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Functional Properties of Mushroom Chitosan Containing β-Glucan: Prevention of Fat Absorption

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INTRODUCTION

With the Westernization of eating habits, the number of individuals consuming excess fat is on the increase, and cholesterol and neutral lipids are at an all-time high. Given this situation, it is critical to reduce fat intake through a properly balanced diet and to exercise regularly. Doing so, however, requires discipline. Persons who are unable to control their diet and obtain sufficient exercise are susceptible to hyperlipemia, whereby fat accumulates in the body, leading to obesity. Furthermore, an overweight condition is said to lead to so-called lifestyle-related diseases, such as arteriosclerosis, hypertension and diabetes.

In order to prevent overweight and obese, peoples try many kind of treatment as the severe meal limitation. However, this treatment is very difficult to continue unless people having strong willing.

This paper analyzes experiment results, and provides an outline of the properties of mushroom chitosan containing β-glucan (chitoglucan), a new product successfully developed by our company, Ricom. It also contains a discussion of chitoglucan's functional properties and its use in a new dietary regimen.

Studies on Mushroom Chitosan

In 1811, the French botanist Braconnot first isolated the compound that would come to be known as chitin. Some 22 years later, in 1823, another French scientist, Odier, noted that chitin could also be found in the structure of insects and certain plants. More recently, chitin has been refined from crab shells and attracted extensive attention.

Edible mushroom market in the world is grown more than 5 million tons per year. Various functional properties were found in the mushroom. But mushroom chitosan remained not to research. The efficient industrial manufacture may be very difficult on the mushroom chitosan.

In 1989, we isolated a natural mushroom extract known as Champignon extract, and began supplying this valuable new raw material for use in health foods, drinks, hospital diets, sweets and pet food.

Following the discovery that Champignon extract played a role in protecting against kidney failure, it was hypothesized that the chitin/chitosan in mushrooms could be one of the beneficial elements, and research was undertaken into the extraction and refinement of mushroom chitosan in order to further exploit the full potential of Champignon extract.

Since 1998, the production and commercialization of mushroom chitosan has been explored using various methods and raw materials, including *shiitake*, *enokitake*, *maitake* and *Agaricus bisporus*. In 2003, we submitted an international patent application.

What is Chitoglucan

Chitoglucan is a new functional dietary fiber that is being used as a raw material.

Mushrooms contain chitin, β-glucan and various other fibrous polysaccharides. They play an important function,

although their roles are often mutuality intertwined within the mushroom structure. Using a special alkaline treatment, chitoglucan can be obtained from mushroom chitin without breaking this structure.

Chitosan obtained from mushrooms offers a major advantage over chitosan obtained from crab and lobster shells in that the β-glucan molecule of mushroom origin can be readily used as a functional dietary fiber.

Properties of Chitoglucan

Chitoglucan is a compound dietary fiber consisting of β-glucan and chitosan.

It is difficult to dissolve in both water and acid, and just can be swelling. These properties of acid-insolubility and swelling in water offer a major difference with chitosan derived from crab sources. It appears that this difference – the presence of β-glucan within the molecule – enables it to slowly combine with large quantities of fat.

Chitoglucan has a kind of properties as surface activation. This kind of property have never observed in the other kind of chitosan or dietary fibers.

Moreover, in vitro experiments have shown that dilute solutions of chitoglucan have a powerful action in wrapping and excrete directly the fat moiety. Chitoglucan taken before a meal, thinly coating the mucous cell membranes of the intestines, has demonstrated an in vivo effect that controls the absorption of fat.

Characteristics of Chitoglucan

Chitoglucan can be used in a wide array of foods since it contains none of the rough taste of crab chitosan. On the contrary, standard quality light yellowish brown chitoglucan features a pleasant natural scent slightly reminiscent of mushrooms. It is available as finely size particle, of which more than 95% passes through 30 mesh, enabling easy mixture with other powders. Humidity is not a factor of significant concern so long as it is handled in an environment of less than 10% humidity. Chitoglucan is a high food fiber material, comprised of 60~70% dietary fiber. Alleviating cause for concern regarding impurities and heavy metals, chitoglucan contains less than 10 ppm Pb, less than 1ppm arsenic in the form of As_2O_3, and less than 10 ppm chromium. In the inspection of microorganisms, fewer than 1,000/g of general bacteria were found, less than 300/g of molds, and the presence of coliform bacilli groups was negative. These data indicate that no specific problem exists. There are no contaminations in the product by examination of agricultural chemicals.

