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Oyster Thief (*Codium Fragile*): A Vital Marine Alga

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Abstract : The seaweed-derived bioactive products are a storehouse of healthy attributes. Researchers across the globe are waking up to tap these unexploited marine sources to develop novel therapeutics. Among the three main divisions of marine macro algae (Chlorophyta, Phaeophyta and Rhodophyta), the marine green algae (Chlorophyta) especially, *Codium* genus has been of public and scientific concern from the last two decades, because some of these are invasive, bloom-forming nature. Among them, *Codium fragile* is the most invasive seaweed in the world. It is believed to be native to Japan and was then unintentionally spread around the world. *Codium fragile*, sponge seaweed and it is well known in the name of "OYSTER THIEF", because it usually attach to oysters and float away, carrying the animals with it. This was the inspiration for the common name "oyster thief". It constitutes useful raw materials for the development of diets or ingredients for human and animal nutrition and has different biological activities with a great potential in pharmaceutical applications. It biosynthesizes sulfated polysaccharides with very distinct structural features. So focus on marine green alga Oyster thief (*Codium fragile*) with an overview of the recent progress of its structurally diverse of bioactive materials/compounds and biological activities.
Keywords : Seaweed, Oyster thief, *Codium fragile,* Polysaccharides, Pharmaceutical applications.

Introduction

Over 90% of the world's living biomass is contained in the oceans, which cover 71% of the earth surface. Among marine organisms, marine algae are relatively simple photosynthetic plants with unicellular reproductive structures. They range from unicellular organisms to non vascular filamentous or thaloid plants[1]. Marine algae are heterogeneous group of plants with a long fossil history. Two major types of algae can be identified: The macroalgae occupy the littoral zone, and the microalgae are found in both bentheic and littoral habitats and also throughout the ocean waters as phytoplankton[2].

Marine macroalgae or Seaweeds are large algae (macro algae) that grow in salt water or marine environments and lack true stems, roots and leaves[2]. Seaweeds commonly grow on coral reefs or in rocky landscape or can grow at great in depths if sunlight can penetrate through the water. Seaweeds play an important and vital role in the marine ecosystem, providing food and shelter for host of creatures, such as seaurchins, lobsters and young fishes. Three major groups of seaweeds are recognized according to their pigments that absorb light of particular wavelengths and give them their characteristic colors of green, brown or red[3].The importance of seaweeds for human consumption is well known since 300 BC in China and Japan[2]. These two countries are the major seaweed cultivators, producers, and consumers in the world. In the Indian Ocean region countries like Malaysia, Indonesia, Singapore, Thailand and Korea, seaweeds are used in salad, jelly, soup etc. However, in India, seaweed consumption is negligible except in the preparation of porridge

from *Gracilaria* species and *Acanthophora* species in coastal states such as Kerala and Tamil Nadu[4]. Seaweeds are rich in soluble dietary fibers, proteins, minerals, vitamins, antioxidants, phytochemicals, and polyunsaturated fatty acids, with low caloric value. They are an excellent source of vitamins A, B_1, B_2, B_3, B_{12}, C, D and E. Their amino acids content is well-balanced and contains all or most of the essential amino acids needed for life and health[5]. Moreover, the active compounds isolated from marine macroalgae exhibit various applications in various fields. Recent times have seen a surge in interest to tap these unexploited marine sources to develop novel therapeutics.

Seaweed species and their products used as a renewable bioresource in pharmaceutical industry and medical field. Medical applications: They show antioxidant[6], anti-viral,[7,8] anti-allergic[9], anti-inflammatory[10,11], anti-cancer[12,13] and anti-coagulant activities[14,15], and Surgical applications as a Prosthetic devices like Alginates are best alternative to prosthetic devices Joint replacements, fracture fixation plates, bone defect fillers, coating artificial hips and heart valves. Alginates derived from *Crocystis, Laminaria, Sargassum* are used in the preparation of biodegradable sutures. *Spirulina* has extremely high chlorophyll content. In fact, taking *Spirulina* ensure rapid healing of damaged tissue, as well as providing a concentrated source of calcium, necessary to maintain the health of teeth and gums. Fucoidan (brown algae) in the pre-surgical patient diet seems to reduce the intensity of blood loss and vascular bed collapse shock during and after surgery. They also have numerous industrial applications: Carrageenan and Alginate obtained from *Chondrus, Gigartina, Crocystis*, *Laminaria*, *Ascophyllum* and *Eucheuma* used in food industry (jellies, jams, salad dressing, stabilization agent for canned food, gelling agent), cosmetic industry, paint industry and Alginate in textile industry (bending and forming or stiff); raw material for artificial fiber, clarifier for wine making, sewage purifier, water softener, separating agent for chemical composition, alkaloid refining agent, emulsifier for paint and plastics industry, bonding agent, stabilizer, lubricant and filling agent The large amount of insoluble carbohydrates in brown seaweeds act as soil[16]. Conditioners (improve aeration and soil structure, especially in clay soils) and have good moisture retention properties. Biomass for fuel: Gracilarial seaweeds are rich in methane. Packaging: Because of their biological biodegradability, the use of bioplastics is especially popular in the packaging sector. Funoran obtained from Gloeopeltis used in paper-making industry, textile industry. Agar obtained from *Gelidium*, *Pterocladiella* and *Gracilaria* used in culture medium for microorganism, food industry, textile industry, paper-making, clarifier for wine making; aids in the making of ultra thin separating film, analytical research (electrophoresis), medicine (gastro-intestinal intolerance and capsules)[16].

Among the three main divisions of marine macroalgae (Chlorophyta, Phaeophyta and Rhodophyta), the marine green algae (Chlorophyta) are truely green with no pigments to mask the chlorophyll. The green algae are diverse from microscopic free-swimming single cells to large membranous, tubular and bushy plants. They are valuable sources of structurally diverse bioactive compounds and remain largely unexploited in nutraceutical and pharmaceutical areas. Marine green algae contributing the most as bioactive materials from Ulvales, including three main genera, *Monostroma*, *Ulva*, and *Enteromorpha*. Algae of these genera are widespread and present large biomasses. *Ulva* and *Monostroma* algae are particularly known for their high nutritional value and health benefits and are usually grown or collected for food consumption. *Codium* and *Caulerpa* algae are additional representative of green seaweeds, which are broadly distributed in tropical seas. The green algae *Codium* are believed to feature certain invasive properties because of their ability to thrive in temperate waters.

The genus *Codium* (Codiaceae, Bryopsidales, Chlorophyta) currently comprises around 125 species, widely distributed through the world's seas with the exception of the Polar Regions, but mainly found in temperate and subtropical areas[17]. Seaweeds of this genus show a broad variation of forms and occur in various habitats. The majority of those belonging to the suborder Bryopsidineae, exhibit no calcification. *Codium* thalli can spread out over hard surfaces as mats, form spheres or grow upright, either unbranched and finger-like, or branched, with cylindrical or flattened branches. *Codium* is found in marine habitats ranging from rocky coasts exposed to full wave-forces to calm lagoons, from intertidal habitats to deep reefs, from arctic to tropical waters and from eutrophic estuaries to nutrient-depleted coral reefs[18]. Some *Codium* species have been of public and scientific concern from the last two decades, because of the invasive, bloom-forming nature. *Codium isthmocladum*, forms harmful blooms on reefs, consequence of increased eutrophication[18]. On the other hand, *Codium* species are used as food for culturing abalone, which are consumed by humans, and are a source of bioactive compounds[18]. Among them *Codium fragile* is the most invasive seaweed in the world.

