CHAPTER 8

Summary and Conclusions
The interest in the application of natural and renewable polymers especially polysaccharides is increasing due to its non toxicity, water solubility, broad range of functional properties, in industrial applications of foods, textiles, paints, cosmetics and pharmaceuticals. The two major categories of natural storage polysaccharides, abundantly available in our region, namely \( \alpha \)-glucans (starch) and \( \beta \)-glucans (xyloglucan) were studied to enhance the commercial utility of the same. A brief overview of the recent developments in the field of polysaccharide nanoparticles, gels and films, scope of the present work and a detailed description of materials, methods and techniques used for the extraction, purification and characterisation of the glucans are given. The important conclusions are

- Starch as an alpha glucan, has many properties which can be utilized in the medicinal field. The medicinal utility of starch from rice and cassava was investigated. A study was undertaken on the unique physico chemical properties of starch from the Njavara rice. So the rice starch was compared with, Chamba rice’s starch which is our staple food. Njavara, unique rice, short duration cultivar grown only in certain pockets in Kerala state, south India and belongs to the family Oryza. This is the only cultivar traditionally used effectively in the Ayurvedic system of medicine in certain specific treatments like \textit{Panchakarma}. It was found that the njavara starch has high gelatinisation temperature, water absorption capacity, solubility, swelling power, pasting properties, high gel strength and better rheological properties. The starch degrades at higher temperature and has a higher enthalpy of gelatinization. The better thermal properties make it useful in products which need to be processed at a higher
temperature. It is because of its high heat holding capacity, the njavara rice is specifically used in *Ayurveda* treatments which needs sustained heat application such as *kizhyi*. Texture profile analysis showed that the properties of both starches are similar, however njavara starch has better springiness and chewiness. Both the starches are of A type X ray diffraction pattern, which is typical of cereal starches and hence are of similar crystalline nature.

- The chemical modification of cassava starch by grafting with long chain fatty acids and utilizing this partially hydrophobic starch to make nanoparticles was accomplished. Nanoparticles loaded with drugs show drug release at right rate and dose at specific sites in the body for certain duration to realize the accurate delivery, which enhances the therapeutic effect and reduces the toxicity and side effects of the drug. Starch grafted with oleic acid was obtained in good yield compared to that grafted with stearic acid (DS 0.1 to 1.0). These starches become slightly hydrophobic and have good thermal properties. Grafted starch has a high swelling power at neutral pH compared to the native starch. This behaviour is useful in drug delivery applications. Starch nanoparticles were prepared by crosslinking the starch molecules with sodium tripoly phosphate and controlled precipitation with a solvent of appropriate dielectric constant and then stabilizing with a surfactant. Controlled release of the drug previously loaded in nanoparticle was studied by using indomethacin as the model drug. This modified cassava starch nanoparticles was found to be an excellent vehicle for the controlled release of drug by slow dissolution.
Xyloglucan is a beta glucan and have applications in food, serving as a thickener and stabilizer, gelling agent, ice crystal stabilizer, and starch modifier, also in cosmetic and pharmaceutical industries. It is a neutral, non-toxic storage polysaccharide (backbone composed of 1, 4- linked β-D-glucopyranose residues) present in the seeds of the tamarind tree (*Tamarindus indica*) which is abundantly available in South India. Pure xyloglucan extracted from tamarind seed as such does not form a gel. Chemical derivatisation methods are employed to improve the properties of these intractable, inexpensive polysaccharides material. Chemical oxidation of xyloglucan was carried out to get dialdehyde xyloglucan. A transparent, colourless gel combining oxidised xyloglucan with chitosan in right proportion, having good compatibility for food and cosmetic applications was developed and named ‘Chitam gel’. The ultra micro structure revealed that, the gel has a fibrous network structure of chitosan intercalated with oxidized xyloglucan as nano sized elongated particles. The transparent gel has superior thermal properties compared to native xyloglucan. The chitam gel is obtained in high yield since 10 g raw material produces 1Kg gel, with 100 fold yield and hence cost effective. The chitam gel had a viscosity of 4100 cP at 28 ±2 °C, is thermostable from -20 to 90 °C. Does not decompose on prolonged exposure to sun light or on exposure to ultra violet rays. It is as a zero calorie gel, not digested by digestive enzymes in humans, and useful as a diet replacement, especially for diabetic patients. It can be incorporated into beverages, Jam, jellies, marmalades, to give the desired viscosity, gel characteristics and mouth feel and also found compatible with edible colours, flavors and artificial sweeteners.
(aspartame, alitame, saccharine). Very stable at acidic to neutral pH of 3 to 7 and hence can also be used as a vehicle for drugs in oral, topical, or wound healing patches. The chitam gel is easy to handle and hygienic and nontoxic. No need for heat or long periods of stirring for the gel preparation since the gel is formed at room temperature and at atmospheric pressure, costly processing equipments are not necessary. The desired final product viscosity can be tailor made from the chitam gel either modifying the percentage of xyloglucan, chitosan ratio or by diluting with suitable media. Apart from supplement functional foods (nutritional care) the chitam gel has applications in the area of cosmetic and personal care products, as a ultraviolet protective agent or as a tissue adhesive which can be used, including haemostasis, wound sealing, tissue engineering or localised drug delivery as capsules & tablets . The food and beverage market is also will benefit by the chitam gel as it has zero calorific value and can be used in beverages, jam, jellies, marmalades and ice creams useful for diabetic patients.

