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ANTIOXIDANT ACTIVITIES OF MARINE ALGAE: A REVIEW

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ABSTRACT

Oxidative stress is the result of an imbalance between pro-oxidant and antioxidant homeostasis that leads to the generation of toxic reactive oxygen species (ROS). The necessity of compounds with antioxidant activity is increasing as it is realized that the formation of reactive oxygen species (ROS) and reactive nitrogen species (RNS) have been linked in the pathogenesis of several human diseases such as atherosclerosis, diabetes mellitus, chronic inflammation, neurodegenerative disorders and certain types of cancer. The antioxidant activity of these compounds are mainly attributed to scavenging activity against superoxide and hydroxyl radicals, chelating ability, quenching singlet and triplet oxygen, and reducing power. It is important to develop, identify and utilize new source of safe and effective antioxidants of natural origin. Recently, much research attention has been focused on the free-radical-scavenging activity of metabolites from marine macro algae. Several studies have investigated the antioxidant activity of natural products in marine and freshwater algae. The marine environment is known as a rich source of chemical structures with numerous beneficial health effects. Among marine organisms, marine algae have been identified as an under-exploited plant resource, although they have long been recognized as valuable sources of structurally diverse bioactive compounds. Here summarized what are the compounds, methods and recent research on antioxidant activities of marine algae.

INTRODUCTION

Oxidative stress is the result of an imbalance between pro-oxidant and antioxidant homeostasis that leads to the generation of toxic reactive oxygen species (ROS). The necessity of compounds with antioxidant activity is increasing as it is realized that the formation of reactive oxygen species (ROS) and reactive nitrogen species (RNS) have been linked in the pathogenesis of several human diseases such as atherosclerosis, diabetes mellitus, chronic inflammation, neurodegenerative disorders and certain types of cancer (Collier *et al.*, 1990; Boynes, 1991). Oxygen free radicals disintegrate DNA, destroy cell membranes, and create havoc among cell's basic enzymatic metabolic processes (Kerr *et al.*, 1991). Among the major causative factors in induction of many chronic and degenerative diseases including atherosclerosis, diabetes mellitus, cancer, Parkinson's disease and immune dysfunction and is involved in aging (Halliwell, 2000; Metodiewa and Koska, 2000, Young & Woodside, 2001).

The formation of cancer cell in human body can be directly induced by free radicals. Furthermore, ionizing radiation, which causes free radicals, is well documented as a carcinogen. Radicals which have one or more unpaired electrons are produced in normal or pathological cell metabolism. Free Reactive oxygen species (ROS) react easily with free radicals to become radicals themselves. ROS are various forms of activated oxygen, which include free radicals such as superoxide anion radicals (O2) and hydroxyl radicals (OH), as well as non-free radical species (H2O2) and the singled oxygen (O2). The antioxidant activity of these compounds are mainly attributed to scavenging activity against superoxide and hydroxyl radicals, chelating ability, quenching singlet and triplet oxygen, and reducing power (Ruberto *et al.*, 2001 ; Athukorala *et al.*, 2006) . Antioxidants are used to preserve food quality mainly by prevention of oxidative deterioration of constituent of lipids. It is important to develop, identify and utilize new source of safe and effective antioxidants of natural origin (Li *et al.*, 2007; Qi

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et al., 2005). There are several synthetic antioxidants such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), Propylgallate (PG) and butylated hydroxyquinone (TBHQ) are commercially available and currently used.

However, the use of these synthetic antioxidants for food or medicine components has been stricted by the toxicity and safety that can lead to the problems of the potential health in human. Due to the reasons, many researchers have tried to find the more effective oxidation inhibitors that may be used as antioxidants for food or medicine compositions without the side effects for the past several years. So many researchers have paid attention to many kinds of natural antioxidants that can be used without toxicity in human.

Algae are found everywhere on earth: in the sea, rivers and lakes, on soil and walls, in animal and plants (as symbionts-partners collaborating together); in fact just about everywhere where there is a light to carry out photosynthesis. The beginnings of algae research were dated in 1768 when Samuel Gottlieb Gmelin developed the work Historia Fucorum. He was followed by W.H. Harvey (1811—1866) who was the first who divided the algae into four divisions, based on their pigmentation. This was the first use of a biochemical criterion in plant systematics. Harvey's four divisions are: Red Algae (Rhodophyta), Brown Algae (Heteromontophyta), Green Algae (Chlorophyta) and Diatomaceae (Dixon, 1973). Seaweeds are marine plants, divided into three categories based on their colors, such as red (4,500 species), green (900 species) and brown (1,000 species).

