

Preparation of Insoluble Dietary Fiber from Forest Waste and Its Physiological Function in Rat Fed High Cholesterol Diets

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Abstract

This study investigated the production of insoluble dietary fiber using forest waste and the dietary effect of manufactured insoluble fiber on physiological function in rat fed high cholesterol diets. Insoluble dietary fiber was prepared from the wood chips of oak (*Quercus mongolica*). The best condition for steam-explosion treatment for the preparation of insoluble dietary fiber was 25 kg/cm³ pressure for 6 minutes. In the chemical analysis of insoluble dietary fiber pretreated by 1% sodium hydroxide solution with steam-exploded wood, α -cellulose content was 61.7% in the insoluble dietary fiber, which contained 7.6% residual lignin. In order to compare insoluble dietary fiber with commercial α -cellulose of physiological function, Sprague-Dawley male rats weighing 100 \pm 10 g were randomly assigned to one normal diet and five high cholesterol diet containing 1% cholesterol. The high cholesterol diet groups were classified as fiber free diet (FF group), 5% commercial α -cellulose diet (5C group), 10% commercial α -cellulose (10C group), 5% insoluble dietary fiber diet (5M group), and 10% insoluble dietary fiber (10M group). The rats were fed ad libitum for 4 weeks. Food intake, weights gain, and food efficiency ratio in high cholesterol groups were higher than those of normal group, but there were no significant differences between the experimental groups. There were not any significant differences in the weights of liver, kidney, and small intestine of insoluble dietary fiber supplemented groups, but weight of cecum in all insoluble dietary fiber group were significantly higher than those of FF group. A gastrointestinal transit time was decreased by supplementation of insoluble dietary fiber. Weight and water contents of feces in the insoluble dietary fiber supplemented groups were significantly higher than those of the FF group. There were not any significant differences in the activities of the glutamic pyruvic transaminase (GPT) and glutamic oxaloacetic transaminase (GOT) between the experimental groups. In conclusion, the manufactured insoluble dietary fiber and commercial insoluble fiber have the same physiological effects. The preparation method of the insoluble dietary fiber from the oak chips suited its purpose.

Key words: insoluble dietary fiber, forest wastes, steam explosion, delignification, gastrointestinal transit time

INTRODUCTION

Dietary fiber is largely classified as soluble and insoluble, of which the soluble dietary fiber includes pectin, gums and soluble hemicellulose while the insoluble fiber is cellulose, lignin and insoluble hemicellulose (1). Compared to the insoluble dietary fiber, the soluble fiber features higher solution-retaining rate which makes it possible to form gel and raise viscosity. For this reason, it has been reported that food staying within the stomach for the longer period of time leads to satiety and delay of nutrient digestion and absorption, therefore, improves anti-diabetic function (2). It has been also reported that when cholesterol and bile acid are absorbed in the intestines, there is a change in both excrement and lipoprotein metabolism which decreases the level of serum

cholesterol and reduces the invasion of coronary cardiovascular diseases and colic cancer (3).

The insoluble dietary fiber is much less influenced by the action of microorganisms in the large intestine and its unfermented residue remains within the intestine. On the other hand, the increase of excrement results in the change in the role of epithelial cells (4,5), and the production of simple fatty acid reduces the level of pH in the large intestine (6) and may have an effect on the prevention of colic cancer (7,8) and the balance of microorganisms in the large intestine (4-9). In addition, it has been reported that pectin, wheat bean and cellulose increase the quantity of fat and protein contained in excrement (10) and have an impact on the morphological change in the intestine's surface or the production of goblet cells which eventually cause the change of glyco-

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lipid metabolism (11).

As stated above, it has been recently shown that dietary fiber is effective in preventing and treating chronic diseases, such as diabetes, cardioangiological disease, colic cancer, fatness and intestinum diverticular disease, and that there has been more increase in its demands so far. Although it is very good for modern people to take dietary fiber, as natural food, with an aspect of physiological activation, they are too busy to take dietary fiber from natural food in society aiming toward more convenience. Accordingly, a variety of products have been developed and used, such as dietary drinks, cereals and low-calorie food, containing a lot of dietary fiber. The common material mainly used to produce these dietary fiber products contains such synthetic dietary fiber as indigestible dextrin and polydextrose, as well as partially hydrolyzed guar gum, carageenan and alginic acid (12). Since it was proven that dietary fiber is quite effective for the prevention of adult diseases, its demand has continued to increase and, therefore, we urgently need to find methods for getting dietary fiber from natural food.

With both an increasing worldwide interest in the environment and a shortage of oil resources, a study has been actively carried out on the chemical use of woody biomass as an energy resource (13,14). The biomass resource features its availability for reproduction and highest set efficiency of solar energy. This woody biomass is a group of cell walls consisting of cellulose and hemicellulose as carbohydrate, and lignin as a phenolic compound, which seem to be usable as food, feed, liquid fuel and raw material for the chemical industry.