Codium fragile (Suringar) Hariot, 1889 (Phylum: Chlorophyta; Class: Chlorophyceae; Order: Codiales; Family: Codiaceae Genus: *Codium*) named for its dark green color and soft, felt-like texture (figure 1). It has earned various names "Dead Man's Fingers" or "Green Sea Fingers" for its swollen, finger-shaped branches that float in the water, These "fingers" consist of plump, rounded branches which may grow up to 1 meter long, which originate from a central fleshy mass commonly referred to as a "spongy, "basal holdfast". It also called as as "felty fingers", "forked felt-alga", "stag seaweed", "sponge seaweed", "green sponge", "green fleece", and "oyster thief".

Figure1: Oyster thief (*Codium fragile*) collected from Rameswaram in August 2015

Codium growth pattern is bush-like structure, in bushels of up to two feet across and it prefers warm water temperatures of approximately 24°C for optimal growth and reproductive success, but tolerates a wide spectrum of water temperature and salinity. Requiring low sunlight, so grows well in light obstructed areas, including developed coastal areas featuring human-wrought structures, beneath and upon which it may attach and thrive in the shade. Table 1 gives the habitat preference of *Codium fragile*. It is one of familiar seaweeds and has been used as edible one from ancient times. Furthermore, it was recorded in the Law of Taiho (AD 701) that it was paid as tax to the Court. Its uses there have been recorded for the treatment of enterobiasis, dropsy and dysuria in Oriental medical textbooks[19].

Table 1: Habitat preference of *Codium fragile* (72)

Habitat	**Status**
Brackish:	
Estuaries	Present
Littoral:	
Coastal areas	Principal habitat
Intertidal zone	Present
Mud flats	Present
Salts marshes	Present
Marine:	
Benthic zone	Principal habitat
Climate:	
Warm temperate climate	Tolerated
Continental climate	Tolerated
Salinity	12 to 40‰
Reproductive salinity	12 to 48‰
Temperature	-2 to 34°C
Reproductive temperature	10 to 24°C
Depth	0 to 15m

Oyster thief as an invasive species

Oyster thief (*Codium fragile*) has made its ways around the world (figure 2), from the Asia Pacific, particularly in the Japan, to mainland Europe. From there, it was introduced to England, to the River Yealm in Devon in 1939, and first discovered at Steer Point. The first recorded sighting in the U.S. occurred in 1957 on Long Island, New York, followed by Boothbay Harbor, Maine in the early 1960s. Since then, *Codium* has spread up and down the east coast of the U.S., heading north to the Gulf of Saint Lawrence, and south to the Carolinas[20]. A full-scale invasion taking place in Massachusetts, where *codium* is washing up in such large quantities in some Cape Cod communities those beaches must be closed to the public and has proven to be a most successful invasive species.

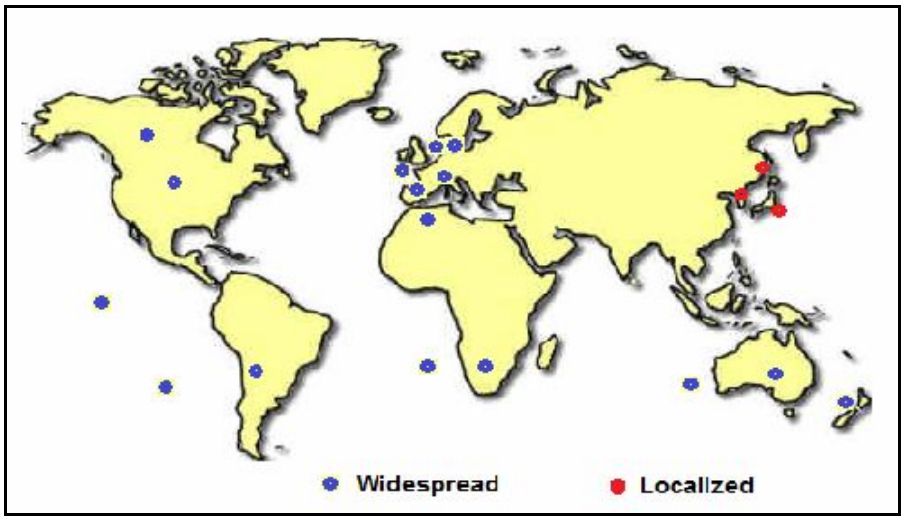

Figure 2: Map showing the distribution of *Codium fragile* around the world

Its main method of transportation from port to port is believed to be on the hulls of ships, or attached to captured shellfish. Human activities including boating and shellfish collection also contribute to dispersion. *Codium fragile* has become well established in wide varieties of climates in its current distribution. This success is due to the seaweed's versatile requirements for sunlight, nutrients, and reproductive conditions. *Codium* grows best in low sunlight, which helps it to thrive even in light obstructed areas, including developed coastal regions featuring human-wrought structures, beneath and upon which it may attach and thrive in the shade. Dead Man's Fingers can reproduce sexually, when a female gamete is fertilized by a male gamete; parthenogentically when the female gamete develops into a new individual without fertilization; or vegetatively when a genetically identical individual is formed from a branch or stem of the parent plant. Table 2 gives the invasion of *Codium fragile* from different countries.

Table 2: Invasion of *codium fragile* from different countries

Country	Distribution	Occurrence	Invasive	References
ASIA				
Japan	Localized	Native	Non invasive	21
Korea	Localized	Native	Non invasive	20
OCENIA				
Australia	Widespread	Exotic	Invasive	22
South Australia	Widespread	Exotic	Invasive	23
Western Australia	Widespread	Exotic	Invasive	24
AFRICA				
Algeria	Widespread	Exotic	Invasive	25
Tunisia	Widespread	Exotic	Invasive	25
EUROPE				
France	Widespread	Exotic	Invasive	25

Italy	Widespread	Exotic	Invasive	23
Ireland	Widespread	Exotic	Invasive	29
Spain	Widespread	Exotic	Invasive	25
NORTH AMERICA				
Long Island, New York	Widespread	Exotic	Invasive	27 28
Eastern Canada	Widespread	Exotic	Invasive	29
Atlantic Canada	Widespread	Exotic	Invasive	30
Nova Scotia	Widespread	Exotic	Invasive	31
California	Widespread	Exotic	Invasive	23
Gulf of Maine	Widespread	Exotic	Invasive	32
SOUTH AMERICA				
Chile	Widespread	Exotic	Invasive	33
New Zealand	Widespread	Exotic	Invasive	25, 34

Biomolecules from Oyster thief

The chemical composition of seaweed varies with species habitat, maturity, salinity, temperature, light intensity, tidal variations and other environmental conditions. Due to their ability to produce polyunsaturated fatty acids (PUFAs), marine algae have attracted considerable attention from researchers all over the world in the last two decades[15]. Lipid extracts obtained from *Codium* species have been shown to exhibit antibacterial, antiviral, antifungal and cytotoxic activities[35]. A variety of sterols, fatty acids and terpenes are responsible for these activities. The crude molecules and fractions from *Codium fragile* consisted mostly of carbohydrates (44.1–80.5%), sulfates (3.2–22.2%) and proteins (3.0–15.7%) with small amounts of uronic acids (1.1–4.2%), and included different levels of mannose (91.3–18.7%), glucose (62.7–8.6%) and galactose (37.5–59.5%) [36]. These molecules contained one or two sub-fractions with molecular weights (Mw) ranging from 148×103 to 4879×103 g/mol. The chemical composition of enzymatic extracts of *C. fragile* from North Atlantic coasts consisted of 11% protein, 31% neutral sugars, 0.8% sulfate, 0.6% uronic acids, and 49% ash[37]. The yield of the crude was slightly lower than those from *C. fragile* that were reported by Tabarsa[36]. This discrepancy might be attributable to differences in the extraction procedures, in particular the extraction temperature.

