



ORYZA OIL & FAT CHEMICAL CO., LTD.

# BROCCOLI SPROUT

Anti-Oxidative Supplement  
Detoxification Activity  
Beauty Function

## ■BROCCOLI POWDER

(Powder, Food Grade)

## ■BROCCOLI SPROUT EXTRACT

(Water-soluble Powder, Food Grade, Cosmetic Grade)

## ■BROCCOLI SPROUT EXTRACT-PC

■ (Water-soluble Powder, Cosmetic Grade)

## ■BROCCOLI SPROUT EXTRACT-PS1

(Water-soluble Powder, Food Grade)

## ■BROCCOLI SPROUT POWDER

(Powder, Food Grade)



ORYZA OIL & FAT CHEMICAL CO., LTD

ver. 6.3 YF

# BROCCOLI SPROUT

## Health Food and beauty Ingredients

### 1. Introduction

The term “preventive medicine” has recently attracted much attention. The concept is based on the idea that an improvement in dietary habits prevents diseases under daily stressful conditions.

Dating back to about ten years ago, a “designer foods” project began nationwide in the US. At the time, excess energy intake was recognized as a causative factor of “cancer” along with various other factors, such as life environment and eating habits. In the hope of suppressing these factors and preventing “cancer”, numerous compounds derived from fruits and vegetables were investigated.

Our company has recently developed a variety of products to contribute to preventive medicine. We selected broccoli since it was found to have a component providing potential anti-cancer effects, and produced powdered broccoli sprouts (broccoli sprout extract) as well as powdered broccoli (Broccoli powder) on a commercial basis.



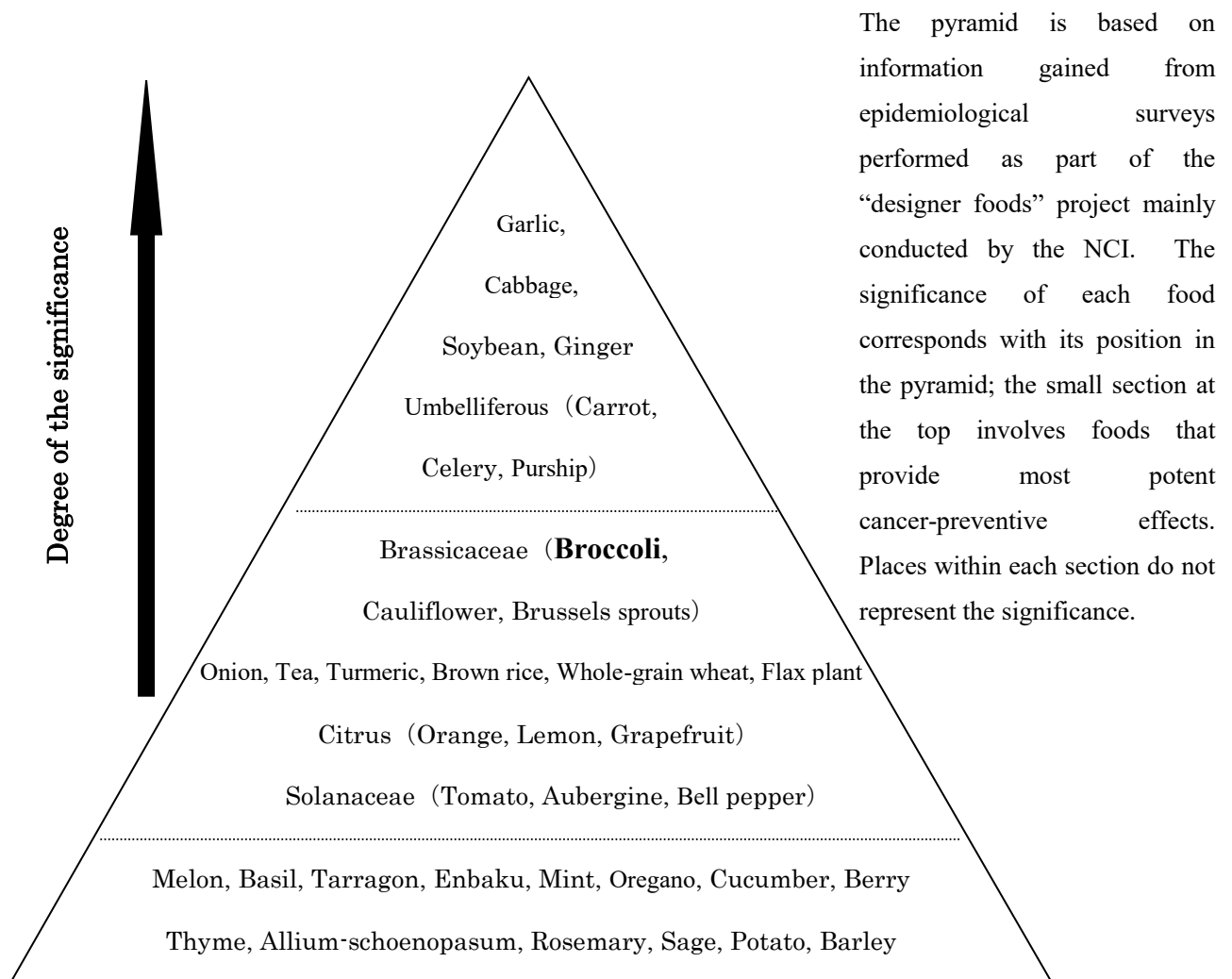
Broccoli



Broccoli Sprout

## 2. What are “designer foods”?

In 1990, the “designer foods” project was established in the US mainly by the National Cancer Institute (NCI). To clarify how food components prevent “cancer”, the project has been conducted with a collaborative effort among various research groups. During the project, epidemiological surveys have demonstrated potential cancer-preventive foods and food components as shown in the pyramid below (Figure 1). The significance of each food corresponds with its position in the pyramid; the small section at the top involves foods that provide most potent cancer-preventive effects.



**Fig. 1 Potential cancer-preventive foods and food components**

### 3. Components of the broccoli powder

#### 3-1 Isothiocyanates and sulforaphane

Volatile allyl sulfides, abundantly found in alliaceous vegetables such as garlic and onions as well as in cruciferous vegetables, are not only sources of the distinctive flavor of these vegetables but an inhibitor of cancer in the skin, colon, rectum, liver as well as the lung and proventriculus of rats. Many studies have recently demonstrated the mechanisms underlying the cancer-preventive effects of allyl sulfides in the alliaceous. Some have suggested the components act as free radical scavengers or inhibit the activation of carcinogen metabolism. On the other hand, other type of sulfur-containing components, “isothiocyanates”, richly found in cruciferous vegetables such as broccoli, radishes, Japanese white radishes and turnips have recently gained considerable attention. “Isothiocyanates” have been demonstrated to inhibit cancer of the esophagus, large intestine, liver, lung and proventriculus in animal studies. Above all, “sulforaphane”, a type of isothiocyanates found in broccoli, has attracted great attention. More recently, this component has been found to prevent carcinogenesis in animal studies by inducing detoxification enzymes to neutralize carcinogens .

