# Bio-function Summary of Marine Oligosaccharides

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**Abstract**

Marine ecosystems can be considered as a rather unexplored source of drugs as well as a surprising reservoir of bioactive oligosaccharide. Marine oligosaccharides could be obtained by hydrolysis of polysaccharide existing in a variety of biomass sources or synthesized by simple oligosaccharides. In recent years, marine oligosaccharides which degraded by polysaccharides are attracting increasing interest in developing potential drugs for chronic diseases. Significant efforts have been made to find and confirm their bioactivity such as anti-tumor, antimicrobial, antioxidant, anti-inflammatory and so on. In this review, the mechanisms of bio-function eliciting kinds of applications of different marine oligosaccharides are summarized.

**Keywords:** Marine drugs, Oligosaccharide, Polysaccharide, Bioactivity

## 1. Introduction

With the increasing understanding of bio-functional properties of marine foods, the utilization of these sources has been improved significantly. During the past decades, the development of new technologies to isolate and identify bioactive materials has progressed. Initial identification of bioactive materials from marine sources paved the way for utilization of various oligosaccharides.

Oligosaccharides are low molecular weight (LMw) carbohydrates in nature, between simple sugars and polysaccharides. They can be obtained by direct extraction from natural sources, or chemical processes such as polysaccharides hydrolyzation or chemical synthesis from disaccharides. The low-viscosity and high-solvency of oligosaccharide at neutral pH especially enhanced their potential use as drugs. Moreover, their biodegradable, non-toxic and non-allergenic properties attracted scientific interest.

<Table 1>

Recent advances show insight into the health benefits of oligosaccharides including enhanced immunity, anticoagulation and anti-tumor properties (Yuan, 2006, pp.228-234; Volpi, 2009, pp.356-367; Zhao, 2006, pp.8779-8787), lower blood cholesterol, blood pressure and blood glucose (Fitton, 2008, p.345; Hou, 2009, pp.46-50), protection against microbial infection and peroxide (Sudarshan, 1992, pp.257-272; Alekshun, 2004, pp.483-489; Xie, 2001, pp.1699-1701). Besides, oligosaccharides play important roles in plant systems, including fertilization, embryogenesis, neuronal development, cell proliferation and metastasis (Pospieszny, 1991, pp.63-68). There has been a growing interest in the health-promoting and disease-preventing role of certain foods over their usual nutritional value. Especially, researches on oligosaccharides in foods and nutrition areas have emphasized on their ability to improve food quality and human health. In this paper, many aspects of mechanisms about how oligosaccharides performed *in vitro* are briefly discussed on three levels, namely molecular, tissue and organ. A recent and somewhat comprehensive list of bioactivities is also provided.

## 2. Effects on immuno-enhancing

The benefits of disease treatments based on the host's own defense system had been verified by clinical trials (Ehrke, 2003, pp.1105-1119; Yuan, 2006, pp.228-234). In the theory of immunology and immunotherapy, the state of immune system plays an essential role in mediating the occurrence or development of disease. Therefore, drug researches in immunopharmacology and oncotherapy are mainly focused on the special chemicals that could potentiate the function of immune system.

### *2.1 Humoural immunity*

Early studies demonstrated that chitosan and COs could inhibit malignant disease by exerting immuno-enhancing effects. Some studies have suggested that the observed bioactivity are not only caused by directly killing malignant cells but also caused by increasing the lymphokines secretion (Yuan, 2006, pp.228-234). Significant resistant effect of hexamer COs on the metastasis of lung carcinoma has been proved *in vivo* experiments on mice. The

mean Mw of COs ranging from 1.5 to 5.5kDa could effectively inhibit the growth of S180 or Uterine cervix carcinoma No. 14 (U14) in tumor cell-bearing mice (Kim, 2008, p.183). Compared to other bio-molecules, COs could exert their immunocompetence via oral administration. That is similar to intraperitoneal injection. Qin (2002, pp.111-117) demonstrated that water-soluble COs mixture prepared with tetramer and pentamer could inhibit the growth of S180 tumor cells in mice after oral or intraperitoneal administration.

Hu (2006, pp.646-650) obtained the gelling and thickening CAOs. These CAOs significantly increased the weight of immune organs such as thymus, which suggests that the CAOs could initiate organ-mediated defense reactions. Results showed that CAOs significantly influenced transplantable tumors, macrophage phagocytosis, quantitative hemolysis of sheep red blood cells (QHS), lymphocyte proliferation, the activity of natural killer cells, the production of IL-2 and TNF-α in S180-bearing mice (Yuan, 2006, pp.228-234). All these experimental indexes demonstrated that CAOs exert defense effects through the regulation of the immune system.

*2.2 Cellular immunity*

The immunomodulatory effect of LO preliminarilly used on mice cDNA microarray suggested that LO and LP1 can be utilized to develop new immuno-potentiating substances or functional alternative medicines. Li and his/her colleagues obtained the JG3 and elucidated its detailed immunomodulatory mechanisms on cell migration using a Chinese hamster ovary cell line (Li, 2009, pp.1033-1038). JG3 could resist to the activity of heparanase, resulting to a marked inhibition of syndecan clustering both *in vitro* and *in vivo*. In addition, JG3 abolished heparanase-driven invasion, inhibited the release of heparan sulfate and repressed subsequent angiogenesis. Thus, JG3 acted as a mimetic substrate and a competitive inhibitor of heparan sulfate in inhibiting major heparanase. All the above functions identified JG3 as a promising new compound for potential drugs (Zhao, 2006, pp.8779-8787).

Since Sugawara (1982, pp.162-171) found that sulfated carrageenan act more effective on human T lymphocyte than carrageenan. CAOs (Mw=1726) showed particular effects on immunological regulation, especially on the phagocytosis ratio and phagocytosis index of macrophage and lysozyme activity of the serum, which might be beneficial for the defense system (Wang, 2004, pp.333-340). Wang suggested that the activity of CAOs differs between *in vitro* and *in vivo*. Tumor formation was markedly inhibited when mouse treated with this 1726 Mw product at a dose of 100 mg $kg^{-1}$. Alginate with average Mw of 230kD can enhance phagocytic activity of macrophages (Fujihara, 1992, pp.343-347; Iizima-Mizui, 1985, pp.59-71). More than 50 alginate lyases with various substrate specificities have been isolated from algae, marine invertebrates and a wide range of micro-organisms (Masuda, 2006, pp.135-144; Iwasaki, 2000, pp.1067-1070). Although the bioactivity mechanisms of alginates and their hydrolysates are still unclear today, it is suggested that the molecular size and molecular conformational properties are strongly related to their bioactivity. While AOs enhanced the growth of human endothelial cells and keratinocyes (Maeda, 1999, Patent No.: US 5866677), AO with the specific Mw have been shown to stimulate the secretion of cytotoxic cytokines from human macrophages (Kurachi, 2005, pp.199-203).

**3. Anti-tumor**

Tumor is one of the most dangerous threats against people's health and life. Although many drugs have been used in treating tumor, the side effect of drugs exceeds the tolerance of many patients. In recent years, more and more researchers have come to the realization that marine organisms hold immense potential as a source of novel molecules and new anticancer agents. Among all those agents, researchers pay more attention on marine oligosaccharides, which showed fewer side effects. These marine oligosaccharides could not only directly kill the tumor cell, but also impair the tumor's nutritional support system by targeting the tumor blood vessel network, activating the immune system and so on.

