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MINI REVIEW

A review of antioxidant potential from seaweeds - extraction, characterization, benefits and applications

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Abstract

Potential natural antioxidant from seaweeds is used to improve the oxidative stability of food. It also functions as an active ingredient. There are three classes of antioxidants consisting of vitamins, carotenoids and polyphenols. Researchers employ a variety of ways to evaluate antioxidant activity, including screening approaches and chemical composition. Solid-liquid extraction pressured liquid extraction, supercritical fluid extraction, and other green extraction techniques are among the seaweed antioxidant extraction methods commonly utilized and reviewed in this article. The aim of the review was to identify the potential source of high antioxidant content and polysaccharides, along with the best extraction method. This overview summarised the composition of seaweed antioxidants, extraction methods and the advantages of antioxidants found in seaweed.

1. Introduction

1.1 Chemical composition (antioxidant/prooxidant substances)

Antioxidant compounds are found in many types of seaweed such as phlorotannins, carotenoids, tocopherols and sulfate polysaccharides. Figure 1 shows the chemical structure of selected antioxidants. Phlorotannins are a class of polyphenol compounds, regularly extracted from seaweeds, especially brown algae, using typical solvent extraction followed by chromatographic purification procedures. Phlorotannins are oligo- or polymer phloroglucinol (1,3,5-trihydroxybenzene) (Steevensz *et al*., 2012; Cotas *et al*., 2020; Batool and Menara, 2020). It is related to the phloroglucinal unit in which naturally occurring phlorotannin is classified. Based on the study of Steevensz *et al.* (2012), phlorotannin molecular weight varies from 0.126 to 650 kDa, although the most frequently observed range is from 10 to 100 kDa.

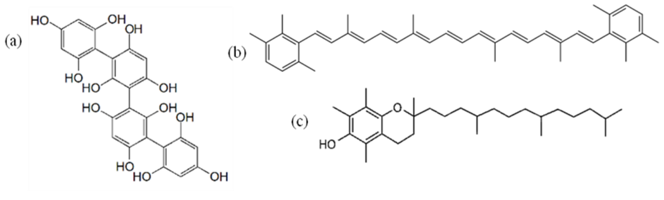

Figure 1. Chemical structure of antioxidant compounds. (a) phlorotannins (b) carotenoids, and (c). tocopherols.

Sabah is known for its outstanding seaweed production resources, particularly in the Semporna region, south of Sabah, the South China Sea Island of Banggi and the Kudat region. These three regions have a very large amount of seaweed cultivation, but only seven major seaweed species seem to be present within those areas. The seven species of seaweed are *Eucheuma, Sargassum, Eucheuma, Caulerpa, Gracilaria, Hypnea, Padina* and *Hydroclathrus* (Cotas *et al*., 2020).

Among the types of seaweed widely commercialized in Sabah are *Eucheuma cottonii* and *Eucheuma spinosunm*. These seaweeds are also widely grown in Malaysia and areas in Southeast Asia (Awalludin *et al.,* 2021). The difference between the two types is that *Eucheuma spinosum* has thorny branches while *Eucheuma cottonii* has no thorny branches. Seaweed is one of the most valuable underwater plants found in some rivers in the world and the types of this seaweed are different from the types of plants in the sea (Anwar *et al.,* 2018; Jianfui *et al*., 2019).

There are two categories of antioxidants based on their origin, which are natural and synthetic. Synthetic oxidation has known to be very efficient, but not all food systems would be protected (Plaza *et al.,* 2010). Synthetic antioxidants such as EDTA, BHA, BHT and tertiary butylhydroquinone are examples of these synthetic antioxidants (Farvin *et al.,* 2013). The advantages of these synthetic antioxidants that make

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them so good are that it has a cheaper production value and food energy stability. However, the disadvantages of using these synthetic antioxidants are high toxicity owing to health risks. In contrast, natural antioxidant has no negative effect on health problems.

Tocopherols and ascorbic acid are examples of natural antioxidants. Farvin *et al.* (2013) also noted that a range of different antioxidants, including phenolic acids, polyphenols, and flavonoids, are found in natural supplements, seasonings, and algae. Table 1 shows the comparison between natural and synthetic antioxidants.