#### 3-2 Active components

Along with sulforaphane cited above, broccoli contains other compounds exhibiting anti-cancer effects, that is, indole-3-carbinol, protocatechuic acid, chlorogenic acid and the carotenoids. Sulforaphane, a component of broccoli, shows potential detoxification activity against carcinogens and endocrine-disrupting chemicals. Broccoli sprouts have increased concentrations of sulforaphane just after budding. Our broccoli sprout extract, produced using a novel concentration technique, contains a high concentration of sulforaphane.

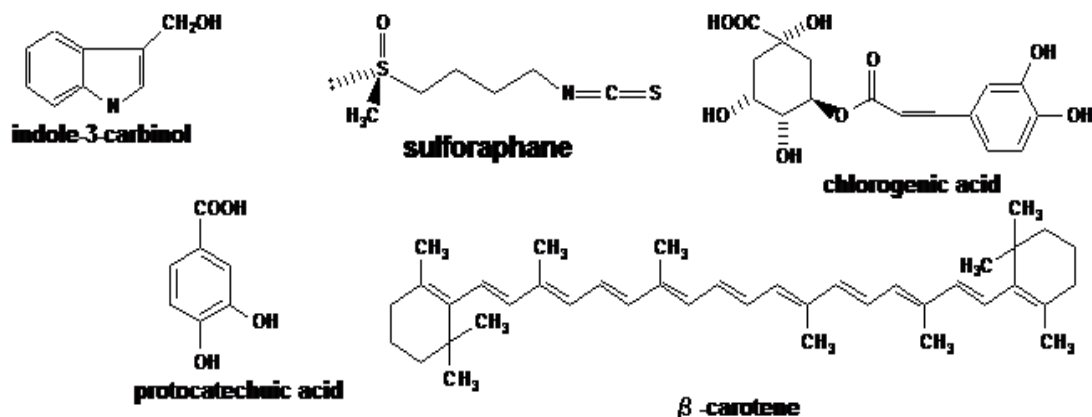


Fig. 2 Structures of major compounds in broccoli

### 3-3 Synthesis of sulforaphane

Sulforaphane is produced from a certain type of glucosinolate that is a family of sulfoglycosides. Glucosinolates are enzymatically degraded by myrosinase to cleave thioglycosidic linkages as shown in Figure 3. The resulting sulfate esters cause intramolecular reactions, yielding isothiocyanates.

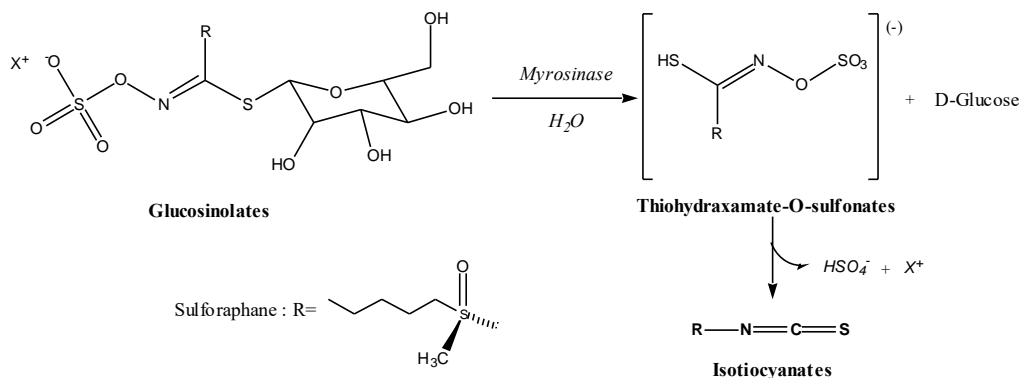


Fig. 3 Synthesis of isothiocyanates from glucosinolates

## 4. Functional activities of broccoli powder

### 4-1 Antioxidant

#### 4-1-1 Comparison of activities: SOD-like activity and DPPH radical scavenging activity

Among daily-consumed vegetables, broccoli has been demonstrated more potent SOD-like activity and DPPH radical scavenging activity. According to Fig. 4, broccoli shows superior activities, particularly, in the assay for SOD-like activity, an index of antioxidant activity, broccoli exhibits the highest value among daily-consumed vegetables.

Table 1 Antioxidant effect of broccoli

Vegetable	SOD-like Activity	DPPH Radical Scavenging Activity
Spinach	24	46
Shungiku	5	65
Cabbage	19	34
<b>Broccoli</b>	<b>28</b>	<b>59</b>
Cauliflower	23	43
Lettuce	3	18
Chinese cabbage	18	19
Welsh onion	14	31
Parsley	18	79

#### 4-1-2 Effect of BROCCOLI SPROUT EXTRACT on intravitam antioxidant in normal healthy adults (oral application)

The effect of BROCCOLI SPROUT EXTRACT-PS1(BSE, contained 1% sulforaphane) on intravitam antioxidant was examined in oral administration clinical trial. Decrease of urinary 8-OHdG level means to inhibit oxidative DNA damage *in vivo*. And decrease of urinary Hexanoyl-Lys (HEL) level means to inhibit lipid oxidation *in vivo*. Measuring these levels is able to discuss intravitam antioxidant. We chose 20 healthy males who have high levels of 8-OHdG and HEL from 30 individuals. And they were randomly divided into three groups: BSE-150 group, BSE-450 group, and control group. Each subject in BSE-150 group and BSE-450 group consumed 150 mg or 450 mg of BROCCOLI SPROUT EXTRACT per day for 3 weeks. Each subject in control group consumed 450 mg of dextrin instead of BROCCOLI SPROUT EXTRACT. For evaluation, urinary 8-OHdG and HEL level were measured at initiation, 1 and 3-week. And on 3-week, questionnaire were performed.

Consequently, urinary 8-OHdG and HEL levels of BSE-150 and BSE-450 group at 3-week were reduced significantly ( $p < 0.05$ ) in comparison with control group (Fig 4). Additionally, urinary 8-OHdG on 3-week was reduced significantly in comparison with initiation ( $p < 0.01$ ). As a result, BROCCOLI SPROUT EXTRACT was recognized to inhibit oxidative DNA damage *in vivo*.

Urinary HEL level of BSE-450 group at 3-week was reduced significantly in comparison with control group ( $p < 0.05$ ), showing an effect to inhibit lipid oxidation *in vivo*.(Fig 5)

To confirm subjective symptoms, the questionnaire was carried out about physical condition, condition of a stomach, fatigue degree, degree of irritation, skin shine, quality of sleep. There were not difference with control group at all.