These organisms constitute a total of 25-30,000 species, with a great diversity of forms and sizes, and that can exist from unicellular microscopic organisms (microalgae) to multi cellular of great size (macroalgae). Recently, much research attention has been focused on the free-radical-scavenging activity of metabolites from marine macro algae. Considerable work has been done on natural products for the presence of nontoxic antioxidants that could be used in chemotherapy. Several studies have investigated the antioxidant activity of natural products in marine and freshwater algae (Fujimoto & Kaneda, 1984; Matsukawa *et al*., 1997; Lim *et al.*, 2002; Xue *et al.*, 2004). Marine algae, like all photosynthesizing plants are exposed to a combination of light and high oxygen concentrations, which lead to the formation of free radicals and other strong oxidizing agents. The elements of the photosynthetic apparatus are vulnerable to photodynamic damage, because polyunsaturated fatty acids are important structural components of the thylakoid membrane (Sukenik *et al.,* 1993). The absence of such damage in seaweeds, in spite of the proximity of the photosynthetically produced oxygen and suitable target within the photosynthetic apparatus suggests that these cells have protective antioxidative mechanism and compounds (Matsukawa *et al.*, 1997; Lim *et al.*, 2002).This review, however, focuses specifically on the antioxidant activities and compounds of marine algae.

Antioxidant compounds from algae

The marine environment is known as a rich source of chemical structures with numerous beneficial health effects. Among marine organisms, marine algae have been identified as an under-exploited plant resource, although they have long been recognized as valuable sources of structurally diverse bioactive compounds. Marine algae are an important source of bioactive ingredients that can be applied to many aspects of processing healthier foods and developing functional protective foods. Seaweeds are known to contain reactive antioxidant molecules, such as ascorbate and glutathione (GSH) when fresh, as well as secondary metabolites, including carotenoids (α- and β-carotene, fucoxanthin, astaxanthin), mycosporine-like amino acids (mycosporine-glycine) and catechins (e.g., catechin, epigallocatechin), gallate, phlorotannins (e.g., phloroglucinol), eckol and tocopherols (α-, χ-, δ-tocopherols) (Yuan *et al.*, 2005). Several prenyl toluquinones were isolated from the brown alga *Cystoseira crinita.* Compounds exhibited potent radical-scavenging effects (Fisch *et al.,* 2003).The Brown alga *Ecklonia stolonifera* collected from South Korea yielded a new phlorotannin, eckstolonol which possessed a potent DEPP radical-scavenging activity (Kang *et al*., 2003). Brown alga *Sargassum thunbergii* afforded a novel chromene, sargothunbergol A, as a free radical scavenger (DPPH assay) (Seo *et al.*, 2007). Two monogalactosyldiacylglycerols were isolated from *S. thunbergii* (Kim *et al.*, 2007). Fucodiphlorethol G, a tetrameric phlorotannin, was isolated from *Ecklonia cava*, and was a strong radical scavenger (DPPH assay) (Ham *et al.*, 2007). Among macroalgal natural antioxidants, terpenoids, phlorotannins, polyphenols, phenolic acids, anthocyanins, hydroxycinnamic acid derivatives, and flavonoids are important (Bandoniene and Murkovic, 2002). Flavonoids have potent, anti-allergic, anti-viral and have free radical scavenging abilities and also provide protection against cardiovascular mortality. However, these synthetic antioxidants have side effects such as liver damage and carcinogenesis (Wichi, 1988). Recently, researchers have isolated various types of antioxidant compounds from different algal species, including fucoxanthin in *Hijikia fusiformis* (Yan *et al.,* 1999); phycocyanin and phycocyanobilin in *Spirulina platensis* and *Aphanizomenon flos-aquae* (Bhat & Madyastha 2000; 2001, Hirata *et al.,* 2000; Romay and Gonzalez 2000; Benedetti *et al*., 2004); phenolic acids, tocopherols, and b-carotene in *Spirulina maxima* (Miranda *et al*. 1998); fatty acids, polyphenols, and phlorotannin in *Sargassum kjellamanianum, S.siliquastrum, Rhodomela confervoides, Symphjocladia latiuscula* and *Kappaphycus alvarezzi* (Yan *et al*., 1996, Lim *et al.,* 2002, Huang and Wang 2004, Fayaz *et al*. 2005); lutein in *Botryococcus braunii* (Rao *et al*. 2006); and carotenoids in *Dunaliella salina* (Ben-amotz and Avron 1988, Neuman *et al.,* 1999, Levy *et al.,* 2000, Herrero *et al*., 2006).

METHODOLOGY

Among the features of marine algae and their substance, several extracts were screened on an antioxidant

capability (Latham, 2008), and their inhibitory activity on lipoxygenase enzyme (Mori *et al.*, 2003)

