

AN *IN-VITRO* STUDY ON PLANT POLYSACCHARIDES WITH FOOD PRESERVATIVE PROPERTIES

Submitted in partial fulfillment of the requirements for the award of
Bachelor of Technology degree in Biotechnology

By

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SCHOOL OF BIO AND CHEMICAL ENGINEERING**

SATHYABAMA

**INSTITUTE OF SCIENCE AND TECHNOLOGY
(DEEMED TO BE UNIVERSITY)**

Accredited with Grade "A" by NAAC

JEPPIAAR NAGAR, RAJIV GANDHI SALAI, CHENNAI - 600 119

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This is to certify that this Project Report is the bonafide work of Rajeshware V (37230066) and Saranya S (37230079) who carried out the project entitled "An *in-vitro* study on plant polysaccharides with food preservative properties" under my supervision from January 2021 to April 2021.

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DECLARATION

We, Rajeshware V (37230066) and Saranya S (37230079) hereby declare that the Project Report entitled 'An *in-vitro* study on plant polysaccharides with food preservative properties' done by us under the guidance of Dr. Valli Nachiyar at Sathyabama Institute of Science and Technology; Jeppiaar Nagar, Rajiv Gandhi Road, Chennai - 600 119 is submitted in partial fulfillment of the requirements for the award of Bachelor of Technology degree in Biotechnology.

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ABSTRACT

Plant polysaccharides are natural biopolymers which are widely distributed in the cells of plants. Polysaccharides are widely distributed in nature and are produced by all organisms including plants, animals, and microorganisms. These molecules have varied biological functions such as storage of energy (starch), cell wall architecture (cellulose), and cellular communication (glycosaminoglycans). These are such large macromolecules that can attain several millions of Daltons molecular weight. Polysaccharides may be homopolymers or heteropolymers, may contain neutral (pentoses and hexoses) or anionic sugars (uronic acid), and may or may not have linked nonsugar compounds. Many plant polysaccharides have been shown good emulsifying and thickening effects, therefore, they are widely used as stabilizers in emulsions. Polysaccharides may function in foods as viscosifying agents, stabilizers, emulsifiers, gelling agents, or water-binding agents. The global market for hydrocolloids, which includes many polysaccharides, is still dominated by plant and algae derived polysaccharides such as starch, galactomannan, pectin, carrageenan and alginate. Polysaccharide is the most abundant organic macromolecules in nature. Some of the microbial polysaccharides commercially in use in food industry are bacterial cellulose, pullulan, xanthan gum, gellan gum, bacterial alginates, levan, and hyaluronic acid. However, extracellular polysaccharides (EPS) are mainly used in food industry as compared to intracellular polysaccharides

Keywords: Plant polysaccharides, food preservative, natural antioxidants

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CHAPTER – 1 INTRODUCTION

1.1 PLANT POLYSACCHARIDE

Plant polysaccharides are structural constituents of the cell walls, so the extraction method depends on the cell wall structure. In general, tea polysaccharides are extracted using solvent extraction (water, acidic, or dilute alkaline solution) after pretreatment to remove the fat, monosaccharide, oligosaccharides, alkaloids and polyphenols in tea materials

The major role of polysaccharides in the food industry is to gel the aqueous phase. Hydrogen bonding and ionic interactions in a large number of polysaccharide molecules result in ordered polysaccharide conformation structure, that is, the gel structure is formed. The properties of the gel are dependent on the polysaccharides involved in the association that form a confirmed structure.

There can be different forms of polysaccharides in aloe. Their content depends on the age of the plant. Aloe contains a soluble fiber fraction, i.e., glucomannan and a hemicellulose component that binds to fibroblast receptors in the cell walls of some plants, enhancing their proliferation. Thus, it accelerates the healing of wounds. Aloe also contains lignin's, which aid the absorption of its components through the skin. In consequence, more collagen is produced when aloe is administered locally or externally. Mucopolysaccharides are a special group of polysaccharides in aloe.

1.2 EXOPOLYSACCHARIDE

B Microbial polysaccharides represent a class of important products that are of growing interest for many sectors of industry. The advantages of microbial polysaccharides over plants polysaccharides are their novel functions and constant chemical and physical properties. A number of common marine bacteria widely distributed in the oceans can produce EPSs.