In this study, BROCCOLI SPROUT EXTRACT is effective to inhibit oxidative DNA damage and lipid oxidation *in vivo*. and this result indicate that BROCCOLI SPROUT EXTRACT-PS1 has antioxygenation *in vivo*. It seems that this function appears after consecutive three weeks taking.

(The final examination is a collaboration with Nippon Milk Community Co.,Ltd.)

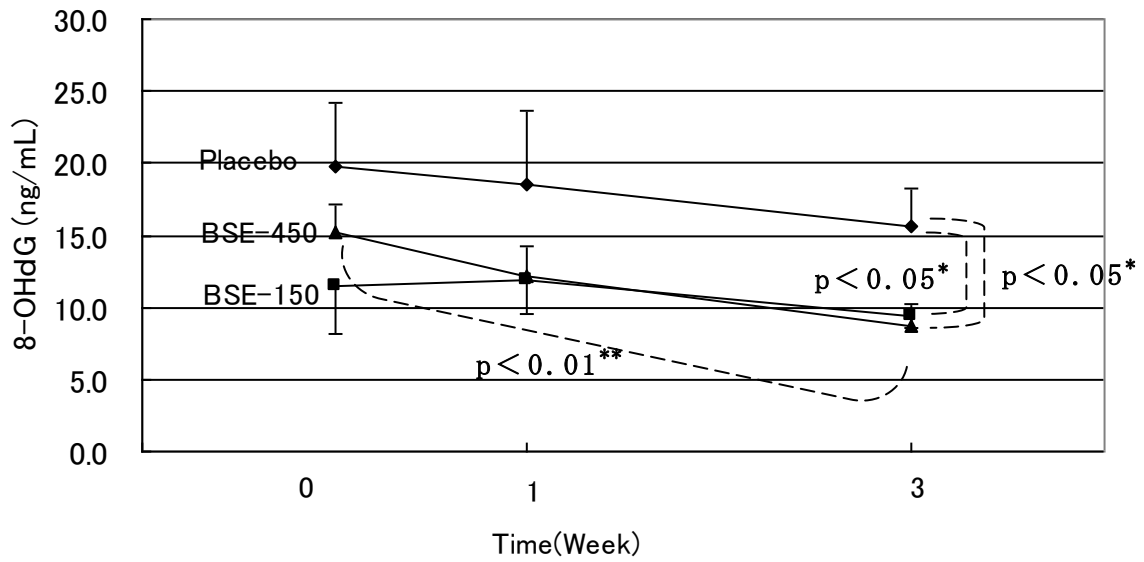


Fig. 4 Effect on urinary 8-OHdG level

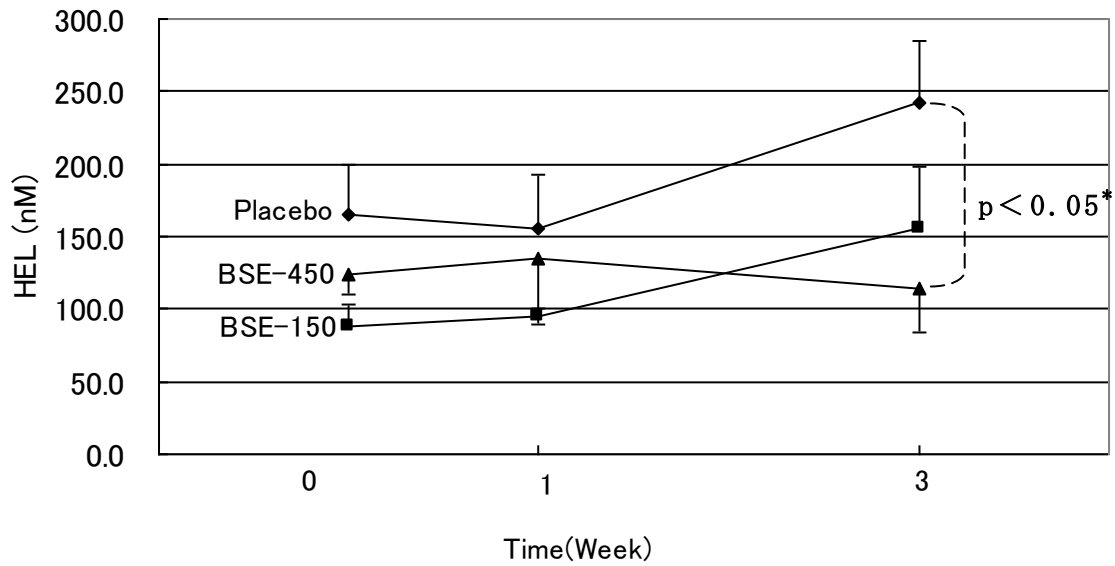


Fig. 5 Effect on urinary HEL level

## 4-2 Anti-cancer effect

### 4-2-1 Inhibitory effect of sulforaphane on rat mammary cancer

Sulforaphane, found in daily-consumed vegetables, induces phase II enzymes without affecting the synthesis of cytochrome P-450. For the investigation of the enzyme induction, structurally related isothiocyanates (ITC) were synthesized, and several norbornyl-ITC were found to induce phase II enzymes by the same mechanisms as sulforaphane[4]. Zhang et al. examined the inhibitory effects of sulforaphane and three types of norbornyl-ITC compounds on rat mammary cancer induced by 9,10-dimethyl-1, 2-benzanthracene (DMBA)[5]. Table 2 shows the results of their experiment in which female rats at 40 days of age were serially administered ITC at a daily dose of 75, 100 or 150  $\mu$  mol for five days, followed by oral administration of 8.0 mg of DMBA dissolved in 1 ml of sesame oil. Carcinogenesis 152 days after DMBA administration (at 202 days of age) was examined. The incidence of cancer was markedly reduced in rats treated with 150  $\mu$  mol of sulforaphane to 0.26, compared with 1.56 in the control. Compound 1 showed potent activity comparable to that of sulforaphane, whereas compounds 2 and 3 had lower activity.

