Novel Approach of Extraction of Tamarind Seeds And Lady's Finger (Okra) Gum As A Binder For SR Tablet Formulation

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Abstract- Tamarind seed polysaccharide (TSP), a natural polysaccharide extracted from tamarind seeds is used in the pharmaceutical, textile and food industries as a mucoadhesive polymer. This work aimed to extract TSP from tamarind seeds Kernel powder of tamarind seeds was slurred into a clear solution, set aside overnight and then centrifuged at 6000 rpm for 20 min to separate all foreign matter Future works will focus on the quantitative analysis, biological activity and possible use of TSP as a drug delivery system.

Okra (Abelmoschus esculentus L.) is a flowering plant of the Malvaceae family which is also known as lady's finger, gumbo, bamya or bania or commonly known as bhindi. Natural polymers have been used in different pharmaceutical formulations. They are easily available, non-toxic, biodegradable and cost effective to be used as pharmaceutical excipients. In present investigation, we have reviewed about method for extraction and characterization of mucilage (Hibiscus esculentus) and further characterized to be used as pharmaceutical excipient. Main focus of review was to study about anti cancer activity of okra mucilage. Different methods for isolation and physicochemical method for characterization was focused. Antioxidant activity as well as IR spectra determination was noted. Okra is rich in phenolic compounds with important biological properties like quartering and Flavones derivatives, Catechin Oligomers and hydroxycinnamic derivatives. Okra is also known for being high in antioxidants activity. Okra has several potential health beneficial effects on some of the important human diseases like cardiovascular disease, type 2 diabetes, digestive diseases and some cancers.

Keywords- Okra, Excipients, Antioxidant and Anticancer, tamarind seed polysaccharide (TSP); Tamarind indica L.; natural polysaccharides

I. INTRODUCTION

Natural polymers or gums have been used in the preparation of release and controlled release drug dosage

forms, because of their great properties, such as biodegradability, non-toxicity, biocompatibility in Nature and swelling when they come in contact with aqueous media. Tamarind (Tamarind indica L.) belongs to the Leguminosae family The oil extracted from its seeds is rich in eicosanoic fatty acids such as palmitic, oleic and linoleic, the highest concentrations corresponding to linoleic acid and palmitic acid, present in 36%-49% and 14%-20%, respectively Tamarind seed polysaccharide (TSP), is a natural branched polysaccharide polymer with a molecular weight of 700-880 kDa . TSP constitutes about 65% of the tamarind seed composition .TSP is composed of a $(1\rightarrow 4)$ β -d-glucan backbone substituted with side chains of α -d-xylopyranose and β -d-galactopyranosyl linked $(1 \rightarrow 2) - \alpha$ -d-xylo-pyranose linked $(1\rightarrow 6)$ to glucose residues. The chemical constituents of TSP are glucose, xylose and galactose in a ratio of 2.80:2.25:1.00 . TSP, regarded as a galactoxylloglucan, is a novel polymer with various properties useful to the textile, food, and pharmaceutical industry. Singh et al. found that tamarind gum was a highly viscous, mucoadhesive and biocompatible natural polymer, which could be used for oral controlled drug release, ocular drug delivery systems and in the design of sustained release drug delivery systems and dosage forms . TSP possesses various features, making it an attractive candidate as a vehicle for ophthalmic medicaments. Mixtures of TSP and hyaluronic acid are employed as artificial tears for ophthalmic application in dry eye syndrome. TSP can be used in drug delivery systems for the ocular administration of hydrophilic and hydrophobic antibiotics. Tamarind seed polysaccharide is composed of pectin with high methoxyl content (6.8%-8.37%), that promotes gel strength and heat stability. It possesses properties of high viscosity, broad pH tolerance, noncarcinogenicity, mucoadhesive nature and biocompatibility. And is insoluble in organic solvents and dispersible in warm water to form a highly viscous gel as a mucilaginous solution . TSP possesses the characteristic property of forming gels with sugar concentrates in a wide pH range that are also not affected by boiling in a neutral aqueous solution, even if boiled for a long period, which makes them superior to fruit pectins. It has been described as a viscosity

enhancer showing mucomimetic and muco adhesive ability to form hydrogels. The individual components of the seeds have not been fully identified and quantitated . Therefore, this project aimed to extract TSP from tamarind seeds from three different sources and characterize its physical and chemical properties for the possible future development of drug delivery systems.