Cellulose accounting for the most part of woody biomass is the main ingredient of high plant's cell walls, including cotton, flax and lumber. A lot of cellulose amounting to several billion tons are produced by photosynthesis every year. Recently, the lumber resources have been used as an important raw material in such various industries as paper manufacturing, textile, chemical engineering and food, while much research also has been intensively done on cellulose all over the world (15,16). The cellulose or hemicellulose, a major ingredient, accounts for about 75% of lumber. If it is possible to optionally separate a carbohydrate from lumber, therefore, a big quantity of natural dietary fiber can be produced and used as a major resource for the dietary fiber.

With a lack of lumber resources, this country is required to carry out a variety of studies on recycling lumber waste. The accumulated total quantity of Korea's domestic lumber resource is 1.4×10^8 ton and the lumber waste available for recycling amounts to 1.96×10^6 ton and thinning lumber to 8×10^4 ton each year. Nevertheless, most of these lumber waste resources are not recy-

clad. The current lumber waste resource is mainly from felling or lumbering fields and they are disposed by burning out. In particular, lumber waste is equivalent in terms of quality to lumber and it can be a useful material if its gathering and controlling are done in proper conditions. In so doing, economic benefits also can be achieved, in addition to the proper disposal of lumber waste.

This study was conducted to find out the methods for effectively separating cellulose from lumber, by pre-treating *Quercus mongolica* and its lumber waste among oak trees using the steam explosion method and delignification treatment which have been well-known as an effective method for the pre-treatment.

While a variety of processed food has been recently used as functional food after the addition of dietary fiber, all the domestic demand for dietary fiber entirely depends on importation and the production of dietary cellulose from natural materials is insufficient because most of this fiber are synthetic dietary fiber. It seems, therefore, that the production of cellulose from forest waste is able to greatly contribute to both economic benefits from import-replacement and recycling of waste materials with respect to the environment.

In this study, accordingly, the processes were established to produce insoluble dietary fiber from forest waste, and its nontoxic and physiological effects were tested to examine the possibility to prepare the insoluble dietary fiber, using forest waste.

MATERIALS AND METHODS

Manufacturing of insoluble dietary fiber materials from forest waste

Analysis of materials and constitutive components: Material used for this study is *Quercus mongolica* from Cheongsong-gun, Gyeongbuk, Korea. Its lumbered wood was chipped by a chipper to the size of $2 \times 2 \times 0.2$ cm and then was pre-treated by a steam explosion equipment unit in this lab (17). The steam explosion treatment has its origin in Masonite processing developed to prepare particle boards, which is designed to expose the materials to $200 \sim 250^\circ\text{C}$ saturated steam for several minutes and then leave them in an outdoor place. This method is accepted as a popular technique, featuring the high efficiency of treatment, the easy follow-up treatment and separation of major ingredients from lumber, the short treating period of time available for consequent mass-treatment, and no environmental pollution.

The steam explosion equipment unit, made of stainless steel SUS-304 & 316 with resistance against high temperature, high pressure, acid and alkali, was designed and prepared by this lab and Korea Research Institute

of Chemical Technology, which consists of a reactor (3 L), a steam generator (56 L), and a reaction-collecting tank (50 L). The conditions for steam explosion treatment are shown in Table 1.

The wood components were analysed and measured, using cold & hot water, solvent, alkali extract, ash content and Klason lignin content, by the general lumber-analysing technique (18) based on the JIS and TAPPI techniques. In addition, each component of steam exploded wood was analysed by the alditol-acetate technique (19).

Manufacturing of insoluble dietary fiber from steam-explosion pretreatment material

Manufacturing of insoluble dietary by sodium hydroxide treatment: 250 mL of 1% sodium hydroxide solution was added to 5 g steam exploded wood within a 500 mL triangular flask, agitated at the indoor temperature for 2 hours, and filtered with a glass filter (1G3). Its residue was washed out with distilled water and 10% acetic acid solution, and then the insoluble dietary fiber was prepared (Fig. 1).

Manufacturing of insoluble dietary by sodium chloride treatment: 250 mL of 1% sodium chloride was added to 5 g steam-exploded wood within a 500 mL triangular flask, agitated at the indoor temperature for 2 hours, and filtered with a glass filter (1G3). Its residue was washed out in such order as distilled water, methanol and acetone, and then insoluble dietary fiber was produced.

Analysis on physical properties of the prepared insoluble dietary fiber: The prepared insoluble dietary fiber was entirely dried by a dryer at the temperature of 105°C. And then, its weight measured after completely drying

and its water content measured by means of the specimen's weight before drying.

$$\text{Water content} = B/A \times 100$$

where, A is the specimen's weight before drying, and B is its weight after drying.

Its physical properties were analysed by the alditol-acetate technique, and its lignin content was measured by the Klason lignin technique. To analyse its contained inorganic substance, on the other hand, the specimen was analysed by the X-ray fluorescence spectrometer (Philips Co., PW2400) (15), and the measurement of α -cellulose content (16) and the analysis on the degree of polymerization according to the CED viscosity method (17) were performed.