**Table 2. Protective effect of sulforaphane and norbornyl isothiocyanates 2, 3, and 4 on incidence and multiplicity of mammary tumors in DMBA-treated female Sprague-Dawley rats**

Treatment group	No. of rats		Tumor incidence % (% of control)	No. of tumors	
	In group	With tumors		Total	Multiplicity (% of control)
<b>Control</b>	25	17	68.0 (100)	39	1.56 (100)
<b>Sulforaphane</b>					
75 $\mu$ mol	20	7	35.0*(51.4)	9	0.45† (28.8)
150 $\mu$ mol	19‡	5	26.3*(38.7)	5	0.26† (16.7)
<b>Compound 2</b>					
75 $\mu$ mol	20	5	25.0*(36.8)	6	0.30† (19.2)
150 $\mu$ mol	20	5	25.0*(36.8)	7	0.35† (22.4)
<b>Compound 3</b>					
(100 $\mu$ mol)	19§	9	47.3 (69.6)	14	0.74† (47.4)
<b>Compound 4</b>					
(100 $\mu$ mol)	20	8	40.0 (58.8)	8	0.40† (25.6)

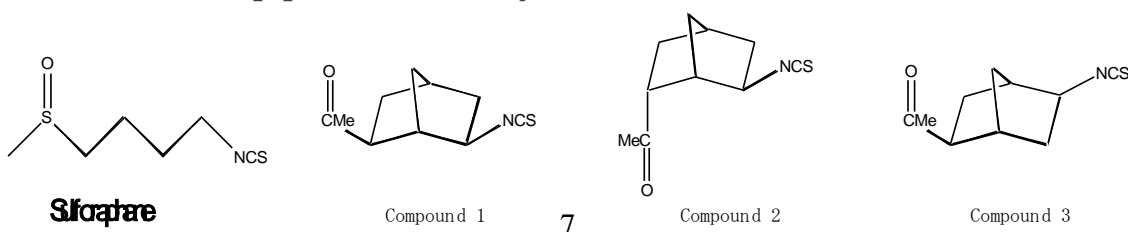
A total of 145 rats were entered into the experiment. Each received 8 mg of DMBA at age 50 days. There were initially 25 controls and 20 animals in each of the six treated groups. The above analysis is based on 143 animals (see below).

\*  $P < 0.05$  for differences from controls (Fisher exact test).

†  $P \leq 0.01$  for differences from controls (Poisson distribution model).

‡ One rat died immediately after gavage and is not included.

§ One rat died without palpable tumors at 167 days and is not included.





#### 4-2-2 Inhibitory effect of BROCCOLI SPROUT POWDER on azoxymethane-induced colonic aberrant crypt foci in rats

In our company, we confirmed the inhibitory effects of BROCCOLI SPROUT POWDER (GBP) on azoxymethane (AOM) induced colonic aberrant crypt foci (ACF) in collaboration with Kanazawa Medical and Gifu University, and reported the results at the 62<sup>nd</sup> General Meeting held by the Japanese Society of Cancer (2003).

##### Method

Thirty-two male F344 rats were used, and divided into the following 5 groups: Group 1: AOM alone (20 mg/kg, once a week, total: twice, subcutaneous injection); Group 2: AOM + 20 ppm GBP; Group 3: AOM + 100 ppm GBP; Group 4: 100 ppm GBP; and Group 5: no treatment. Food containing GBP was administered for 4 weeks starting from 1 week before AOM administration. The rats were sacrificed 4 weeks after the start of the experiment, and ACF was counted (Fig.6).

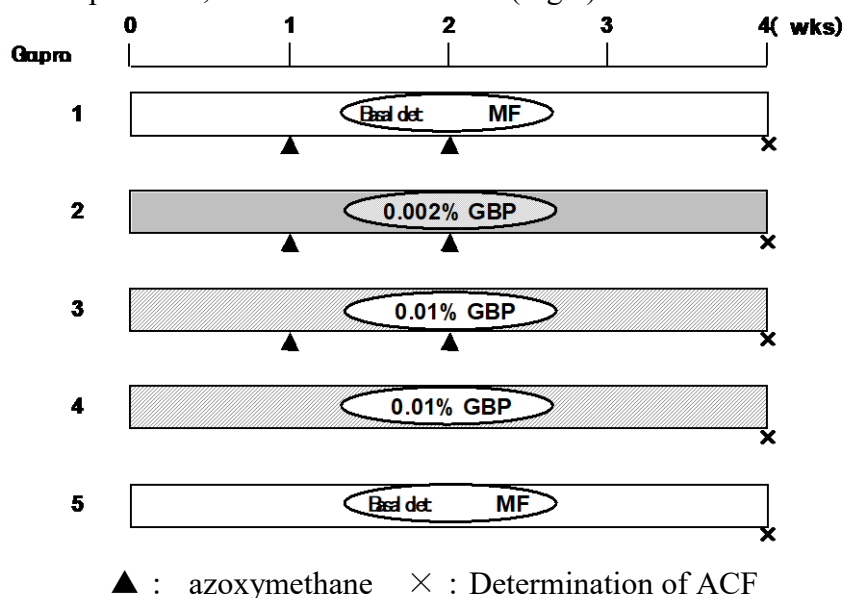


Fig. 6 Experimental protocol

##### Result

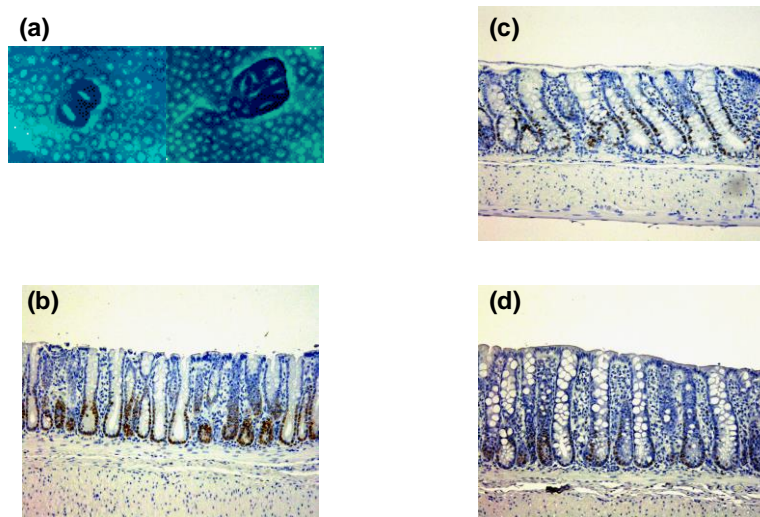
The number of ACF lesions was  $106 \pm 10$ /colon in Group 1,  $56 \pm 11$ /colon in Group 2, and  $64 \pm 23$ /colon in Group 3. The values in Group 2 and 3 in which GBP was administered were significantly lower than that in Group 1 ( $p < 0.001$ ). In Group 4 and 5 without AOM treatment, no ACF lesion developed (Table 3). Immunohistochemical Staining of the colonic mucosa of rat in Group 1, 2, and 3 are shown in Fig. 7.

**Table 3 Effect of GBP on AOM-induced ACF formation**

Group No.	Treatment (No. of rats examined)	Incidence (%)	Total no. of ACF/colon	Total no. of Acs/colon	No. of aberrant Crypts/focus
1	AOM alone (8)	8/8 (100%)	106 ± 10	220 ± 3	2.07 ± 0.07
2	AOM+0.002%GBP (8)	8/8 (100%)	56 ± 11 *	94 ± 16 *	1.67 ± 0.09 *
3	AOM+0.01%GBP (8)	8/8 (100%)	64 ± 23 *	106 ± 39 *	1.66 ± 0.06 *
4	0.01%GBP (4)	0/4 (0%)	0	0	0
5	None (4)	0/4 (0%)	0	0	0

Mean ± SD.