DPPH radical scavenging activity

A rapid, simple and inexpensive method to measure antioxidant capacity of algae involves the use of the free radical, 2, 2-Diphenyl-1-picrylhydrazyl (DPPH). DPPH is widely used to test the ability of compounds to act as free radical scavengers or hydrogen donors, and to evaluate antioxidant activity of algae as foods. It has also been used to quantify antioxidants in complex biological systems in recent years. The DPPH method can be used for solid or liquid samples and is not specific to any particular antioxidant component, but applies to the overall antioxidant capacity of the sample Yan and Chen (1995). Briefly, 50 µl of 0.16m M 2, 2-Diphenyl-1-picrlyhydrazyl (DPPH) solution in methanol were added to 50 µl aliquot of sample. The mixture was vortexed for 1 min and kept at room temperature for 30 min in the dark. The absorbance of all the sample solutions was measured at 517 nm. The scavenging effect (%) was calculated by using the formulae given by Duan *et al.*, 2006) % Inhibition = ((Blank-Test)/Blank) * 100, Blank and control samples were performed. Shimada *et al*. (1992) DPPH solution was prepared at the concentration of 0.1 mM in ethanol. During the assay, the 1 ml of test solution (concentration of 0.5-3.5 mg/ml) was mixed with 1ml DPPH solution. The mixture was incubated in dark place for 30 min at 25°C. After standing for 30 min, absorbance was recorded at 517 nm by UV Beckman spectrophotometer (Beckman Coulter). The percentage of DPPH free radicals scavenging activity I= [1-(A1-A2)/A0] × 100%. A0, A1 and A2 are the absorbance of the control (without test solution), the presence of the test solution, and without DPPH, respectively. (Blois, 1958) Different concentration of sample 50, 100 and 150µl (0.25,0.5 &0.75 mg) of the extracts were taken in the test tubes. 3.0ml of 0.1mM DPPH in ethanol was added to each tube and incubated in dark at room temperature for 30minutes. The absorbance was read at 517nm using UV-visible spectrophotometer. BHT (butylated hydroxy toluene) was used in standard calibration. I % = [Abs (control) - Abs (sample)] x 100 Abs (control)

Superoxide anion radical scavenging

The superoxide radicals were generated by a pyrogallol autoxidation as described by (Jing and Zhao, 1995). Briefly, 9 mL of Tris-HCl buffer solution (50 m mol/L, pH=8.2) was added into a test tube, and contents was incubated in a water bath at 25°C for 20 min. A volume of 40 µL of pyrogallol solution (45 mM/L of pyrogallol in 10 m M/L of HCl), which was also pre-incubated at 25°C, was added to the aforementioned test tube. The mixture was incubated at 25°C for 3 min and then a drop of ascorbic acid was dripped into the mixture promptly to terminate the reaction. The absorbance at 420 nm was marked as *A0*. The same procedure was carried out with extracts instead of ascorbic acid and the absorbance was marked *A1*. A blank was run and noted as A2 and the scavenging percentage was calculated using the following formula:

% scavenging activity (I) = (*A*0–(*A*1–*A*2)) /A0*100

Estimation of reducing power

Reducing power of crude methanolic extract was determined as described by (Oyaizu 1986). Briefly, 1.0 ml of methanol containing different concentration of sample was mixed with 2.5 ml of phosphate buffer (0.2 M, pH 6.6) and 2.5 ml potassium ferricyanide (1%). Reaction mixture was incubated at 50°C for 20 min. After incubation, 2.5 ml of TCA (10%) was added and centrifuged (650 g) for 10 min. From the upper layer, 2.5 ml solution was mixed with 2.5 ml distilled water and0.5 ml FeCl3 (0.1%). 300 µl of the content was transferred into a microtitre plate and absorbance was measured at 695 nm. Increased absorbance indicates increased reducing power. The results were expressed in Ascorbic acid equivalents.

Total antioxidant activity

Total antioxidant activities of crude methanolic extract and fractions were determined as described by (Prieto *et al.*, 1999). Briefly, 0.3 ml of sample was mixed with 3.0 ml reagent solution (0.6 M sulfuric acid, 28mM sodium phosphate and 4 mM ammonium molybdate). Reaction mixture was incubated at 95°C for 90 min under water bath. 300 µl of the content was transferred into a micro titre plate and absorbance was measured at 695 nm. Total antioxidant activity was expressed as the number of equivalents of ascorbic acid in milligram.

Superoxide Anion Radical Scavenging Activity Assay

The superoxide anion radical scavenging activity of the test solution was determined by the method of (Nagai *et al.*, 2002) The volume of 0.48 ml of 0.05 M sodium carbonate buffer (pH10.5), 0.02 ml of 3 mM xanthine, 0.02 ml of 3 mM EDTA, 0.02 ml of 0.15% bovine serum albumin, 0.02 ml of 0.75 m MNBT and 0.02 ml of the test solution (concentration of 0.5-3.5mg/ml) were mixed together. After pre-incubating at 25°C for10 min, the reaction was started by adding 6 mU XOD and carried out at 25°C for 20 min. And the reaction was stopped by adding 0.02 ml of 6 mM CuCl2 after 20 min. The absorbance at 560 nm was measured, and the scavenging activity of the hot-water extract

Scavenging activity = (1-A1/A0) × 100%. A0 and A1 are the absorbance of the control (without test solution) and the presence of the test solution, respectively.