Physiological function of preparation of insoluble dietary fiber of rat fed high cholesterol diets

Experimental animals and diet: Sprague-Dawley male rats weighing 100 ± 10 g were purchased from KRITC (Taejon, Korea). Rats were individually housed in stainless steel cage in a room with controlled temperature (20~23°C) and lighting (alternating 12 h periods of light and dark). They were randomly assigned to groups of one normal diet and five high cholesterol diet containing 1% cholesterol. The high cholesterol diet groups were classified to fiber free diet (FF group), 5% commercial α -cellulose diet (5C group), 10% commercial α -cellulose (10C group), 5% insoluble dietary fiber diet (5M group), and 10% insoluble dietary fiber (10M group). The rats were fed ad libitum for 4 weeks. This experimental design was approved by the committee of Catholic university of Daegu for care and use of laboratory animals (Table 2).

Measurements of body weight, and the food efficiency ratio: The body weight was measured regularly at the same time every other day throughout the experimental period. The efficiency ratio was calculated by dividing the body weight by the dietary intake during the experimental period.

Collecting samples and sample preparation: The livers were excised, washed in 0.9% of NaCl, frozen rapidly in liquid nitrogen, stored at -80°C, then prepared for the experiment.

Measurement of gastrointestinal transit time: The 0.5% carmine red (Sigma Chem. Co., C1022) solution, used as a marker, was added to individual experimental diets and fed in the fourth week of the experiment, and then the excrement of the marker was checked. The period of time from the beginning of experimental diets' feeding to the marker's first appearance in the excrements was regarded as the gastrointestinal transit time.

Measurement of activity of GOT, GPT: The activity

Table 1. Steam explosion conditions of wood chips

Species	Material No.	Steam explosion condition	
		Pressure (kgf/cm ²)	Time (min)
<i>Quercus mongolica</i>	EQ 25-3	25	3
	EQ 25-6	25	6
	EQ 25-9	25	9

EQ: Exploded oak wood *Quercus mongolica* fischer.

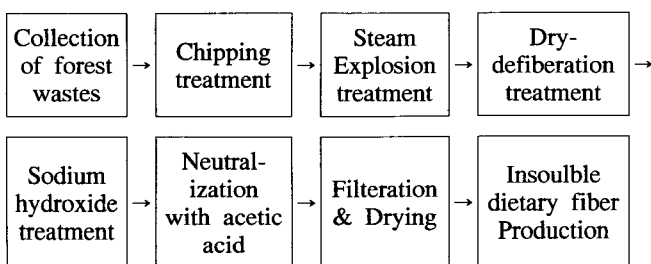


Fig. 1. Production scheme of insoluble dietary fiber prepared from forest wastes.

Table 2. Classification of experimental groups according to different insoluble dietary fiber and levels of rats fed high cholesterol (%)

Ingredients	Groups ¹⁾	Normal	High cholesterol diet				
			FF	5C	10C	5M	10M
Starch ²⁾		65	68.8	63.8	58.8	63.8	58.8
Casein ³⁾		15	15	15	15	15	15
Salt mixture ⁴⁾		4	4	4	4	4	4
Vitamin mixture ⁵⁾		1	1	1	1	1	1
Corn oil ⁶⁾		5	5	5	5	5	5
Sucrose ⁷⁾		5	5	5	5	5	5
Sodium cholate ⁸⁾		-	0.25	0.25	0.25	0.25	0.25
Cholesterol ⁹⁾		-	1	1	1	1	1
Commercial insoluble fiber ¹⁰⁾		-	-	5	10	-	-
Manufactured insoluble fiber ¹¹⁾		-	-	-	-	5	10
Total (%)		100	100	100	100	100	100

¹⁾Normal: basal diet.

FF: basal diet + 1% cholesterol + fiber free.

5C: basal diet + cholesterol + 5% commercial insoluble fiber.

10C: basal diet + 1% cholesterol + 10% commercial insoluble fiber.

5M: basal diet + cholesterol + manufactured insoluble fiber.

10M: basal diet + 1% cholesterol + 10% manufactured insoluble fiber.

²⁾Pung jin Chem. Co., Seoul, Korea.

³⁾Lactic casein, 30 mesh, New Zealand Dairy Board, Wellington, N.Z.

⁴⁾AIN-76 likeness (g/kg mixture): Calcium phosphate, dibasic (CaHPO₄ · 2H₂O) 500, Sodium chloride (NaCl) 74, Potassium citrateminohydrate (K₃C₆H₅O₇ · H₂O) 220, Potassium sulfate (K₂SO₄) 52, Magnesium oxide (MgO) 24, Manganous carbonate (45~48% Mn) 3.5, Ferric citrate (16~17% Fe) 6, Zinc carbonate (70% ZnO) 1.6, Cupric carbonate (53~55% Cu) 0.3, Potassium iodate (KIO₃) 0.01, Sodium selenite (Na₂SeO₃ · 5H₂O) 0.01, Chromium potassium sulfate [CrK(SO₄)₂ · 12H₂O] 0.55, filled up to 1,000 with sucrose.