Natural polymers are obtained from the plant sources. They are high molecular weight and water-soluble polymers composed of monosaccharide units and united by glucosidal bonds .Some of the known polymers are pectin, guar gum, acacia, locust bean gum, tamarind gum, okra gum, etc. The uses of natural gums have increased a lot nowadays, due to their biodegradability in the body, their nontoxic nature and sometimes they provide a rate retarding effect on the release of drugs in a particular dosage form. Okra (Abelmoschus esculentus) is the only vegetable crop of significance in the Malvaceae family and is very popular in the Indo-Pak subcontinent. It is an oligo purpose crop, but it is usually consumed for its green tender fruits as a vegetable in a variety of ways. These fruits are rich in vitamins, calcium, potassium, and other mineral matters. The mature okra seed is a good source of oil and protein has been known to have superior nutritional quality. Okra seed oil is rich in unsaturated fatty acids such as linoleic acid which is essential for human nutrition. They are also known as ladies finger. It is used as a binder and produces tablet formulations with good and optimum physicochemical properties. It also retards the release of drug, increasing the half-life of a successfully used in controlled/sustained release tablet formulations and is also a hydrophilic polymer. It is also used as retardant, disintegrant, suspending agent, and matrix forming material. It is easily available and is quite economical. Being a natural polymer, it exhibits the property of biodegradation and mucoadhesion. Okra gum produces high viscosity mucilage at low concentrations. In continuation with the ongoing research on okrabased formulations, the major objective of the present investigation was to prepare to formulate, develop, and evaluate the compression. coated tablet using okra gum extracted from okra as binder along with synthetic hydrophilic polymers like various grades of hydroxypropyl methylcellulose (HPMC) and compare their various parameters. It is repotted that Mucilage from okra contains significant levels of protein, carbohydrate, neutral sugars, minerals and other complex polysaccharides and medically reported to be linked with anticancer, antimicrobial, hypoglycemic, anti-ulcer activities.

• TSP Extraction Procedure :-

• Tamarind Seed Preparation

Tamarind seeds taken from paddy farmland, with pulse tissue, were separated from their pulse by hand, then the seeds were washed with tap water and dried in an oven at 100 °C for 30 min. The seeds were allowed to cool down to room temperature and then lightly ground for 0.5-1 min in a blender to separate the brown peels from the kernel seeds. The kernel seeds were finally ground into powder with a blender.

• TSP Extraction Method 1.

Method A:

Cold distilled water (200 mL) was added to TSP powder (20 g) to prepare a slurry. The slurry obtained was poured into boiling distilled water (800 mL) and then boiled for 20 min on a hot plate to give a clear solution that was stored overnight. The thin clear solution was further centrifuged at 6000 rpm for 20 min to separate all the foreign matter. The supernatant was separated and poured into excess 95% ethanol with continuous stirring. The obtained precipitate was collected using a stainless sieve, and dried in an oven at a temperature 50 °C for 4 h. The dried polymer was stored in a desiccator. In the same way, tamarind seed powder, waste from the export tamarind juice industry and were extracted using the procedure mentioned above. Only tamarind seeds taken from paddy farmland, were extracted by Accelerated Solvent Extraction (ASE) using methanol as a solvent, following by ethanol, at a temperature of 100 °C for 30 min to give methanol extract (7.51%) and ethanol extract (3.31%).

Method B:

Tamarind seed powder (50 g) was defatted using hexane and 20 g of the defatted seed powder were taken as the starting material and subjected to the process described as Method A.

• Characterization of TSP

- The structure of the TSP was analyzed by NMR using an AVANCE 400 instrument in D₂O.
- TSP samples from the three sources were subjected to FT-IR spectroscopy as KBr pellets in a range of 4500–500 cm⁻¹ on a Fourier-transform infrared spectrophotometer. xtraction, purification and polysaccharides characterization of extraction Seeds of Tamarindus indica was collected and dried in sunlight. After that seeds were crushed into powder and boiled with water at 45 °C xtraction, purification and characterization of extraction Seeds of Tamarindus polysaccharides indica was collected and dried in sunlight. After that seeds were crushed into powder and boiled with

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METHOD

Isolation of gum from tamarind seedTake the tamarind seeds and peel out the outer cover and obtaining the white part of seeds and crush them. The crushed seeds of Tamarindus indica were soaked

in water for 24 h and then take the muslin cloth and the soaked seeds were put into it for the release of gum from it. The marc was removed from the gum and

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TSP Extraction Method:

Isolation of gum from tamarind seed.