*3.1 Killing tumor cells*

Some specific oligosaccharides such as sialylLewisX or sialylLewisA on the tumor cell surface can interact with other marine oligosaccharides, resulting to death of the tumor cell. This anti-tumor activity has been proved by mild hydrochloric acid hydrolysis of Carrageenan sulfated polysaccharides from *Kappaphycus* striatum (Yuan, 2005, pp.7-13). Both Carrageenan sulfated polysaccharides and oligosaccharide fractions (F1) were investigated in three human neoplastic cell lines (KB, BGC, and Hela). The bioassay results showed that F1 exhibited relatively higher anti-tumor activity against the three cancer cells than polysaccharides.

*3.2 Inhibiting the growth of vessel*

It is now well established that solid tumor growth is critically dependent on the growth of new vessels from pre-existing blood vessels surrounding the tumor, a process named angiogenesis (Douglas, 1996, pp.353-364; Zetter, 1998, pp.407-424). A variety of potential targeting drugs shows high toxicity. However, it has been indeed

identified in preclinical study that the combined treatment of anti-tumor drugs with oligosaccharides compromising the tumor vasculature can decrease the toxicity effects.

Ma (2008, p.e3774) was devoted to study the Marine-Derived oligosaccharide sulfate (MdOS), which show high anti-angiogenic activity both *in vitro* and *in vivo*. Tube formation assay, rat aortic ring method and chicken chorioallantoic membrane (CAM) assay made it a promising agent for future evaluation in PTK-associated cancer therapy. MdOS not only exhibited anti-angiogenic activity in a PTK-dependent manner, but also acted as a broad-spectrum PTK inhibitor, MdOS inhibited HER2, EGFR, VEGFR, PDGFR, c-Kit, FGFR1 and c-Src, with little impact on FGFR2 at an enzymatic level; meanwhile, it suppressed HER2, EGFR, VEGFR2 and downstream molecules of Erk1/2 at cellular settings. Deters (2008, pp.197-204) investigated the effects of COs (DP2, DP3, DP4, DP5 and DP7) on epithelial cells, tissue and on the induction of physiological processes in two different test systems. Results obtained from both test systems indicated that the COs increased the barrier function of skin cells by induction of cell differentiation and also induced secretion of mucin from intestinal epithelial cells. One LMw, highly sulfated λ-CAOs with anti-angiogenesis were obtained from depolymerization of carrageenan. Significant inhibition of vessel growth was observed in chicken CAM and human umbilical vein endothelial cell. The λ-CAO as a potential angiogenesis inhibitor down-regulates intracellular matrix metalloprptrinases (MMP-2) expression on endothelial cells (Chen, 2007, pp.6910-6917). Several anti-tumor oligosaccharides (GSO) include sulfated disaccharide and neoagarotetraose have been discovered in the Delesseriaceae (*Ceramiales, Rhodophyta*) (Osumi, 2002, pp.1441-1444). These oligosaccharides were prepared by spray dried and proved the physiological activity by reducing the number of revertants colonies of *Salmonella typhimurim* TA98 or TA100 and mildly inhibiting angiogenesis on the CAM. GSO was assumed to have a desmutagenic activity and no harmful side effects such as blood coagulation of fibrinoysis. GSO with high safety owed various physiological activity, which is expected to apply in future food and medical supplies.

Partish (1999, pp.3433-3441) found that sulfated oligosaccharides could reduce tumor metastasis by inhibiting heparanase activity and/or by simultaneously inhibiting angiogenesis via blocking angiogenic growth factor action. COs (DP5 to DP8) prevented the growth of tumor cells and acted as inhibitors of angiogenesis *in vitro* (Harish Prashenth, 2007, pp.117-131). *In vivo*, COs (DP about 19) increased IL-12 and interferon-α levels, while levels of IL-1 and TNF were reduced (Kim, 2006, pp.439-446). Enoki (2008, Patent No.: US 2009/0099101 A1) found the carcinostatic activity and inhibitory activities on gastric cancer, liver cancer, bladder cancer and other tumor cells.

## 4. Anti-inflammatory

Research showed that oligosaccharides influence cell immunity, humoral immunity and inflammation process in autoimmune diseases in many aspects and have reliable curative effects on autoimmune diseases such as rheumatoid arthritis. It can also be used as a ligand to identify the position of cells.

### *4.1 Suppression of inflammatory cytokines*

The research that has led to the discovery of physiological actions related to AOs, such as an anti-inflammation activity by the suppression of prostaglandin E2 (PGE2), TNF-α, IL-1 and IL-6, has been carried out by Takara Shuzo Co, Ltd (Boon, 2002, pp.1432-1459). The suppression of such inflammatory cytokines not only reduced the production of heme oxygenase (HO-1), but also exerted preventive and therapeutic effects *in vivo* models of chronic inflammation. Enoki (2002, Patent No.: US 2003/0158250 A1) identified activities of AOs including: (1) suppression of the overproduction of nitric monoxide (NO), which is involved in the enhancement of the inflammatory response; (2) enhancement of the production of carbon monoxide, which suppresses the inflammatory response; (3) suppression of the production of TNF-α, which is deeply involved in the worsening of rheumatism. It is thus concluded that AOs, produced by hydrolyzing agar, is useful as a new dietary ingredient that improves joint function or as a new medicine against chronic rheumatoid arthritis.

### *4.2 Increasing epithelial cells internalization*

Since bilirubin has attracted attention as a physiological compound with antioxidant properties, the mechanism has been studied involving the elimination of activated oxygen species and prevention of lipid per-oxidation (Neuzil, 1993, pp.281-284). Thus, at inflammatory sites it may remove activated oxygen species and prevent worsening of the lesions (Boon, 2002, pp.1432-1459). Wiring of the class II PapG adhesion at Escherichia coli (*E. coli*) to the carbohydrate moiety of globoseries glycolipids in the human kidney is a key step in the development of pyelonephritis and a form of urinary tract infection. An assay for the quantification of the binding of the class II PapG to oligosaccharides has been developed, which indicated that the position of oligosaccharides appeared to be the most promising inhibitors against pyelonephritis (Larsson, 2003, pp.2255-2261).

Yoon (2004, pp.611-623) summarized several theories, which were proposed to explain the failure of lung mucosal defense and the prevalence of *Pseudomonas aeruginosa* (*P. aeruginsa*) in the cystic fibrosis (CF) lung. Based on the hypothesis that airway infection is mediated mainly by the direct binding of bacteria to the surface of airway epithelial cells, the cystic fibrosis transmembrane conductance regulator (CFTR) present on the apical surface of lung epithelia was proposed to act as a pattern-recognition receptor, while LPS-core oligosaccharide was identified as the ligand for internalization by airway epithelial cells (Currie, 2003, pp.671-680).

## 5. Antioxidant

Several mechanisms including hydroxyl radical scavenging, superoxide radical scavenging, erythrocyte hemolysis inhibiting, metal chelating activities, and anti-lipid per-oxidation have been studied to identify the antioxidant activity of oligosaccharides of marine origin (Kochkina, 2000a, pp.208-211).