Fatty acids are important components in the cellular structure, take part in the production of some hormones and bile salts, and have important regulatory functions for cellular activity and gene expression. In addition they are related to the prevention and treatment of atherosclerosis, thrombosis, arthritis and cancer, mainly through their metabolic conversion into prostaglandins, thromboxanes and leucotriens. The nutritional composition of the *Codium fragile*, including amino acid and fatty acid contents along with tocol and carotenoid contents showed a high range of protein contents (13.7-10.8%), amino acid contents (1879.6-1417.7 mg/100 g dry algae) and a low content in lipids (0.7-15.0%) [38]. They reported that most abundant fatty acids in *C. fragile* are linolenic, oleic, and linoleic acid. β-Tocopherol and α-tocopherol (677.8 and 453.5 mg/g lipid, respectively) were found in it. In addition, β-carotene was the principal carotenoid found 197.9 and 113.7 mg/g dry algae. Recently, in the northern Chile, 19 fatty acids are detected in *Codium fragile*[39]. They found linolenic acid to be the main fatty acid, making up 24.6% of the total fatty acid content, followed by palmitic (17.74%), stearic (17.38%) and (12.25%) oleic acids.

Cell wall polysaccharides

Green seaweeds from Dasycladales and Bryopsidales including *Codium* are called “Giant-cell” algae because their habits comprise continuous multinucleated siphons lacking cross cell walls and cell plate formation, unless at the septa formed in the basal part of reproductive structures[40]. Polysaccharides of green seaweeds, are chemically and physicochemically different from those of land plants, and may have special physiological effects on the human body. Cell walls of many green algal groups remain uncharacterized, with

the exception of the glycoprotein-rich Volvocacean walls[41].

Codium has developed unique cell wall architecture, when compared, not only with that of vascular plants, but also with other related green seaweeds and algae[42]. The first structural study about the polysaccharides from *Codium* was carried out by Love and Percival[43] who found that the water extracts from *C. fragile* contained galactose and then arabinose as major monosaccharide constituents, but also small amounts of xylose, rhamnose, glucose, and mannose. This product contained 22% of sulphate, and minor amounts of uronic acids. It also contained 25% of protein and it was supposed to be a proteoglycan. Glucose and mannose were eliminated by periodate oxidation and were considered to arise from contaminant polymers. The resulting product was fractionated by anion exchange chromatography obtaining a fraction with similar quantities of galactose and arabinose, and important amounts of sulphate that would be in part, linked to C-2 or C-3 of the arabinose units. Partial acid hydrolysis and fractionation of the oligosaccharides, allowed to isolate a neutral fraction containing 3-*O*-β-L-arabinopyranosyl-L-arabinose and 3-*O*-β-D-galactopyranosyl-D-galactose and an acidic fraction containing galactose 4-sulphate and galactose 6-sulphate. This pioneer study gave a first clear idea about the structure of these polysaccharides.

Sulphated Galactans

There are several reports regarding the isolation of pyruvylated sulfated galactans from related species, such as Codium yezoense and Codium isthmocladum[44]. In addition, pyruvylated arabinosulfated galactans were reported as macromolecular components for the cell walls of *C. fragile*[42]. Thus, these highly pyruvylated and sulfated galactan might be a characteristic structure of sulfated polysaccharide from Codium species. A pyruvylated sulfated galactan isolated from *Codium fragile*[44] consisting of 3-linked, 3, 6-linked, and non-reducing terminal D-galactose with pyruvate and sulfate groups.

The sulphated polysaccharides extracted with water from *C. fragile* from the Atlantic coast of Patagonia were studied[13]. The room temperature water extracts from this seaweed showed galactose and arabinose as major sugar components. As no fractionation was carried out, it was not possible to deduce if this extracts contained arabinogalactans or a mixture of arabinans and galactans. However, in general terms, the galactan moieties in this extracts were similar to the galactan from *C. yezoense*.

Galactan sulphates from *C. fragile* of two different locations, Puerto Deseado (Santa Cruz), Argentina (F1 and F6), and Kamakura, Japan (FG) were studied at the same time by Estevez[42]; Ciancia[45]; Fernandez[46]; Kasulin[30]. Table 3 gives the structural units deduced from data published in these papers. These galactans showed the same structural units found for other galactans from *Codium* species, with the exception of F1 where structural data suggests the presence of terminal 3, 4-pyruvylated β-D-galactose 6-sulphate units. In addition, there are important differences in the percentages of the structural units: (1) most of the 3,6-linked galactose units are sulphated in FG, while they are not sulphated in F1 and in F6 only half of these units are sulphated; (2) there are not only 3,4-pyruvylated galactose terminal units, but also significant amounts of 3-linked 4,6-pyruvilated β-D-galactose units in F1 and F6, while the latter units are absent in FG. These results show that although similar, galactans from different *Codium* species have important interspecific differences.[42, 45, 48] also important differences between galactans of the same species but from different locations have been observed. Besides, galactans F1 and F6, fractions from the same seaweed and location are also distinct (Table 3).

Table 3: Structural Units Found in Sulphated Galactans from *Codium fragile* from two different locations

Structural Units	Argentina		Japan
	F1	F6	FG
→3)β-d-Gal*p* (1–	25	26	19
→3)β-d-Gal*p* 4S(1→	3	6	8
3,4Pyr-β-d-Gal*p*(1→	28	36	40
→3,6)β-d-Gal*p*(1→	24	12	7
→3,6)β-d-Gal*p* 4S(1→	-	10	25
→3)β-d-Gal*p*4,6-pyr(1→	8	-	11
3,4Pyr-β-d-Gal*p* 6S(1→c	15	-	-

A Nomenclature used: Galp: galactopyranose; S: sulphate; Pyr: pyruvate. sSmall amounts of other sugers were considered to drive from contaminent polymers. cSpeculative unit, deduced from contaminent polymers. Cspeculative unit, deduced from the desulphation pattern, '-'data deficiency.