Functional of Chitoglucan

Chitoglucan boasts various beneficial functional properties, including a cholesterol repressive action and the blocking of fat absorption. This phenomenon has a powerful dietary effect, aiding in the prevention of hyperlipemia. A compound food fiber of low molecular weight can be expected to promote bifidus bacterium growth activity in the intestinal environment. High molecular weight moiety of chitoglucan has swelling activity as dietary fiber like properties.

Chitoglucan is expected to provide substantial immunity boosting properties beyond the capabilities of conventional crab chitosan, given that it contains polysaccharides which have anti-cancer properties found in many mushrooms, and due to the presence of β-glucan found in the fiber element.

The Effect of Chitoglucan on Cholesterol (Animal Study)

Fat (cholesterol) intake was investigated in Wistar male rats of five weeks of age. Two groups of rats were freely given high cholesterol feed for a period of 3 weeks. One group consist 8 rats. This group was given "high

cholesterol feed" (control group), while the other was given "high cholesterol feed added with 5% chitoglucan."

Blood sampling was performed once a week during the period of free intake, and the total serum cholesterol concentration was measured. The results are shown in Fig. 1.

As for the control group that took only high cholesterol feed, after 1 week, the total serum cholesterol concentration increased to 243±101 mg/dL; and after 2weeks, to 304±125 mg/dL−about 5 times the initial value (68±14 mg/dL). On the other hand, in comparison with the control group, the group receiving supplementary chitoglucan showed low numerical values. After 1, 2 weeks, the cholesterol increased to only 83±15 mg/dL and 118±51 mg/dL, respectively, compared with an initial value of 67±12 mg/dL.

Therefore, in high cholesterol feed for rats, the absorption of fat containing cholesterol was controlled by the supplementation of chitoglucan, and it appears that a rise in total serum cholesterol concentration was thereby controlled.

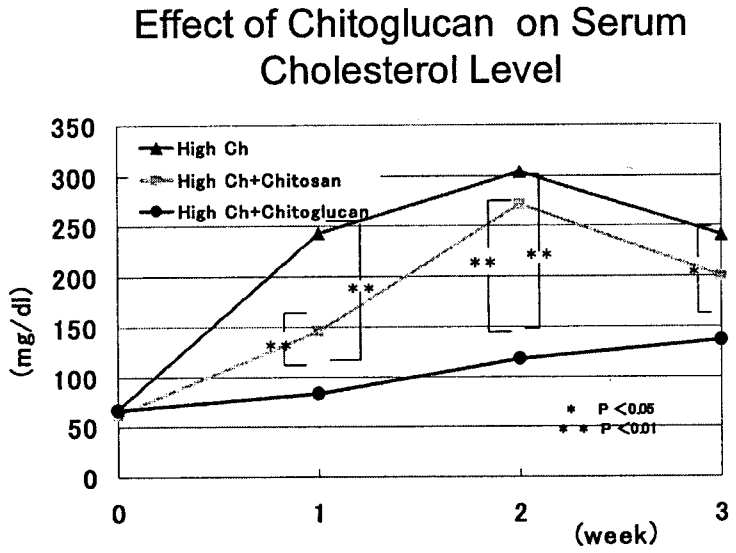

Fig. 1. Effect of chitoglucan on serum cholesterol level.

Clinical Trial on the Effect of Chitoglucan on Fat Absorption

A clinical trial on fat intake was conducted on 10 subjects, who were divided into a control group (4 people), an after-meal medication group (3 people), and a before-meal medication group (3 people). The subjects were required to fast from 21 o'clock on the previous day, and serum samples were collected before the subjects broke their fast on the test day. High-fat food (cream corn potage with added butter and lard to fat content level of 40 g, an energy quantity of 437 kcal) was administered at 9 o'clock, and serum samples were collected each hour following, measuring for serum neutral lipid concentration.

In the group receiving after-meal medication, one gram of chitoglucan was administrated with water promptly after the high-fat food intake; and for the group receiving before-meal medication, 0.3 g of chitoglucan was administrated with water 20 minutes prior to the intake of high-fat food.

The results are shown in Fig. 2.

The lines in the figure show the rise in concentration from the initial value (prior to eating) of the serum neutral lipid concentration.