### *5.1 Radical scavenging activity*

Mouy (2007, pp.646-654) studied the antioxidant activity of three marine oligosaccharides including AOs, COs, and FOs and drew the conclusion that they exhibited different activities using various *in vitro* assays. AOs had the highest scavenging hydroxyl radical activity among the three marine oligosaccharides, while COs and FOs indicated good chelation. COs had the highest scavenging superoxide radical and inhibiting erythrocyte hemolysis activity and it also showed significantly antioxidant activity in per-oxidation of anti-lipid. Later, Wang (2004, pp.333-340) proved the anti-oxidative activity of three products of AOs (AOs-1, AOs-2, AOs-3) in scavenging hydroxyl free radical, superoxide anion radical and inhibiting lipid per-oxidation, which indicated that the anti-oxidative activity was closely related to the molecular mass as well as to the substitute groups. *In vivo*, Chen (2005, pp.103-111; 2006, pp.31-43) carried out a study in order to evaluate the antioxidant activity of AOs with different DPs and they established a relationship between activity and DPs. Different levels of antioxidant activities of AOs with various DPs were observed, and their scavenging ROS capability was associated with the improvement of the cell viability.

Another derivative of AOs is the AOSC, which could inhibit the toxicity induced by amyloid beta protein in both rat cortical cells and human neuroblastoma cells via the mechanism of inhibition of apoptosis, the reduction of intracellular $Ca^{2+}$ and the generation of ROS (Hu, 2004, pp.248-255). Later, Wang (2007a, pp.96-102) observed the effect of AOSC on SH-SY5Y cell line and demonstrated that AOSC not only inhibited the reactive phenotype of astrocytes and blocked cellular oxidative stress, but also reduced the production of TNF-α and IL-6 and prevented the influx of $Ca^{2+}$. Therefore, all these results may identify AOSC as a potentially therapeutic compound for Alzheimer's disease (AD).

The COs or AOs could rescue the loss of cell viability and prevent cell apoptosis that induced by ROS. Since COs or agarohexaose (one of AOs) could effectively protect human embryonic hepatocytes (L02 cells) cells against oxidative stress, they could also bring the benefit of liver-protection (Chen, 2007, pp.44-47). Xu (2009, pp.1-7) investigated the protective effect of COs against $H_2O_2$-induced oxidative stress on L02 and its scavenging activity against the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical *in vitro*. Je (2004, pp.381-387) also investigated scavenging activities of COs against DPPH, hydroxyl, superoxide and carbon-centered radicals using electron spin resonance (ESR) spin-trapping technique. Yan (2006, pp.178-184) studied the protective effect of COs, d-glucosamine ($GlcNH_2$) and N-acetyl-d-glucosamine (GlcNAc) on $CCl_4$-induced hepatotoxicity. The mechanisms illustrated that the antioxidant defensive system in the body was strengthened to counteract the $CCL_4$-induced damage. Results suggested that pretreatment with COs, $GlcNH_2$ and GlcNAc can efficiently protect mice against $CCl_4$-induced toxicity.

Ngo also have obtained series of COs (NA-COs) from crab chitin and demonstrated their reducing power and scavenging activity (Ngo, 2010, pp.161-177). The radical-scavenging activity of NA-COs is increasing in a dose-dependent manner, showing different $IC_{50}$ values for different groups (Xie, 2001, pp.1699-1701). The mechanisms of ROS damage on organisms mainly relate to structural change of DNA, especially mtDNA such as inducing base deletion, base modification, single and double-strand breakage and inter-stranding DNA cross links in nuclear DNA (Yuan, 2009, pp.1339-1345). Furthermore, high occurrence of oxidized protein markers has been reported in numerous diseases (Marcinkiewicz, 2003, pp.471-479). This may happen because the residues of lysine, proline, arginine and histidine in membrane proteins are highly susceptible to oxidative attack by ROS and leads to accumulation of their carbonyl moieties. This process may play an important role in maintaining cellular integrity and normal biological functions. The inhibitory effect of NA-COs on oxidation cell membrane proteins was demonstrated by Ngo (2009, pp.188-198) though exposing mouse macrophage membranes to hydroxyl radicals. Betting (2009, pp.250-259) illustrated that NA-COs exhibited the inhibitory effect on the oxidative

damage of DNA from human lymphoma U937, as well as the radical-scavenging effect in human fibrosarcoma cells (HT1080).

*5.2 Regulation of enzyme level*

Yoon (2009, pp.869-872) studied the inhibitory activities against cholinesterase (acetylcholinesterase (AChE) and butyrylcholinesterase) of COs with different substitution groups, including aminoethyl (AE), dimethylaminoethyl (DMEM) and diethylaminoethyl (DEAE). These derivatives perform potent activities against AChE and show no effect on butyrylcholinesterase. AE-COs was shown to be non-competitive while DMAE-COs and DEAE-COs were shown to be competitive against AChE, which could exert some effort on both central and autonomic nervous system (e.g. preventing AD). In addition, COs with different Mw and degrees of deacetylation could suppress the level of AChE protein expression and AChE activity induced by Ab25–35 in PC12 cells (Lee, 2009, pp.860-862). Besides, Wang (2007, pp.96-102) illustrated the importance of oligosaccharide structure and charge on influencing Gial derived neurotrophic factor (GDNF) activity as well as the potential use of oligosaccharides in treating neurological disorders such as Parkinson's disease.

Ngo (2009, pp.188-198) investigated the antioxidant activity of COs on pancreatic islet cells in diabetic rats induced by streptozotocin, which they showed that COs drastically affect the activity of superoxide dismutase (SOD) and diminute the content of malondialdehyde. According to the morphological investigation in the pancreatic cells, COs exerted effects by reduction of islets, loss of pancreatic cells and nuclear pyknosis of pancreatic cells. It has been further suggested that chitosan inhibits the uptake of dietary lipids by increasing the thickness of the intestinal lumen boundary layer (Furda, 1990, pp.67-82). A proposal, which was supported by numerous animal experiments identify that COs possess various biological activities and can be used in diabetes treatment (Yuan, 2009, pp.1339-1345).

Chen (2007, pp.44-47) demonstrated the increasing activities of CAO on SOD and catalase, suggesting that CAO was effective in promoting the anti-oxidation ability and eliminating danger from free radicals. Wang (2004, pp.333-340) found that the oral administration of CAOs was advantageous to promote the activity of antioxidant enzymes, which might play an important role on antitumor mechanism.

NA-COS owed a broad inhibitory extent like affecting myeloperoxidase (MOP), membrane protein oxidation and DNA protection activity; scavenging cellular radical and mediating induction of intracellular glutathione (GSH) level (Wartman, 2008, pp.469-470). MPO, the most abundant protein in neutrophils, promotes the conversion of $H_2O_2$ and chloride into HClO, which is the most potent oxidant acting against pathogens and is directly related to the release of ROS (Sosa, 1991, pp.489-496). It is the ROS that oxidize thiols in protein, react with unsaturated fatty acids of membrane lipid and destabilize the integrity of the cell membrane. Ngo (2008, pp.228-232) identified NA-COs as a potent inhibitor, which can inhibit MPO activity and control oxidation of cellular bio-molecules in neutrophils (Park, 2004, pp.17-22; Rajapakse, 2005, pp.562-569). Lee investigated angiotensin converting enzyme (ACE) inhibitory activity of hetero-COs and showed that the activity was depended on the degree of deacetylation. Inhibition of ACE may act an important role in anti-atherosclerosis and antihypertensive (Lee, 2004, pp.41-47; Sang-Pill, 1998, pp.1476-1479).

