1. What is ‘SEANOL’?

The powers of seaweeds have been known for thousands of years for their ability to prolong life, prevent disease, and impart beauty and health. The product, SEANOL (phlorotannin), is extracted from freshly harvested brown seaweed of “Ecklonia cava” from the clean Jeju island coast of Republic of Korea. This extract contains the life essential properties of marine phlorotannin, fucodian, alginates, laminarans, organic minerals and etc. The active component of SEANOL is marine phlorotannins called “PPC” which has unique physical and biological properties. Its unique structure which cannot be found in terrestrial plants distinguishes itself from well-known tea and grape phlorotannin in addition to sharing general characteristics such as antioxidant activities. Notable biological activities of PPC which are hardly found in other types of phlorotannin are its strong in vitro activities related with vasodilatation and erectile function.

• Marine Phlorotannin

Marine algae, like other photosynthesizing plants, are exposed to a combination of light and oxygen that leads to the formation of free radicals and other strong oxidizing agents. However the absence of oxidative damage in the structural components(polyunsaturated fatty acids) of seaweeds and their stability to oxidation during storage suggest that their cells have protective antioxidative defence systems. The significant class of the phlorotannin compounds from marine plants is “Phlorotannins” or algal phlorotannin compounds. They have a wide range of molecular sizes (400 to 400,000 Da) and can occur in variable concentrations (0.5-20% of the dry weight) in brown algae. Phlorotannins are commonly believed to have defensive or protective functions, e.g., against herbivores, bacteria, fungi, fouling organisms and UV-B radiation, to function in wound healing processes, or in cell-wall construction.

The potential biological effects associated with certain algal components have prompted increased use of seaweed in manufacturing as raw or semi-processed products in the food industry. Of the dietary phytochemicals, it has been suggested that phlorotannin prevent oxidative damage to important biological membranes and to plant foods.

Powerful antioxidant activity was recently discovered in certain members of brown seaweeds and this finding has resulted in an enormous increase in research on sea vegetables’ metabolites and their activity against free radicals. The fruits of this research have shown that certain phaeophytes contain highly active phlorotannin called polyphloroglucinols that possess free radicals neutralizing activity. Many algal species contain polyphloroglucinol phenolics (phlorotannins) and in several cases the antioxidant activity of algae could be due to these compounds.(J. Sci., Food., Agric. 81, 530-534, 2001)
Recently Japanese scientists discovered that phloroglucinol compounds possess 5-lipoxigenase activity and inhibit the action of $\alpha_2$-macroglobulin and $\alpha_2$-plasmin which play a role of the suppressing control in the fibrinolytic enzyme system. (Chemistry Letters, 739-742, 1985)$^2$

SEANOL contains various kinds of polyphloroglucinol phenolics (phlorotannins) like Eckol, Dieckol, Bieckol, Phlorotannin A, Phlorofurofukoeckol, (Figure. 1) etc. This substance is founded in phaeophytes commonly and hold various physiological activities such as antioxidant activity (Fisheries Science 62(6), 923-926, 1996)$^3$ and good thrombolytic activities.

![Chemical structures of phloroglucinol compounds](image)

**Figure1. Isolated polyphloroglucinol compounds in VNP001**

- Alginate:

  Alginate is a naturally occurring biopolymer that is finding increasing applications in the biotechnology industry. Alginate has been used successfully for many years in the food and beverage industry as a thickening agent, a gelling agent and a colloidal stabilizer.

  The major matrix component of brown seaweeds is an alginate consisting of alternating sequences of (1,4)-$\beta$-D-mannuronic acid, its C5 epimer $\alpha$-(1,4)-L-guluronic acid and 20-30 units of the uronic acids mentioned.

  Alginate also has several unique biological activities that lowers blood pressure; remove cholesterol in digestive system; acts as a natural absorbent of radioactive elements, heavy metals and free radicals. (Nutrition Research, Vol.20, No. 4, 585-598, 2000)$^4$
• Fucoidan:

Fucoidan is one of the main anionic polysaccharides of marine algae and is extracted from common brown algae. Fucoidan is primarily composed of (1,2)-α-L-fucose-4-sulphate with branching or a sulphate ester group on C3, and contain very small quantities of D-xylose, D-galactose, D-mannose and uronic acids. Several biological activities have been attributed to the fucoidans such as anticoagulant, antithrombotic, antiinflammatory, antitumoral, contraceptive, and antiviral activities.(Thrombosis Research, 99, 623–634, 2000)5

• Laminarin:

Laminarins, reserve polysaccharides found in brown algae, are composed of (1,3)-β-D-glucose with some (1,6)-linkages and in which some of the reducing ends are replaced by mannitol.

• Minerals:

Seaweed mineral content is higher than that of land plants and animal products. In most land vegetables, ash content ranges from 5 to 10 g/100 g dry weight, e.g. potato 10.4, carrot 7.1, and tomato 7.1. Sweet corn has a lower content (2.6%), while brown seaweeds have a exceptionally high mineral content (20-35%) compared with land plants. Thus edible marine seaweeds may be an important source of minerals, since some of these trace elements are lacking or very minor in land vegetables.(Food Chemistry, 79, 23-26, 2002)6
2. Production of “SEANOL”

1) PLANT MATERIALS

The brown seaweed *Ecklonia cava* was collected off in Jeju island of Korea. The freshly collected whole seaweed was washed with water and then air-dried, and milled.

2) EXTRACTION

SEANOL is extracted from *Ecklonia cava* by ethanol solvent extraction. A batch process is utilized for the extraction of SEANOL from *Ecklonia cava*. The extraction process is optimized when the ratio of 25% ethanol to *Ecklonia cava* is 12kg :1kg, the slurry is constantly stirred, and the temperature of the ethanol is greater than 50°C.

