression of the products for consumers, please refer to health-promoting regulation and drug legislation

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\* The contents of this catalogues may be changed without prior notice.

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