\**p* <0.001



**Fig. 7 Morphology and PCNA immunohistochemistry of ACF**

- (a) ACF on methylene-blue-stained colonic mucosa of a rat in group 1
  - (b) PCNA immunohistochemistry of “normal appearing” crypts of a rat from group 1
  - (c) PCNA immunohistochemistry of “normal appearing” crypts of a rat from group 2
  - (d) PCNA immunohistochemistry of “normal appearing” crypts of a rat from group 3
- Original magnifications, (a) x4; and (b)-(d) x20

**Conclusion**

GBP inhibited the development of AOM-induced rat ACF. Furthermore, sulforaphan contained in GBP has been reported to induce antidotal enzymes and inhibit production of free radicals; this substance may inhibit carcinogenesis in the colon.

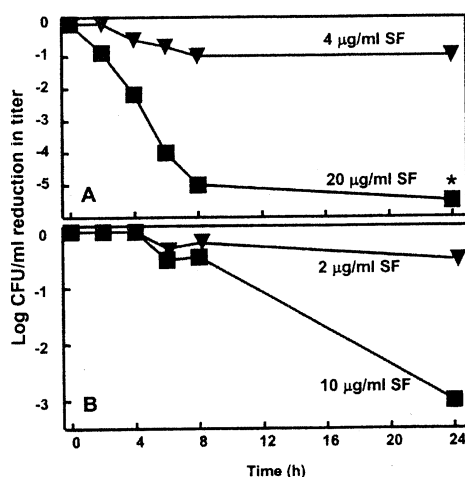
### 4-3 Inhibitory effect on *Helicobacter pylori*

#### 4-3-1 Inhibitory effect of sulforaphane on *Helicobacter pylori*

Gastric infection with *Helicobacter pylori* is a cosmopolitan problem, and is especially common in developing regions where there is also a high prevalence of gastric cancer. These infections are known to cause gastritis and peptic ulcers, and dramatically enhance the risk of gastric cancer.

At neutral pH, sulforaphane exhibited high bacteriostatic activity against all 48 strains tested, with MIC values ranging from 0.06 to 8  $\mu$ g/ml [mean=2.5  $\mu$ g/ml; median=2  $\mu$ g/ml; MIC<sub>90</sub> (MIC at which growth of 90% of strains is inhibited)=4  $\mu$ g/ml]. Thus, sulforaphane was significantly more potent than other natural compounds that have been tested, such as resveratrol from grape skins and red wine (MIC<sub>90</sub>=25  $\mu$ g/ml), allixin from garlic bulbs (MIC<sub>90</sub>=25  $\mu$ g/ml), protolichesterinic acid from the lichen *Cetraria islandica* (MIC<sub>90</sub>=32  $\mu$ g/ml), and epigallocatechin gallate from tea (MIC<sub>90</sub>=32  $\mu$ g/ml). Moreover, sulforaphane exhibited a high inhibitory activity (MIC<sub>90</sub>=4  $\mu$ g/ml) against both the clarithromycin-resistant strains (n=17; MIC<sub>90</sub> for clarithromycin, 32  $\mu$ g/ml) and the metronidazole-resistant strain (n=14; MIC<sub>90</sub> for metronidazole, 64  $\mu$ g/ml) tested.

Having established the bacteriostatic activity of sulforaphane against *H. pylori*, we next evaluated its bactericidal potency by using a time-to-kill assay with one reference strain (26695) and one clinical isolate (LBN201). The effect was nearly always concentration dependent (Fig. 8)



**Fig. 8 Bactericidal potency of sulforaphane on two strains of *H. pylori***  
(A, LBN201, clinical isolate; B, 26695, reference strain)

SF: sulforaphane \* : below the limits of detection

### 4-3-2 Inhibitory effects of broccoli sprouts on *Helicobacter pylori*

A study group of Tsukuba University reported that ingestion of broccoli sprouts, a food containing sulforaphan, relieved atrophy of the gastric mucosa related to loading with a high salt diet in *H. pylori*-infected mice at the 9th Meeting held by the Society of *Helicobacter* in 2003.

#### **Method**

A high salt diet (7.5%NaCl) or a standard diet (0.25%NaCl) was given to *H. pylori* (SS1)-infected 6-week-old mice for 2 months. To some mice, broccoli sprouts containing 2.5 mM sulforaphan were given. We investigated histological changes in the gastric mucosa (Updated Sydney System), DNA damage (8OhdG content), and expression of TNF- $\alpha$ , IL-1 $\beta$ , and IL-8 (real-time PCR).

#### **Result**

The high salt diet increased expression of TNF- $\alpha$ , IL-1 $\beta$ , and IL-8 in the gastric mucosa of the *H. pylori*-infected mice, accelerating deterioration of atrophy.

Administration of broccoli sprouts decreased the number of *H. pylori* bacteria, and relieved high salt diet-related atrophy of the gastric mucosa.

#### **Conclusion**

These results suggest that ingestion of broccoli sprouts, a food containing sulforaphan, relieves atrophy of the gastric mucosa in patients with *H. pylori* infection, preventing the development of gastric cancer.

### 4-3-3 Inhibitory effects of broccoli sprouts on *Helicobacter pylori*

According to a result of a research group of Tsukuba University, 9 of the 25 *H. pylori* infected people who took broccoli sprouts (contained 250 mg sulforaphane precursor) for eight weeks, became false-negative in the HpSA (*H. pylori* Stool Antigen) test <sup>7)</sup>.

## 4-4 Skin Lightening Effect

### 4-4-1 Inhibition of tyrosinase (*in vitro*)

Tyrosinase is the enzyme responsible for skin hyperpigmentation in the production of melanin through dopa-quinone pathway. *In vitro* studies confirmed that Broccoli Sprouts Extract demonstrated a dose-dependent inhibitory effect against tyrosinase (Fig. 9).

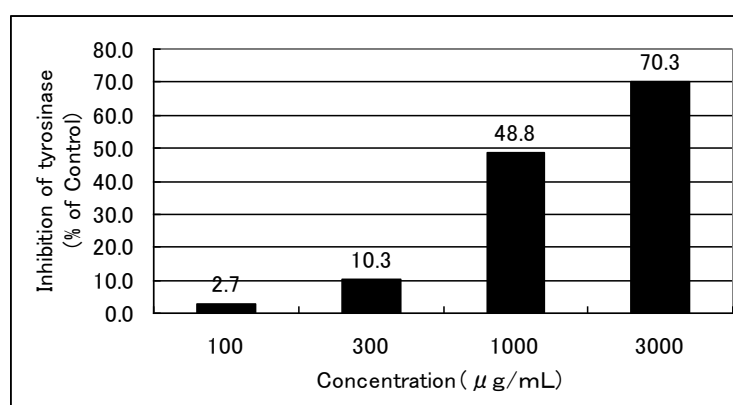


Fig.9 The Effect of Broccoli Sprouts Extract on tyrosinase

### 4-4-2 Inhibition on B16 melanoma cells

Further experiment was prompted using B16 melanoma cells to evaluate the skin lightening effect of Broccoli Sprouts Extract. As shown in Fig. 10, Broccoli Sprouts Extract demonstrated a dose-dependent inhibitory effect against melanin production in B16 melanoma cells. Therefore, Broccoli Sprouts Extract is preventive against skin hyperpigmentation.