Sulphated Arabinans

In a screening of inhibition of thrombin by sulphated polysaccharides isolated from green algae, arabinans were extracted from *C. fragile*. No structural details were determined[49]. These give an idea that sulphated arabinans are usual components of *Codium* cell walls. However, there was a lot of confusion due to reports with very different structures, but no detailed information. The room temperature water extracts from *C. fragile* showed important amounts of arabinose. Methylation and desulphation-methylation analyses showed that 3-linked 2, 4-disulphated arabinopyranose was the major structural unit. No evidence of the presence of arabinose in the furanose form was found. From the NMR spectra, it was suggested that these residues were β-anomers[45].

Mannans

Linear β-(1 → 4)-D-mannans are the major fibrillar component of cell walls from *Codium* species[45]. The insoluble mannan obtained from *C. fragile* after exhaustive water extraction was studied by degradation with a β-mannanase and analysis of the resulting oligosaccharides; confirming the linear nature of this structure and the β-(1 → 4)-linkages, no evidence of side chains were found[46]. On the other hand, soluble mannan structures were found to be important components of the hot-water extracts from *C. fragile*. The structure of the backbone was clearly established, being identical to that of the fibrillar component. The reason of the high solubility of these mannans in hot water was not clearly established. The average molecular weight for those from *C. fragile* was 39-49kDa and no side chains were detected. Recently, a very different structure was proposed for a mannan isolated from a water extract of *C. fragile* collected from the coast of Sokcho, Gangwon province; Korea, comprising 3-linked α-D-mannose units[50]. This structure is completely different to those isolated and characterised by many other researches for the same species from various locations[42, 43, 51]. However the evidence presented by the authors could be subject of different interpretations.

Hydroxyproline-rich Glycoproteins(HRGPs)

Arabinogalactan proteins (AGPs) and extensins are hydroxyproline-rich glycoproteins (HRGPs) usually present in plant cell walls[52]. There are contradictory data about the exact functions of AGPs in a broad variety of processes as plant growth and development, plant defence, cell proliferation, cell expansion, cell differentiation, cell extension and somatic embryogenesis. Although they have also been detected in some Chlorophycean and Charophyceae green algae, the first seaweed that was found to contain HRGPs was *C. fragile*[42] by immunolabeling using antibodies against specific cell wall HRGP epitopes (AGPs and extensins) and by reaction with the β-glucosyl Yariv reagent. In addition, a unique furanosic α-arabinosyl structure was detected in an arabinose-rich fraction obtained by fractionation of a room temperature water extract of the same seaweed, consisting in a 5-linked arabinan branched at C-3. 3-Linked and 3, 6-linked β-D-galactose units were also detected. Although the latter units could be part of the major galactan biosynthesised by this seaweed, altogether, these results showed the presence in this fraction of AGP-like structures.

Bioactivities of Oyster thief

A number of seaweed species are used as traditional medicines, foods and healthcare in various regions of the world. A green seaweed Oyster thief (*C. fragile*) is consumed by humans, used as a food additive, and an anti-helminthic, and to treat fever, especially in children, with fever accompanying pain, cough, and cold sweat, with no side effects. It is also used as an herbal medicine in China to treat many urinary diseases, dropsy, and helminthiasis[11]. As material sources of herbal medicine in oriental countries, most of those medicinal effects of *C. fragile* are directly or indirectly related to anti-inflammatory action of the seaweed. In China, it is also used as anticancer, antipyretic and helminthic agents in Chinese traditional medicine. It also has antiviral and anticoagulant properties. Algal lectins, affect blood clotting and fibrinolysis, from *Codium* spp are routinely used in biochemical studies[53].

Anticoagulant activity

Thrombosis is a leading cause of morbidity and mortality throughout the world. Thrombolytic agents are important for both the prevention and treatment of thrombosis. The hot water extracts containing sulfated

polysaccharides (SPs) from *C. fragile* collected from Jeju Island in Korea[54], showed prolonged activated partial thromboplastin time (APTT), which suggests inhibition of intrinsic factors and increased intrinsic pathway-dependent clotting times. Jurd were confirmed the anticoagulant activities of SPs from *C. fragile* using coagulation techniques and chromogenic substrate assays[47]. The room-temperature water extracts containing polysaccharides from *Codium fragile* were also studied[55], they reported that it contains highly sulfated and substituted with pyruvic acid ketals and showed a dual haemostatic effect: prevented coagulation, but induced platelet aggregation. Anticoagulant activity and platelet aggregation were higher in the samples with polysaccharides richer in sulfate.

Recently, Codiase a new bi-functional fibrinolytic serine protease having thrombolytic, anticoagulant, and antiplatelet activities purified from *Codium fragile*[56]. The molecular weight of the enzyme was estimated to be 48.9 kDa and The N-terminal sequence was found to be APKASTDQTLPL, which is different from that of other known fibrinolytic enzymes. From Fibrin plate assays they revealed that it was able to hydrolyze fibrin clot either directly or by activation of plasminogen. Codiase effectively hydrolyzed fibrin and fibrinogen, preferentially degrading α- and Aα chains, followed by γ–γ, and γ-chains. The in vitro and in vivo studies revealed that codiase reduces thrombosis in concentration-dependent manner. Codiase was found to prolong activated partial thromboplastin time and prothrombin time.

Anti-inflammatory activities

The anti-inflammatory activities of the *C. fragile*, and its mechanism in macrophages induced by Peptidoglycan (PGN) were studied[57]. They reported that treatments of macrophages with 100 ug/ml of ethanol extract of *Codium fragile* (EECF) inhibited PGN-induced IL-6, NO and PGE_2 production in a dose-dependent manner as well as expression of inducible NO synthase (iNOS) and cyclooxygenase (COX)-2. These findings may help elucidate the mechanism by which EECF modulates RAW 264.7 cell activation under inflammatory conditions. Ho-Dong[58] were also studied the anti-inflammatory characteristics of EECF mediated by the regulation of inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 (COX-2) using LPS-stimulated murine macrophage RAW 264.7 cells. CFE significantly inhibited LPS-induced NO and PGE2 production in a dose-dependent manner and suppressed the expression of iNOS and COX-2 proteins in LPS-stimulated RAW 264.7 is mediated through the NF-κB-dependent transcriptional downregulation of iNOS and COX-2, with no cytotoxicity.

Recently, The Dichloromethane, ethanol, and boiling water extracts of *Codium fragile*, were tested for anti-inflammatory, antipyretic, and analgesic activities in mice[59]. The dichloromethane and ethanol extracts inhibited inflammatory symptoms of mouse ear edema and erythema by 74% or higher. These extracts also demonstrated for inhibition of pyrexia, similar to that of acetyl salicylic acid and they were isolated the Eicosapentaenoic acid from the *C. fragile* as the main active anti-inflammatory compound.

Immunostimulatory activities

A pyruvylated sulfated galactan isolated from *C. fragile* collected from Japan, was found to stimulate nitric oxide (NO) production by inducing iNOS at the mRNA and protein levels and induce the mRNA expression of several cytokines, such as IL-1 β, IL-6, IL-10, and TNF-α[44]. These SPs were suggested to possess potent immunostimulatory activities by activating macrophages while preventing potential detrimental inflammatory effects from excessive macrophage activation. The molecular characteristics and immunostimulating Activity of *Codium fragile* collected from Korea were also studied[50]. They reported that the molecules of one or two sub-fractions contained molecular weights (Mw) ranging from 148×103 to 4879×103 g/mol. The crude fractions stimulated RAW 264.7 cells to produce considerable amounts of pro-inflammatory mediator nitric oxide (NO) and cytokines. The treatment of sample molecules facilitated the degradation and phosphorylation of mitogen-activated protein kinases (MAPK) in RAW264.7 cells, suggesting that they might stimulate RAW264.7 cells through the activation of NF-B and MAPK pathway.