Fig.1.1 Tamarind Seed

Isolation of gum from tamarind seed Take the tamarind seeds and peel out the outer cover and obtaining the white part of seeds and crush them. The crushed seeds of Tamarindus indica were soaked in water for 24 h and then take the muslin cloth and the soaked seeds were put into it for the release of gum from it. The marc was removed from the gum and equal quantity of absolute ethyl alcohol was added to precipitate the gum and the gum was separated by filtration. The marc was not discarded because it was sent for multiple extractions with decreasing quantity of extracting solvent i.e., water with the increase of number of extractions. The isolation was continued until the material was free of gum. The separated gum was dried in hot air oven at temperature 40°C. Then the dried gum was powdered and stored in airtight containers at room temperature. xtraction, purification and characterization of polysaccharides extraction Seeds of Tamarindus indica was collected and dried in sunlight. After That seeds were crushed into powder and boiled with water at 45 °C



Fig.2.0 Tamarind Seeds Gum.

Physicochemical characterization of gum.

 Identification tests for carbohydrates, proteins, mucilage and gums: Aqueous solution of extracted gum was used for chemical characterization. Test for carbohydrates, proteins, mucilage, alkaloids, fats, tannins, amino acids and gums were perform according to standard procedure.

- Organoleptic evaluation of isolated gum: The isolated gum was characterized for organoleptic properties such as color, odor, taste, fracture and texture.3
- Solubility behavior gum: One part of dry gum powder was shaken with different solvents and the solubility was determined.
- pH of gum: The gum was weighed and dissolved in water separately to get a 1% w/v solution. The pH of solution was determined using digital pH meter

TSP Extraction Method 3

Isolation of Protein and Polysaccharide:

The proteins of defatted TKP (10 gm) were extracted twice using 100 ml of 0.1 M NaOH for 5 mins at 40 C. The extract was then centrifuged at 3000 rpm for 15 mins. The residue (Tamarind Seed Polysaccharide – TSP) remaining after extraction was washed with alcohol and dried at 370 C.

TSP Extraction Method 4



Fig.4.1 Tamarind Kernel Powder

Protein fractionation:

- The proteins of commercial Sample of defatted tamarind kernel powder (TKP) were extracted according to their solubilities in different solvents.
- Defatted TKP flour (1.0 g) was extracted twice with 10.0 ml
- Distilled water for 30 min at room temperature.
- The extract Was then centrifuged at 3000 rpm for 30 min and the Supernatant was used for the determination of a water-soluble Protein (albumin).

- The residue was then extracted successively in a similar manner with 10 ml of 1.0 M NaCl, 70% ethanol and 0.2 % NaOH.
- The supernatant of each Extract was collected separately and used to estimate salt (globulin), alcohol (prolamin) or alkali (glutelin) soluble Fraction.
- The residue remaining after successive extractions .
- Represents the insoluble proteins and polysaccharide.

TSP Extraction Method 5

- Polysaccharides Extraction.
 - 1. Seeds of Tamarindusindica was collected and dried in sunlight.
 - 2. After that seeds were crushed into powder and boiled with water 50 ml at 45 °Cdto extract the polysaccharides.
 - 3. After boiling for 12 h the supernatant liquids were collected and stored in cool place.
 - 4. After the liquids become cooled acetone 10 ml was added and freeze at -4 $^{\circ}$ C.
 - 5. Freeze materials then lyophilized to extract out the Tamarind seed polysaccharides.
- EXTRACTION AND ISOLATION OF LADY'S FINGER (OKRA)OF MUCILAGE:-

Okra Extraction Method 1:-

- Okra (Abelmo schusses culentus) was obtained from local market of Pune, India. Collected okra was carefully washed and dried Under shade for 24 h, further dried at 30–40°C until constant weight was obtained.
- Size was reduced through grinder.
- Powdered Fruit passed through sieve no. #22 and stored it in air tight container for further use.