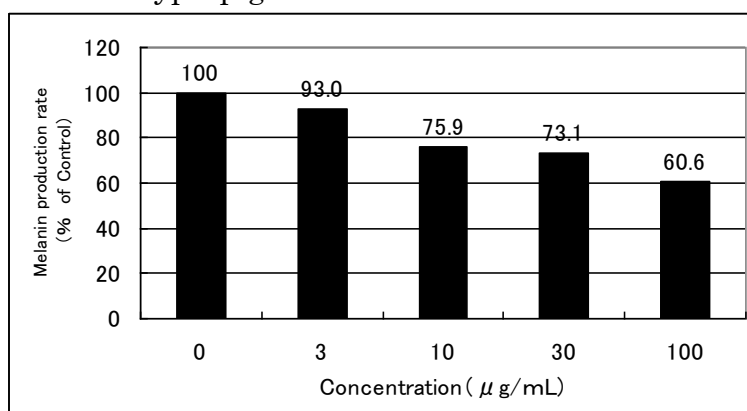


Fig.10 The Effect of Broccoli Sprouts Extract on Melanocyte Growth

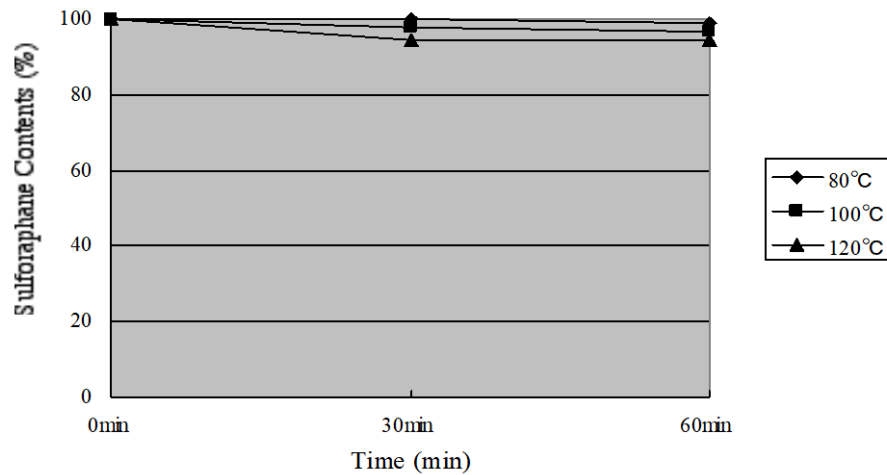
**References**

- 1) T. Osawa, H. Ohigashi, K. Yoshikawa (Functional Food for Cancer Prevention) 1999.
- 2) *J. Agric. Food Chem.* 1996, 1014-1021
- 3) *Bull. Agric. chem. Soc. Japan* 1959, 555-556
- 4) *J. Med. Chem.*, 1994, 170
- 5) *Proc. Natl. Acad. Sci. USA*, 1994, 3147-3150
- 6) *Proc. Natl. Acad. Sci. USA*, 2002, 7610-7615
- 7) *J. New. Rem. & Clin.*, 2005, 979-980

## 5. Stability of broccoli products

### 5-1 Thermal resistance

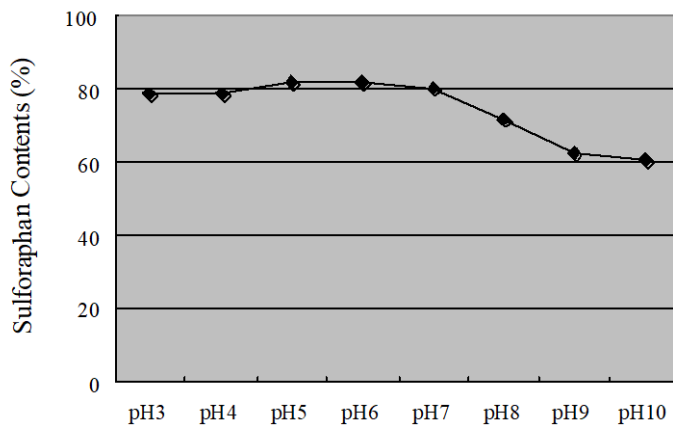
The pyrolysis of Broccoli Products does not occur at a normal food processing temperature for 60 minutes.



### 5-2 pH stability

Sulforaphans in Broccoli Products remains stable especially at neutral to acid field of pH.

※ The sulforaphane concentration in 90% ethanol solution (pH 6 unregulated) was set 100%



## 6. Daily dosage of BROCCOLI SPROUT EXTRACT & POWDER

BROCCOLI SPROUT EXTRACT	90-180 mg/day
BROCCOLI SPROUT EXTRACT-PS1	180-360 mg/day
BROCCOLI SPROUT POWDER	150-300 mg/day





## 7. Nutrition facts

Items Analyzed	BROCCOLI SPROUT EXTRACT	BROCCOLI SPROUT EXTRACT-PS1	BROCCOLI SPROUT POWDER	BROCCOLI POWDER
Water	2.2g/100g	1.1g/100g	2.2g/100g	2.7g/100g
Protein	8.7g/100g	8.7g/100g	45.2g/100g	32.4g/100g
Fat	0.25g/100g	0.3g/100g	12.5g/100g	6.9g/100g
Ash	2.55g/100g	2.6g/100g	5.4g/100g	6.2g/100g
Sugar	—	—	13.5g/100g	21.5g/100g
Available carbohydrate	86.3g/100g	87.3g/100g	—	—
Energy	383kcal/100g	387kcal/100g	347kcal/100g	278kcal/100g
Dietary Fiber	0.15g/100g	0.2g/100g	21.2g/100g	30.3g/100g
Sodium	16mg/100g	14mg/100g	14.4mg/100g	236mg/100g
Iron	0.2mg/100g	—	—	10.5mg/100g
Calcium	66mg/100g	—	—	264mg/100g
Potassium	790mg/100g	—	—	2.42g/100g
Zinc	0.6mg/100g	—	—	4.67mg/100g
Vitamin A	0.6 μg/100g 以下	—	—	537 μg/100g
Vitamin B1	0.28mg/100g	—	—	0.81mg/100g
Vitamin B2	0.37mg/100g	—	—	1.04mg/100g
Vitamin C	1mg/100g 以下	—	—	526mg/100g
Vitamin E	0.1mg/100g 以下	—	—	7.4mg/100g
Vitamin U	1mg/100g 以下	—	—	58mg/100g
Pantothenic Acid	2.42mg/100g	—	—	4.53mg/100g
Folic acid	12 μg/100g	—	—	0.39mg/100g
Analysis Examination Report	<p>○Tested by: SRL, Inc. ○Research result issue number: 200506210021 200603240020</p> <p>○Tested by:Japan Food Research Center Foundation ○Research result issue number: 306030747 - 001</p> <p>The value from water to dietary fiber is calculated from analysis value of broccolis sprout extraction written in above reports. The value from sodium to folic acid is measured value.</p>	<p>It is calculation value from analysis value of broccolis sprout extraction.</p>	<p>○Tested by:Japan Food Research Center Foundation ○Research result issue number: 301100594 - 001</p>	<p>○Tested by:Japan Food Research Center Foundation ○Research result issue number: 301110593 - 001 302040067 - 001 302030274 - 001, 002 302030274 - 003</p>