1.2 Benefits of antioxidant

To preserve food quality, as well as personal care, antioxidants play a significant role. In the health industry, the presence of antioxidants is crucial in the prevention of cancer and tumours, blood vessel narrowing, premature ageing, and so much more (Rani, 2017). Antioxidants also inhibit oxidation reactions by binding to free radicals and highly reactive molecules, so cell damage can be prevented. Oxidation reactions with free radicals often occur in protein molecules, nucleic acids, lipids, and polysaccharides.

In the food industry, antioxidants can be used to prevent the oxidation process that subsequently causes damage to food materials such as rancidity, change in colour and aroma, and other physical damage. Antioxidant acts as an inhibitor of lipid peroxidation to prevent lipid peroxidation in food (Plaza *et al.,* 2010). Lipid peroxidation is a chemical reaction that mostly occurs in food that, during handling and preparation generates acidified, harmful scents and are toxic, thereby affecting the quality of food ingredients.

Consuming adequate quantities of antioxidants is essential to minimize the chances of developing degenerative diseases such as cardiovascular disease, cancer, atherosclerosis, osteoporosis, and some other degenerative diseases. Consumption of food that contains antioxidants can improve the immunological status and prevent degenerative disease due to ageing. Optimal adequacy of antioxidants is required by all age groups (Plaza *et al.,* 2010; Anwar *et al.,* 2018; Jianfui *et al.,* 2019).

Phytochemical compounds are natural substances found in plants that give a distinctive taste, aroma and colour to plants. Hence, some of the properties of these phytochemical compounds are functional as an antioxidant, boosts the immune system, and maintain blood pressure, cholesterol, and sugar levels in our body (Steevensz *et al.,* 2012; Cotas *et al*., 2020). Table 2 shows the selected chemical composition found in many types of seaweed such as phlorotannins, carotenoids, tocopherols and sulfate polysaccharides.

1.3 Antioxidant compounds

Chemically, antioxidant compounds are known as the giving electrons compounds (electron donors). Biologically, antioxidants are substances that can counterbalance or decrease harmful oxidants by transferring electrons to a particular oxidant compound. Thus, it is possible to decrease the production of these oxidant compounds (Santos-Sánchez *et al*., 2019). Antioxidants also protect the body from free radicals by inhibiting or bringing down the problem caused by oxygen (Kurutas, 2016). To maintain a healthy biological system, the human body needs to have a balance antioxidative defence system in the form of endogenous and exogenous antioxidants. In the presence of excessive free radicals, continuous demand for exogenous antioxidants is needed to prevent oxidative stress (Bouayed and Bohn, 2010).

The unknown side effects of synthetic antioxidants cause the search for natural antioxidants as an alternative option. Phenolics have various biological impacts such as antioxidant agents for the reduction of free radicals, metal chelating, and dampening the development of singlet oxygen and electron donors (Moukette *et al.,* 2015). Flavonoids are one of the phenolic compounds present in vegetables and fruits (Rodríguez-García *et al.,* 2019). Flavonoids are shown to have enormous potential among diseases caused by radical scavengers in past years (Panche *et al.,* 2016). For example, it was found that flavonoids have neuroprotective and cardioprotective effects that involve several clinical trials in the treatment of Alzheimer's disease (Kim and Park, 2021).

Table 1. Comparison between natural and synthetic antioxidant.

Origin	Antioxidant type	
	Natural	Synthetic
Lipid oxidation	No negative effect	Very efficient but not all food systems would be
Example	Tocopherols and ascorbic acid	EDTA, BHA, BHT and tertiary butylhydroquinone
Benefits	Skin cells and on human skin (including reduction in pigmentation and bruising, activation of collagen production, keratinization refinement, and anti-	Low labour value and better food stability

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Table 2. Selected chemical composition found in using many types of seaweeds