⁵⁾ALN-76 likeness (mg/kg mixture): Thimin-HCl 600, Riboflavin 600, Pyrdoxine · HCl 700, Nicotinic acid (nicotinamide in equivalent) 3,000, D-calcium pantothenate 1,600, Folic acid 200, D-biotin 20, Cyanocobalamin (vitamin B₁₂) 1, Retinyl palmitate or acetate (vitamin A) as stabilized powder to provide 400,000 IU vitamin A activity or 120,000 retinol equivalent, DL- α -tocopheryl acetate 5,000 IU, Cholecalciferol (100,000 IU, may be in powder form) 2.5, Menaquinone (vitamin K, menadione) 5, filled up to 1,000 with sucrose.

⁶⁾Dong Bang Oil Co., Seoul, Korea. ⁷⁾Sam Yang Co., Seoul, Korea.

⁸⁾Sigma Chem. Co., St. Louis, Missouri, U.S.A. ⁹⁾Sigma Chem. Co., St. Louis, Missouri, U.S.A.

¹⁰⁾Sigma Chem. Co. CMC (Sodium carboxyl methyl cellulose, non-nutritive fiber), St. Louis, Missouri, U.S.A.

¹¹⁾Manufactured α -cellulose from the forest wastes.

of GOT and GPT in the serum was measured by the method of Reitman & Frankel (20).

Statistical analysis: The data were analyzed by ANOVA, and when a significance was identified, the differences between the groups were evaluated using Tukey's HSD test (21).

RESULTS AND DISCUSSION

Manufacturing of insoluble dietary fiber from forest waste

Constitutive components of materials: Table 3 and 4 show the result analysed on the chemical constituents and sugar composition of *Quercus mongolica*. It was shown that the result of a general analysis on materials is similar to that of an analysis on components. An analysis on sugar composition showed that hemicellulose seems to mainly consist of the residue of xylose. Because the residue of arabinose and galactose comes from ara-

Table 3. Chemical compositions of wood

Characters	<i>Quercus mongolica</i> (%)
Cold water extractives	2.0
Hot water extractives	4.8
Ethanol-benzene extractives	2.6
Alkali (1% NaOH) extractives	24.2
Klason lignin	20.8
Ash	0.6

Table 4. Sugar composition of wood measured by alditol-acetate method

Species	Sugar composition (%)					
	Ara. ¹⁾	Xyl. ²⁾	Man. ³⁾	Gal. ⁴⁾	Glu. ⁵⁾	Rham. ⁶⁾
<i>Quercus mongolica</i>	2.7	29.8	1.6	1.2	64.6	T ⁷⁾

¹⁾Ara.: Arabinose ²⁾Xyl.: Xylose ³⁾Man.: Mannose ⁴⁾Gal.: Galactose ⁵⁾Glu.: Glucose ⁶⁾Rham.: Rhamnose ⁷⁾T: Trace, below 0.1%.

binogalactan and no galactoglucomannan exists in the hemicellulose of broad-leaved lumber, on the other hand,

it seems that the residue of mannose comes from glucomannan (22).

Constitutive components of steam exploded wood: An analysis on the constitutive components of steam exploded wood was done by measuring the content of lignin and sugar composition, and the result of analysis is as shown in Table 5.

It was shown that there is difference in the content of constitutive components for steam exploded wood according to its treating conditions (Table 1). It appeared that the longer the period of time steam explosion treating under the same pressure, the more the reduction of the residual content of xylose, particularly, which is the major component of hemicellulose. The reason could be that a kind of pentose sugar, such as xylan contained in a broad-leaved tree, is dehydrogenated by high temperature and acid resulting from acetyl group in lumber. As a result, it seems that there is a relative decrease in the content of hemicellulose, while a relative increase appears in the content of glucose residue in carbohydrate.

It appeared, on the other hand, that the longer the period of time treating lignin under the same pressure, the more the increase in its content. The reason must be that, as stated above, the harder conditions for treating steam explosion cause carbohydrate to change and be removed so that there is a relative increase in the content of lignin. On the other hand, it was reported that carbohydrate is coated by a sudden cooling effect when lignin, melted by the high temperature in the process of steam explosion pre-treatment, is discharged to the air (23). For this reason, it seems that why the content of lignin is increased by the harder conditions for steam explosion treating is partly influenced by the coating effect of lignin.

Finally, the result of a visual examination and analysis on the constitutive components of steam explosion treating agent showed that even the treatment for about 6 minutes is enough to achieve the defiberation effect of the steam exploded wood. It was shown that when its

economic values (steam explosion treating time and treating water content) are taken into consideration, the condition for steam explosion pre-treatment best suited to the production of dietary fiber is to perform the treatment for 6 minutes under the pressure of 25 kg/cm². Accordingly, all of the following tests were carried out centering around the best-conditioned specimen.