It is well-known that increased GSH level protects cell against death either by elimination of free radicals or by conjugation with toxicants (Sánchez-Reus, 2005, pp.48-56). Ngo (2009, pp.188-198) studied the detect time and concentrations of NA-COs at incubation, which showed that the intracellular GSH level was different with time and dose.

**6. Anticoagulation**

Glycosaminoglycans were extracted from the dry tissue of marine clam *Scapharca inaequivalvis*, composed of dermatan sulfate (DS) and heparin sulfate. DS was further depolymerized and separated into oligosaccharides. The most prominent generated oligosaccharides comprised the repeating unit Hex-GalNAc$SO_4$, thus confirming the results obtained by disaccharide analysis. DS was calculated to possess a high heparin cofactor II activity fairly similar to that of several DS samples purified from porcine and bovine tissues (Volpi, 2009, pp.356-367).

Pomin (2005, pp.1376-1385) employed a nonspecific approach to cleave the linear sulfated fucan and got the LMw derivatives, which were employed to determine the requirement for interaction of this polysaccharide with heparin cofactor II and achieve complete thrombin inhibition. Fucan has the similar structural and anionic characteristics with heparin. Fukuta (2008, pp.499-503) illustrated that fucan and fucan-derived oligosaccharides have similar ability to stimulate production of hepatocyte growth factor (HGF) as heparin and heparin-derived oligosaccharides. Via mechanisms of increasing HGF by fucan or its oligosaccharide derivates, various injuries and disease can be avoided. The fucan chains of marine origin consist of repeating oligosaccharide units differing

in the number and arrangement of sulfate groups. Their anticoagulant activity has been shown to depend not only on their Mw, but also on the distribution of sulfate groups in the repeating units (Parish, 1999, pp.3433-3441). Sulfated COs was reported to possess anticoagulant activity *in vitro* (Park, 2004, pp.529-533). The agglutination of red blood cells by chitosan is dependent on the molecular size and other physical characters such as the ionic attraction between negatively charged red blood cell membranes and positively charged groups in chitosan (Kim, 2005, pp.357-368).

## 7. Antimicrobial and Anti-infection

Antibiotics used in the treatment of infectious diseases have provided an immeasurable benefit to human health, but an alarming increase in resistance of bacteria caused great concern about the future of antimicrobial therapy. The dwindling options of anti-infective treatments have raised renewed interest in research and development of novel strategies to prevent infection. The pharmaceutical industry has responded by exploiting natural resources to obtain drugs with expanded spectrums of antibacterial activity during past decades.

### *7.1 Antibacterial*

The antibacterial activity of COs has been researched deeply. Jeon (2000, pp.133-141) obtained the enzymatic production of COs with a high DP. Oligosaccharides which were obtained from the reactor system showed antibacterial activity and completely inhibited the growth of *E. coli* at 0.5% concentration. Eaton (2008, pp.1128-1134) suggested that the antimicrobial effects are strongly dependent on the target micro-organism (e.g. Gram-negative or Gram-positive bacterium) and the Mw of oligosaccharides. The LMw COs had only a short-lived effect on the cell viability, but even so the cells were weakened when treated with COs (Wang, 2007, pp.917-920).

The different structures of COs that contribute to the antibacterial activity have also been studied. COs-GTMAC was obtained by Kim, which exhibited 80% growth inhibition against *Streptococcus mutans*, whereas the COs showed 10% growth inhibition (Kim, 2003, pp.23-27). It was found that antimicrobial activity of the COs could be tremendously enhanced by the introduction of quaternary ammonium function. CSO-OA nanoparticles which were prepared by the method of sonication have been identified as good antibacterial activators and inhibited the bleeding caused by *E. coli* and *Staphylococcus aureus* infection (Huang, 2009, pp.321-327). Since CFTR was proposed to act as a pattern-recognition receptor on the surface of lung epithelia, Pier (1996, pp.64-67) identified LPS-core oligosaccharide as the ligand for internalization by airway epithelial cells. The degree of deacetylation and the number of amino groups have been proven as major factors in antibacterial activity (Moon, 2007, pp.989-998; Kim, 2005, pp.357-368).

Oligosaccharides can alter permeability characteristics of microbial cell membrane and further prevent the entry of materials or cause leakage of cell constituents that finally leads to bacterial death (Sudarshan, 1992, pp.257-272). Choi (2001, pp.553-557) carried out a study and exhibited the morphological change of bacterial pathogen *Aggregatibacter actinomycetemcomitans* (*A. actinomycetemcomitans*) using scanning electron microscope and transmission electron microscopy images. The exposure of *A. actinomycetemcomitans* to the COs resulted in the disruption of cell membranes and that could be considered for the treatment of periodontal diseases associated with this pathogen.

The similar results were observed by Eaton (2008, pp.1128-1134), which have shown that COs had antimicrobial activity and were able to stimulate immune response. Dou (2007, pp.209-213) found that COs up-regulated the functions of resting rabbit neutrophils by increasing *in vitro* viability, the production of reactive nitrogen intermediates-NO as well as the production of reactive oxygen intermediates $O^{2-}$. The down-regulate functions of PMA-activated neutrophils have been proved to be none dose-dependent with COs. Lee (2009, pp.327-333) examined the antibacterial activity of water-soluble COs against *Vibrio vulnificus*, as well as their inhibitory effects against *in vitro V. vulnificus*-induced cell cytotoxicity, which proved the strong activity in intestinal epithelial cells.

### *7.2 Antiviral*

The administration of COs on different leaf surfaces can decrease the number of local necroses, which is virus-induced infection (Pospieszny, 1989, pp.29-31). It is the hypersensitivity response of host plants that improve the plant resistance against viral infections (Hirano, 1989, pp.3065-3066). COs is also considered as a candidate for an inactivated mucosal influenza vaccine. COs stimulate functional activity of macrophages and increase the generation of active oxygen species, which lead to viral destruction (Bacon, 2000, pp.57-64). Another possible mechanism of antiviral activity of COs is explained in relation to interactions between protein receptors on viral coat and blood leucocytes (Sosa, 1991, pp.489-496). Furthermore, anionic derivatives of COs results to

lower antiviral activity while increasing the degree of deacetylation improves antiviral activity. All results indicate the positive relationship between positively charged groups of COs and antiviral activity, which explicates those cationic charges of amino groups of COs may have additional functions to activate the immune and defense systems (Kurita, 1998, pp.117-120).

Carrageenan has been reported to have anti-HIV activity, but its strong anticoagulant activity is considered to be an adverse reaction (Yuan, 2005, pp.7-13). Yamada examined O-acylated carrageenan with different Mw and sulfate contents and the result showed that anti-HIV activity was increased by depolymerization and sulfation with low anticoagulant activity (Yamada, 1997, pp.51-55; Yamada, 2000, pp.115-120).

To explore the structural basis of anti-HIV-1 action, size-defined oligosaccharides were prepared. Liu (2005, pp.501-510) studied a series of homogeneously sized sulfated polynabburonate (SPMG) fragments and evaluated their capacity to bind rgp120. Competitive inhibition and stoichiometric analyses disclose that SPMG oligosaccharides bind to multiple binding sites on gp120. Consistent with binding data, a positive correlation exists between the size of SPMG oligosaccharides and their anti-HIV activity.