Antimicrobial and Antioxidant activities

The methanol, dichloromethane and hexane extracts of *Codium fragile* collected from Izmir coast, Turkey, were tested for antimicrobial and antioxidant activities[60]. Antioxidant effects were evaluated by hydroxyl radical scavenging assay (deoxyribose degradation assay) and β-carotene bleaching assay. These extracts indicated relatively little antioxidant activities, as compared to commercial antioxidants. They were also analyzed the composition of the essential oil of *C. fragile* by GC and GC-MS. Twenty four compounds

were identified and n-tricosane (11.88 %) was determined as major component. They showed weak antibacterial activity against all gram positive bacteria tested, except methicillin-oxacillin resistant *Staphylococcus aureus* ATCC 43300. Similarly, all the extracts of *C. fragile* showed weak antimicrobial activity on tested organisms. At the same time seaweeds from the Red Sea Hurghada, Egypt, were also screened for their antibacterial activities against both grampositive bacteria (*Staphylococcus aureus* NCIMB 50080 and *Bacillus cereus*) and gramnegative bacteria (*Escherichia coli* NCIMB 50034, *Enterococcus feacalis* NCIMB 50030, *Salmonella* sp. and *Pseudomonase aeruginosa*) [61]. Ethyl acetate extracts and methanolic extracts of *C. fragile* showed higher antibacterial activities than other members of the tested algae. Recently methanolic extracts *of Codium fragile* and some other species from the Aegean coast of Turkey were also studied for their inhibitor activity against pathogenic microbes (*Staphylococcus aureus, Staphylococcus epidermidis, Salmonella typhimurium, Enterobacter aerogenes, Escherichia coli, Enterococcus faecalis and Escherichia coli* O157:H7) [62]. Against the extracts of all the tested marine algae, *S. aureus* was the most sensitive bacteria since it was inhibited by most of the extracts. On the other hand, the highest inhibitor activity was shown to *Enterobacter faecalis* by the extract of *Codium fragile*.

The methanolic and aqueous extracts of *C. fragile* at room temperature were investigated for their antioxidant activities from Portugal[63]. They reported that these are not highly anti-oxidative in nature. But the aqueous extract of *Codium fragile* collected from Jeju Island at 70°C, exhibited the highest scavenging activities against $O_2^{\cdot}$, $HO^{\cdot}$, H_2O_2 and DPPH free radicals.

Antiviral activities

A highly branched galactan from *C. fragile* inhibited the replication of HSV-2, possibly by interfering in the early steps such as virus adsorption and penetration into host cells[48]. They studied that, it suppressed virus production most efficiently when added at the same time as virus infection and throughout the incubation thereafter. Pretreatment of host cells showed no inhibitory effects, and less anti-HSV-2 effect was observed when added only during viral infection. It maintained antiviral activity at higher levels when added to the medium even after 6 h of virus infection. In addition, it was found to make the virion lose its infectivity. In contrast, it did not show anti-influenza A virus effect. The antiviral activity of enzymatic extracts of *C. fragile* from North Atlantic coasts were also investigated[37]. They examined the enzymatic hydrolysates exhibited significant activity against Herpes simplex virus (HSV-1) with EC50 of 77.6–126.8 µg/mL, at a multiplicity of infection (MOI) of 0.001 ID50/cells without cytotoxity (1–200µg/mL).

Anticancer activities

The Clerosterol isolated from *Codium fragile* and studied the cytotoxic effects and mechanism of action in A2058 human melanoma cells[65]. They reported that, Clerosterol inhibited the growth of A2058 cells with an IC50 of 150µM and induced apoptotic cell death, as evidenced by DNA fragmentation, an increase in the number of sub-G1 hypodiploid cells and the presence of apoptotic bodies. Clerosterol treatment caused the loss of mitochondrial membrane potential. Alterations in the expression of apoptosis-associated proteins in response to clerosterol treatment included upregulation of Bax, downregulation of Bcl-2 and activation of caspases 3 and 9. The pan-caspase inhibitor treatment attenuated the expression of the active form of Caspases and cell death induced by clerosterol.

Since the anti-angiogenic therapy has becoming a promising approach in the prevention of cancer and related diseases, the anti-angiogenic effect of siphonaxanthin from *Codium fragile* was examined by using cell culture model systems and ex vivo approaches in human umbilical vein endothelial cells (HUVECs) and rat aortic ring, respectively[66]. Siphonaxanthin significantly suppressed HUVEC proliferation ($p < 0.05$) at the concentration of 2.5M(50% as compared with control) and above, while the effect on chemotaxis was not significant. It suppressed the formation of tube length by 44% at the concentration of 10M, while no tube formation was observed at 25M, suggesting that it could be due to the suppression of angiogenic mediators. The ex vivo angiogenesis assay exhibited reduced microvessel outgrowth in a dose dependent manner and the reduction was significant at more than 2.5M. These results imply a new insight on the novel function of siphonaxanthin in preventing angiogenesis related diseases.

Other activities of Oyster thief

The effects of sulfate and protein contents as well as molecular weights of the sulfated glycoproteins (NF2) from *Codium fragile* on the immunomodulation were studied[67]. NF2 was not able to stimulate

RAW264.7 cells to release NO without its protein moiety, which was essential to activate NF-κB pathway through the degradation and phosphorylation of IκB-α and the subsequent translocation of p65/p50 complex in the cell nucleus. In addition, the proteins in NF2 were required to trigger MAPK pathway for the phosphorylation of ERK1/2, p38, and JNK1/2 as well as the nuclear translocation of c-JUN and c-FOS. However, the protein moiety itself could not activate RAW264.7 cells, thus the complex formation of the polysaccharide and protein moieties in NF2 was pivotal to stimulate macrophage cells.

The binding of a sulphated arabinogalactan from *C. fragile* to IL-2, IL-7 and gamma interferon (INF- γ) were investigated[68]. This polysaccharide appears to bind cytokines to a similar extent as λ-, κ- and ι-carrageenans. Sulphated polysaccharides could be useful for cytokine-based therapies, as they help to concentrate cytokine molecules close to their site of secretion and also protect them against proteolytic degradation.

Codium fragile from Gulf of california exhibited significant antifouling activity with minimum inhibitory concentration (MIC) between 1-10μg/ml^{-1} against marine microalgae *Rhodosorus magnei, Neorhodella cyanea*and and *Prymnesium calathiferu*[69]. Table 4 gives the bioactivities of extracts of *Codium fragile* from different geographical locations.