Determination of Total phenolic compounds

Phenolic compounds act as free radical scavengers, reducing agents and metal chelators, and thus effectively inhibit lipid oxidation.Total phenolic content was determined with Folin & Ciocalteau reagent according to the method described by (Singleton *et. al.*, 1999) using gallic acid as standard

Determination of Total Flavonoids

Total Flavonoids content was determined by the method described by (Zhishen *et. al.*, 1999)

RESULTS AND DISCUSSIONS

Various antioxidant activities of different algae

Seaweeds have received special attention as a source of natural antioxidants (Matsukawa *et al* . 1997) .*Ecklonia cava*, a kind of brown seaweed is plentifully produced in Jeju Island in Korea (30,000 tons per year), is not available in Europe; Many researchers have reported that Ecklonia species exhibits radical scavenging activity (Kang *et al*. 2004; Kang *et al*. 2003),, it has been reported that total polyphenolic compounds in *E. cava* are richer than in other brown seaweeds (Heo *et al*., 2005; Heo *et al.,* 2003). These polyphenolic compounds of brown seaweeds have been called as phlorotannins. And the phlorotannin components of *E. cava* that are phenolic secondary metabolites such as eckol (a closed-chain trimer of phloroglucinol), 6, 6-bieckol (a hexamer), dieckol (a hexamer), phlorofucofuroeckol (a pentamer) and triphlorethol-A have been known to be related to the biological activities (Kang *et al*., 2005a; 2005b).

Free radical scavenging (DPPH-decolorization method)

and inhibition of lipidperoxidation (Fe2+/Ascorbate) in three species of seaweeds (*Sargassum dentifolium*, *Laurencia papillosa and Jania corniculata* (Egyptian isolates) were evaluated. Three species of these seaweeds were collected from Defressoar (Suez Canal, Egypt) during spring 2004; Maximum free radical scavenging activity was exhibited by higher concentration of dichloromethane extract of *S. dentifolium* followed by *L. papillosa* and *J. corniculata*. Also, higher concentration of dichloromethane extract of *L. papillosa* had the maximum anti-lipid peroxidation activity followed by *S. dentifolium* and *J. corniculata*.

Padina minor Yamada is a brown alga found in abundance at the coastal area of the Gulf of Thailand and the Andaman Sea. The aqueous extract of *P. minor* (Aq. *P*) was found to show interesting pharmacological properties such as hypotensive activity (Amornlerdpison *et al*. 2007) and gastroprotective effect (Amornlerdpison *et al*., 2009). Aq. *P* exhibited antioxidant activity when tested by DPPH, ABTS•+ and lipid peroxidation assays (Amornlerdpison *et al*., 2007). Extracts from *Padina pavonica*, a brown marine alga, can be useful in the improvement of firmness of the skin applied or by softening the look of lines and wrinkles particularly on the face and/or hands (Isabelle and Hani, 2007).

Consuming seaweeds as sea vegetables has been a long tradition in the Far East and Pacific while the principal use of seaweeds in Western countries has been as source of thickening and gelling agents for different industrial applications including uses in foods (Jimenez-Escrig; Sanchez-muniz, 2000; Nagai and Yukimoto, 2003). Moreover, seaweeds are known to contain several compounds having health protective effects (Burtin, 2003). The potential antioxidant compounds in brown algae have identified as fucoxantine in *Hijikia fusidormis* and phylopheophytin in *Eisemia bicyclis* (Kuda *et al*, 2005a). Brown algae (Phaophyceae) like any one of the three groups of seaweeds differ from the other groups with regards to the reserve and cell wall polysaccharides. *Sargassum boveanum* is a marine species and its presence in the coastal waters of the Persian Gulf is confirmed (Algae Base, 2007; Sohrabipour and Rabii, 1990).

Recently (Henry *et al.,* 2000) reported the antioxidant activities of 29 commercially available C-8-C-24 saturated and unsaturated fatty acids. Most of the unsaturated fatty acids tested showed good antioxidant activities (Henry *et al.,* 2000). A literature survey revealed that most sea weeds are very rich in fatty acids, especially in the lipophilic extracts (Li *et al.,* 2002). (Huang & Wang 2004) Lipophilic extracts from 16 species of seaweeds collected along the Qingdao coastline were screened and evaluated for their antioxidant activities (AA) using the *â*-carotene-linoleate assay system. The diethyl ether soluble extracts of all selected seaweeds exhibited various degrees of antioxidative efficacy in each screen. The highest antioxidant capacities among the tested samples were observed for *Rhodomela confervoides* and *Symphyocladia latiuscula* and were comparable with that of the well-known antioxidant butylated hydroxytoluene and greater than that of propyl gallate. Marine green algae viz., Ulva sp, are an important food source in many south-east Asian countries. *Ulva fasciata* and *Ulva lactuca* are usedin soups and salads, and have been reported to possess antioxidant and antibacterial properties.