Mazumder (2002, pp.87-95) got a high Mw sulfated galactan and proved its antiviral activity against herpes simplex virus 1 and 2 in bioassays, which is likely due to an inhibition of the initial virus attachment to the host cell. Oligosaccharides are also effective in preventing several phage infections. The inhibition of the bacteriophages' replication involved in several different mechanisms such as structural change of phage particles or inhibition of the receptor-recognizing structures on the phage particles (Kochkina, 2000a, pp.208-211; 2000b, pp.217-219).

*7.3 Antifungal*

COs could react with negatively charged groups of fungi and then exhibited the antifungal activity against both mold and yeast-like fungi. The inhibition mechanism of $CO_S$ against fungi is similar to that against the bacteria. The reaction between COs and negatively charged groups on the cell surface directly interferes with the growth and normal physiological functions of fungi, suggesting that the antifungal activity is correlated with the charge distribution of COs (Hirano, 1989, pp.3065-3066). The inhibitory activities of lsozyme and chitinase were lower than that of LMw-chitosan and COs. This result strongly suggests that the depolymerized products of chitosan are effective for growth inhibition (Uchida, 1988, pp.22-29). Chitosan acts in the growth inhibition in several fungi by inducing plant chitinase activity (Kurosaki, 1986, pp.1587-1591).

## 8. Other functions and applications

Marine oligosaccharides and their derivatives have shown various functions and have been used in many fields including food industry, cosmetics, biomedicine, agriculture, environmental protection, wastewater management (Zhang, 2003, pp.51-56; Yvin, 1999, Patent No.: US 5980916; De la Fuente & Penadés, 2006; Giordano, 2006, pp.511-530; Kurita, 2006, pp.203-226; Kim, 2005, pp.357-368).

## 9. Conclusions

Marine oligosaccharides possess various biological activities and potential applications in drugs. Many studies were carried out to search the bioactivity of marine oligosaccharides, though the detailed molecular mechanisms were not provided. It is hard to explain how exactly these molecules exert their activity. Therefore, future research should be directed towards the understanding of molecular level details, which may provide an insight into the unrevealed molecular level functions of oligosaccharide in future applications.

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## References

Akiyama, H., Endo, T., Nakakita, R., Murata, K., Yonemoto, Y., & Okayama K. (1992). Effect of depolymerized alginates on the growth of bifidobacteria. *Bioscience, biotechnology, and biochemistry,* 56(2), 355-356.

Alekshun, M. N., & Levy S. B. (2004). Targeting virulence to prevent infection: to kill or not to kill? *Drug Discovery Today: Therapeutic Strategies, 1*(4), 483-489.

Bacon, A., Makin, J., Sizer, P. J., Jabbal-Gill, I., Hinchcliffe, M., Illum, L., Chatfield, S., & Roberts M. (2000). Carbohydrate biopolymers enhance antibody responses to mucosally delivered vaccine antigens. *Infection and immunity, 68*(10), 57-64.

Bakunina, I. Y., Nedashkovskaya, O. I., Alekseeva, S. A., Ivanova, E. P., Romanenko, L. A., Gorshkova, N. M., Isakov, V. V., Zvyagintseva, T. N., & Mikhailov V. V. (2002). Degradation of fucoidan by the marine proteobacterium Pseudoalteromonas citrea. *Microbiology, 71*(1), 41-47.

Betting, D. J., Mu, X. Y., Kafi, K., McDonnel, D., Rosas, F., Gold, D. P., & Timmerman J. M. (2009). Enhanced immune stimulation by a therapeutic lymphoma tumor antigen vaccine produced in insect cells involves mannose receptor targeting to antigen presenting cells. *Vaccine, 27*(2), 250-259.

Boon, P., & Ben-Menachem E. (2002). Abstracts for Oral Sessions. *Journal of Neurology, 249*, 1432-1459.

Chaki, T., Kajimoto, N., Ogawa, H., Baba, T., & Hiura N. (2007). Metabolism and calcium antagonism of sodium alginate oligosaccharides. *Bioscience, biotechnology, and biochemistry, 71*(8), 1819-1825.

Chen, H. M., Ma, H. H., Zhang, L., Lin, W., & Yan X. J. (2007b). Agoaro-oliosaccharide enhances the antioxidant property of liver cell in vitro and vivo. *Oceanologia Et Limnologia Sinica, 38*(3), 44-47.

Chen, H., Yan, X., Lin, J., Wang, F., & Xu W. (2007a). Depolymerized Products of [lambda]-Carrageenan as a Potent Angiogenesis Inhibitor. *Journal of Agricultural and Food Chemistry, 55*(17), 6910-6917.

Chen, H., Yan, X., Zhu, P., & Lin J. (2006). Antioxidant activity and hepatoprotective potential of agaro-oligosaccharides in vitro and in vivo. *Nutrition Journal, 5*, 31-43

Chen, H. M., & Yan X. J. (2005). Antioxidant activities of agaro-oligosaccharides with different degrees of polymerization in cell-based system. *Biochimica et Biophysica Acta (BBA)-General Subjects, 1722*(1), 103-111.

Choi, B. K., Kim, K. Y., Yoo, Y. J., Oh, S. J., Choi, J. H., & Kim C. Y. (2001). In vitro antimicrobial activity of a chitooligosaccharide mixture against Actinobacillus actinomycetemcomitans and Streptococcus mutans. *International journal of antimicrobial agents, 18*(6), 553-557.

Currie, A. J., Speert, D. P., & Davidson D. J. (2003). Pseudomonas aeruginosa: role in the pathogenesis of the CF lung lesion. *Thieme Medical Publishers, 24*(6), 671-680.

De la Fuente, J. M., & Penadés S. (2006). Glyconanoparticles: Types, synthesis and applications in glycoscience, biomedicine and material science. *Biochimica et Biophysica Acta (BBA)-General Subjects, 1760*(4), 636-651.

Deters, A., Petereit, F., Schmidgall, J., & Hensel A. (2008). N-Acetyl-D-glucosamine oligosaccharides induce mucin secretion from colonic tissue and induce differentiation of human keratinocytes. *Journal of Pharmacy and Pharmacology, 60*(2), 197-204.

Dou, J., Tan, C., Du, Y., Bai, X., Wang, K., & Ma X. (2007). Effects of chitooligosaccharides on rabbit neutrophil*s* in vitro. *Carbohydrate Polymers, 69*(2), 209-213.

Douglas, H., & Judah F. (1996). Patterns and emerging mechanisms of the angiogenic switch during tumorigenesis. *Cell, 86*, 353-364.

Durmaz Y. (2007). Vitamin E ([alpha]-tocopherol) production by the marine microalgae *Nannochloropsis oculata* (Eustigmatophyceae) in nitrogen limitation. *Aquaculture, 272*(1-4), 717-722.

Eaton, P., Fernandes, J. C., Pereira, E., Pintado, M. E., & Xavier Malcata F. (2008). Atomic force microscopy study of the antibacterial effects of chitosans on Escherichia coli and Staphylococcus aureus. *Ultramicroscopy, 108*(10), 1128-1134.

Ehrke M. J. (2003). Immunomodulation in cancer therapeutics. *International immunopharmacology, 3*(8), 1105-1119.

Enoki, T., Sagawa, H., Tominaga, T., Nishiyama, E., Koyama, N., Sakai, T., Yu, F. G., Ikai, K., & Kato I. (2008). Drugs, foods or drinks with the use of algae-derived physiologically active substances. Patent No.: US 2009/0099101 A1.