Antioxidant	Chemical Composition	Reference(s)
Phlorotannins	• Oligo- or polymers of phloroglucinol (1,3,5-trihydroxybenzene)	Salehi *et al.* (2019)
	• Total Phenolic Compounds can be used to estimate the content of phlorotannins in brown algae • Identified phlorotannins (a species rich in these compounds) - liquid chromatography coupled to a mass spectrometer (LC-MS)	Jönsson *et al.* (2020)
	• Low molecular weight phlorotannins from *F. vesiculosus*	Heffernan *et al.* (2015)
Carotenoids	• Prevent human diseases such as cardiovascular diseases, cancer and other chronic diseases	Sathasivam and Ki (2018)
	• Allenic carbon bond, 5,6-monoepoxide in the molecule, and is known as an efficient quencher of singlet oxygen in photooxidation • Prooxidants under high oxygen pressure to promote lipid oxidation	Kuczynska *et al.* (2015)
	• Pigments from plant materials extracted using solid phase extraction (SPE) and analyzed by high-performance liquid chromatography (HPLC)	Scigalski and Kosobucki (2020)
Tocopherols	• Fat-soluble antioxidant; food industry owing to their efficient radical scavenging activity • Antioxidant activity. The order of the four tocopherol isomers in bulk oil is $\delta > \gamma > \beta > \alpha$ • Lipids can be extracted with a chloroform: methanol solvent mixture • Tocopherols and tocotrienols can be detected using liquid chromatography system equipped with a fluorescence detector	Plaza *et al.* (2010)
Sulfate Polysaccharides	• Anticoagulant, antitumor, cancer preventive, antimicrobial, anti-inflammatory and antioxidant • Physical stability to emulsion systems • Fucoidan and laminarans of brown algae, carrageenan of red algae, and ulvan of green algae • Negative-ion electrospray tandem MS with collision-induced dissociation • To obtain direct information on the structural heterogeneity from the brown alga *Hizikia fusiforme*	Wang *et al.* (2012)

The oxidation process does not only occur in the human body but can also occur in food. The most dietary component that is easily oxidized is fat. Antioxidants are compounds added to fat or fatty food to prevent the occurrence of the oxidation process to extend the freshness and palatability of these foods. The antioxidant compound act as an inhibitor to inhibit auto-oxidation. The oxidizing properties of the phenolic compounds play a role in neutralizing their free radicals (Kaurinovic and Vastag, 2019).

Nuts, vegetables, fruits, chocolate, and tea are the source of flavonoids. Apart from that, flavonoids are also available as a supplement in the form of powder, capsule, or extract. Currently, dietary supplements are commonly available in the market as extract powder. In addition, green tea extract contains catechins (flavonol monomers), black tea extract contains theaflavin and arubigins, and extracts of bilberry, elderberry, black currants, anthocyanin-rich grapes are also available. Phytochemical compounds are found in various types of vegetables and fruits. These compounds have health benefits and make the body healthier and stronger (Panche *et al.,* 2016; Rodríguez-García *et al.,* 2019; Kaurinovic and Vastag, 2019). The aim of the review is to identify the potential source of high antioxidant content and polysaccharides, along with the best extraction method.

2. Extraction techniques and application usage

2.1 Methods used to extract bioactive compounds from seaweeds

Several methods can be used to extract bioactive compounds from seaweed as shown in Figure 2. Table 3 summarizes the extraction efficiency and the extract composition affected by the parameters from which the extraction process is performed.

Solid-liquid extraction (SLE) using solvent is the most widely used process. There are many disadvantages to conventional solvent bioactive compounds extraction as it involves the use of high amounts of organic solvents, longer extraction time, selectivity issues and non -targeted co-extraction or interference compounds (Scigalski and Kosobucki, 2020; Awalludin *et al.,* 2021). Therefore, the traditional SLE process has been refined using many environmentally friendly technologies for

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the extraction of bioactive compounds from plant sources, including supercritical fluid extraction (SFE) and pressurized liquid extraction (PLE) (Plaza *et al.,* 2013). The SFE and PLE methods use less hazardous solvents, which apply green extraction techniques that cause a lower carbon footprint. In addition, these methods also offer other advantages such as high selectivity, high extraction effectiveness and low extraction time (Sarkar *et al.*, 2022).