Antioxidant effect was observed with a sulfoglyco lipid fraction isolated from *Porphyridum creuntum* (Berge, *et al.,* 2002). Extracts from several macro algae harvested in Spain (Jiménez-Escrig, Jiménez-Jiménez, Pulido and Saura-Calixto, 2001), Korea (Han, Lee, & Sung, 1999), China (Yan, Nagata and Fan, 1998) and Japan have demonstrated antioxidant activity *in vitro*. The extracts of macroalga *Taonia atomaria* exhibited high radical-scavenging activity due to the compounds stypodiol and stypoldione (Mayer & Lehmann, 2000). Sargaquinoic acid from brown macroalga S*argassum macrocarpum* has been found to possess antioxidant activity (Tsang and Kamei, 2004), and therefore may be a potential food supplement. (Sadati *et al.,* 2011) Investigated possible antioxidant activity and total phenolic contents of three brown algae species (Sargassum swartzii, Cystoseira myrica, Colpomenia sinuosa) collected from Asaloye-Niband marine protected area of the Persian Gulf and found *S. swartzii* could be potential rich source of natural antioxidants which lots of them are known as phenolic compounds. Brown-algal polyphenols phlorotannins worked as antioxidants, antibacterial and anti-algal compounds (Kuda *et al.,* 2007; Shibata *et al.,* 2006). Phlorotannins purified from several brown algae have been reported to possess strong antioxidant activity which may be associated with their unique molecular skeleton. Phlorotannins from brown algae have up to eight interconnected rings. They are therefore more potent free radical scavenger than other polyphenols derived from terrestrial plants, including green tea catechins, which only have three to four rings (Hemat, 2007).

CONCLUSION

In conclusion, marine algae are a valuable natural source of antioxidant agents. Research is a crucial part of the response to new and emerging diseases. A sustained, forward-thinking applied research programs would enable scientists to identify the weak links in the amour of emerging diseases, create novel ways to fight free radicals, and evaluate the preventive power of new approaches. Algal species as alternative materials to extract natural antioxidative compounds have attracted much attention of biomedical scientists. Presenting the antioxidant activities and compounds of algae will lead the researchers for future research. Extraction of bioactive natural compounds from seaweeds is desired, but little has happened in this area to systematically study their potentiality. The priority for the next decades should be focused in the development of new alternative compounds and/or the recovery of natural molecules that would allow the consistent and proper control of many Reactive oxygen species (ROS) related diseases. Complacency and delay will have major detrimental effects on future public health. Seaweeds may be an answer to unsolved and growing global problems, a novel untapped source to combat various disorders. The mentioned microalgae exhibited various antioxidant activities against ROS and it would be an excellent candidate as a natural antioxidant source which can be applied in food and pharmaceutical industry. Detailed information and data on these activities need to be undertaken with individual species. There are number of challenges ahead like isolation of the antioxidant components present in the algae also *In vivo* testing on Human beings and further in large-scale controlled studies.

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References

Algae base – http://www.algaebase.org, 2007.

Berg, L.A. Bustamante, M. 1974. Heat treatment and meristem culture for the production of virus-free bananas. Phytopathology., 64: 320–322.

Amornlerdpison, D., Peerapornpisal, Y., Rujjanawate, C., Taesotikul, T., Nualchareon, M and Kanjanapothi, D. 2007. Hypotensive Activity of Some Marine Algae. J. Sci. Res. Chula, section T: 363-368.

Amornlerdpison, D., Peerapornpisal, Y., Taesotikul, T., Noiraksar, T., and Kanjanapothi D., 2009. Gastro protective activity of *Padina minor* Yamada. Chiang Mai J. Sci., 36 : 92-103.

Amornlerdpison, D., Peerapornpisal, Y., Rujjanawate, C., Taesotikul, T., Nualchareon, M and Kanjanapothi, D. 2007. Antioxidant activity of Padina minor Yamada, KMITL Sci. Tech. *J*., 7(s1):1-7.

Athukorala, Y., Nam, K. and Jeo, Y. 2006. Anti proliferative and antioxidant properties of an enzymatic hydrolysate from brown alga *Ecklonia cava*. Food Chem. Toxicol., 44: 1065-1074.

Bandoniene ,D., Murkovic, M. 2002. On-line HPLC–DPPH screening method for evaluation of radical scavenging phenols extracted from apples (*Malusdomestica* L.). Journal of Agricultural and Food Chemistry., 50: 2482–2487.

Boynes, J.W. 1991 .Role of oxidative stress in development of complication in diabetes. Diabetes., 40: 405-411.

Benedetti, S., Benvenuti F., Pagliarani, S., Francogli, S., Scoglio. S. and Canestrari, F. 2004. Antioxidant properties of a novel phycocyanin extract from the blue-green alga *Aphanizomenon flosaquae*. Life Sci., 75:2353–2362.

Berge, J.P., Debiton, E, Dumay, J., Durand, P. and Barthomeuf, C. 2002. *In vitro* anti-inflammatory and anti-proliferative activity of sulfolipids from the red alga *Porphyridium cruentum*. Journal of Agricultural and Food Chemistry., 50: 6227–6232.

Ben-Amotz, A., Avron, M.1988. The wavelength dependence ofmassive of carotene synthesis in *Dunaliella bardawil*. J. Phycol., 25:178–183.

Bhat, V .B., Madyastha, K.M. 2000. C-phycocyanin: apotent peroxyl radical scavenger *in vivo* and *in vitro*. Biochem. Biophys. Res. Commun., 275:20–25.

