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Overview of Extraction Methods for Extracting Seaweed and its Applications

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Abstract: Seaweed is a carbohydrate polymer of alginate, agar, and carrageenan which can be extracted and purified as hydrocolloids. Each carbohydrate has different physicochemical properties and hydrocolloids can form gels when dispersed in water. To achieve targeted hydrocolloids according to specific purposes and functions, conventional extraction methods, and several green extraction methods for extracting seaweed have been proposed and discussed. The conventional extraction method is in the form of solvent extraction (SLE), while the green extraction includes: microwave-assisted extraction (MAE), ultrasound-assisted extraction (UAE), supercritical fluid extraction (SFE), pressurized solvent extraction (PSE), and reactive extrusion which automatically selectively presented as a promising method for extracting hydrocolloids in seaweed. These methods have been applied by taking into account the recovery rate, time, volume, temperature, pH, ratio, and type of solvent, as well as the type of method. This study discusses extraction methods that can extract bioactive compounds from seaweed with high yields and are economical and environmentally friendly.

Keywords: Extraction; Gel; Hydrocolloid; Seaweed

Introduction

Seaweed is a macroalga that does not have roots, stems, and leaves. This function is replaced by the thallus and holdfast at the bottom of the seaweed. These macroalgae include bioactive polysaccharides (Benslima et al., 2021). Seaweed is also a plant organism Rhodophyta (red seaweed), Phaeophyta (brown seaweed), and Chlorophyta (green seaweed) (Khalil et al., 2017; Jönsson et al., 2020). The main products are alginate carbohydrates, agar, and carrageenan which can be extracted as gelling hydrocolloids (Jönsson et al., 2020; Singh et al., 2018). Seaweed hydrocolloids can be extracted using various separation methods (Vuai, 2022).

Extraction is the process of separating substances from solids or liquids using a solvent, where the solvent will extract the required substance without extracting other ingredients. Extraction is used to separate substances from reaction mixtures or natural sources for batch or continuous purification of substances (Budhiraja, 2004). The extraction process occurs due to adding a specific volume of solvent (Christian, 2004).

The extraction process is the first and vital step in extracting seaweed. The choice of method and extraction efficiency is influenced by the seaweed species being analyzed, the type of solvent, temperature, time, pH, environmental impact, cost, quantity, and desired properties (Godlewska et al., 2017; Lim et al., 2021; Jacobsen et al., 2019). Seaweed extract contains components of polysaccharides, proteins, unsaturated fatty acids, pigments, and minerals (K, Mg, Ca, and Na) (Godlewska et al., 2016). Seaweed contains flavonoid and polyphenolic compounds, namely phlorotannin, bromophenol, flavonoid, and phenolic terpenoids (Kristanto et al., 2021). Phenol compounds are very difficult to isolate quantitatively because they tend to react with other compounds. However, it can be extracted at a lab scale (Cotas et al., 2020).

Conventional extraction using hot water to produce seaweed extract has been carried out. This method is environmentally friendly and does not require organic solvents (Khalil et al., 2017; Godlewska et al., 2016). Extraction with dilute acid or alkali (KOH, NaOH) is also possible (Rodriguez-Jasso et al., 2011; Wolle & Conklin, 2018). In addition to soxhlet extraction, TLC-

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based approaches and continuous liquid-liquid chromatography (HSCCC) for extracting pigments and isolating seaweed compounds have also been optimized (Cotas et al., 2020; Rajauria & Abu-Ghannam, 2013; Xia et al., 2019). Solid-liquid extraction (SLE) has the disadvantages of using large amounts of organic solvents, long extraction times, selectivity, and co-extraction of interfering compounds (Jacobsen et al., 2019). Solvent extraction (LLE) involves significant amounts of organic solvents that produce hazardous waste, so using small amounts of organic solvents and biodegradable materials is a green extraction option (Sanagi et al., 2016).

Efficient and environmentally friendly green extraction methods such as microwave-assisted extraction (MAE), ultrasound-assisted extraction (UAE), supercritical fluid extraction (SFE), pressurized solvent extraction (PSE), and reactive extrusion have been able to increase the yield of seaweed hydrocolloid extraction. Apart from the lack of use of organic solvents, selectivity, lower temperature, and short extraction time are the advantages of these methods (Vijayan et al., 2014; Han et al., 2010). The combination of these methods also resulted in better seaweed extract. Environmentally friendly and economical, fast, sensitive, and accurate extraction methods are preferred over conventional extraction (Lim et al., 2021; Gullón et al., 2020).