Enoki, T., Yamashita, S., Nishimura, K., Sagawa, H., & Kato I. (2002). Agents correcting gene expression regulatory error. Patent No.: US 2003/0158250 A1.

Fitton, J. H., Irhimeh, M. R., & Teas J. (2008). 14 Marine Algae and Polysaccharides with Therapeutic Applications. In Barrow, C. J., & Shahidi, F. Marine nutraceuticals and functional foods. (p. 345). USA: CRC Press.

Fujihara, M., & Nagumo T. (1992). The effect of the content of D-mannuronic acid and L-guluronic acid blocks in alginates on antitumor activity. *Carbohydrate Res*earch*, 224*, 343-347.

Fukuta, K., & Nakamura T. (2008). Induction of hepatocyte growth factor by fucoidan and fucoidan-derived oligosaccharides. *Journal of Pharmacy and Pharmacology, 60*(4), 499-503.

Furda, I., & Brine C. J. (1990). Interaction of dietary fiber with lipids--mechanistic theories and their limitations. *Advances in Experimental Medicine and Biology, 270,* 67-82.

Giordano, A., Andreotti, G., Tramice, A., & Trincone A. (2006). Marine glycosyl hydrolases in the hydrolysis and synthesis of oligosaccharides. *Biotechnology Journal, 1*(5), 511-530.

Harish Prashanth, K. V., & Tharanathan R. N. (2007). Chitin/chitosan: modifications and their unlimited application potential--an overview. *Trends in food science & technology, 18*(3), 117-131.

Hirano, S., & Nagao N. (1989). Effects of chitosan, pectic acid, lysozyme, and chitinase on the growth of several phytopathogens. *Agricultural and Biological Chemistry, 53*(11), 3065-3066.

Hou Q. H., Song W. D., Wang H., & Ji L. L. (2009). Antidiabetic Effect of Thallus laminariae Oligosaccharide on Type 2 Diabetic Rats. *Journal of Zhanjiang Ocean University, 29*(004), 46-50.

Hu, X., Jiang, X., Aubree, E., Boulenguer, P., & Critchley A. T. (2006). Preparation and In Vivo Antitumor Activity of κ-Carrageenan Oligosaccharides. *Pharmaceutical Biology, 44*(9), 646-650.

Hu, J., Geng, M., Li, J., Xin, X., Wang, J., Tang, M., Zhang, J., Zhang, X., & Ding J. (2004). Acidic oligosaccharide sugar chain, a marine-derived acidic oligosaccharide, inhibits the cytotoxicity and aggregation of amyloid beta protein. *Journal of pharmacological sciences, 95*(2), 248-255.

Huang, L., Cheng, X., Liu, C., Xing, K., Zhang, J., Sun, G., Li, X., & Chen X. (2009). Preparation, characterization, and antibacterial activity of oleic acid-grafted chitosan oligosaccharide nanoparticles. *Frontiers of Biology, 4*(3), 321-327.

Iizima-Mizui, N., Fujihara, M., Himeno, J., Komiyama, K., Umezawa, I., & Nagumo T. (1985). Antitumor activity of polysaccharide fractions from the brown seaweed Sargassum kjellmanianum. *The Kitasato archives of experimental medicine, 58*(3), 59-71.

Iwasaki, K., & Matsubara Y. (2000). Purification of alginate oligosaccharides with root growth-promoting activity toward lettuce. *Bioscience, biotechnology, and biochemistry, 64*(5), 1067-1070.

Je, J. Y., Park, P. J., & Kim S. K. (2004). Free radical scavenging properties of hetero-chitooligosaccharides using an ESR spectroscopy. *Food and Chemical Toxicology, 42*(3), 381-387.

Jeon, Y. J., & Kim S. K. (2000). Production of chitooligosaccharides using an ultrafiltration membrane reactor and their antibacterial activity. *Carbohydrate Polymers, 41*(2), 133-141.

Kim, D. E., Lee, E. Y., & Kim H. S. (2009). Cloning and characterization of alginate lyase from a marine bacterium Streptomyces sp. ALG-5. *Marine Biotechnology, 11*(1), 10-16.

Kim, S. K., Rajapakse, N., & Shahidi F. (2008). 7 Production of Bioactive Chitosan Oligosaccharides and Their Potential Use as Nutraceuticals. In Barrow, C. J., & Shahidi, F. *Marine nutraceuticals and functional foods*. (pp. 183). USA: CRC Press.

Kim, K. H., Kim, Y. W., Kim, H. B., Lee, B. J., & Lee D. S. (2006). Anti-apoptotic activity of laminarin polysaccharides and their enzymatically hydrolyzed oligosaccharides from Laminaria japonica. *Biotechnology letters, 28*(6), 439-446.

Kim, S. K., & Rajapakse N. (2005). Enzymatic production and biological activities of chitosan oligosaccharides (COS): A review. *Carbohydrate Polymers, 62*(4), 357-368.

Kim, J. Y., Lee, J. K., Lee, T. S., & Park W. H. (2003). Synthesis of chitooligosaccharide derivative with quaternary ammonium group and its antimicrobial activity against Streptococcus mutans. *International journal of biological macromolecules, 32*(1-2), 23-27.

Kochkina, Z. M., & Chirkov S. N. (2000b). Influence of chitosan derivatives on the development of phage infection in the Bacillus thuringiensis culture. *Microbiology, 69*(2), 217-219.

Kochkina, Z. M., & Chirkov S. N. (2000a). Effect of chitosan derivatives on the reproduction of coliphages T2 and T7. *Microbiology, 69*(2), 208-211.

Kurachi, M., Nakashima, T., Miyajima, C., Iwamoto, Y., Muramatsu, T., Yamaguchi, K., & Oda T. (2005). Comparison of the activities of various alginates to induce TNF-α secretion in RAW264. 7 cells. *Journal of infection and chemotherapy,* 11(4), 199-203.

Kurita K. (2006). Chitin and chitosan: functional biopolymers from marine crustaceans. *Marine Biotechnology, 8*(3), 203-226.

Kurita K. (1998). Chemistry and application of chitin and chitosan. *Polymer Degradation and Stability, 59*(1), 117-120.

Kurosaki, F., Tashiro, N., & Nishi A. (1986). Induction of chitinase and phenylalanine ammonia-lyase in cultured carrot cells treated with fungal mycelial walls. *Plant and cell physiology, 27*(8), 1587-1591.

Larsson, A., Ohlsson, J., Dodson, K. W., Hultgren, S. J., Nilsson, U., & Kihlberg J. (2003). Quantitative studies of the binding of the class II PapG adhesin from uropathogenic Escherichia coli to oligosaccharides. *Bioorganic & medicinal chemistry, 11*(10), 2255-2261.

Lau, J. M., McNeil, M., Darvill, A. G., & Albersheim P. (1985). Structure of the backbone of rhamnogalacturonan I, a pectic polysaccharide in the primary cell walls of plants. *Carbohydrate research, 137*, 111-125.

Lee, B. C., Kim, M. S., Choi, S. H., Kim, K. Y., & Kim T. S. (2009a). In vitro and in vivo antimicrobial activity of water-soluble chitosan oligosaccharides against Vibrio vulnificus. *International journal of molecular medicine, 24*(3), 327-333.