Physical properties of prepared insoluble dietary fiber: In this study, sodium hydroxide and sodium chloride were taken into consideration for chemical treatment required to prepare the insoluble dietary fiber from forest waste. Steam exploded wood was treated for 6 minutes under the pressure of 25 kg/cm² which appeared to be the optimal condition, and then allowed to go through the above procedures for chemical treatments after accepted as standard materials. The result of an analysis on the components of chemically treated specimen is as shown in Table 6.

The water-collecting rate after chemical treatment showed that the sodium hydroxide-treated specimen was higher than the sodium chloride treated one. The content of lignin residue was that the sodium hydroxide-treated specimen accounted for 7.6% which was high. As a result of analyses on the composition of carbohydrate, it was shown that the specimen had the purity of insoluble dietary fiber equivalent to its products on the market. Based on only these analytic results, therefore, it was possible to make certain that the sodium chloride treatment was better than the sodium hydroxide treatment, with respect to their performance, and that its result was better than the insoluble dietary fiber products on the market. Among chemical pre-treatments, in general, the treatment effect of sodium hydroxide, an alkali swelling agent, was helpful for both the swelling of lumber cellulose and the extension of internal surface width (24). It was apparent, however, that a lot of low molecular weight lignin was eluted and removed, considering that the lignin content of steam explosion treating material for this study accounted for 30%. Moreover, a lot of car-

Table 5. Sugar composition and lignin content of steam exploded wood

Species	Material No.	Steam explosion condition		Sugar composition: ¹⁾ (%)						Klason lignin ²⁾ (%)
		Pressure (kgf/cm ²)	Time (min)	Ara. ⁴⁾	Xyl. ⁵⁾	Man. ⁶⁾	Gal. ⁷⁾	Glu. ⁸⁾	Rham. ⁹⁾	
<i>Quercus mongolica</i>	EQ 25-3 ¹⁰⁾	25	3	0.7	18.6	1.6	T	78.9	T ³⁾	28.3
	EQ 25-6 ¹¹⁾	25	6	0.5	11.4	1.4	T	86.7	T ³⁾	33.5
	EQ 25-9 ¹²⁾	25	9	0.4	5.9	1.0	T	92.4	T ³⁾	38.0

¹⁾Based on total carbohydrates. ²⁾Based on exploded wood. ³⁾T: Trace, below 0.1%.

⁴⁾Ara.: Arabinose. ⁵⁾Xyl.: Xylose. ⁶⁾Man.: Mannose. ⁷⁾Gal.: Galactose. ⁸⁾Glu.: Glucose. ⁹⁾Rham.: Rhamnose.

¹⁰⁾EQ 25-3: Steam explosion condition of oak wood (25 kgf/cm², 3 min).

¹¹⁾EQ 25-6: Steam explosion condition of oak wood (25 kgf/cm², 6 min).

¹²⁾EQ 25-9: Steam explosion condition of oak wood (25 kgf/cm², 9 min).

Table 6. Yield, lignin content and sugar analysis of manufactured insoluble dietary fiber and commercial insoluble dietary fiber

Materials	Yield (%)	Klason lignin (%)	Sugar composition (%)					
			Ara. ¹⁾	Xyl. ²⁾	Man. ³⁾	Gal. ⁴⁾	Glu. ⁵⁾	Rham. ⁶⁾
Commercial insoluble dietary fiber ⁷⁾	-	2.0	0.4	7.1	1.6	-	90.9	-
Manufactured insoluble dietary fiber with sodium hydroxide treatment	59.2	7.6	0.4	9.7	1.3	-	88.6	-
Manufactured insoluble dietary fiber with sodium chlorite treatment	56.3	1.2	-	4.3	0.9	-	94.8	-

¹⁾Ara.: Arabinose. ²⁾Xyl.: Xylose. ³⁾Man.: Mannose. ⁴⁾Gal.: Galactose. ⁵⁾Glu.: Glucose. ⁶⁾Rham.: Rhamnose.

⁷⁾Sigma Chem. Co. CMC (Sodium carboxyl methyl cellulose, non-nutritive fiber), St. Louis, Missouri, U.S.A.

bohydrate also seemed to be removed with the elution of lignin because the water-collection rate was about 59%.

As a delignification treating technique used generally for lumber, the sodium chloride treatment had the water-collecting rate similar to that of the sodium hydroxide treatment, however, while there was a significant difference in the content of lignin residue. Based on this fact, it could be assumed that lignin was resolved selectively as compared with the sodium hydroxide treatment.

Table 7 shows the results of the α -cellulose content, polymerization degree and elementary analysis of both the prepared insoluble dietary fiber and its fiber products on the market.

The α -cellulose purity of the sodium chloride treated specimen accounted for 87.5% which was highest, while its polymerization degree was 530.7 which was lowest. As a result of an elementary analysis, both the insoluble dietary fiber on the market and the sodium chloride treated and prepared dietary fiber tended to have a high level of the residual chlorine content, while not any chlorine residue existed in the sodium hydroxide treated specimen. In addition, it was observed that the sodium chloride treated had a relatively low degree of polymerization, but its chemical properties were similar with those of insoluble dietary fiber products on the market.