Considering the recent trends in the increasing applications of chitosan, an animal derived cationic polysaccharide, which vegetarians does not prefer, in drug and gene delivery, we envisaged to synthesise a plant derived cationic polysaccharide from tamarind kernel xyloglucan. This was accomplished by a facile synthetic strategy viz. amination of xyloglucan. The resultant product has gelling property and emits blue fluorescence in aqueous medium and green fluorescence in solid state. At very low concentration in aqueous medium, it forms a self assembled nano particles of size of ~60 nm and this unique property have great applications in delivery of drugs, nucleic
acids etc. The product showed good thermal properties and higher melting point compared to the non aminated xyloglucan. The crystalline nature decreases after amination as seen from the x-ray diffraction pattern. This property can be used for a variety of applications such as in drug targeting and diagnostics, since the material is non toxic, plant derived, low cost and abundantly available. This cationic xyloglucan hydrogels can also be used as a substitute for the animal derived chitosan for gene therapy, tissue engineering scaffolds and in various other novel applications.

- The film forming property of the xyloglucan and its composites were studied and good quality films were produced. Xyloglucan as such showed a good film forming property. The xyloglucan- chitosan composites forms films with good tensile strength, however the tensile strength decreases with increase in the concentration of xyloglucan. The contact angle measurements showed that xyloglucan- chitosan blend film was more hydrophobic in nature. High melting point of this blend film indicates its strength. Controlled release of the drug previously loaded in the films was studied by using streptomycine as the model drug. The films can be used for the preparation of therapeutic patches, drug release and in food industry.

**Future prospects:**

The studies on the preparation of composite polysaccharide micro/nano particles and films for novel applications were accomplished to a large extent and the new findings will pave way for renewed interest and more research in this area. The unique properties of njavara rice starch such as high thermal, pasting and rheological
properties over the commonly available chamba rice starch, was found to be the main reason for its specific use in ayurveda treatments. There may be other important properties of this njavara rice, such as antioxidant property which brings about many health benefits, may have to be investigated in future.

Starch as such is very hydrophilic and is not suitable for controlled release applications. Fatty acid grafted starch nano particle was found to be a good vehicle for the controlled oral drug delivery. This grafted starch become slightly hydrophobic and has good thermal properties and high swelling power at neutral pH compared to the native starch, which may useful not only in drug delivery but also for a variety of applications in personal care products, and cosmetics, food and beverage. Starch nanoparticles were stabilized by crosslinking with sodium tripoly phosphate, but there may be other options to cross link and stabilize the starch nano particles. Many new surfactants and solvents may be used for this purpose. The storage stability of nano particles in both emulsion as well as powder forms are to be carried out in detail. Novel methods of making nano particles in large scale have to be developed, so that the availability of nano particles are enhanced, which can be used in many more fields.

Today’s consumers in the cosmetic, food and beverage market are increasingly interested in healthy life styles, a trend which has produced a rising demand for health oriented products. Cosmetic and personal care products manufacturers claim that there is promising trend for transparent products for instance those which use clear formulation techniques in their gels and emulsions. The synthesis of a transparent,
colourless, nontoxic, biodegradable, biocompatible gel from oxidized xyloglucan and chitosan co-polymer having good thermal, antimicrobial and texture properties was accomplished. Hence the gels which is crystal clear, colourless, not thermoreversible, non toxic, biodegradable, biocompatible and having an ordered structure and from renewable resources and hence cost effective are in great demand globally. In addition, its antimicrobial and texture properties are very promising and hence the colourless, odourless, non toxic transparent gel will find use in specialty foods and in cosmetic applications.

Development of many more conjugates of a variety of non-starch polysaccharide molecules like the present xyloglucan –chitosan composite, to give colourless transparent gels, having good compatibility for food, cosmetic and other applications may be happening in the near future.

Cationic polysaccharides have wide applications in drug and gene delivery. Attempts have been made to make a plant derived cationic polysaccharide from xyloglucan. The resultant blue fluorescent gel will find use in new areas like biotronics and fluorescent labeling applications in biological area. Polycations and negatively charged nucleic acids can spontaneously form nanocomplexes by electrostatic interaction. Polycationic vector reduces the electrostatic repulsion between DNA and cell surface by neutralizing the negative charge and also protects it from enzymatic digestion by nucleases in serum and extra cellular fluids.
Xyloglucan as such showed a good film forming property. Xyloglucan chitosan composite forms a film with good tensile strength and is hydrophobic in nature and released streptomycin. Chemically modified xyloglucan gels and films with different functional properties will have significant applications in cosmetic, food and medical field.

Thus the xyloglucan from the tamarind seed kernel was found to be a versatile polysaccharide for further modifications to enhance its utility. Japan is using our tamarind kernel powder for a variety of functional properties and in India it is far under utilized. It is urgently needed to develop value added products from tamarind kernel. Further work can be done to develop tailor made particles, gels and films from this type of natural non toxic polysaccharides, which are cheap and abundant in nature, amenable to derivatisation, to impart new properties, and for better utilization of this natural resource.