Carbohydrate Polymers 98 (2013) 89–94

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

journal homepage: www.elsevier.com/locate/carbpol

Preparation and characterization of mucilage polysaccharide for biomedical applications

G. Archana[a], K. Sabina[a], S. Babuskin[a], K. Radhakrishnan[a], Mohammed A. Fayidh[a], P. Azhagu Saravana Babu[a], M. Sivarajan[b], M. Sukumar[a,*]

[a] Centre for Biotechnology, A.C. Tech., Anna University, Chennai 600025, India
[b] Chemical Engineering Division, Central Leather Research Institute, Adyar, Chennai 20, India

ARTICLE INFO

Article history:
Received 13 February 2013
Received in revised form 5 April 2013
Accepted 17 April 2013
Available online xxx

Keywords:
Carbohydrates
Polysaccharide
Swelling index
Emulsifying capacity
Abelmoscus esculentus
Gracilaria corticata

ABSTRACT

In the present investigation, the polysaccharide/mucilage from waste of *Abelmoscus esculentus* by modification in hot extraction using two different solvents (Acetone, Methanol) were extracted, characterized and further compared with seaweed polysaccharide for their potential applications. The percentage yield, emulsifying capacity and swelling index of this mucilage were determined. The macro algae and okra waste, gave high % yield (22.2% and 8.6% respectively) and good emulsifying capacity (EC% = 52.38% and 54.76% respectively) with acetone, compared to methanol (11.3% and 0.28%; EC% = 50%) (PH = 7) while swelling index was greater with methanol than acetone extracts respectively. The infrared (I.R.) spectrum of the samples was recorded to investigate the chemical structure of mucilage. Thermal analysis of the mucilage was done with TGA (Thermal Gravimetric Analyzer) and DSC (Differential Scanning Calorimeter) which showed both okra and algal polysaccharide were thermostable hydrogels.

1. Introduction

Mucilage and gums are known as rich sources of polysaccharides since ancient times. They are widely used in the pharmaceutical & food industries as thickeners, water retention agents, and emulsion stabilizers, suspending agents, binders etc. Apart from their role in finished medicinal products, newer uses have been found in the preparation of cosmetics, textiles and paper industry. The okra plant *Abelmoschus esculentus* (L.) Moench, a native plant from Africa, is now grown in now commonly other areas such as Thailand, the Middle East India and the southern states of the USA. The optimum yield of okra is approximately estimated as 6.6 t ha^{-1}. Its water extracts are thick and slimy due to the presence of polysaccharides and are used to thicken soups and stews. Commercially these mucilage polysaccharides are known as hydrocolloids based on their wide range of functional properties. The okra or "gumbo" pod as a whole is used as vegetable for human consumption. But the upper part of the okra pods (upper crown head) are commonly removed or cut and thrown prior to cooking as waste (Al-Barak & El-Said, 2010). This part of okra pod also contains mucilage which gives its slimy viscous characteristic. Carbohydrates are mainly present in the form of mucilage (Sengkhamparn, Bakx, et al., 2009). Okra pods specially when heated produce more sticky mucus. This mucilage has a good potential for pharmaceutical applications such as diluents, binder, pharmaceutical excipients over synthetic excipients, disintegrant in tablets, thickeners in oral liquids, protective colloids in suspensions, film forming agents in transdermal and periodontal films and gelling agents in gel base for topical application due to ease of application for better percutaneous absorption. The main components of this mucilage are galactose (25%), rhamnose (22%), galacturonic acid (27%) and amino acids (11%) (Sengkhamparn et al., 2010). Previous studies showed okra fruit rhamnogalacturonans polysaccharide component increased cell proliferation. Savello et al. (1980), showed potential application of okra mucilage as a plasma replacement or blood volume expander. Similarly the phycocolloid polysaccharides obtained from some families of red macro algae (Rhodophyta), mainly Gracilariaceae and Gelidiaceae (Armisen, 1995) are widely used in pharmaceutical, cosmetics and food industry. Agar is essentially a mixture of the neutral polymer agarose, pyruvated agarose and sulfated galactans (Alves, Caridade, Mano, Sousa, & Reis, 2010). The ability of seaweeds to produce secondary metabolites of antimicrobial value, such as volatile components (phenols, terpenes) was well investigated in previous studies (Shukla, Kumar, Prasad, Reddy, & Jha, 2011). The objective of this study is to use a simple algal mucilage polysaccharide extraction procedure for extraction of mucilage polysaccharide from okra waste, thereby evaluate & compare the properties of crude mucilage polysaccharides for their potential industrial applications.

* Corresponding author. Tel.: +91 44 22358385; fax: +91 44 2235 0299.
E-mail address: sukumaractech@gmail.com (M. Sukumar).

0144-8617/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.carbpol.2013.04.062

The aim of the present study was to use a different extraction method for okra mucilage polysaccharide (Distantina, Wiratni, Fahrurrozi, & Rochmadi, 2011) and to compare, investigate its properties with that of marine algal polysaccharide for their potential role in development of biopolymer for tissue engineering.