Microwave-assisted extraction for active compounds in seaweed extraction where the radiation energy is efficient in dissolving is selective and fast (Rodriguez-Jasso et al., 2011; Thodhal Yoganandham et al., 2019). Subcritical fluid extraction is also demanding for phytochemical compounds (Kristanto et al., 2021). Both of these methods limit the use of solvents, and even microwave extraction can be used as a pre-treatment before hot water extraction and during subcritical fluid extraction (Torres et al., 2021). Enzymatic extraction is able to maintain the structure and function of seaweed polysaccharides (Rhein-Knudsen et al., 2015), while ultrasonic extraction does not use chemical solvents and saves time compared to enzymatic depolymerization (Torres et al., 2021). Other extraction methods have also been developed but have not been able to quantify seaweed (Manns et al., 2014). Green extraction is more practical in extracting seaweed hydrocolloids without chemicals (Yew et al., 2016). Seaweed was extracted with cocoa pod ash as a substitute for potassium hydroxide by comparing the extraction yield, composition, and the effect of temperature and time on the molecular weight and viscoelasticity of the material (Rhein-Knudsen et al., 2015). The extraction process becomes more desirable if it is carried out without alkaline treatment and alcohol precipitation (Webber et al., 2012).

Extraction has been used to extract bioactive compounds. For this purpose, seaweed is extracted, filtered, jellified, frozen, thawed, and dried (Din et al., 2019). This stage uses organic solvents, suc as ethanol, cyclohexane, chloroform, methanol, methanol-toluene, petroleum ether, ethyl acetate, dichloromethane, and butanol. However, ethanol is preferred because of its low cost (Godlewska et al., 2017; Cotas et al., 2020). Hydrolysis using sulfuric acid is also fast for catalyst concentration, temperature, reaction, and the amount of seaweed. Liquid seaweed extract can also use alkaline hydrolysis and hot water (Jang et al., 2021; Arioli et al., 2015). Hot water is used to extract seaweed hydrocolloids, except for alginates which require hot alkali or dilute acid as solvent. This extraction converts the insoluble alginate salt into water-soluble (Sterner & Gröndahl, 2021)

Alginate, agar, and carrageenan are hydrocolloids extracted from seaweed. Apart from being a hydrocolloid, alginate is also an emulsifier, thickener, and gelling agent. Agar and carrageenan can be made using hot water, but their rheology is weak, so alkaline treatment is carried out to optimize the results (Trica et al., 2019). Seaweed extract has also been used as a liquid biostimulant to increase plant tolerance to salinity and drought. This biostimulant also affects the bioactive content of plants through the absorption and assimilation of nutrients (Hernández-Herrera et al., 2014; Hernández-Herrera et al., 2016; Raj et al., 2018; Kocira et al., 2020; Kocira et al., 2019). In addition to biostimulants, seaweed extract has now been used as biosynthesis of metal nanoparticles to reduce waste efficiently and environmentally friendly manner. This extract also functions as an inhibitor for mild steel corrosion in an acidic environment and removes heavy metals from electroplating waste through precipitation, coagulation, ion exchange, and solvent extraction (Wolle & Conklin, 2018; Mahdavi et al., 2013; Thilagavathi et al., 2019; Subramani & Sindhu, 2012). Water-soluble seaweed gel extract can also be used to precipitate metal ions in water (Chen et al., 2018). This paper will examine the various extraction methods used to extract seaweed and their use in various chemical applications. State the objectives of the work and provide an adequate background, avoiding a detailed literature survey or a summary of the results.

Method

The study analysis of the extraction method to extract seaweed was carried out through document studies in the form of textbooks and article reviews directly related to the extraction, especially the extraction of seaweed. The analysis in the form of a literature review was carried out on 2 textbooks associated with the notion of extraction and 48 articles discussing conventional extraction methods and green seaweed extraction.

Table 1. Extraction methods

Extraction methods	Types	Principles
Conventional Extraction	Solid-Liquid Extraction (SLE).	Using organic solvents with a certain extraction time.
Green Extraction	Microwave-Assisted Extraction (MAE), Ultrasound-Assisted Extraction (UAE), Supercritical Fluid Extraction (SFE), Pressurized Solvent Extraction (PSE), and Reactive Extrusion	Using a small amount of solvent under specific conditions with a fast extraction time.