Fig.1.1 Okra Fruit Powder.

Okra Extraction Method 2:-

Mature okra stem

Cutting into pieces

Pounding and soaking In water (1:10, v/v), 6 h

Filter through double \rightarrow Plant Layer muslin cloth residue

Flocculation of mucilage \rightarrow Solvent with ethanol (50:50, v/v)

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Mucilage recovery-Chlorophyll, etc. And washing with Acetone (100%)

↓ Air drying. ↓

Mucilage powder

Okra Extraction Method 3:-

- Elsewhere, powdered fruit kept in 500ml of distilled water.
- Heated with stirred continuous at 60°C.
- For approximately4h.
- Concentrated solution has filtrated through muslincloth
- Cool at 4°C-6°C.

Okra Extraction Method 4:-

- Extracted gum has isolated in acetone.
- This allows filtration through muslin cloth.
- Washed with acetone and the mucilage filtrated through muslin cloth.

- Pressed mucilage was further dried to constant weight at 35–45°C in hot air oven.
- Hard mucilagecake was grinded and sieved through sieve # 22, stored indessicator for further used.



Fig.2.1 Okra Fruit Powder

Okra Extraction of mucilage Method 5:-

- Hibiscus esculentus fruit were used for isolation of mucilage.
- Firstly Fruit were washed with water to clean it from dirt if any and grinded into in a mixer.
- The material obtained was soaked in warm water for 4 h, boiled for 2 h and kept aside for 2 h for release of mucilage into water.
- After a period of 2 h material was squeezed in a muslin bag to remove the mark from the filtrate.

Isolation of mucilage:-

Equal volume of ethyl alcohol was added to filtrate to precipitate the mucilage, the mucilage was separated, dried in oven at about 45°C, powdered and passed through sieve # 80. The powdered mucilage was stored in desicator until further use.

Okra Extraction of mucilage Method 6:-

- Okra pods (without seeds) were sliced and immersed in water at room temperature.
- After 12h, with the aid of a muslin cloth, the solid was separated from the liquid fraction (filtrate).
- Three volumes of ethanol were added to the filtrate and the liquid was slowly stirred by handling until mucilage was precipitated.

- The mucilage was dried for 12h at 300 oC in an oven, pulverized to a fine powder with the aid of a grinder and passed through sieves (mesh 100 and mesh 325).
- The resultant fine powder was stored in an amber recipient until the moment of use.

Characterization method of okra gum physicochemical characterization of okra mucilage:

The aqueous extract was mixed with Molisch's reagent followed by addition of sulfuric acide.

Bulk density:

Accurately weighed amount of the dried gum (10 g) was taken and kept in a bulk density apparatus. The volume was noted. Hence, the bulk density was calculated using the formula: Bulk density=Weight of the powder/bulk volume.d. The violet color ring appeared at junction, showing the presence of carbohydrates.

Solubility behavior:

The mucilage was sundried that led to the formation of a powdery material, which was shaken with different solvents such as water, alcohol, and acetone, and solubility was further determined

II. SUMMARY & CONCLUSION

Okra (Abelmoschus esculentus L.) is Natural polymers which have been used in different pharmaceutical formulations. They are easily available, non-toxic, biodegradable and cost effective to be used as pharmaceutical excipients. In the present aspect of the study was to evaluate the efficacy of okra gum that has been used as a tablet binder. It is easily available and inexpensive. Okra gum as a binder produces tablet formulations with good physicochemical properties and good candidate for sustained release formulations. The tamarind seed, which is considered waste, can be converted into a useful agricultural byproduct through the extraction of the polysaccharide. Tamarind polysaccharides could be used to replace commercial pectin, reducing import bills and foreign currency expenditure. Research has been successfully carried out. The polysaccharides have been isolated and extracted. Characterization for various physiochemical properties and phytochemical properties was carried out. In the desired range, other properties such as pH, solubility, flow property, bulk density, etc. were found. In addition, FTIR, SEM, DSC was also performed for their physicochemical characterization. In

SEM, TSP powder shows two types of particles, smaller particles with rough rounded edges and larger particles with a smooth surface irregular shape while, DSC shows a sharp exothermic peak at 350°C, which shows its crystalline nature. The study also predicted that polysaccharide extracted could be used as a gelling agent in various pharmaceutical preparations.

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