In recent years, there has been a growing interest in isolating new exopolysaccharides

(EPSs)-producing bacteria from marine environments, particularly from various extreme marine environments. Many new marine microbial EPSs with novel chemical compositions, properties, and structures have been found to have potential applications in fields such as adhesives, textiles, pharmaceuticals, and medicine for anticancer, food additives, oil recovery, and metal removal in mining and industrial waste treatments, etc. General information about the EPSs produced by marine bacteria, including their chemical compositions, properties, and structures, together with their potential applications in the industry.

1.3 ALOE BOTANICAL CHARACTERIZATION

It is a short-stemmed perennial that grows to a height of 60–100 cm. Aloe plants have thick, green or grey and green fleshy, sword-shaped leaves. The leaf edges have triangular thorns at their edges. The flower shoot, which grows in the summer, is built from numerous, pendulous bell-shaped pink and orange flowers. When the plant sheds blossom, it gives fruit in the form of bags. A flesh and pulp obtained from aloe leaves differ in composition and properties. Aloe flesh can be obtained by peeling leaves and then washing and squeezing them carefully. This procedure gives pure flesh without bitter aftertaste or strong laxative properties. Aloe flesh is light green with jelly-like consistency. It is composed of water (96%) and dry matter (4%), which contains protein (6.86%), fat (2.91%), dietary fiber (73.35%), ascorbic acid (0.004%) and ash (16.88%). Apart from flesh, aloe pulp contains epidermis. It is not washed nor filtered, consequently having strong laxative properties due to the content of aloin.



Figure 1.1 aloe vera plant

1.4 PLANT POLYSACCHARIDE IN PRODUCTION

Plant polysaccharides have achieved industrial production and are widely used in food, medicine, cosmetics, chemicals, and other fields. Biological activity and gel properties are two important properties of polysaccharide macromolecules. For their special physiological activities, such as immunoregulation, antitumor activity, anti-AIDS, and antioxidation effect, polysaccharides are applied in health foods as raw materials or ingredients, such as lentinan, Ganoderma lucidum, and dendrobium polysaccharides, etc. Furthermore, polysaccharides also have good gel property and are used as thickeners, emulsifiers, and stabilizers in goods, such as carageen and konjac gum. Aloe vera have an anti-inflammatory effect and support lipid and carbohydrate metabolism, helping to maintain normal sugar and cholesterol levels in blood and normal body weight. When aloe is applied externally, it accelerates the regeneration of the damaged skin. Aloe contains antioxidants, which may increase the shelf-life and nutritional value of food; therefore, it is widely used in cosmetic, pharmaceutical and food industry. An antioxidant activity was shown for leaf's skin, flowers and gel of aloe.



Figure 1.2 aloe vera fresh produce

1.5 ALOE VERA LEAF

The innermost part of the leaf is a clear, soft, moist, and slippery tissue that consists of large thin-walled parenchyma cells in which water is held in the form of a viscous mucilage. Therefore, the thick fleshy leaves of aloe plants contain not only cell wall carbohydrates such as cellulose and hemicellulose but also storage carbohydrates such as acetylated mannans.



Figure 1.3 aloe leaf

1.6 ALOE VERA LEAF

Processing of the leaf pulp has become a large worldwide industry. In the food industry, it has been used as a source of functional foods and as an ingredient in other food products, for the production of gel-containing health drinks and beverages. In the cosmetic and toiletry industry, it has been used as base material for the production of creams, lotions, soaps, shampoos, facial cleansers, and other products. In the pharmaceutical industry, it has been used for the manufacture of topical products such as ointments and gel preparations, as well as in the production of tablets and capsules. Important pharmaceutical properties that have recently been discovered for both the A. vera gel and whole leaf extract include the ability to improve the bioavailability of co-administered vitamins in human subjects

1.7 THERAPEUTIC EFFECTS OF ALOE

A. vera has been used for many centuries for its curative and therapeutic properties, and although over 75 active ingredients from the inner gel have been identified, therapeutic effects have not been correlated well with each individual component. Many of the medicinal effects of aloe leaf extracts have been attributed to the polysaccharides found in the inner leaf parenchymatous tissue, but it is believed that these biological activities should be assigned to a synergistic action of the compounds contained therein rather than a single chemical substance.

1.8 ALOE EMPLOYED IN DRUG ADMINISTRATION

Due to its absorption enhancing effects, A. vera gel may be employed to effectively deliver poorly absorbable drugs through the oral route of drug administration. Furthermore, the dried powder obtained from A. vera gel was successfully used to manufacture directly compressible matrix-type tablets. These matrix-type tablets slowly released a model compound over an extended period of time and thereby showing potential to be used as an excipient in modified release dosage forms.