Result and Discussion

Extraction of seaweed hydrocolloids is a slow process using hot water to extract seaweed, except for alginates which require hot alkali as a solvent. Through alkaline extraction, the alginate salt is converted into water-soluble. Carrageenan and agar can also be extracted in hot water, but their rheology is weak. Therefore, alkali is added to overcome and optimize the extraction process. This method is also effective for seaweed gel formation (Rhein-Knudsen et al., 2015). Different seaweed sources and extraction conditions will also produce hydrocolloids with varying gel properties (Chew et al., 2018). Currently, the hydrocolloid extraction method using chemical solvents is still used.

Table 2. Aims of seaweed hydrocolloid extraction

Extraction Process	Extraction Aims	Seaweed hydrocolloids		
		Agar	Carrageenan	Alginate
Cleaning	To remove dirt, salt and other contaminants.	-	-	-
Alkali	For gel formation through the conversion of unstable sulfate molecules to 3,6-anhydro-L-galactopyranose (3,6-AG).	-	-	
Formaldehyde	To remove pigment and increase alginate yield.			-
Hydrochloric acid	To convert insoluble alginate salts (Mg, K, etc.) into soluble salts.			-
Hot water and	To obtain compounds by optimizing temperature, time,	-	-	
Alkali (extraction)	pH, concentration, etc.	-	-	-
Neutrally	To neutralize the extract by removing excess solvent.	-	-	-
Filtering	To remove the residue of the extract and get the pure compound.	-	-	-
Drying and grinding	To get a dry product according to the desired shape.	-	-	-

Extraction with hot alkali aims to convert alginate salts (Mg, Ca, K, etc.) into soluble sodium alginate. Before alkaline extraction, seaweed can be added with formalin and hydrochloric acid. The addition of formalin aims to remove color and odor pigments, while the addition of HCl aims to change the salts that are not soluble in water into soluble salts (Bertagnolli et al., 2014). However, this method causes depolymerization of the alginate, thereby reducing the molecular weight and viscosity of the alginate (Chee et al., 2011).

Precipitation of hydrocolloids is done by adding alcohol, calcium chloride (CaCl2), or hydrochloric acid (HCl). Carrageenan and agar can also be precipitated by the addition of potassium chloride (KCl) during filtration, which is prepared by heating (75°C) using potassium hydroxide (KOH) to increase gel strength (Khalil et al., 2017). This process is then washed, dried, and ground into a powder. This process can be continued by heating (95–110 °C) to dissolve the gel and recovered by alcohol precipitation or gel pressing (Rhein-Knudsen et al., 2015).

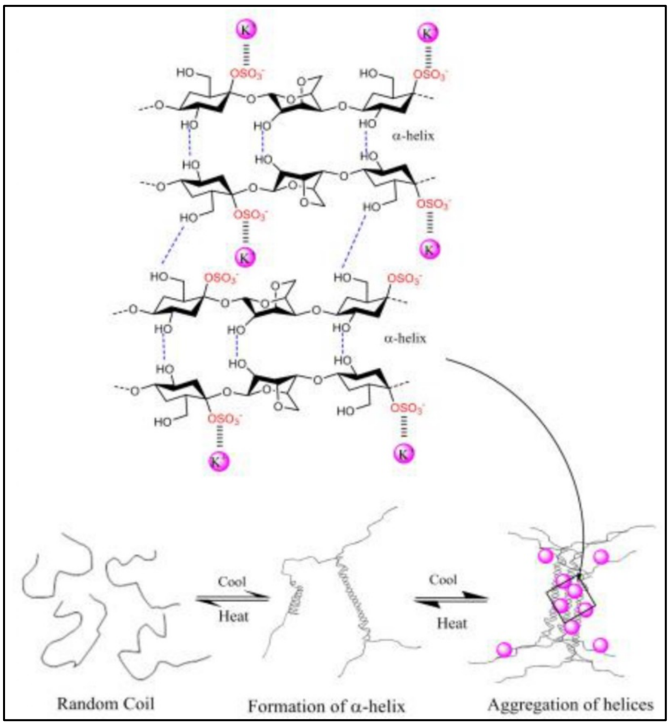

Figure 1. Reaction of carrageenan gel with potassium ions.

Solvent Extraction (Conventional Extraction)

Seaweed has high water content, so extraction needs to begin with determining the water content. Pre-treatment like size reduction and addition of acid or alkali is also carried out before the extraction process. In conventional extraction, seaweed carrageenan is boiled to form a gel in the presence of potassium chloride and then rinsed to remove residual alkali, dried, and ground. Agar extraction was also carried out using the same way using lye and hot water by optimizing time, temperature, and alkali concentration to increase gel strength (Jönsson et al., 2020).