It was shown, as described above, that the content of lignin residue appeared to be high in the process of manufacturing the insoluble dietary fiber, and that its α -cellulose purity was relatively lower than that of insoluble dietary fiber products on the market. Nevertheless,

lignin is also used for dietary fiber so that it must be not significant. Finally, the following is the processing diagram best suited to the production of insoluble dietary fiber, based on the results of an analysis on components after chemical treatment and the best-suited conditions for steam explosion pre-treatment.

Physiological function in rats fed the prepared insoluble dietary fiber

Weight gain and food efficiency ratio: Table 8 shows the rat's food intake, weight gain and food efficiency ratio during the period of experiment. Both the food intake and weight gain were higher in the groups fed high cholesterol diets than those in the normal group, but there was no difference between the groups fed high cholesterol diets. Compared to the normal group, the group of fed fiber free diet (FF group) showed some

Table 8. Effect of insoluble dietary fibers on food intake, body weight gains and food efficiency ratio (FER) in rats fed high cholesterol

Groups	Food intake	Body weight gain	FER
	(g/body wt)	(g)	
Normal	24.50 ± 0.61 ^{1)a2)}	168.0 ± 7.0 ^a	0.29 ± 0.03 ^{NS}
FF	25.93 ± 0.28 ^b	185.2 ± 5.3 ^b	0.29 ± 0.02
5C	26.70 ± 0.37 ^b	181.0 ± 8.1 ^b	0.29 ± 0.01
10C	27.29 ± 0.73 ^b	183.2 ± 5.2 ^b	0.30 ± 0.02
5M	26.34 ± 0.81 ^b	184.5 ± 6.9 ^b	0.29 ± 0.01
10M	27.43 ± 0.95 ^b	183.2 ± 5.8 ^b	0.31 ± 0.01

¹⁾All values are mean ± SE (n=10).

²⁾Values within a column with different superscripts are significantly different at p<0.05 by Tukey's test.

The experimental conditions are the same as Table 2.

Table 7. α -Cellulose content, degree of polymerization and inorganic compound analysis of manufactured insoluble dietary fiber and commercial insoluble dietary fiber

Materials	α -Cellulose content (%)	Degree of polymerization	Inorganic compound			
			Si	Ca	K	Cl
Commercial insoluble dietary fiber	84.2	635.9	0.297	1.949	-	2.154
Manufactured insoluble dietary fiber with sodium hydroxide treatment	61.7	604.2	0.392	2.926	-	-
Manufactured insoluble dietary fiber with sodium chlorite treatment	87.5	530.7	0.301	2.013	-	2.174

increase in the food efficiency ratio, however, which did not show any difference between the kinds or densities of dietary fiber. The change was same in both of the prepared dietary fiber-fed group and the group fed the dietary fiber product on the market.

This result agrees with the report of Chang and Hong (25) that there was no any remarkable difference in the weight gains of rats between the kinds of dietary fiber when added. This fact implies that the insoluble dietary fiber is less effective although it reduces the energy density in diets due to its physical and chemical properties which are different from the soluble dietary fiber.

Organ weight: Table 9 shows the weight of the liver, kidneys, small intestine and appendix by each unit. The experiment by the kinds of dietary fiber showed that the liver weight was remarkably heavier for all the groups fed the fiber free diet (FF group) than that of the normal group, but there was no any remarkable difference between the groups fed high cholesterol diets and the kidneys' weight also did not show any significant difference between the experimental groups. The small intestine weight showed no difference between the normal group and fiber free diet (FF group), but was remarkably heavier for all of the dietary fiber-fed groups. This fact is similar to the report of Lupton and Morin (26) about the increase in the small intestine weight according to the intake of soluble or insoluble dietary fiber. The appendix weight showed more increase in the experimental group than that of the normal group. In particular, there was more significant increase in the 5% prepared and on the market dietary fiber-fed 5C and 5M groups than that of fiber free diet (FF group). Adam et al. (27) reported that the increase of the appendix weight was caused by the change of possibility to produce short chain fatty acid when the dietary fiber was fermented in the large intestine by the appendix's microorganism.

It seems, therefore, that the increase in the weight of both the small intestine and appendix is a physiological

Table 9. Organ weights of rat fed insoluble dietary fibers (g/100 g body weight)

Groups	Liver	Kidney	Intestine	Cecum
Normal	2.96 ± 0.27 ^{1)a2)}	0.69 ± 0.05 ^{NS}	2.02 ± 0.07 ^a	0.57 ± 0.05 ^a
FF	5.06 ± 0.10 ^b	0.68 ± 0.03	2.26 ± 0.19 ^{ab}	0.69 ± 0.05 ^a
5C	5.19 ± 0.14 ^b	0.65 ± 0.01	2.49 ± 0.07 ^b	0.86 ± 0.06 ^b
10C	5.16 ± 0.14 ^b	0.68 ± 0.02	2.41 ± 0.14 ^b	0.61 ± 0.07 ^{ab}
5M	4.97 ± 0.10 ^b	0.66 ± 0.01	2.54 ± 0.15 ^b	0.79 ± 0.11 ^b
10M	5.04 ± 0.15 ^b	0.66 ± 0.02	2.58 ± 0.09 ^b	0.67 ± 0.05 ^{ab}

¹⁾All values are mean ± SE (n=10).