Table 4: Bioactivities of extracts of *Codium fragile* from different geographical locations

Biological Property	Type of extract / Compounds	Study area	Reference
Anti-angiogenesis	Siphonaxanthin from acetone extract	Japan	66
Cytotoxicity	Clerostrerol	Korea	65
Anti inflammatory, Antipyretic, and Analgesic activities	Dicholromethane, ethanol and hot water extracts	Korea	59
Anti-inflammatory	Ethanolic extracts	Korea	58
	Methanol extract	Korea	70
	Ethanol extract	Korera	57
	Sulfated glycoproreins from aquous extract	Korea	32
Antiviral	Enzymatic extracts	Canada	37
	Methanol extracts	Korea	71
	Sulfated galactan from Aquous extract	Japan	48
Hemostasis	Polysaccharides from aquous extract	Argentina	55
Anticoagulant	Sulfated polysaccharides from Hot water extract	Korea	54
Thrombolytic, anticoagulant and antiplatelet activities	Codiase, a bi-functional fibrinolytic enzyme from aqueous extract	Korea	56
Antioxidant and antimicrobial	methanol, dichloromethane and hexane extracts	Turkey	60
Antioxidant	Methanol and aqueous extract	Portugal	63
Antibacterial	Ethyl acetate and methanolic extract	Egypt	59
	Methanolic extracts	Turkey	62
Immunostimulation	Sulphated galactan	Japan	44
	Anionic macromolecules from aquous extract	Korea	67
Antifouling	Ether extract	California	69

Ecological role

Codium fragile grows along intertidal zones close to shore, or along the shore in caverns and tidepools. It prefers areas that are unlikely to freeze, and grows well in sheltered areas including harbors and bays. It is a source of food for many invertebrate species, though not usually a primary food source. *Codium* provides an

important service for the Alesia seaslug, which takes chloroplasts from its siphonous branches, and stores them in a gland where the choroplasts help to produce the slime used by the slug in locomotion. In new habitats, *Codium* displaces native seagrasses and seaweeds, disrupting life for the multitude of fish and invertebrate species they contain[70].

Conclusion

Seaweeds have been used as natural materials from which to extract bioactive substances over the past 20 years because of their widespread distribution and large biomass. They are usually collected for food consumption and especially known for their high nutritional value and health benefits. Marine green algae remain unexploited among the three main divisions of macroalgae (*i.e.*, Chlorophyta, Phaeophyta, and Rhodophyta). But it has been recently taken up because of their many active ingredients, particularly those that used for medical purposes. However, many different mechanisms of action have been proposed but the structure of the active compounds was usually not elucidated. Recently, it has been carried out using purified fractions.

Codium species have drawn attention due to their high anticoagulant activity. Among them *Codium fragile* (Oyster thief), have vital functions for human health and nutrition. Recent studies have also reported that it has antioxidant, antiviral, anti cancer activities and mainly using as remedies for inflammation-related symptoms, anticoagulant and platelet disaggregation properties, and lack of toxicity make it a potential agent for thrombolytic therapy. It provides different bioactive compounds of great chemical diversity and different structural units with distinct bioactivities from various places in different seasons. Therefore, further intensive studies are needed to exploit its new compounds for maximum therapeutic potential in the field of medicinal and pharmaceutical sciences.

References

1. Dembitsky V.M, Rezankova H, Rezanka T, Hanus L.O., Variability of the fatty acids of the marine green algae belonging to the genus *Codium*, Biochemical Systematics and Ecology, 2003, 31, 1125-1145.
2. El Gamal A.A., Biological importance of marine algae, Saudi Pharmceutical Journal, 2010, 18, 1–25.
3. Druehl L. Pacific Seaweeds. Harbour Publishing, Madeira Park, B.C. Canada. 2000, 122.
4. Dhargalkar V.K, Pereira N., Seaweed: Promising plant of the millennium. Scitific Culture, 2005, 71, 60-6.
5. Khotimchenko S.V, Vaskovsky V.E, Titlyanova T.V., Fatty acids of marine algae from the Pacific coast of North California, Bot Mar, 2005, 45, 17–22.
6. Indu. H, Seenivasan. R.., In vitro antioxidant activity of selected seaweeds from southeast coast of india, Intenational journal of pharmacological science, 2013, 5(2), 474-484.
7. Kim K.Y, Garbary D.J., Photosynthesis in Codium fragile (Chlorophyta) from a Nova Scotia estuary: responses to desiccation and hyposalinity. Marine Biology, 2007, 151, 99-107.
8. Angélica Ribeiro Soares, Marcela C.S. Robaina, Gabriella S. Mendes Thalia S. L. Silva, Lísia M. S. Gestinari, Odinéia S. Pamplona, Yocie Yoneshigue valentine Carlos R. Kaiser, Maria Teresa Villela romano., Antiviral activity of extracts from Brazilian seaweeds against herpes simplex virus, Brazilian Journal of Pharmacognosy, 2012, 22(4), 714-723.
9. Artan M, Li Y, Karadeniz F, Lee S.H, Kim M.M, Kim S.K., Anti-HIV-1 activity of phloroglucinol derivative, 6,6'-bieckol, from *Ecklonia cava*, Bioorganic and Medicinal Chemistry, 2008, 16, 7921-7926.
10. Ana Iris Frías Vázquez, Carlos Manuel Dutok Sánchez, Neivys García Delgado, Ana María Suarez Alfonso, Yúlica Santos Ortega , Hiran Cabrera Sánchez., Anti-inflammatory and analgesic activities of red seaweed *Dichotomaria obtusata*, Brazilian Journal of Pharmaceutical Sciences, 2011, 47, 111-118.
11. Kang J.W., Illustrated Encyclopedia of Fauna & Flora of Korea: Marine Algae, Samhwa Press, Seou 1968, 85-90.
12. Park H.K, Kim J, Nam T.J., Induction of apoptosis and regulation of erbb signaling by laminarin in HT-29 human colon cancer cells, Int. J. Mol. Med., 2013, 32, 291-295.
13. Kazlowska K, Victor Lin H.T, Chang S.H, Tsai G.J., In vitro and in vivo anticancer effects of sterol fraction from red algae Porphyra dentata. Evid. Based Complement, Alternaternative and complementary Medicine, 2013, 4(9), 38-69.