Solid-liquid extraction (SLE) using organic solvents has been widely applied for the extraction of bioactive compounds, where the extraction results are highly dependent on the solvent or solvent mixture used. Binary solvents containing water and organic solvents have proven to be efficient, for example, using ethanol: water (70:30 v/v) or 80% ethanol to precipitate proteins and phenolic compounds. Solvent extraction has several disadvantages: large amounts of organic solvents, long extraction times, and selectivity problems. So it is necessary to optimize SLE through the development of extraction techniques that are more efficient and environmentally friendly to increase extraction yields, use less solvent, extraction at lower temperatures, and time efficiency to improve extraction performance (Christian, 2004).

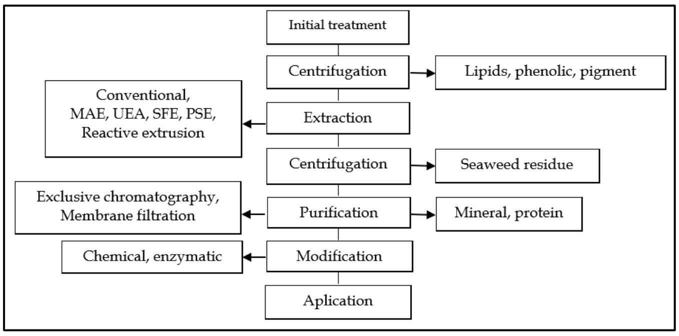

Figure 2. Extraction of seaweed polysaccharides.

Green Extraction

The innovation and design of "green method" extraction is currently an attractive extraction technique in the chemical field based on economic, quality and environmentally friendly aspects that allow for extracting bioactive compounds using less solvent (Khalil et al., 2012; Tatke & Jaiswal, 2011).

Tabel 3. Seaweed Extraction Methods

Extraction methods	Description	Advantages	Disadvantages
Conventioal Extraction	The extraction process depends on a particular solvent's solubility of the target compound. Extraction is also carried out by adding acid or base.	The method has been tested.	Long extraction time, using a lot of solvents, acids and bases resulted in the degradation of polysaccharides.
Microwave-Assisted Extraction (MAE)	MAE uses electromagnetic radiation where temperature, pressure, time, seaweed/water ratio can be changed to maximize yield.	Short extraction time with relatively small amount of solvent.	High heat causes the degradation of polyphenols.
Ultrasound-Assisted Extraction (UAE)		The extraction time is short. The method works at low temperatures using a relatively little solvent.	Degradation and changes in the structure of polysaccharides.
Supercritical Fluid Extraction (SFE)		CO was removed from the extract easily and did not cause structural degradation of the compound.	High pressure to retain solvent which has a negative effect on the compound.
Pressurized Solvent Extraction (PSE)	PSE as pressurized liquid/fluid extraction (PLE/PFE), accelerated solvent extraction (ASE), high-pressure solvent extraction (HPSE)	The method is similar to Soxhlet extraction, but the solvent has high extraction properties.	The combination of high temperature and pressure to increase the solubility and diffusion of the solvent is difficult.
Enzyme-Assisted Extraction (EAE)	EAE uses enzymes for degradation. Cellulose and hemicellulose enzymes were used to extract the molecules.	Does not use organic solvents but high yield.	Extraction results depend on the time, pH, and enzyme temperature. Efficiency depends on the nature of the enzyme.

Microwave-Assisted Extraction (MAE)

Microwave-Assisted extraction (MAE) is extraction using a microwave system that causes heat directly into the matrix through friction and collisions between molecules resulting in rapid heating. MAE has advantages such as short extraction time, less solvent, and better extraction ability at lower costs than conventional extraction methods. MAE has been used to extract hydrocolloids from red and brown seaweeds (Khalil et al., 2017). Factors such as wave frequency, extraction time, solvent concentration, matrix characteristics, solid-to-liquid ratio, extraction pressure, solvent properties, and temperature must be streamlined during extraction (Han et al., 2010).

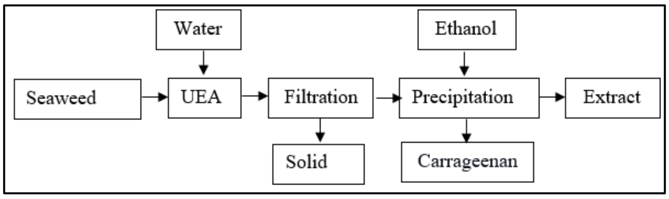

Figure 3. Ultrasound-assisted extraction of carrageenan.