Bhat, V.B., Madyastha, KM. 2001. Scavenging of peroxynitrite by phycocyanin and phycocyanobilin from *Spirulina platensis*: protection against oxidative damage to DNA. Biochem. Biophys. Res. Commun, 285:262–266.

Blois, M.S. 1958. Antioxidant determinations by the use of a stable free radical. Nature, 181: 1199-1200.

Burtin, P. 2003. Nutritional value of seaweeds. Electronic J. Environmental Agric. Food Chem. (ISSN: 1579-4377), 2: 498-503.

Collier, A., Wilson, R., Bradley, H., Thomson, J.A. and Small, M. 1990. Free radical activity is type 2 diabetes. Diabetic Med., 7: 27-30.

Dixon, P.S. 1973. Biology of the Rhodophyta. *Oliver & Boyd*, Edinburgh, p. 285.

Duan ,X.J, Zhang, W.W., Li X.M., and Wang, B.G. 2006. Evaluation of antioxidant property of extract and fractions obtained from a red alga, *Polysiphonia urceolata*. Food Chem., 95: 37-43.

Fayaz, M., Namitha, K.K., Chidambara Murthy, K .N., Mahadeva Swamy, M., Sarada, R., Salma K., Neuman, I., Nahum, H. and Ben-Amotz, A. 1999. Prevention ofexercise-induced asthma by a natural isomer mixture of betacarotene.Ann. Allergy Asthma Immunol., 82:549–553.

Fujimoto, K., Kaneda. T. 1984. Separation of antioxygenic (antioxidant) compounds from marine algae. Hydrobiologia, 116: 111–113.

Kang, K., Park Y., Hwang, H.J., Kim, S.H, Lee, J.G and Shin, H.C. 2003. Antioxidative properties of brown algae polyphenolics and their perspectives as chemopreventive agents against vascular risk factors. Arch. Pharm. Res., 26: 286–293.

Kerr, M .E, Bender, C.M and Monti, E.J. 1991. An introduction to oxygen free radicals. Heart and Lung., 25 : 200-209.

Kim, Y.H., Kim, E.H., Lee, C., Kim, M.H. and Rho, J.R. 2007. Two new monogalactosyl diacylglycerols from brown alga *Sargassum thunbergii*. Lipid., 42: 395–399.

Fisch, KM., Bohm ,V., Wrightand, A.D and Konig, G.M. 2003. Antioxidative meroterpenoids from the brown alga *Cystoseira crinita*. J. Nat. Prod., 66l: 968–975.

Han, K.H, Lee, E. J., and Sung, M.K. 1999. Physical characteristics and antioxidative capacity of major seaweeds. Journal of Food Science and Nutrition., 4: 180–183.

Halliwell, B., 2000. The antioxidant paradox. Lancet., 355: 1179-1180.

Ham, YM., Baik, J.S., Hyun, J.W and Lee, N.H. 2007. Isolation of a new phlorotannin, fucodiphlorethol G, from a brown alga *Ecklonia cava*. Bull. Korean Chem. Soc., 28: 1595–1597.

Hirata, T., Tanaka, M., Ookie, M., Tsunomura, T., Sakaguchi, M. 2000. Antioxidant activities of phycocyanobilin prepared from *Spirulina platensis*. J. Appl. Phycol., 12:435–439.

Herrero, M., Jaime, L., Martı´n,-A´ lvarez, P.J., Cifuentes, A. and Iba´n˜ez, E. 2006. Optimization of the extraction of antioxidants from *Dunaliella salina* microalga by pressurized liquids. J. Agric. Food.Chem., 54:5597–5603.

Heo, S.J., Lee, KW., Song, C.B and Jeon, Y.J. 2003. Antioxidant activity of enzymatic extracts from brown seaweeds. Algae., 18: 71–81.

Hemat, R. A .S. 2007. Fat and muscle dysfunction. In R. A. S. Hemat (Eds), Andropathy. Dublin, Ireland: Urotext., 83-85.

Henry, G.E., Momin, R.A., Nair, M .G. and Dewitt, D .L, 2002. Antioxidant and cyclooxygenase activities of fatty acids found in food. J. Agric. Food Chem ., 50: 2231-2234.

Huang, H.L., Wang, B.G. 2004. Antioxidant capacity and lipophilic content of seaweeds collected from the Qingdao coastline. J Agric Food Chem., 58: 4993-4997.

Isabelle, H. A., Sanchez-Muniz, F. 2000. Dietary Fiber from edible seaweeds: chemical structure, physicochemical properties effects on cholesterol metabolism. Nutr. Res., 20: 585-598.

Jing TY, Zhao XY, 1995. The improved pyrogallol method by using terminating agent for superoxide dismutase measurement. Prog. Biochem. Biophysics., 22: 84-86.

Jiménez-Escrig, A., Jiménez-Jiménez, I., Pulido, R. and Saura-Calixto, F. 2001. Antioxidant activity of fresh and processed edible seaweeds. Journal of Science Food and Agriculture., 81: 530–534.