14. Pushpamali W.A, Nikapitiya C, Zoysa M.D, Whang I, Kim S.J, Lee J., Isolation and purification of an anticoagulant from fermented red seaweed *Lomentaria catenata*, Carbohydrate Polymers, 2008, 73, 274-9.
15. Jos´e Manuel Estevez, Paula Virginia Fern´andez, Luciana Kasulin, Paul Dupree, and Marina Ciancia., Chemical and in situ characterization of macromolecular components of the cell walls from the green seaweed *Codium fragile,* Glycobiology., 2009, 19(3), 212–228.
16. Ramani Gade, Siva Tulasi M, Aruna Bhai V. Seaweeds: A Novel Biomaterial. International Journal of Pharmacy and Pharmaceutical Sciences, 2013, 5(2), 0975-1491.
17. Oliveira-Carvalho M.F, Oliveira M.C, Barreto-Pereira S.M, Verbruggen H., Phylogenetic analysis of Brazilian *Codium* species, including the new species *C. Pernambucensis*, European Journal of Phycology, 2012, 47, 355-365.
18. Verbruggen H, Leliaert F, Maggs C.A, Shimada S, Schils T, Provan J., Species boundaries and phylogenetic relationships within the green algal genus *Codium* (Bryopsidales) based on plastid DNA sequences, Molecular Phylogenetics and Evolution, 2007, 44, 240-254.
19. Yuko Ohta, Kyoko Hayashi, Toshimitsu Hayashi, Jung-Bum Lee., Immunostimulating effects of a sulfated galactan from *Codium fragile*, Carbohydrate Research., 2010, 345, 1452-1454.
20. Chavanich S, Harris L.G, Je JongGeel, Kang RaeSeon., Distribution pattern of the green alga Codium fragile (Suringar) Hariot, 1889 in its native range, Korea, Aquatic Invasions, 2006, 1(3), 99-108.
21. Provan J, Murphy S, Maggs C.A., Tracking the invasive history of the green alga Codium fragile ssp. Tomentosoides, Molecular Ecology, 2005, 14(1), 189-194.
22. Centre for Research on Introduced Marine Pests (CRIMP), Introduced species survey, Port of Albany, Western Australia. CRIMP Technical Report Series, CSIRO, 1997, 41.
23. Trowbridge C.D., Ecology of the green macroalga Codium fragile (Suringar) Hariot 1889: Invasive and non-invasive subspecies, Oceanography and Marine Biology, 1998, 36, 1-64.
24. Justin I. Mcdonald, John M, Huisman, Felicity N. Hart, Rainbo RM. Dixon, John A, Lewis., The first detection of the invasive macroalga Codium fragile subsp. Fragile (Suringar) Hariot in Western Australia, Bioinvasions Records, 2015, 4(2), 75-80.
25. Guiry M.D, Guiry G.M., Algaebase. World-wide electronic publication, National University of Ireland, Galway. Http://www.algaebase.org, searched on 20 July 2009.
26. Brodie J, Maggs C.A, John D.M, Green seaweeds of Britain and Ireland, British Phycological Society, London, 2007, 242.
27. Bouck G.B, Morgan E., The occurrence of *Codium* in Long Island waters, Bulletin of the Torrey Botanical Club, 1957, 84(5), 384-387.
28. Malinowski K.C, Ramus J., Growth of the green alga Codium fragile in a Connecticut estuary, Journal of Phycology, 1973, 9,102-110.
29. Hubbard C.B, Garbary D.J., Morphological variation of Codium fragile (Chlorophyta) in eastern Canada, Botanica Marina, 2002, 45, 476-485.
30. Kusakina J, Snyder M, Kristie D.N, Dadswell M.J., Morphological and molecular evidence for multiple invasions of Codium fragile in Atlantic Canada, Botanica marina, 2006, 49, 1-9.
31. Kim K.Y, Garbary D.J., Photosynthesis in Codium fragile (Chlorophyta) from a Nova Scotia estuary: responses to desiccation and hyposalinity, Marine Biology, 2007, 151, 99-107.
32. Mathieson A.C, Dawes C.J, Harris L.G, Hehre E.J. Expansion of the Asiatic green alga Codium fragile subsp. Tomentosoides in the Gulf of Maine, Rhodora, 2003, 105(921), 1-53.
33. González A, Santelices B., A dichotomous species of Codium (Bryopsidales, Chlorophyta) is colonizing northern Chile, Revista Chilena de Historia Natural, 2004, 77, 293–304.
34. Trowbridge C. Establishment of the green alga *Codium fragile* ssp. Tomentosoides on New Zealand rocky shores: current distribution and invertebrate grazers. Journal of Ecology, 1995, 83, 949-965.
35. Caccamese S, Azzolina D, Furnari G, Cormaci M, Grasso S. Antimicrobial and antiviral activities of some marine algae from eastern Sicily. Botanica Marina, 1981, 24, 365-367.
36. Mehdi Tabarsaa, Supatra Karnjanapratuma, myounglae Cho, Jin-Kyung Kimc, sangguan You. Molecular characteristics and biological activities of anionic macromolecules from *Codium fragile*. International Journal of Biological Macromolecules, 2013, 59, 1-12.
37. Garima Kulshreshtha, Anne-Sophie Burlot, Christel Marty, Alan Critchley, Jeff Hafting, Gilles Bedoux, Nathalie Bourgougnon and Balakrishnan Prithiviraj., Enzyme-Assisted Extraction of Bioactive Material from *Chondrus crispus and Codium fragile* and Its Effect on Herpes simplex Virus (HSV-1), Marine Drugs, 2015, 13, 558-580.