Lee, P., & Kim S. K. (2004). Preparation of hetero-chitooligosaccharides and their antimicrobial activity on Vibrio parahaemolyticus. *Journal of microbiology and biotechnology, 14*(1), 41-47.

Li, Q., Liu, H., Xin, X., Pan, Q., Wang, L., Zhang, J., Chen, Q., Geng, M., & Ding J. (2009). Marine-derived oligosaccharide sulfate (JG3) suppresses heparanase-driven cell adhesion events in heparanase over-expressing CHO-K1 cells. *Acta Pharmacologica Sinica*. 30, 1033-1038.

Liu, H., Geng, M., Xin, X., Li, F., Zhang, Z., Li, J., & Ding J. (2005). Multiple and multivalent interactions of novel anti-AIDS drug candidates, sulfated polymannuronate (SPMG)-derived oligosaccharides, with gp120 and their anti-HIV activities. *Glycobiology, 15*(5), 501-510.

Ma, L. J., Zhang, Y., Bu, N., & Wang S. H. (2010). Alleviation effect of alginate-derived oligosaccharides on Vicia faba root tip cells damaged by cadmium. *Bulletin of Environmental Contamination and Toxicology*, *84*, 161-164.

Ma, J., Xin, X., Meng, L., Tong, L., Lin, L., Geng, M., & Ding J. (2008). The marine-derived oligosaccharide sulfate (MdOS), a novel multiple tyrosine kinase inhibitor, combats tumor angiogenesis both in *vitro* and in *vivo*. *Plos One, 3*(11), e3774.

Maeda, H., Shimizu, K., Miyakawa, Y., Kawada, E., Yamamoto, K., Ohara, H., Sawa, S., Fujii, Y., & Ito, M. (1999). Method and system for producing poly (lactic acid). Patent No.: US 5866677.

Marcinkiewicz, J., Olszowska, E., Mak, P., & Olszowski S. (2003). Collagen type II modification by hypochlorite. *Acta Biochim Polon, 50*(2), 471-479.

Masuda, M., Kawaguchi, S., Abe, T., Kawamoto, T., & Suzuki M. (2006). Additional analysis of chemical diversity of the red algal genus Laurencia (Rhodomelaceae) from Japan. *Phycological Research, 50*(2), 135-144.

Mazumder, S., Ghosal, P. K., Pujol, C. A., Carlucci, M. J., Damonte, E. B., & Ray B. (2002). Isolation, chemical investigation and antiviral activity of polysaccharides from Gracilaria corticata (Gracilariaceae, Rhodophyta). *International journal of biological macromolecules, 31*(1-3), 87-95.

Moon, J. S., Kim, H. K., Koo, H. C., Joo, Y. S., Nam, H., Park, Y. H., & Kang M. I. (2007). The antibacterial and immunostimulative effect of chitosan-oligosaccharides against infection by Staphylococcus aureus isolated from bovine mastitis. *Applied microbiology and biotechnology, 75*(5), 989-998.

Mouy, H., Liy, M. A. N., & Guan H. (2007). In vitro antioxidative activities of three marine oligosaccharides. *Natural Product Research, 21*(7), 646-654.

Neuzil, J., & Stocker R. (1993). Bilirubin attenuates radical-mediated damage to serum albumin. *FEBS letters, 331*(3), 281-284.

Ngo, D., Kim, M., Qian, Z. J. I., Jung, W., Lee, S. H., & Kim S. E. K. (2010). Free radical-scavenging activity of low molecular weight chitin oligosaccharides lead to antioxidant effect in live cells. *Journal of Food Biochemistry, 34*(s1), 161-177.

Ngo, D. N., Lee, S. H., Kim, M. M., & Kim S. K. (2009). Production of chitin oligosaccharides with different molecular weights and their antioxidant effect in RAW 264.7 cells. *Journal of Functional Foods, 1*(2), 188-198.

Ngo, D. N., Kim, M. M., & Kim S. K. (2008). Chitin oligosaccharides inhibit oxidative stress in live cells. *Carbohydrate Polymers, 74*(2), 228-234.

Osumi, Y., Kawai, M., Amano, H., & Noda H. (2002). Physiological activities of oligosaccharides derived from marine algae Porphyra yezoensis Porphyran. *Fisheries science-Tokyo, 68*, 1441-1444.

Parish, C. R., Freeman, C., Brown, K. J., Francis, D. J., & Cowden W. B. (1999). Identification of sulfated oligosaccharide-based inhibitors of tumor growth and metastasis using novel in vitro assays for angiogenesis and heparanase activity. *Cancer research, 59*(14), 3433-3441.

Park, P. J., Je, J. Y., & Kim S. K. (2004b). Free radical scavenging activities of differently deacetylated chitosans using an ESR spectrometer. *Carbohydrate Polymers, 55*(1), 17-22.

Park, P. J., Je, J. Y., Jung, W. K., Ahn, C. B., & Kim S. K. (2004a). Anticoagulant activity of heterochitosans and their oligosaccharide sulfates. *European Food Research and Technology, 219*(5), 529-533.

Pier, G. B., Grout, M., Zaidi, T. S., Olsen, J. C., Johnson, L. G., Yankaskas, J. R., & Goldberg J. B. (1996). Role of mutant CFTR in hypersusceptibility of cystic fibrosis patients to lung infections. *Science, 271*(5245), 64-67.

Pomin, V. H., Pereira, M. S., Valente, A. P., Tollefsen, D. M., Pavao, M. S. G., & Mourao P. A. S. (2005). Selective cleavage and anticoagulant activity of a sulfated fucan: stereospecific removal of a 2-sulfate ester from the polysaccharide by mild acid hydrolysis, preparation of oligosaccharides, and heparin cofactor II-dependent anticoagulant activity. *Glycobiology, 15*(4), 369-381.

Pomin, V. H., Valente, A. P., Pereira, M. S., & Mourao P. A. S. (2005). Mild acid hydrolysis of sulfated fucans: a selective 2-desulfation reaction and an alternative approach for preparing tailored sulfated oligosaccharides. *Glycobiology, 15*(12), 1376-1385.

Pospieszny, H., Chirkov, S., & Atabekov J. (1991). Induction of antiviral resistance in plants by chitosan. *Plant Science, 79*(1), 63-68.

Pospieszny, H., & Atabekov J. G. (1989). Effect of chitosan on the hypersensitive reaction of bean to alfalfa mosaic virus. *Plant Science, 62*(1), 29-31.

Qin, C., Du, Y., Xiao, L., Li, Z., & Gao X. (2002). Enzymic preparation of water-soluble chitosan and their antitumor activity. *International journal of biological macromolecules, 31*(1-3), 111-117.

Rajapakse, N., Mendis, E., Byun, H. G., & Kim S. K. (2005). Purification and in vitro antioxidative effects of giant squid muscle peptides on free radical-mediated oxidative systems. *The Journal of Nutritional Biochemistry, 16*(9), 562-569.

Sánchez-Reus, M. I., Peinado, II, Molina-Jiménez, M. F., & Benedí J. (2005). Fraxetin prevents rotenone-induced apoptosis by induction of endogenous glutathione in human neuroblastoma cells. *Neuroscience research, 53*(1), 48-56.

Sang-Pill, H., Myung-Hee, K., Se-Wook, O., Chan-Kyu, H., & Yong-Hyun K. (1998). ACE inhibitory and antihypertensive effect of chitosan oligosaccharides in SHR. *Korean Journal of Food Science and Technology, 30*(6), 1476-1479.