Kang, H.S., Chung, HY., Kim, JY., Son, BW. Jung, H.A. and Choi, J.S. 2004. Inhibitory phlorotannins from the edible brown alga Ecklonia stolonifera on total reactive oxygen species (ROS) generation.Arch. Pharm. Res., 27: 194–198.

Kang, H.S., Chang, H.Y, Jung, J.H., Son, B.W and Choi, J.S. 2003. A new phlorotannin from the brown alga Ecklonia stolonifera. Chem. Pharm. Bull., 51: 1012–1014.

Kang, K.A., Lee, K.H., Chae, S.W., Zhang, R., Jung, M.S., Lee, Y.G., Kim, S.Y and Kim. H.S, 2005b. Eckol isolated from Ecklonia cava attenuates oxidative stress induced cell damage in lung fibroblast cells. FEBS letters., 579:6295–6304.

Kang, K.A., Lee, K.H., Chae, S.W., Koh, Y.S., Yoo, B.S, Kim, J.H., Ham, Y.M., Baik, J.S, Lee, N.H. and Hyun, J.W.2005a. Triphlorethol-A from Ecklonia cava protects V79-4 lung fibroblasts against hydrogen peroxide induced cell damage. Free Radic Res., 39:883–892.

Kuda, T., Tsunekawa, M., Goto, H. and Araki. Y. 2005a. Antioxidant properties of four edible algae harvested in the Noto Peninsula, Japan. J. Food Compos. Anal., 18: 625-633.

Kuda, T., Kunii, T., Goto, H., Suzuki, T. and Yano, T. 2007. Changes of radical-scavenging capacity and ferrous reducing power in chub mackerel Scomber japonicus and Pacific saury Cololabis saira during 4°C storage and retorting, Food Chem., 103:900-905.

Latham, H. 2008. Temperature stress-induced bleaching of the coralline alga *Corallina officinalis*: a role for the enzyme bromoperoxidase. Biosci. Horizon., 1 : 104–113.

Levy, Y., Haya Zaltsberg, H., Ben-Amotz, A., Kanter, Y and Aviram. M. 2000. Dietary supplementation of a natural isomer mixture ofbeta-carotene inhibits oxidation of LDL derived from patients with diabetes mellitus. Ann. Nutr. Metab., 44:54–60.

Li, X, C., Fan, X., Han, L. J, Yan, X. J and Lou, Q.X. 2002. Fatty acids of common marine macrophytes from the yellow and bohai seas. Oceanol. Limnol. Sinica., 33: 215-223.

Lim ,S.N., Cheung, P.C.K., Ooi, V.E.C. and Ang .P.O. 2002. Evaluation of antioxidant activity of extracts from brown seaweed, *Sargassum siliquastrum*. J. Agric. Food Chem., 50: 3862–3866.

Li, A. H., Cheng, K., Wong, C., King-Wai, F., Feng, C. and Yue, J. 2007. Evaluation of antioxidant capacity and total phenolic content of different fractions of selected microalgae. Food Chem., 102: 771–776.

Matsukawa, R., Dubinsky, Z., Kishimoto E., Masaki, K., Masuda, Y., Takeuchi, T, Chihara., M., Yamamoto, Y., Niki, E. and Karube, I. 1997. A comparison of screening methods for antioxidant activity in seaweeds. J. Appl. Phycol., 9: 29-35.

Mayer, A. M .S., Lehmann, V. K. B 2000. Marine pharmacology in 1998: Marine compounds with antibacterial, anticoagulant, anti-inflammatory, anthelmintic, antiplatelet, antiprotozoan, and antiviral activities; with actions on the cardiovascular, endocrine, immune, and nervous systems; and other miscellaneous mechanisms of action. The Pharmacologist. , 42: 62–69.

Mori, J., Matsunaga, T., Takahashi, S., Hsegaula, C., Saito, H..2003. Inhibitory activity on lipid peroxidation of extracts from marine brown alga. Phytother. Res., 17: 549–55.

Metodiewa, D., Koska, C., 2000. Reactive oxygen species and reactive nitrogen species: relevance to cyto

(neurons) toxic events and neurologic disorders. An overview. Neurotox Res., 1: 197- 233.

Miranda, M .S., Cintra, R. G., Barros, S .B and Filho, J .M. 1998. Antioxidant activity of the microalga *Spirulina maxima*. Braz. J. Med. Biol. Res., 31:1075–1079.

Nagai, T., Sakai. M., Inoue, R., Inoue, H and Suzuki, N.2002. Antioxidative activities of some commercially honeys, royal jelly, and propolis. Food Chemistry., 75: 237-240.

Nagai, T., Yukimoto, T.2003. Preparation functional properties of beverages made from sea algae. Food Chem., 81: 327-332.

Oyaizu, M. 1986. Studies on product of browning reaction prepared from glucose amine. Jpn. J. Nutr., 44: 307-315.

Prieto, P., Pineda, M and Aguilar, M. 1999. Spectrophotometric Quantitation of Antioxidant Capacity through the Formation of a Phospho molybdenum Complex: Specific Application to the Determination of Vitamin E. Anal. Biochem., 269: 337-341.