²⁾Values within a column with different superscripts are significantly different at p < 0.05 by Tukey's test.

The experimental conditions are the same as Table 2.

adoption phenomenon when adigestive organ and its function is influenced by the intake of non-digestive diets. Then, there was no difference in the change between the prepared and on the market dietary fiber.

Observation of gastrointestinal function improvements

Change of gastrointestinal transit time: The gastrointestinal transit time was observed in the second and fourth week, respectively, to examine the gastrointestinal function improvements from the prepared insoluble dietary fiber. The result is as shown in Fig. 2. Compared to the fiber free diet (FF group), all of the experiment groups showed remarkable reduction and the dietary fiber-fed group had the reduced gastrointestinal transit time in both the second and fourth week when its density is thick while there was no difference between the prepared dietary fiber-fed group and on the market dietary fiber-fed group.

The result is consistent with the report of Kelsay et al. (28) that there was remarkable reduction in the gastrointestinal transit time when 5% cellulose and guar gum were added to diets. Accordingly, both the prepared and on the market dietary fiber encouraged the *bifidus* to be so actively produced in the intestine that its peristalsis also can be activated by the produced *bifidus* (29); hence, the gastrointestinal transit time seems to be reduced.

Excrement amount and its water content: The excrement amount and its water content are as shown in Table 10. The wet weight of the excrement amount showed 42% increase in FF group, the non-fiber dietary fiber-fed group, as compared with the normal group. In addition,

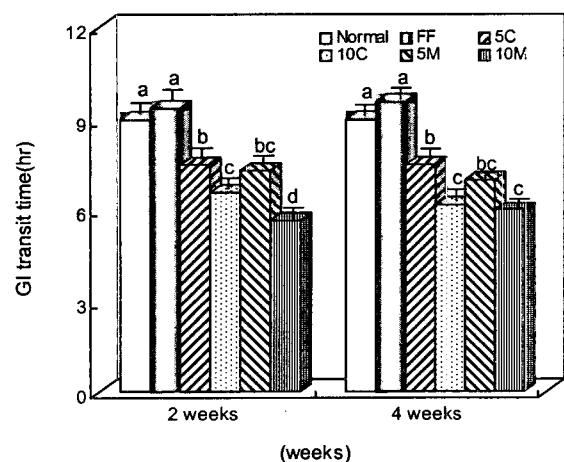


Fig. 2. Effects of insoluble dietary fibers on gastrointestinal transit time in rats fed high cholesterol diets. Bars with different letters are significantly different at p < 0.05 by Tukey's test (mean ± SE). The experimental conditions are the same as Table 2.

Table 10. Weights and water contents of feces in rats fed cholesterol diets with insoluble dietary fibers (g/day)

Groups	Wet weight	Dry weight	Water content
Normal	2.01 ± 0.10 ¹⁾²⁾	1.83 ± 0.02 ^a	0.17 ± 0.01 ^a
FF	1.16 ± 0.02 ^b	1.15 ± 0.14 ^b	0.02 ± 0.01 ^b
5C	2.52 ± 0.23 ^c	2.32 ± 0.12 ^c	0.20 ± 0.02 ^c
10C	3.97 ± 0.11 ^d	3.42 ± 0.31 ^d	0.55 ± 0.02 ^d
5M	2.63 ± 0.58 ^c	2.42 ± 0.33 ^c	0.22 ± 0.03 ^c
10M	4.01 ± 0.35 ^d	3.49 ± 0.08 ^d	0.52 ± 0.02 ^d

¹⁾All values are mean ± SE (n=10).

²⁾Values within a column with different superscripts are significantly different at p < 0.05 by Tukey's test.

The experimental conditions are the same as Table 2.

there was a remarkably increase of 116%, 180%, 126% and 244% in the 5C, 10C, 5M and 10M groups, respectively. The more the dietary fiber-fed amount, then, the more the excrement amount. It also was observed that these trends were similar with the dry weight content of excrement. The water content of excrements remarkably increased in the fiber free diet (FF group) as compared to the normal group and it significantly increased in all of the experimental dietary fiber-fed groups, too, as compared with the fiber free diet (FF group). The thicker the density of dietary fiber, the more the water content of excrements. Also, there was no difference in these trends between the prepared dietary fiber-fed group and on the market dietary fiber-fed group.