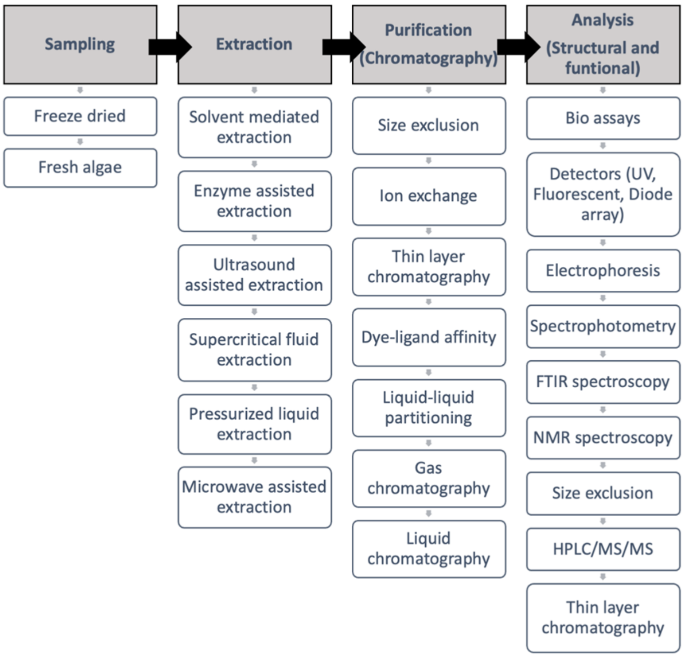

Figure 2. Various extraction method of seaweed samples.
Source: Batool and Menaa (2020)

2.2 Application of antioxidants from seaweed extract

The applications of antioxidative activity in seaweed extract can be seen in some industrial products such as sunscreen cream and fish oil (Santos-Sánchez *et al.,* 2019). According to Moukette *et al*. (2015), UV radiation is one of the leading causes of skin damage and is an essential aspect of the cosmetic industry. With the addition of seaweed extract that contains antioxidants, cosmetics products with UV protection properties have been developed to retain moisture and skin softness, provide nutrition, and prevent premature ageing (Farvin and Jacobsen, 2013).

Red seaweed contains antioxidant compounds which can inhibit the penetration of strong UV light into the tissues or cells (Rodríguez-García *et al.,* 2019). Salehi *et al*. (2016)'s finding reported that *Eucheuma cottonii* and *Sargassum* sp. with a ratio of 1: 1 exhibited strong antioxidant activity with IC_{50} value of 83.4 µg/mL. Several compounds are known to have very strong antioxidants activity if the IC_{50} value is <50 µg/mL; Antioxidant activity is considered strong if the IC_{50} value is 50-100 µg/mL; moderate if IC_{50} 100-150 µg/mL, and weak if the IC_{50} is 150-200 µg/mL (Anwar *et al.,* 2018; Rodríguez-García *et al.,* 2019).

The effectiveness of solar cream preparations is based on the sun protection factor (SPF) value to determine the protection period for the skin (Saravana *et al.,* 2018). Uwineza *et al*. (2020) mentioned that the ability of sun cream in protecting the skin is divided into several categories, including minimal protection (SPF 2-4), moderate protection (SPF 4-6), extra protection (SPF 6-8), maximum protection (SPF 8-15), and ultra-protection (SPF > 15). Sari *et al*. (2019)'s studies reported that additional ethyl acetate extract of *Eucheuma cottonii* into sunscreen cream provided maximum inner protection to the skin with an SPF value of 8.8.

Besides *Eucheuma cottonii*, Sari *et al*. (2019)'s findings also reported the capability of the ultra-protection effect of *Turbinaria conoides* in protecting the skin with an SPF value of 16.7. Fish oil, one of the fisheries products contain highly unsaturated fatty acids which are prone to oxidation and this lipid oxidation can be reduced by the addition of antioxidants. According to Kurutas (2016)'s study, antioxidants have low activation energy which assists in releasing one hydrogen atom to the fat radical to inhibit the additional oxidation process.