1.9 ALOE VERA BIOACTIVE COMPONENTS

Aloe contains a large amount of bioactive compounds, such as flavonoids, terpenoids, lectins, fatty acids, anthraquinones, mono- and polysaccharides (pectin's, hemicelluloses, glucomannan), tannins, sterols (camp sterol, β -sitosterol), enzymes, salicylic acid, minerals (calcium, chromium, copper, iron, magnesium, manganese, potassium, phosphorus, sodium and zinc) and vitamins (A, C, E, β -carotene, B1, B2, B3, B6, choline, B12, folic acid). Aloe contains numerous enzymes such as: alkaline phosphatase, amylase, brady kinase, catalase, lipase, protease, creatine phosphokinase, carboxypeptidase, cellulose, and oxidase. Superoxide dismutase is the most important and active enzyme found in aloe. There is diversified content of phenolic compounds in individual morphological parts of aloe. The content of phenolic compounds in aloe's leaf epidermis is higher than in flowers.

CHAPTER – 2

LITERATURE SURVEY

2.1 POLYSACCHARIDES

Polysaccharides are widely distributed in nature and are produced by all organisms including plants, animals, and microorganisms. These molecules have varied biological functions such as storage of energy (starch), cell wall architecture (cellulose), and cellular communication (glycosaminoglycans) (Ates, 2015; Held et al., 2015). These are such large macromolecules that they can attain several millions of Daltons molecular weight (Braun et al., 2001; Delattre et al., 2011). Polysaccharides may be homopolymers or heteropolymers, may contain neutral (pentoses and hexoses) or anionic sugars (uronic acid), and may or may not have linked Non sugar compounds (Aluwihare and Repeta, 1999). Although the majority of the polysaccharides currently being used in the industry are of plant and seaweed origin, polysaccharides from microorganisms with novel and unique physical properties have recently emerged as important biopolymers. Nowadays, polysaccharides of microbial origin have proved themselves as new polymeric materials that are competing economically with other natural gums, such as Arabic gum and carrageenan produced by plant and marine algae, respectively (Milani and Maleki, 2012; Shit and Shah, 2014). Use of microbial polysaccharides has several advantages as compared to already available synthetic and plant polysaccharides

2.2 ISOLATION AND IDENTIFICATION OF POLYSACCHARIDES

Various types of a given polysaccharide may be produced by controlling the source and isolation procedure. EPS are isolated from culture broths after fermentation by an appropriate organism. The crude extracts consist of mixtures of proteins, polysaccharides, and secondary metabolites. Purification is necessary to obtain products that are suitable for human or animal consumption or for use in food, pharmaceuticals, and other industries (Beutler, 2009; Baiano, 2014). Analytical

ultracentrifugation has provided valuable information on physical properties of polysaccharides in solution, mainly because of the technique's diversity, absolute nature, and inherent fractionation ability without the requirement of columns and membranes for separation (Harding, 2005; Oberlerchner et al., 2015). Size-exclusion chromatography, anion-exchange chromatography, gas– liquid chromatography, and high-performance liquid chromatography are also useful for isolation and characterization of polysaccharides (Francisco et al., 1998; Harding 2005; Khan et al., 2007; Muffler and Ulber, 2005).

2.3 ALOE VERA

Aloe vera is a common succulent plant that has been used for centuries regarding their healing properties and health benefits. Nowadays, scientific investigations on its gel have gained more attention because of its interesting antioxidant and antimicrobial properties. Also, the food industry encounters the need to preserve safety and quality of fresh produce; fruits and vegetables are in high demand due to their reported health benefits, and fresh-cut products are a new trend that meets the restless needs of the society. Edible coatings are an effective way to maintain freshness of these products, extend their shelf life, and even act as an alternative to modified atmosphere packaging to be used in conventional packaging.

Aloe vera gel is a natural hydrocolloid, composed mainly by polysaccharides, that has been applied in the last years on fruits and vegetables. It can act as a semipermeable barrier for gases and water vapor, decreasing the respiration and ripening processes of the fruit, thus maintaining weight, firmness and valuable compounds. Its antioxidant and antimicrobial properties make it also an interesting material for increasing the shelf-life of fruits and vegetables. This review aims to describe the preparation and preservation of Aloe vera gel as well as the properties and compounds that are effective against oxidation and microbial growth. Moreover, the recent findings of its use – with or without additives – as an edible coating on fruits and vegetables have been widely detailed, showing that Aloe vera gel is a promising preservative method in this industry.