Sosa, M. A. G., Fazely, F., Koch, J. A., Vercellotti, S. V., & Ruprech R. M. (1991). N-Carboxymethylchitosan-N, O-sulfate as an anti-HIV-1 agent. *Biochemical and Biophysical Research Communications, 174*(2), 489-496.

Sudarshan, N. R., Hoover, D. G., & Knorr D. (1992). Antibacterial action of chitosan. *Food Biotechnology, 6*(3), 257-272.

Sugawara, I., & Ishizaka S. (1982). Polysaccharides with sulfate groups are human T-cell mitogens and murine polyclonal B-cell activators (PBAs): I. Fucoidan and heparin. *Cellular Immunology, 74*(1), 162-171.

Volpi, N., & Maccari F. (2009). Structural characterization and antithrombin activity of dermatan sulfate purified from marine clam Scapharca inaequivalvis. *Glycobiology, 19*(4), 356-367.

Wang, J., He, S., Xie, S., Xu, L., & Gu N. (2007b). Probing nanomechanical properties of nickel coated bacteria by nanoindentation. *Materials Letters, 61*(3), 917-920.

Wang, S., Li, J., Xia, W., & Geng M. (2007a). A marine-derived acidic oligosaccharide sugar chain specifically inhibits neuronal cell injury mediated by-amyloid-induced astrocyte activation in vitro. *Neurological research, 29*(1), 96-102.

Wang, J., Jiang, X., Mou, H., & Guan H. (2004). Anti-oxidation of agar oligosaccharides produced by agarase from a marine bacterium. *Journal of Applied Phycology, 16*(5), 333-340.

Wartman, D., & Weinstock M. (2008). Are we overemphasizing sun avoidance in protection from melanoma? *Cancer Epidemiology Biomarkers & Prevention, 17*(3), 469-470.

Xie, W., Xu, P., & Liu Q. (2001). Antioxidant activity of water-soluble chitosan derivatives. *Bioorganic & Medicinal Chemistry Letters, 11*(13), 1699-1701.

Xu, Q., Ma, P., Yu, W., Tan, C., Liu, H., Xiong, C., Qiao, Y., & Du Y. (2009). Chitooligosaccharides Protect Human Embryonic Hepatocytes Against Oxidative Stress Induced by Hydrogen Peroxide. *Marine Biotechnology*, 1-7.

Xu, X., Iwamoto, Y., Kitamura, Y., Oda, T., & Muramatsu T. (2003). Root growth-promoting activity of unsaturated oligomeric uronates from alginate on carrot and rice plants. *Bioscience, biotechnology, and biochemistry, 67*(9), 2022-2025.

Yamada, T., Ogamo, A., Saito, T., Uchiyama, H., & Nakagawa Y. (2000). Preparation of O-acylated low-molecular-weight carrageenans with potent anti-HIV activity and low anticoagulant effect. *Carbohydrate Polymers, 41*(2), 115-120.

Yamada, T., Ogamo, A., Saito, T., Watanabe, J., Uchiyama, H., & Nakagawa Y. (1997). Preparation and anti-HIV activity of low-molecular-weight carrageenans and their sulfated derivatives. *Carbohydrate Polymers, 32*(1), 51-55.

Yan, Y., Wanshun, L., Baoqin, H., Bing, L., & Chenwei F. (2006). Protective effects of chitosan oligosaccharide and its derivatives against carbon tetrachloride-induced liver damage in mice. *Hepatology Research, 35*(3), 178-184.

Yokose, T., Nishikawa, T., Yamamoto, Y., Yamasaki, Y., Yamaguchi, K., & Oda T. (2009). Growth-Promoting Effect of Alginate Oligosaccharides on a Unicellular Marine Microalga. *Nannochloropsis oculata*. *Bioscience, biotechnology, and biochemistry, 73*(2), 450-453.

Yonemoto, Y., Tanaka, H., Yamashita, T., Kitabatake, N., Ishida, Y., Kimura, A., & Murata K. (1993). Promotion of germination and shoot elongation of some plants by alginate oligomers prepared with bacterial alginate lyase. *Journal of Fermentation and Bioengineering, 75*(1), 68-70.

Yoon, N. Y., Ngo, D. N., & Kim S. K. (2009). Acetylcholinesterase inhibitory activity of novel chitooligosaccharide derivatives. *Carbohydrate Polymers,* 78(4), 869-872.

Yoon, S. S., & Hassett D. J. (2004). Chronic Pseudomonas aeruginosa infection in cystic fibrosis airway disease: metabolic changes that unravel novel drug targets. *Expert Review of Anti-Infective Therapy, 2*(4), 611-623.

Yuan, W. P., Liu, B., Liu, C. H., Wang, X. J., Zhang, M. S., Meng, X. M., & Xia X. K. (2009). Antioxidant activity of chito-oligosaccharides on pancreatic islet cells in streptozotocin-induced diabetes in rats. *World Journal of Gastroenterology: WJG, 15*(11), 1339-1345.

Yuan, H., Song, J., Li, X., Li, N., & Dai J. (2006). Immunomodulation and antitumor activity of [kappa]-carrageenan oligosaccharides. *Cancer letters, 243*(2), 228-234.

Yuan, H., & Song J. (2005). Preparation, structural characterization and in vitro antitumor activity of kappa-carrageenan oligosaccharide fraction from Kappaphycus striatum. *Journal of Applied Phycology, 17*(1), 7-13.

Yvin, J. C., Levasseur, F., Malo, S., Hud'Homme, F., & De la Mer S. J. (1999). Use of laminarin and oligosaccharides derived therefrom in cosmetics and for preparing a skin treatment drug. Patent No.: US 5980916.

Zetter P. B. R. (1998). Angiogenesis and tumor metastasis. *Annual Review of Medicine, 49*(1), 407-424.

Zhang Z., & Jiang, X. (2003). Bioactivities of Oligosaccharides and the Potential Application of Marine Oligosaccharides. *Chinese Journal of Marine Drugs, 22*(3), 51-56.

Zhao, H., Liu, H., Chen, Y., Xin, X., Li, J., Hou, Y., Zhang, Z., Zhang, X., Xie, C., & Geng M. (2006). Oligomannurarate sulfate, a novel heparanase inhibitor simultaneously targeting basic fibroblast growth factor, combats tumor angiogenesis and metastasis. *Cancer research, 66*(17), 8779-8787.

Table 1. Abbreviation name of oligosaccharide

| Abbreviation | Full name | Species |
|---|---|---|
| CO/ NA-CO | Chitosan oligosaccharides | Crustacea |
| COs-GTMAC | Cos-glycidyl trimethylammonium chloride | |
| CO-OA | Acid-grafted chitosan oligosaccharide | |
| LO | Laminarin oligosaccharides | Laminaria japonica Aresch |
| JG3 | Marine-derived oligosaccharide sulfate | |
| AO | Alginate oligosaccharide | |
| AOSC | Acidic alginate oligosaccharide sugar chain | |
| ADO | Alginate-derived Oligosaccharides | |
| FO | Fucoidan oligosaccharides | |
| CAO | Carrageenan oligosaccharides | Chondrus crispus |
| λ-CAO | λ-carrageenan oligosaccharides | |
| GSO | | Rhodophyta |


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