Eismann *et al.* (2020)'s finding explained that one seaweed from Chlorophyceae, *Ulva lactuca* can inhibit fish oil oxidation at 0.1% concentration. Antioxidant activity test on *Ulva lactuca* ethanol extracts using the DPPH method reported a strong value of antioxidant activity with IC_{50} around 50 ppm (Chidambararajan *et al*., 2019). According to Rivero-Cruz *et al*. (2020) study,

Table 3. Different extraction methods to extract bioactive compounds from seaweeds

Extraction Method	Compound Extracted	Reference(s)
Solid-Liquid Extraction (SLE)	The extractant aspect has a significant effect on total phenolic compounds (TPC) and the composition of other bioactive compounds such as pigments, polysaccharides, proteins, and peptides, including on the activity of antioxidants.	Farvin and Jacobsen (2013)
Pressurized Liquid Extraction (PLE)	Phenolic and other bioactive compounds have been successfully separated through PLE from natural specimens.	Plaza *et al.* (2010); Plaza *et al.* (2013); Saravana *et al.* (2018)
Supercritical Fluid Extraction (SFE)	This method is suitable for lipophilic materials and has excellent selectivity. Use to extract heat-sensitive compounds such as carotenoids and pigments.	Uwineza *et al.* (2020)

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a material was said to have powerful antioxidant activity if they have the IC_{50} value of 50- 100 ppm. Apart from *Ulva lactuca*, a study by Cotas *et al*. (2020) explained that *Sargassum* sp. including brown seaweed has antioxidant compounds which help to inhibit fish oil oxidation.

The addition of 1% ethanol extract of *Sargassum* sp. was reported able to inhibit the oxidation of fish oil emulsion. Omidi *et al.* (2018)'s findings on oilemulsion reported that the presence of methanol extract of *Sargassum* sp. acts as antioxidant and assists in the lower oxidation process that occurs in the fish oil emulsion with the peroxide value of 59.1 meq/kg when compared to the control sample without antioxidant (308.5 meq/kg) under the high-temperature exposure of 500°C for a period of 24 hrs. Peroxide value measurement is commonly used to measure the level of peroxides and the hydroperoxides that formed in the early stages of the fat oxidation process. A high peroxide number indicates the fat or oil has undergone an oxidation process (Esfarjani *et al.,* 2019).

Antioxidants extracted from seaweed are also used as plant biostimulants and feed additives. Commercial seaweed liquid extracts have been studied for the effect on the growth and antioxidant activities of *Capsicum annuum* (Ashour *et al.*, 2021). The resulting liquid extract was found to have a high content of phytochemicals such as phenolics (101.67 mg/g) and flavonoids (2.60 mg/g), which showed good antioxidant activity (54.52 mg/g) and DPPH inhibition of 70.33%. Furthermore, the incorporation of seaweed into animal diets increases antioxidant activity, improves the gut environment, and enhances meat quality (Michalak *et al*., 2022). Dietary supplementation of seaweeds was also found to increase animals' immunity to stresses like diseases and boost their productivity.

3. Conclusion

Phlorotannins, pigments, tocopherols and polysaccharides are antioxidant compounds found in some species of seaweeds. Seaweeds are the potential source of antioxidant compounds, particularly brown seaweed because of their high content of phlorotannins, mainly present in *A. nodosum* and *Fucus* sp. However, the cultivation of *Fucus spp*. is poor, leading to limited availability in terms of antioxidant viability. *S. japonica* is a seaweed with high antioxidant content. The rich source of polysaccharides can be found in seaweed *Laminaria* spp. However, *Fucus* spp. is still high in phlorotannin compared to *Laminaria* spp. Many extraction methods are used to extract active compounds from seaweeds, with the most used technique being the SLE method. Various in-vitro tests such as the radical return of DPPH and the chelating ability of metals are used to evaluate the antioxidant properties of extracts. To examine the antioxidant potential of seaweeds, *in vitro* antioxidant screening along with TPC is commonly used. However, for the food system, it should not be extrapolated directly using *in vitro* antioxidant activity alone.

Conflict of interest

The authors declare no conflict of interest.

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