2. Materials and method

2.1. Sample collection and preparation

Seaweeds species (*Gracilaria corticata*) was collected from mandapam coast in Gulf of Mannar, Tamilnadu southeast coast of India at a latitude 9°45′N and longitude 79°0′E on low tide during December 2011. Collected samples were washed with tap water to remove epiphytes and other marine organisms. The samples were transported to the laboratory in sterile polythene bags. In the laboratory, samples were rinsed with tap water and were shade dried and powdered in a mixer grinder. Similarly okra bio waste (upper crown head) was collected from canteen, Anna University, Chennai, & rinsed with tap water and was shade dried. Dried okra wastes were broken to powder form in an electrical mixer. It was then sieved into particle sizes 0.5–4 mm and stored in glass bottles for further use. All reagents used in the experiment had analytical purity.

2.2. Extraction

The mucilage polysaccharides were extracted by the method followed by Distantina et al. (2011). 5 g of clean, dried seaweed and okra powder sample was soaked in distilled water for 15 min. After soaking, the water was separated from the seaweed by filtration. Firstly, a known amount of solvent (methanol, acetone; (1/50 g/mL)) was heated in a beaker as an extractor which emerged in a water bath equipped by a stirrer. If the temperature of solvent reached 85 °C (Distantina et al., 2011), the samples then were added into solvent, and the time of extraction started was counted. The speed of stirrer was set constant at 275 rpm. The constant ratio of seaweed weight to solvent volume (1/50 g/mL) was maintained by adding hot water. The extraction was stopped after 45 min. Filtrate was separated from residue using filter cloth and immediately poured into 3 volumes of cold (5 °C) acetone (90% w) which caused precipitation of polysaccharides. The precipitation was done for 30 min with stirring gently by hand. The precipitated polysaccharides were collected and oven dried at 50–60 °C to a constant weight. The experiments were carried out with different solvent such as acetone and some parameters namely the yield, chemical structure, swelling index, emulsifying capacity of the extracted polysaccharides were determined.

2.3. Chemical analysis

2.3.1. Organoleptic evaluation of polysaccharide

Both okra and seaweed polysaccharide were subject to qualitative evaluation based on the study of morphological and sensory profile by the organs of sense (skin, eye, tongue, nose and ear) and by macroscopic evaluation which includes color, odor, taste, size, shape and special feature, like touch, texture etc. The organoleptic characters of both the mucilage were found acceptable. The crude polysaccharides showed low solubility due to the presence of impurities, high molecular weight or large molecules and insoluble matters. Full solubility is beneficial from the view point of appearance and texture. Therefore, it is necessary to achieve the maximum solubilization in order to maintain the functionality in biopolymer development as reported earlier (Amid & Mirhosseini, 2012) and presented in Table 1

Table 1
Organoleptic evaluation of selected polysaccharide.

Parameter	*Abelmoscus esculentus*	*Gracilaria corticata*
Color	Cream	White
Odor	Honey	Odorless
Taste	Tasteless	Tasteless
Shape	Irregular	Irregular
Touch and texture	Hard and rough	Hard and rough
Solubility	Slightly soluble	Slightly soluble

2.4. Percentage yield

The percentage yield was calculated based on the amount of dry powder sample used for the extraction process and the amount of dry water soluble mucilage/polysaccharide obtained after the extraction. The percentage yield (mg/g dry extract) was calculated using the following formula as mentioned below,

$$\%\,\text{yield} = \frac{\text{wt. of dried mucilage obtained}}{\text{wt of power taken}} \times 100$$

2.5. Swelling index

In this study, 1.0 g of dry water soluble mucilage/polysaccharide was placed in a 100-mL stopper graduated cylinder. The initial volume of the dry mucilage/polysaccharide was measured. 2-mL of alcohol (95%) was added for good dispersion and then distilled/demineralized water was added to sufficient quantity to yield 100-mL of uniform dispersion. The viscous solution was stored at room temperature and the sediment volume of the swollen mass was noted after 24 h. The swelling ratio was calculated by determining the ratio of the swollen volume to the initial bulk volume using the formula.

$$S = \frac{V_2}{V_1}$$

where S is the swelling index;V_2 is the volume occupied by the gum prior to hydration; V_1 is the volume occupied by gum after hydration.

2.6. Emulsifying capacity

Mucilage powder was used to determine the emulsion capacity. The samples were accurately weighed (1.0 g), dissolved in 50 ml distilled water, and added with 50 ml refined oil (Thanatcha & Pranee, 2011). Then the emulsion was prepared by homogenizing the above mixture for 1 min and centrifuged with 4100 × g for 5 min. Finally measured the height of emulsified layer compared with the height of whole layer and calculated the emulsion capacity by the following equation.

$$\text{Emulsifying capacity}\,\% = \frac{\text{Height of emulslfied layer}}{\text{Height of whole layer}} \times 100$$

2.7. Infrared spectroscopy (IR)

A potassium bromide disk of each of the dried mucilage was prepared, and the infrared spectra recorded (Perkin-Elmer 720) between 4000 and 650 cm^{-1}. By referring to wavelengths of polysaccharides from previous research, the result showed the chemical groups corresponding the wave number of bonds cm^{-1}. Apart from usual bands for hydroxyl (3500 cm^{-1}) and ester carbonyl (1730 cm^{-1}) groups, Nitrogen of protein (1550 cm^{-1}), amide deformation (1630 cm^{-1}) and ether group (1220 cm^{-1}) can be distinguished. The IR spectra of crude seaweed extracts were also determined.