## 8. Acute toxicity and safety

### 8-1 Residual agricultural chemicals of BROCCOLI SPROUT EXTRACT

BROCCOLI SPROUT EXTRACT is conformed to regulation stipulated for 447 residual agricultural chemical compounds. No residual agricultural chemicals detected as confirm by test trustee.

Test trustee : Masis Co. Ltd.

Data : August 10, 2006

Report No. : 6900

### 8-2 Residual agricultural chemicals of BROCCOLI SPROUT POWDER

Assayed Items	Results	Detection Limits	Assay Method
Chlorpyrifos	Not Detected	0.01ppm	Gas Chromatography
Parathion	Not Detected	0.05ppm	Gas Chromatography
Methamidphos	Not Detected	0.05ppm	HPLC-MS
Fenvalerate	Not Detected	0.02ppm	Gas Chromatography
BHC	Not Detected	0.02ppm	Gas Chromatography
DDT	Not Detected	0.02ppm	Gas Chromatography
Aldrin	Not Detected	0.01ppm	Gas Chromatography
Dieldrin	Not Detected	0.01ppm	Gas Chromatography
Endrin	Not Detected	0.01ppm	Gas Chromatography
Diazinon	Not Detected	0.05ppm	Gas Chromatography
Parathion	Not Detected	0.05ppm	Gas Chromatography
Marathion	Not Detected	0.05ppm	Gas Chromatography

Tested by: Japan Food Research Center Foundation

Research results issue number: 302070278-001(Chlorpyrifos-Fenvalerate)

Research results issue number: 301100594-001(BHC-Marathion)

### 8-3 Residual agricultural chemicals of BROCCOLI POWDER

Assayed Items	Results	Detection Limits	Assay Method
Spinosad	Not Detected	0.01ppm	HPLC-MS
Chlorpyrifos	Not Detected	0.01ppm	Gas Chromatography
Parathion	Not Detected	0.05ppm	Gas Chromatography
Methamidophos	Not Detected	0.05ppm	HPLC-MS
Iprodione	Not Detected	0.05ppm	HPLC-MS
Fenvalerate	Not Detected	0.02ppm	Gas Chromatography
Procymidone	Not Detected	0.01ppm	Gas Chromatography
Flufenoxuron	Not Detected	0.05ppm	HPLC-MS

Tested by: Japan Food Research Center Foundation  
 Research results issue number: 202061354-001

### 8-4 Acute toxicity

Five weeks old mice were orally given BROCCOLI SPROUT POWDER (5000mg/kg) and then fed a laboratory chow for two weeks. No toxic effects were observed, thus the LD<sub>50</sub> (mouse) is more than 5000mg/kg.

## 9. Commercial Application

Applications	Examples
Dried Foods	Soup, Dried noodles, Seasoning, Pasta, Cereal, Oatmeal, and Topping for pizza.
Confectionery	Candies, Gum, Cookies, Pudding, Jelly, Yogurt, Chocolate
Snacks	Rice crackers, Cookies, and Wafers.
Fermentative Foods	Bread and Yogurt
Others	Health foods, Nutraceutical foods, and Functional foods

## 10. Packaging

BROCCOLI SPROUT EXTRACT

3kg Interior packaging: aluminium-coated plastic bag

Exterior packaging: cardboard box

**BROCCOLI SPROUT EXTRACT-PS1**

5kg Interior packaging: aluminium-coated plastic bag

Exterior packaging: cardboard box

**BROCCOLI SPROUT POWDER**

5kg Interior packaging: aluminium-coated plastic bag

Exterior packaging: cardboard box

**BROCCOLI POWDER**

10kg Interior packaging: aluminium-coated plastic bag

Exterior packaging: cardboard box

**BROCCOLI SPROUT EXTRACT-PC**

1kg Interior packaging: aluminium-coated plastic bag

Exterior packaging: cardboard box

## 11. Storage

Store in cool, dry place. Avoid humidity.

## 12. Expression

&lt; Food &gt;

**BROCCOLI SPROUT EXTRACT,****BROCCOLI SPROUT EXTRACT-PS1**

Expression : Broccoli Sprout Extract

**BROCCOLI SPROUT POWDER**

Expression : Broccoli Sprout Powder

**BROCCOLI POWDER**

Expression : Broccoli Powder

&lt; Cosmetic &gt;

**BROCCOLI SPROUT EXTRACT-PC**

INCI Name : Brassica Oleracea Italica(Broccoli) Sprout Extract , Dextrin

※Please refer to your nation's standard.

**PRODUCT STANDARD**

## PRODUCT NAME

**BROCCOLI SPROUT EXTRACT**

(Food)

This product is the powder extracted with aqueous ethanol from germinated broccoli (*Brassica oleracea* var. *italica*). It contains minimum 2.0% of sulforaphane and 5.0% of glucoraphanin.

This powder is water-soluble.

<u>Appearance</u>	Slight yellowish powder with slight unique aroma.	
<u>Glucoraphanin</u>	Min. 5.0 %	(Sulforaphan amount (GC Method) × 2.46*)
<u>Sulforaphane</u>	Min. 2.0 %	(GC Method)
<u>Loss on Drying</u>	Max. 10.0 %	(Analysis for Hygienic Chemists , 1g, 105°C, 2h)
<u>Purity Test</u>		
(1) Heavy Metals (as Pb)	Max. 10 ppm	(Sodium Sulfide Colorimetric Method)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 1 ppm	(Standard Methods of Analysis in Food Safety, The Third Method, Apparatus B)
<u>Standard Plate Counts</u>	Max. 1 × 10 <sup>3</sup> cfu/g (Analysis for Hygienic Chemists)	
<u>Moulds and Yeasts</u>	Max. 1 × 10 <sup>2</sup> cfu/g (Analysis for Hygienic Chemists)	
<u>Coliforms</u>	Negative	(Analysis for Hygienic Chemists)

<u>Composition</u>	<u>Ingredients</u>	<u>Contents</u>
	Broccoli sprout extract	50 %
	<u>Maltodextrin</u>	50 %
	Total	100 %

2.46\*: 436 (Molecular Weight of Glucoraphanin) ÷ 177 (Molecular Weight of Sulforaphane)

**PRODUCT STANDARD**

PRODUCT NAME

**BROCCOLI SPROUT EXTRACT-PS1**

(Food)

This product is the powder extracted with aqueous ethanol from germinated broccoli (*Brassica oleracea* var. *italica*). It contains minimum 1.0% of sulforaphane and 2.5% of glucoraphanin.