2.4 ANTIOXIDANTS

Antioxidants are those substances which at low concentration inhibits the oxidizable substrate oxidation. In vitro antioxidant potential was reported by the methanol extracts of leaf epidermis and flower of *A. vera* (López et al. 2013). The radical scavenging activity of most Aloe species is due to presence of chromone glycosides.

It is noteworthy that the flower extract contains phenolic compounds such as gentisic acid, quercitrin and epicatechin. These phytochemicals are proved to highly significant as an antioxidant agent which will eventually help in the inhibition of diabetic cells and proliferation of normal cells (López et al. 2013). Furthermore, the flower extract of *A. vera* has been reported to possess anti-inflammatory effects due to the combination of extracted phytochemicals, which will also be beneficial in their antidiabetic effects (Vazquez et al. 1996, Nejatzadeh-Barandozi and Enferadi 2012).

2.5 FOOD PRESERVATION

A wide range of foods is used in the form of colloids. Emulsions of oil in water are an important component of a group of food products in which oils or lipids, water, and other ingredients are processed to form sauces, spreaders, and so forth. Some of the emulsion-containing food systems are marketed in dried form and are reconstituted, just before use. Such emulsion food products require polysaccharides for stabilization (Akbarzadeh et al., 2012, 2013).

Xanthan is an important polysaccharide, which has found many applications in dry mix food products. It prevents the constituents from reverting to their original separate phases and maintains long-term stability, which is very essential for food products (Ahmed and Varshney, 2011; Siracusa et al., 2008). Salad dressings, relishes, and yogurts in which polysaccharides are used as stabilizers are of relatively low pH. Thus, polysaccharide being used for this purpose must be acid stable (Cho et al., 2006; Gaaloul et al., 2009; Rafe et al., 2013).

The major role of polysaccharides in the food industry is to gel the aqueous phase.

Hydrogen bonding and ionic interactions in a large number of polysaccharide molecules result in ordered polysaccharide conformation structure, that is, the gel structure is formed (Banerjee and Bhattacharya, 2012; Brunchi et al., 2014; Saha and Bhattacharya, 2010; Sman, 2013). The properties of the gel are dependent on the polysaccharides involved in the association that form a confirmed structure.

CHAPTER – 3

AIM AND SCOPE

3.1 AIM

The primary aim of this investigation is to study the food preservative properties of aloe vera in an *in-vitro*, artificial, liquid medium supplemented with varying concentrations of lead. The secondary aim is to study associated direct growth kinetics and the effect of the heavy metal, lead, on the growth rate retardation of *B.monneri*.

3.2 OBJECTIVES

Below objectives were outlined before undertaking this study, these are:

- To extract Aloe vera and purify it
- To optimize the leaf gel extract and obtain 95 % ethanol leaf gel
- To identify the presence of flavonoids & polyphenols by exposing the LGE to diethyl ether, ethyl acetate.
- To analyze the various substances in LGE using Gas chromatography and mass spectrometry.
- To obtain antioxidant assay

CHAPTER – 4

MATERIALS AND METHODS

4.1 MATERIALS

4.1.1 Plant Materials

A long leaf was taken from a single parent plant

4.1.2 Chemicals

1. 95% ethanol
2. 3-phenylbutyric acid (25 mg/50 mL)
3. sodium acetate buffer (0.125 M)
4. β -Glucuronidase (30 μ L)
5. ethyle acetate
6. di ethyl acetate
7. liquid nitrogen
8. bis trifluoroacetamide (BSTFA, 100 μ L)
9. folin's reagent
10. sodium carbonate (7.5%, w/v)
11. H₂O
12. 10% (w/v) AlCl₃
13. 5% (w/v) NaNO₂
14. 1 M NaOH

4.1.3 Apparatus

1. 250 ml beakers
2. 250 ml conical flasks
3. 30ml test tubes
4. test tube racks
5. centrifuge tubes
6. eppendorf tubes

7. 50-100 ml cylinder
8. 1-5 ml glass pipettes
9. 0.2-1 ml glass pipettes
10. 2-1000 µl micropipettes
11. 250-500 ml standard measuring flasks

4.1.4 Instruments

1. Autoclave
2. Incubator
3. Electronic Balance
4. Calorimeter
5. Homogeniser
6. U-V Spectrophotometer
7. Refrigerator
8. Vortex Mixer
9. Centrifuge
10. GC-MS

4.2 METHODOLOGY

4.2.1 PREPARATION OF ALOE EXTRACT

Whole, freshly cut aloe vera leaf was taken from single parent plant which was home grown from uncultivated farmlands located near mambakkam in the month of December 2020. the inner leaf gel was removed, homogenized, freeze-dried, and stored at -2°C until analysis. This was termed the leaf gel extract (LGE) for the purpose of this study. Approximately half of the LGE was used for the preparation of a 95% ethanol extract as described previously. This was termed the 95% ethanol leaf gel extract (ELGE).