As stated above, it seems that the increased excrement amount and the reduced gastrointestinal transit time reduce the absorption time of glycolipid, therefore, which can have an effect on the glycolipid metabolism of high cholesterol diets-fed rats. Especially, the insoluble dietary fiber is less influenced by the microorganism in the intestine and, therefore, remains unfermented in the large intestine. Stephen and Cumming (30) reported, therefore, that the matrix of the dietary fiber remains intact within the large intestine so that the weight and amount of excrement can be increased effectively. Park and Joo (31) also reported that the excrement amount of the group fed wheat with the largest dietary fiber was largest among rice, unpolished rice, barley, wheat, and wheat flour.

The activity of glutamic pyruvic transaminase (GPT) and glutamic oxaloacetic transaminase (GOT) in serum: The activity of GOT for the liver tissue's mortification and GPT for its state and hypertrophy was measured to examine the prepared dietary fiber's physiological harmfulness, which is shown in Fig. 3. The activity of GOT and GPT appeared that there was more increase in the FF group, the non-fiber diets-fed group, than those of the normal group, while there was not any significant difference between the experiment groups, as compared with the FF group. Although there was an increase in

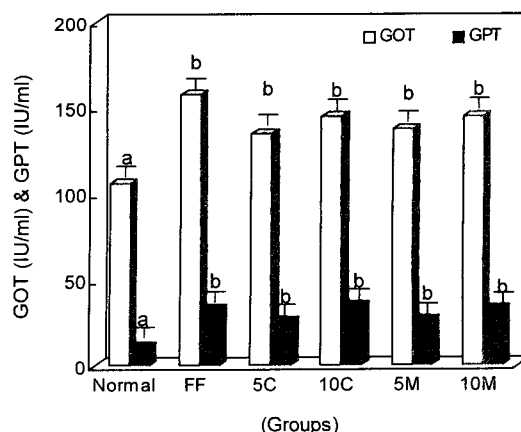


Fig. 3. Effects of insoluble dietary fibers on serum GOT and GPT activities in rats fed high cholesterol diets. Bars with different letters are significantly different at p < 0.05 by Tukey's test (mean ± SE). The experimental conditions are the same as Table 2.

the activity of GOT and GPT by the high cholesterol diets, accordingly, no any extra increase appeared in the activation by the insoluble dietary fiber; thus, it was shown that the prepared insoluble fiber did not contain any toxic substance.

The above examination of physiological function in rats showed that adding the prepared insoluble dietary fiber to high cholesterol diets was so effective in reducing the gastrointestinal transit time and improving the gastrointestinal function, due to increased excrement amount, that it could be helpful for prevention of constipation.

In conclusion, the insoluble dietary fiber, cellulose, was effectively prepared by both steam exploded, known as an effective treatment method, and delignification, a pre-treatment method, with *Quercus mongolica* of oak tree featuring the largest forest waste. When compared with the insoluble dietary fiber products on the market, also, the prepared dietary fiber showed similar physiological effects in the gastrointestinal function improvement and physiological non-toxicity of high cholesterol-fed rats; hence, it is apparent that this study's insoluble dietary fiber has been so excellently prepared in the currently developed manner that it can be used effectively in the future.

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SUMMARY

This study was conducted to establish the process to

prepare high-purity insoluble dietary fiber treated chemically with *Quercus mongolica* having the largest forest waste in Korea, feed 5% and 10% insoluble dietary fiber product on the market and prepared dietary fiber mixed with 1% high cholesterol diets to rats for 4 weeks to examine the physiological function of insoluble dietary fiber prepared from the forest waste, and observe the gastrointestinal function improvement and physiological non-toxicity.

The effective insoluble dietary fiber was prepared by the delignification treatment of washing and dehydrating with 1% sodium hydroxide solution after steam explosion treatment of *Quercus mongolica*. The result of experiments with rats designed to examine the physiological function showed that the dietary intake, weight gain and dietary efficiency of high cholesterol-fed rats were influenced by the insoluble dietary fiber. While the dietary fiber did not have any impact on the weight of the liver, kidneys and small intestine, the appendix weight increased in the groups fed the dietary fiber product on the market and the prepared dietary fiber as compared to the normal group and non-fiber diets-fed group. When the gastrointestinal transit time was observed in the second and fourth week of experiment, respectively, all the insoluble dietary fiber-fed groups showed the reduced gastrointestinal transit time as compared to the non-fiber diets-fed group. The excrement amount increased in the non-fiber diets-fed groups, compared with the normal group and the more the fed amount of dietary fiber, the more the excrement amount. The water content of excrements remarkably increased in the fiber free diets-fed group as compared with the normal group and the thicker the density of dietary fiber, the more the water content of excrement, compared to the group fed fiber free diets. The activation of GOT and GPT in serum was not influenced by the insoluble dietary fiber. There was no difference in the physiological function between the dietary fiber product on the market and the prepared dietary fiber. In conclusion, the physiological similarity was found between the insoluble dietary fiber product on the market and the prepared insoluble dietary fiber. It is apparent, therefore, that the production method of insoluble dietary fiber newly developed by this study is excellent.

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