This powder is water-soluble.

<u>Appearance</u>	Slight yellowish powder with slight unique aroma.	
<u>Glucoraphanin</u>	Min. 2.5 %	(Sulforaphane amount (GC Method) × 2.46*)
<u>Sulforaphane</u>	Min. 1.0 %	(GC Method)
<u>Loss on Drying</u>	Max. 10.0 %	(Analysis for Hygienic Chemists ,1g, 105°C, 2h)
<u>Purity Test</u>		
(1) Heavy Metals (as Pb)	Max. 10 ppm	(Sodium Sulfide Colorimetric Method)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 1 ppm	(Standard Methods of Analysis in Food Safety)
<u>Standard Plate Counts</u>	Max. 1 × 10 <sup>3</sup> cfu/g (Analysis for Hygienic Chemists)	
<u>Moulds and Yeasts</u>	Max. 1 × 10 <sup>2</sup> cfu/g (Analysis for Hygienic Chemists)	
<u>Coliforms</u>	Negative	(Analysis for Hygienic Chemists)

<u>Composition</u>	<u>Ingredients</u>	<u>Contents</u>
	Broccoli sprout extract	50 %
	<u>Maltodextrin</u>	<u>50 %</u>
	Total	100 %

2.46\*: 436 (Molecular Weight of Glucoraphanin) ÷ 177 (Molecular Weight of Sulforaphane)

**PRODUCT STANDARD**

PRODUCT NAME

**BROCCOLI SPROUT POWDER**

(Food)

This product is the powder of germinated broccoli (*Brassica oleracea* var. *italica*). It contains minimum 1.2% of sulforaphane and 3.0% of glucoraphanin..

<u>Appearance</u>	Slight yellowish powder with slight unique aroma	
<u>Glucoraphanin</u>	Min. 3.0 %	(Sulforaphane amount (GC Method) × 2.46*
<u>Sulforaphane</u>	Min. 1.2 %	(GC Method)
<u>Loss on Drying</u>	Max. 5.0 %	(Analysis for Hygienic Chemists ,1g, 105°C, 2h)
<u>Purity Test</u>		
(1) Heavy Metals (as Pb)	Max. 10 ppm	(The Japanese Standards of Food Additives)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 1 ppm	(Standard Methods of Analysis in Food Safety, The Third Method, Apparatus B)
<u>Standard Plate Counts</u>	Max. 1×10 <sup>3</sup> cfu/g	(Analysis for Hygienic Chemists)
<u>Moulds and Yeasts</u>	Max. 1×10 <sup>2</sup> cfu/g	(Analysis for Hygienic Chemists)
<u>Coliforms</u>	Negative	(Analysis for Hygienic Chemists)
<u>Composition</u>	<u>Ingredient</u>	<u>Content</u>
	Broccoli sprout	100 %

2.46\*: 436 (Molecular Weight of Glucoraphanin) ÷ 177 (Molecular Weight of Sulforaphane)

PRODUCT STANDARD

PRODUCT NAME

**BROCCOLI POWDER**

(Food)

This product is the powder of broccoli (*Brassica oleracea var. italica*). It contains minimum 200 ppm of sulforaphane and 500 ppm of glucoraphanin.

<u>Appearance</u>	Green powder with lightly unique smell	
<u>Glucoraphanin</u>	Min. 500 ppm	(Sulforaphane amount (GC Method) × 2.46*
<u>Sulforaphane</u>	Min. 200 ppm	(GC Method)
<u>Loss on Drying</u>	Max. 10.0 %	(Analysis for Hygienic Chemists, 1g, 105°C, 2h)

Purity Test

(1) Heavy Metals (as Pb)	Max. 10 ppm	(Sodium Sulfide Colorimetric Method)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 1 ppm	(Standard Methods of Analysis in Food Safety, The Third Method, Apparatus B)

Standard Plate Counts      Max. 1×10<sup>3</sup> cfu/g      (Analysis for Hygienic Chemists)

Moulds and Yeasts      Max. 1×10<sup>2</sup> cfu/g      (Analysis for Hygienic Chemists)

Coliforms      Negative      (Analysis for Hygienic Chemists)

<u>Composition</u>	<u>Ingredient</u>	<u>Content</u>
	Broccoli	100 %

2.46\*: 436 (Molecular Weight of Glucoraphanin) ÷ 177 (Molecular Weight of Sulforaphane)



**PRODUCT STANDARD**

## PRODUCT NAME

**BROCCOLI SPROUT EXTRACT-PC**

(Cosmetic)

This product is the powder extracted with aqueous ethanol from germinated broccoli (*Brassica oleracea* var. *italica*). It contains minimum 2.0% of sulforaphane and 5.0% of glucoraphanin.

This powder is water-soluble.

<u>Appearance</u>	Slight yellowish powder with slight unique aroma.	
<u>Glucoraphanin</u>	Min 5.0 %	(Sulforaphan amount (GC Method)×2.46*)
<u>Sulforaphane</u>	Min. 2.0 %	(GC Method)
<u>Loss on Drying</u>	Max. 10.0 %	(Analysis for Hygienic Chemists, 1g, 105°C, 2h)
<u>Purity Test</u>		
(1) Heavy Metals (as Pb)	Max. 10 ppm	(The Second method of The Japanese Standards of Quasi-Drug Ingredients)
(2) Arsenic (as As <sub>2</sub> O <sub>3</sub> )	Max. 1 ppm	(The Third method of The Japanese Standards of Quasi-Drug Ingredients)
<u>Standard Plate Counts</u>	Max. 1 × 10 <sup>2</sup> cfu/g	(Analysis for Hygienic Chemists)
<u>Moulds and Yeasts</u>	Max. 1 × 10 <sup>2</sup> cfu/g	(Analysis for Hygienic Chemists)
<u>Coliforms</u>	Negative	(Analysis for Hygienic Chemists)

<u>Composition</u>	<u>Ingredients</u>	<u>Contents</u>
	Brassica Oleracea Italica (Broccoli) sprout extract	50 %
	<u>Maltodextrin</u>	<u>50 %</u>
	Total	100 %



**ORYZA OIL & FAT CHEMICAL CO., LTD.** striving for the development of the new functional food materials to promote health and general well-being.

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