38. Jaime Ortiz, Edgar Uquiche, Paz Robert, Nalda Romero, Vilma Quitra, Catherine Llantn., Functional and nutritional value of the Chilean seaweeds Codium fragile, Gracilaria chilensis and Macrocystis pyrifera, European Journal of Lipid Science and Technology, 2008, 110, 1-10.
39. Ortiz J.E, Uquiche P, Robert N, Romero V, Quitral, Llantén C. Functional and nutritional value of the Chilean seaweeds Codium fragile, Gracilaria chilensis and Macrocystis pyrifera, European Journal of Lipid Science and Technology, 2009, 111, 320-327.
40. Ender F, Godl K, Wenzl S, Sumper M., Evidence for autocatalytic cross-linking of hydroxiproline-rich glycoproteins during extracellular matrix assembly in Volvox, Plant Cell, 2002, 14, 1147-1160.
41. Hallman, A., The pherophorins, common, versatile building blocks in the evolution of extracellular matrix architecture in Volvocales, Plant Journa, 206, 45, 292-307.
42. Estevez J. M., Fernández P. V., Kasulin L., Dupree P., and Ciancia M., Chemical and in situ characterization of macromolecular components of the cell walls from the green seaweed *Codium fragile,* Glycobiology, 2009, 19(3), 212–228.
43. Love J, Percival E., The polysaccharides of the green seaweed *Codium fragile*, part II. The water-soluble sulphated polysaccharides, Journal of the Chemical Society, 1964, 3338-3345.
44. Lee J.B, Ohta Y, Hayashi K, Hayashi T., Immunostimulating effects of a sulfated galactan from *Codium fragile*, Carbohydr. Res., 2010, 345,1452-1454.
45. Ciancia M, Quintana I, Vizcarguenaga MI, Kasulin L, Dios A, Estevez J.M., Polysaccharides from the green seaweeds *Codium fragile* and *C. Vermilara* with controversial effects on hemostasis, International Journal of Biological Macromolecules, 2007, 41, 641649.
46. Ferda Yilmaz Koz N, Ulku Karabay Yavasoglu, Zeliha Demire, Atakan Sukatar And Guven Ozdemir. Antioxidant and Antimicrobial Activities of Codium fragile (Suringar) Hariot (Chlorophyta) Essential Oil and Extracts. F. Asian Journal of Chemistry, 2009, 21 (2), 1197-1209.
47. Jurd K.M, Rogers D.J, Blunden G., Mclellan D.S., Anticoagulant properties of sulfated polysaccharide and a proteoglycan from Codium fragile ssp atlanticum, Journal of Applied Phycology, 1995, 7, 339-345.
48. Ohta Y, Lee J.B, Hayashi K, Hayashi T., Isolation of sulphated galactan from *Codium fragile* and its antiviral effect, Biological and Pharmaceutical Bulletin, 2009, 32, 892-898.
49. Hayakawa Y, Hayashi T, Lee J.B, Srisomporn P, Maeda M., Ozawa T, Inhibition of thrombin by sulphated polysaccharides isolated from green seaweeds, Biochimica et Biophysica Acta, 2000, 1543(1), 86-94.
50. Mehdi Tabarsaa, Supatra Karnjanapratuma, myounglae Cho, Jin-Kyung Kimc, sangguan You., Molecular characteristics and biological activities of anionic macromolecules from *Codium fragile*, International Journal of Biological Macromolecules, 2013, 59, 1-12.
51. Chanzy H.D, Grosrenaud A, Vuong R, and Mackie W., The crystalline polymorphism of mannans in plant cell walls and after recrystallisation, Planta, 1984, 161, 320-329.
52. Albersheim P, Darvill A, Roberts K, Sederoff R, and Staehelin A., Plant cell walls: from chemistry to biology. New York, USA: Garland Science. Taylor and Francis Group. 2011, 430.
53. Albertus J Smith., Medicinal and pharmacologiacal uses of seaweed: A review, Journal of applied phycology, 2004, 16, 245-262.
54. Athukorala Y, Lee K.W, Kim S.K, Jeon Y.J., Anticoagulant activity of marine green and brown algae collected from Jeju Island in Korea, Bioresource Technology, 2007, 98, 1711-1716.
55. Marina Ciancia, Irene Quintana, Mar´ıa Isabel Vizcarg¨u´enaga, Luciana Kasulin, Agustina de Dios, Jos´e Manuel Estevez, Alberto Sa´ul Cerezo., Polysaccharides from the green seaweeds *Codium fragile* and *C. Vermilara* with controversial effects on hemostasis, International Journal of Biological Macromolecules, 2007, 41, 641-649.
56. Jun-Hui Choi, Kumar Sapkota, Se-Eun Park, Seung Kim, Sung-Jun Kim., Thrombolytic, anticoagulant and antiplatelet activities of codiase, a bi-functional fibrinolytic enzyme from Codium fragile, Biochimie, 2013, 95(6), 1266-1277.
57. Han Sinhee, Kim Youngguk, Lee Suhuan, Park Chungberm, Han Seungwon, Jang Hyejin, Lee Hyojeong, Park Seongcheol, Kim Hyesung, Lee Youngseob, And Kwon Dongyeul., Anti-inflammatory activity of *Codium fragile* in macrophages induced by Peptidoglycan, Natural Product Sciences, 2010, 16(3), 153-158.
58. Ho-Dong Yoon, Eun-Ji Jeong, Ji-Woong Choi, Min-Sup Lee, Myoung-Ae Park, Na Young Yoon, Yeon-Kye Kim, Deuk Moon Cho, Jae-Il Kim, Hyeung-Rak Kim., Anti-inflammatory Effects of

Ethanolic Extracts from *Codium fragile* on LPS-Stimulated RAW 264.7 Macrophages via Nuclear Factor kappab Inactivation, Fisheries and Aquatic Sciences, 2012, 14(4), 267-274.
59. Ji-Young Kang, Quoc-Hai Luyen, Mohammed Nurul Absar Khan, Jae-Suk Choi., In vivo Anti-inflammatory, Antipyretic, and Analgesic Activities of the Aquaculturable Green Seaweed Codium fragile Extracts in Mice, Journal of Life Science, 2012, 22(6), 852-856.
60. Ferda Yilmaz Koz N, Ulku Karabay Yavasoglu, Zeliha Demire, Atakan Sukatar And Guven Ozdemir., Antioxidant and Antimicrobial Activities of Codium fragile (Suringar) Hariot (Chlorophyta) Essential Oil and Extracts, F. Asian Journal of Chemistry., 2009, 21 (2), 1197-1209.
61. Saleem W.M, Galal H, Nasr El-deen F., Screening for antibacterial activities in some marine algae from the red sea (Hurghada, Egypt) Salem, African Journal of Microbiology Research, 2011, 5(15), 2160-2167.
62. Turkey Evrim Taşkin, Ergün Taşkin, Mehmet Öztürk., Inhibitor Activities of some Seaweed from the Aegean Coast, Journal of Applied Biological Sciences, 2011, 5(1), 11-15.
63. Rathinam raja, Shanmugam Hemaiswarya K, Arunkumar Isabel S, Carvalho., Antioxidant activity and lipid profile of three seaweeds of Faro, Portugal. Braz, Journal of Botany, 2015, 1-9.
64. Soo-Jin Heo, Sun-Heui Cha, Ki-Wan Lee, Somi K. Cho, You-Jin Jeon., Antioxidant Activities of Chlorophyta and Phaeophyta from Jeju Island, Algae, 2005, 20(3), 251-260.
65. Areum Daseu Kim, Youngki Lee, Sang-Hyuck Kang, Gi Young Kim, Hye Sun Kim and Jin Won Hyun., Cytotoxic Effect of Clerosterol Isolated from *Codium fragile* on A2058 Human Melanoma Cells, Marine Drugs, 2013, 11, 418-430.
66. Ponesakki Ganesana, Kiminori Matsubarab, Takeshi Ohkubоc, Yukihisa Tanakac, Kenji Nodaa, Tatsuya Sugawaraa, Takashi Hirataa., Anti-angiogenic effect of siphonaxanthin from green alga, *Codium fragile*. Phytomedicine, 2010, 17, 1140-1144.
67. Mehdi Tabarsa, Geun-Mook Park, Il-Shik Shin, eunjung Lee, Jin-Kyung Kim, sangguan., Structure-Activity Relationships of Sulfated Glycoproteins from Codium fragile on Nitric Oxide Releasing Capacity from RAW264.7 Cells, Marine Biotechnology, 2015, 17 (3), 266-76.
68. Nika K, Mulloy B, Carpenter B, Gibbs R. Specific recognition of immune cytokines by polysaccharides from marine algae, European Journal of Phycology., 2003, 38, 257-264.
69. Ruth Noemí Águila–Ramírez, Anabel Arenas–González, Claudia Judith Hernández–Guerrero, Bárbara González–Acosta, José Manuel Borges–Souza, Benoit Véron, Josephine Pope and Claire Hellio, Antimicrobial and antifouling activities achieved by extracts of seaweeds from Gulf of California, Mexico. Hidrobiológica, 2012, 22(1), 8-15.
70. Massoumeh Farasata, Ramazan-Ali Khavari-Nejada., Antioxidant Activity, Total Phenolics and Flavonoid Contents of some Edible Green Seaweed from Northern Coasts of the Persian Gulf, Iranian Journal of Pharmaceutical Research, 2014, 13(1), 163-170.
71. Hudson J.H, Kim M.K, Lee R.E, Dewreede, Hong Y.K., Antiviral compounds in extracts of Korean seaweeds: Evidence for multiple activities. Journal of Applied Phycology, 1999, 10, 427-434.
72. NIMPIS (2009). Codium fragile ssp fragile general information. National Introduced Marine Pest Information System. Web publication (http://www.marinepests.gov.au/nimpis, date of access: 21/07/2009).

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