4.2.2 ETHYL ACETATE / DIETHYL ETHER EXTRACTION

The internal standard, 3-phenylbutyric acid (25 mg/50 mL), was added to 25 mg of finely ground LGE and ELGE, followed by the addition of 1 mL of sodium acetate buffer (0.125 M). β -Glucuronidase (30 μ L) was added, and the sample was vortexed and incubated overnight at 37°C. The sample was extracted with 6 mL of ethyl acetate followed by 3 mL of diethyl ether. The organic phase was collected after each extraction via centrifugation. The organic phase from each extraction was pooled and dried under nitrogen. The dried extract was derivatized with bis(trimethylsilyl)trifluoroacetamide (BSTFA, 100 μ L), trimethylchlorosilane (TMCS, 20 μ L), and pyridine (20 μ L) at 70°C for 30 min. After cooling, 0.1 μ L of the extract was injected into the GC-MS via splitless injection.

4.2.3 GAS CHROMATOGRAPHY –MASS SPECTROMETRY

The gas chromatograph was equipped with an SE-30 capillary column (Agilent), a split/splitless injection piece (250°C), and direct GC-MS coupling (260°C). Helium (1 mL/min) was used as the carrier gas. The oven temperature program for analyzing the ethyl acetate/diethyl ether extract was an initial oven temperature of 40°C and was maintained for 2 min, followed by a steady climb to 350°C at a rate of 5°C/min. For the fatty acid analysis, an initial oven temperature of 50°C was maintained for 1.5 min and then allowed to increase to 190°C at a rate of 30°C/min. The oven temperature was maintained at 190°C for 5 min and then allowed to increase to 220°C at a rate of 8°C/min. The oven temperature was again maintained for 2 min and finally ramped to 230°C at a rate of 3°C/min and maintained for 24 min at this temperature.

4.2.4 ANTIOXIDANT ASSAY

TOTAL POLYPHENOL ASSAY

The total polyphenol content of the extracts were determined according to the Folin-Ciocalteu procedure [20]. Briefly, 10 mg of finely ground LGE or ELGE was dissolved in 200 μ L of H₂O in a test tube followed by 1 mL of Folin-Ciocalteu's reagent. This was allowed to stand for 8 min at room temperature. Next, 0.8 mL of sodium carbonate

(7.5%, w/v) was added, mixed, and allowed to stand for 30 min. Absorption was measured at 765 nm (Shimadzu UV-1601 spectrophotometer, Kyoto, Japan). The mean total phenolic content ($n = 3$) was expressed as milligrams of gallic acid (Sigma-Aldrich) equivalents per 100 g of wet and dry mass (mg of GAE/100 g) (standard deviation (SD)).

TOTAL FLAVONOID ASSAY

The total flavonoid content was measured using the $AlCl_3$ colorimetric assay [21] with some modifications. Briefly, 10 mg of LGE or ELGE was dissolved in 1 mL of H_2O , to which 60 μL of 5% (w/v) $NaNO_2$ was added. After 5 min, 60 μL of a 10% (w/v) $AlCl_3$ was added. In the sixth minute, 400 μL of 1 M $NaOH$ was added, and the total volume was made up to 2 mL with H_2O . The solution was mixed well, and the absorbance was measured at 510 nm against a reagent blank. Concentrations were determined using a catechin (Sigma-Aldrich) solution standard curve. The mean total flavonoid content ($n = 3$) was expressed as milligrams of catechin equivalents (CE) per 100 g of wet and dry mass (mg of CE/100 g) (SD).