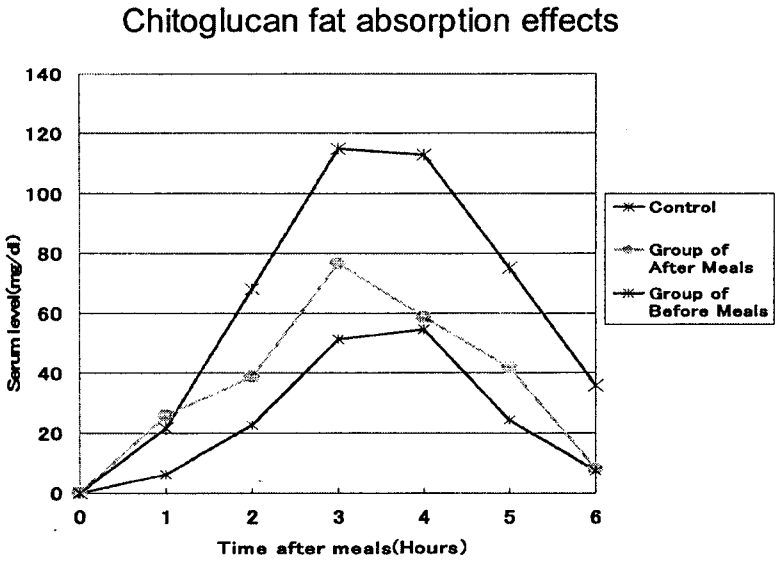

Fig. 2. Effect of chitoglucan on fat absorption.

The peak of the rise in concentration was about 3～4 hours after the meal for all groups.

Comparing the rise in concentration of the control group with the group receiving after-meal medication, the group receiving after-meal medication showed lower values for all but the first hour－a situation that continued throughout the 2～6 hour period following consumption.

Comparing the group receiving before-meal medication with the control group and the group receiving after-meal medication, the group receiving before-meal medication showed the lowest value throughout the 1～6 hour period.

These results show that when chitoglucan is given after a meal, it appears the fat binding activity of chitoglucan (the repression of absorption of the fat) appears two hours after the meal. When chitoglucan is given before a meal, it coats the intestinal mucous membrane by passing through rapidly from stomach being swelling. For consequence in spite of the lower dose of chitoglucan before meals than after meals, the better effect was obtained in the case of administration of chitoglucan before meals.

On the other hand, when the area under the line showing concentration of blood neutral lipid (AUC) is 100 for control group not to administrate chitoglucan, AUC value of the group receiving before-meal and after-meal are 60 and 40, respectively. This means that the rate of prevention of fat absorption in the case of 1 g administration after-meals is 40% and 60% in the case of 0.3 g administration before-meal, respectively.

Chitoglucan is utilized by intestinal bacteria, such as bifidus bacterium, but for the most part is excreted as undigested food fiber.

Coating Diet Method

The coating diet is an entirely new method that takes places in the intestines. With the coating diet method, chitoglucan is consumed with a favorite food 30 minutes prior to mealtime, dramatically repressing the absorption of fat contained in the food consumed at mealtime, because the chitoglucan has coated the insides of the intestines. This method controls the rise of neutral lipids and cholesterol in the blood, thereby preventing the accumulation of body fat. Accordingly peoples of overweight and obese can easily make diet by the activation of fat utilization and by the decrease of fat absorption.

When the anxiety that accompanies the need for unbearable meal limitations is eliminated, anyone can attain healthy diet goals without reserve and without having to fight their appetite. Indeed, the coating diet method has no rebound action by the limitation of diet indicating that peoples can perform readily the final object to stop overweight and obese.

Furthermore, when chitoglucan is taken during and after meals, fat is caught as small lumps, preventing the excessive accumulation of body fat by controlling diffusion and absorption.

CONCLUSION

Needless to say, lipids are an important source of physical energy. When the intake balance gets out of control, however, excessive consumption leads to overproduction of fat and various lifestyle-related diseases. The fat absorption blocking effect of chitoglucan mentioned in this paper is considered to be useful enough to promote immediate long-term benefits, enabling persons who are anxious about excessive fat intake to happily enjoy their meals once again.

Although our chitoglucan research remains insufficient, it reveals various beneficial properties. In forthcoming research, we hope to study properties such as the activity of body fat combustion, the growth of bifidus bacterium, declines in blood pressure and the activation of immunity.

REFERENCES

1. Gamayurova, et al. 1999. Synthesis of soluble derivatives of chitin-glucan complex. Chemistry and Computational Simulation. *Butlerov Communicatios* No.1.
2. Shabrukova, et al. 2001. Study of the nature of chitin-glucan complex. Chemistry and Computational Simulation. *Butlerov Communicatios* No.4.
3. Shida, et al. 1981. Structure of the alkali-insoluble skeltal glucan of *Lentinus edodes*. *J-Biochem-Tokyo* 90: 1093-1100.
4. Hartland, et al. 1994. The linkage of (1-3)-beta-glucan to chitin during cell wall assembly in *Saccharomyces cerevisiae*. *Yeast* 10: 1591-1599.