Ultrasound-Assisted Extraction (UAE)

Ultrasound-Assisted extraction (UAE) is more efficient than conventional extraction and some other green extractions. Ultrasound allows greater solvent penetration into the sample by increasing the contact surface between the solid and liquid phases. As a result, the solute will quickly diffuse from the solid state to the solvent (Grosso et al., 2015). This method significantly improves the extraction yield with more effective mixing, faster energy and mass transfer, selective and faster response to the extraction process. Carrageenan extraction showed promising results with the UAE method, even the modified brown seaweed showed the same results as carrageenan (Han et al., 2010).

Supercritical extraction method (SFE)

The supercritical extraction method (SFE) is used to remove unwanted materials from the product. During the extraction process, temperature and pressure quickly penetrate into the material so that a pure extraction result is obtained. The water content of the material, fluid, solvent flow rate, temperature, pressure, and particle size must be considered regarding extraction efficiency (Heng et al., 2013). This method can be applied to the extraction of seaweed hydrocolloids by modifying the temperature, pressure, or addition of solvents that can prevent the degradation of the extract (Khalil et al., 2017).Using carbon dioxide as a solvent is very effective in extracting bioactive compounds to prevent the degradation of hydrocolloids at high temperatures (Jacobsen et al., 2019). Some of the advantages of SFE: using non-hazardous solvents, extraction time efficiency, extract results have good quality, and the solvent does not pollute the environment (Kristanto et al., 2021).

Pressurized solvent extraction (PSE)

Pressurized solvent extraction (PSE) is extraction using water as a solvent. The solvents propane, dimethyl ether, methanol, and ethyl acetate can also be used, wherein ethyl acetate is more effective (Jacobsen et al., 2019; Han et al., 2010). This method has a high extraction efficiency and a small volume of solvent with a short extraction time, where the combination of temperature and pressure can increase the solubility and diffusion of the solvent into the material, thereby increasing the extraction yield. PSE can extract seaweed hydrocolloids, especially polyphenols and fucoidan (Khalil et al., 2017; Jacobsen et al., 2019).

Enzyme-Assisted Extraction (EAE)

Enzymatic extraction aims to minimize the use of chemical solvents so that the hydrocolloid does not undergo degradation due to high alkaline concentrations (Ale et al., 2011; Ale & Meyer, 2013). Temperature, pH, enzyme substrate, and type of solvent (water or buffer) must be considered to increase the efficiency of seaweed extraction (Rhein-Knudsen et al., 2015). This method can also change the materials not soluble in water to be soluble in water, so it is significant for seaweed gel formation with a yield of 28.65% (Khalil et al., 2017). In addition to using microorganisms, enzymatic extraction can be done by combining carrageenan with cellulose to extract the protein in seaweed (Rhein-Knudsen et al., 2015).

Table 4. Green extraction methods research results

Extraction methods	Research results
Microwave-assisted extraction (MAE)	- Extracts of carrageenan and agar were obtained without further purification at optimum conditions. - Alginate was extracted from brown seaweed through parameter optimization. - Significantly increase yield at low temperature.
Ultrasound-assisted extraction (UAE)	- Accelerates the extraction of high molecular weight phenolics from brown seaweed.
Supercritical extraction method (SFE)	- High purity carrageenan yield
Pressurized solvent extraction (PSE)	- Calcium alginate matrix was obtained from sodium alginate solution pre-treated with CO.
Enzyme-assisted extraction (EAE)	- Extracted from solvent-free seaweed.

Alternative Method: Reactive Extrusion

Alkaline extraction is thought to affect the rheological properties of hydrocolloids, with several disadvantages: long time to achieve optimal extraction results, the volume of reactants and high water usage. Reactive extrusion tends to reduce the use of time, water, and sodium carbonate as reactants but still produces high-quality alginate hydrocolloids. The extraction yield, purity, and rheological properties of the extracts obtained by reactive extrusion were comparable to those of conventional batch extraction. This method is limited to alginate extraction, especially in method modeling and alginate optimization. The efficiency of this method still needs to be tested to extract hydrocolloid agar and seaweed carrageenan (Khalil et al., 2017).

Conclusion

The choice of extraction method needs to consider factors, including recovery rate, temperature, pH, time, volume and type of solvent, and method. Solvent extraction is a conventional method for separating seaweed hydrocolloids, which at high temperatures and long extraction times can cause degradation of polysaccharides. On the other hand, hydrocolloid extraction can be carried out using green extraction methods, which are efficient, short time, and consume less solvent. This method can potentially extract bioactive seaweed compounds efficiently and effectively with maximum results.

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