CHAPTER 5

RESULTS AND DISCUSSIONS

5.1 CONCENTRATION OF PHENOLIC COMPOUNDS IN LGE AND 95% ELGE

The compounds identified and their quantities in the A. vera LGE and ELGE are summarized. Of all the compounds identified, the groups of compounds best described for their health benefits are the phenolic acids/polyphenols. Apart from these, various alkanes, pyrimidines, alkaloids, organic acids were also identified. One would, however, expect a far larger variety of alcohols to occur in Aloe and in far higher concentrations. For better extraction of these, headspace isolation by simultaneous purging should be used as described previously. However, by employing this method, one would extract far less of the other biologically important health-associated compounds. Therefore, to accomplish the aims of our study, alternative extraction procedures were used as described under the 'Methods' section using ethyl acetate/diethyl ether.

Table 1

Concentrations of GC-MS identified compounds from LGE and 95% ELGE

Compound	Concentration (ppm)		ELGE (per dry mass ELGE)
	LGE (per dry mass LGE)	ELGE (per dry mass LGE)	
Phenolic acids/polyp henols			
Phenol	14.32	30.12	1.3×10^4

5.2 CONCENTRATION OF TOTAL POLYPHENOLS AND FLAVONOIDS

Consequently, those polyphenol/phenolic compounds identified in lower concentrations may also be of value. Due to the fact that the majority of the phenolic acids/polyphenols identified in A. vera leaf gel in Table 1 are antioxidants and these compounds as a group occur in the highest concentrations, one would expect these to contribute to the majority of the antioxidant capacity measured in these extracts in Table 2. When

interpreting the data of this nature, one should keep in mind that using the concentrations of these antioxidant compounds alone is insufficient criteria for making predictions of individual contributions to oxidative stress. As previously described, this is due to the fact that the concentrations of individual polyphenol antioxidants are not the only factor influencing antioxidant capacity; the structural arrangements (number and position of hydroxyl groups, double bonds, and aromatic rings) of these compounds also play a role.

Table 2

Concentrations of total polyphenols, flavonoids

Compound	LGE (dry mass)	LGE (wet mass)	ELGE (expressed as dry mass ELGE)		ELGE (expressed as dry mass LGE)	ELGE (expressed as wet mass LGE)
Total polyphenols (mg of GAE/100 g $\pm$ SD)	78.2 $\pm$ 4.03	2.70 $\pm$ 0.14	413 $\pm$ 9.88	26.8 $\pm$ 0.63	0.93 $\pm$ 0.02	
Total flavonoids (mg of CE/100 g $\pm$ SD)	5.3 $\pm$ 0.38	0.19 $\pm$ 0.01	33.6 $\pm$ 1.98	2.15 $\pm$ 0.13	0.08 $\pm$ 0.003	

CHAPTER – 6

SUMMARY AND CONCLUSIONS

Production of value-added biochemicals from microbial biomass serves a better alternative to harsh chemical synthesis processes that employ expensive, hazardous, and nonrenewable raw materials. Although a biopolymer due to its functional characteristics may prove its market potential, unless the polymer is produced economically it cannot find its proper place. If there is any change in feedstock utilization for microbial polysaccharide, it requires intensive research activities for the application of innovative concepts on a large scale. These research activities include all novel supplies and pretreatments, as well as fermentation and downstream processing techniques. Choice of appropriate feedstock for polysaccharide production depends on the metabolic needs of the microorganisms. Various phenolic acids/polyphenols, phytosterols, fatty acids, indoles, alkanes, pyrimidines, alkaloids, organic acids, aldehydes, dicarboxylic acids, ketones, and alcohols were identified and quantified. Due to the presence of the antioxidant polyphenols, indoles, and alkaloids, the *A. vera* leaf gel shows antioxidant capacity as confirmed by ORAC and FRAP analyses. Both GC-MS and spectrophotometric analyses show the non-flavonoid polyphenols to contribute to the majority of the total polyphenol content. Another important aspect for microbial polysaccharides production is that physical and chemical properties of the polymers are largely determined by the cultivation conditions and the biomass used as a feedstock. Hence, the characterization of the microbial polysaccharides, as well as the mechanism of its biosynthesis, is very important in searching suitable biomass resources. Consistency in product quality and yield are important for any high-value biopolymer. This can be ensured to a large extent by preventing the metabolic byproducts. Thus, chemically defined media are usually preferred over complex media. Microalgae facilitate the use of carbon dioxide as a cheap, simple, and abundant carbon source and thus microalgae as a better feedstock for polysaccharides production is being considered. With more knowledge and awareness about the importance of microalgal polysaccharides, currently studies are focused on optimizing the fermentation conditions for polysaccharide production.

CHAPTER 7

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