200kg of dried *Ecklonia cava* is added manually to the agitated extraction vessel. Once the *Ecklonia cava* has been transferred, the vessel is sealed and 2400 kg of 25% ethanol is charged. The ethanol/Ecklonia cava slurry is heated to reach a temperature of 50°C and the temperature of the vessel is maintained at 50°C for an additional 1 hour. After the extraction phase is completed, slurry is filtered.

3) CONCENTRATION & POWDERING

The filtrate is concentrated in vaccum to 15% extracted solution and the ethanol vapour is condensed to a temperature of 5°C by cooling liquid through the tube side of the heat exchanger. The recovered solvent is stored for future usage and concentrated solution is dried by spray-dried method or freeze-dried method.
3. Physical properties of ‘SEANOL’

‘SEANOL’ is a dietary ingredient with the following physical properties, all of which make it very easy to handle.
(1) Typical taste and flavor of seaweed
(2) Dark brown powder
(3) Stable at 40°C for 180 days
(4) Relatively heat-stable under high temperature conditions

<table>
<thead>
<tr>
<th>Content</th>
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<tr>
<td>Phlorotannin</td>
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<tr>
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<tr>
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<td>20</td>
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<tr>
<td>moisture</td>
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4. The safety of ‘ecklonia cava’, the Raw material of “SEANOL”, as a food

1) BIOGEOGRAPHY OF ECKLONIA CAVA

The brown algae *ecklonia cava* is large brown seaweed belonging to the family Laminariaceae of the order Laminariales (Kelp). This seaweed forms the main canopy near the low-water mark to a depth of 5-10m.

The kelp *Ecklonia cava* has a pinnate blade, with a stipe which can attain lengths of 1.5 m or more. These sea plants form dominant subtidal, seaweed forests which can be found in far east Asia (central and southern Japan and Jeju island of Republic of Korea). New blades and juvenile plants of *Ecklonia cava* usually begin to appear in December. From this time, the length and weight of plants increases gradually until both reach a peak in August. Some plants initiate sporeling formation during this period and become fully reproductive in December; mature blades start to decay thereafter. However, secondary plants are also found to grow near to the base of the stalk. The kelp beds are found to persist on rocky shores throughout the year although the resident biomass may change seasonally.

2) HISTORY OF USE

Seaweeds are cooked and dried for common food in Asia since ancient times. More recently marine algae have utilized in far east Asia as raw materials in the manufacture of many seaweed food products, such as jam, cheese, wine, tea, soup and noodles.

There are many different type of brown seaweeds that have been eaten in far east asia such as *Undaria pinnatifida*, *Eisenia bicyclis*, *Laminaria japonica*, *Hizikia fusiforme*, *Ecklonia cava*, etc. The *Ecklonia cava* called as “Kamtae” in Korea and “Kajime” in Japan is mainly used for raw material to produce alginate but in southern Japan and Jeju island of Republic of Korea has been eaten directly since ancient times. The evidence of edible use in Japan is founded in Wajima city. Dried Kajime(Japanese character : かじめ) is sold in traditional seafood market (http://www.e-shokuzai.co.jp/details/konbu/0112072022.html), (http://www.wajima.gr.jp/kojima/asaitimise.htm) and their cooking method is well described (http://www.syamojiya.co.jp/recipe/kajime.htm). Powdered Kajime is also used as a food additives for giving a brown color to ‘mannan’ which is traditional food in Japan and Korea(http://www.marine-science.co.jp/esyohin/eseaweed.html) and its usage is described in the web(http://www5a.biglobe.ne.jp/~smorita/page/content1/kajime.htm).

The other edible usage of ecklonia cava is described in Japanese patent (Japanese patent number: 55-144871).

In Korea, ‘Kamtae’ is mainly exported to Japan for food additive and the market size is estimated
about 10 million dollars and more detail description is founded in (http://goodsinfo.ejeju.net/itemview.asp?LanguageType=2&ID=221)\textsuperscript{13}. The edible usage of ecklonia cava in Korea is described at the published paper(On the components of edible marine algae : J. Korean Agricultural Chemistry Society Vol. 14, No. 3, Dec. 1971)\textsuperscript{14} and Korea Food & Drug Administration is officially approved to use of *Ecklonia cava* for food\textsuperscript{15}.
5. Toxicity study following 4-week oral administration of ‘SEANOL’ in rat

Composition of Test Material (SEANOL)

SEANOL was repeatedly administered orally with doses of 2000, 667 and 222 mg/kg/group for 4 weeks to evaluate toxicity in rats (10 SD rats, male and female each). Following is the summary of results.

(1) No mortality was observed in response to the test article.
(2) General clinical sign following test article administration were not observed.
(3) No change in body weight was observed in both male and female groups.
(4) Test article per se did not induce the difference in the amount of food and water consumption in all groups.
(5) In all groups, there were no abnormal signs in ophthalmoscopic test.
(6) In all groups, urinalysis did not detect any symptoms relevant to test article toxicity.
(7) In all groups, no toxic effect was observed with hematological tests.
(8) In all groups, no toxic effect was observed with serum biochemistry tests.
(9) No specific abnormality was found in autopsy.
(10) No significant difference of organ weighs was found between groups.
(11) No toxic effect was found in pathological anatomy examination following test article administration.

According to the results, it is considered that no toxic effect was found in rats following 4-week repeated oral administration of SEANOL. Also, it is considered that NOAEL is 2000 mg/kg/day and above.