Qi, H., Zhao, T., Zhang, Q., Li, Z., Zhao, Z and Xing R. 2005. Antioxidant activityof different molecular weight sulfated polysaccharides from *Ulva pertusa* Kjellm (Chlorophyta), Appl. Phycol., 17: 527–534.

Rao, A.R., Sarada, R., Baskaran, V. and Ravishankar, G.A. 2006. Antioxidant activity of Botryococcus braunii extract elucidated invitro models. J. Agric. Food. Chem., 54:4593–4599.

Romay, C., Gonzalez, R.. 2000. Phycocyanin is an antioxidant protector of human erythrocytes against lysis by peroxyl radicals.J. Pharm. Pharmacol., 52:367–368.

Ruberto, G., Baratta, M .T., Biondi, D. M and Amico. V. 2001; . Antioxidant activityof extracts of the marine algal genus Cystoseira in a micellar model system. J.Appli. Phycol., 13: 403-407.

Sadati, N., Khanavi, M., Mahrokh, A. and Nabavi, S.M.B. , Sohrabipour, J ., Hadjiakhoondi, A. 2011. Comparison of Antioxidant Activity and Total Phenolic Contents of some Persian Gulf Marine Algae. Journal of Medicinal Plants., 10 : 73-79.

Seo, Y., Park. K.E., Nam Bull, T.J. 2007. Isolation of a new chromene from the brown alga *Sargassum thunbergii*. Korean Chem. Soc., 28: 1831–1833.

Shimada, K., Fujikawa, K., Yahara, K and Nakamura.T. 1992.. Antioxidative properties of xanthan on the autoxidation of soybean oil in cyclodextrin emulsion. Journal of Agriculture and Food Chemistry., 40: 945-948.

Shibata, T., Hama, Y., Miyasaki, T., Ito, M and Nakamura, T. 2006. J Appl. Phycol., 18: 787.

Singleton, V.L, Orthofe, R, Lamuela- Raventos, R.M. 1999. Analysis of total phenols and oxidation substrates and antioxidants by means of Folin – Ciocalteau reagent. Methods Enzymol. , 299: 152 – 177.

Sohrabipour, J., Rabii, R, 1990. A list of marine algae of seashores of Persian Gulf Oman Sea in the Hormozgan province. Iran. J. Bot., 8:131-162.

Sukenik, A., Zmora, O and Carmeli, Y.1993. Biochemical quality of marine unicellular algae with special emphasis lipid composition: II. Nannochloropsis sp. Aquaculture., 117: 313–326.

Subbarao, P.V., Ravishankar, G.A. 2005. Chemical composition, iron bioavailability, and antioxidant activity of *Kappaphycus alvarezzi*(Doty). J. Agric. Food. Chem., 53:792–797.

Tsang, C., Kamei, K .Y.2004. Sargaquinoic acid supports the survival of neuronal PC12D cells in a nerve growth factor-independent manner. European Journal of Pharmacology., 488: 11–18.

Taga, M.S., Miller, E.E and Pratt, D.E. 1984.. Chia seeds as a source of natural lipid antioxidants. J. Am. Oil Chem. Soc., 61: 928-931.

Wichi, H.P. 1988. Enhanced tumor developmentby butylated hydroxyanisole (BHA) from the properties of effect on fure stomach and oesophagelaquamoua epithelium. Food and Chemical Toxicology., 26: 727-723.

Xue, Z., Xue, CH., Li, Z.J., Cai, Y.P., Liu, H.Y and Qi, H.T. 2004. Antioxidant and hepatoprotective activities of low molecular weight sulfated polysaccharide from *Laminaria japonica*. J. Appl. Phycol., 16: 111–115.

Yan X J, Li X C, Zhou CX and Fan X, 1996;. Prevention of fishoil rancidity by phlorotannins from *Sargassum kjellmanianum*. J. Appl. Phycol 8:201–203.

Yan, G.C., Chen, H.Y. 1995;. Antioxidant activity of various tea extracts in relation to their antimutagenecity. J. Agric. Food Chem., 43: 27-37.

Yan, X., Nagata, T and Fan, X, 1998. Antioxidative activities in some common seaweeds. Plant Foods for Human Nutrition., 52: 253–262.

Yuan, Y.V., Bone, D.E., Carrington, M.F.2005. Antioxidant activity of dulse (*Palmaria palmata*) extract evaluated *in vitro*. Food Chemistry., 91:485–494.

Yan, X.J., Chuda, Y., Suzuki, M and Nagata ,T. 1999. Fucoxanthin as the major antioxidant in *Hijikia fusiformis*, a common edible seaweed. Biosci. Biotechnol. Biochem., 63:605–607.

Young, I.S., Woodside, J.V. 2001. Antioxidants in health and disease. J Clin Pathol. 54: 176-186.

Zhishen, I., Mengcheng, T and Jianming ,W. 1999. The determination of flavanoids contents in mulberry and their scavenging effects on Super radicals. Food Chem., 64: 555 -559.

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