Extraction, Modification and Evaluation of *Lepidium sativum* Seed Mucilage

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***Abstract*—The effectiveness of medications relies not just on the active ingredients and manufacturing procedures, but also on the efficacy of the excipients. The conventional notion of an excipient as a mere component excluded from the active substance has significantly evolved from being viewed as an 'inert' and inexpensive carrier to becoming a crucial part of the formulation. This article focuses on the development of new excipients, various types of already-existing excipients and some novel uses for already-existing excipients. Excipients works as adding bulk to the formulation to maintaining stability of the formulation. Natural excipients are easily found in nature and are widely used in formulations. Natural excipients are frequently employed due to their low cost, reduced toxicity and less adverse effects. In present times, polymer applications have been much highlighted in diverse field of science, technology and pharmaceutical industry. Its applications extend from adhesives, coatings, film to electronic devices, biomedical devices. Polymer modification provides modified material properties such as increased or decreased binding, disinfectant, gelling and solubility and other physical properties. Natural polymers are the polymers that present in nature and are readily available. Lepidium sativum is member of Brassicaceae family. Lepidium sativum seed mucilage extraction has many pharmaceutical applications. Extraction and modification of these seed extract has pharmaceutical excipient property. The low production cost, hydrophilic, biocompatible and biodegradable characteristic along with good rheological properties make it an excellent alternative to common synthetic plastic materials.**

***Index Terms*—Cellulose, Cellulose modification, Excipient, Garden Cress seed, *Lepidium sativum*, Polymer.**

I. INTRODUCTION

The effectiveness of the excipients affects the quality of medications in addition to the active ingredients and pharmaceutical procedures[1].It was assumed that the use of chemicals as excipients in dosage form formulation had no impact on the drug's effectiveness[2].

In order to ensure dose, stability and bioavailability, almost every drug dosage form contains some sort of excipient. The pharmaceutical products now on the market use about 1000 excipients from over 40 functional categories[3].

The term *excipients* is believed to have originated from the Latin word *excipere* (Dorland's Medical Dictionary, 1974). This word essentially means to exclude, collect, or accept. Excipients are essential to the manufacturing of stable dosage forms, their administration and the medication development process. Excipients are components of a medication that are therapeutically inactive. These excipients have significance in transforming a pharmacologically active substance into an efficient pharmaceutical product with improved therapeutic potential[4].

Any additive must have specific desirable properties in order to be used to a formula.

Properties of excipients:

- Unreactive
- Inert
- Inactive
- Available easily
- Stable against environmental factors
- Economic
- Compatible
- Pure

Advantages of Excipients:

- Improves stability
- Reduce cost
- Improve bioavailability
- Increases solubility
- Improves organoleptic properties
- Control drug release
- Increase patient compliance

Disadvantages of pharmaceutical excipients

- Variation in quality

- Regulatory challenges
- Impact on environment
- Interaction with APIs
- Toxic and allergic reactions

Scope of Excipients:

Pharmaceutical excipients play a crucial role in the development and manufacturing of pharmaceutical products. Here are some potential future scopes of pharmaceutical excipients:

1. Personalized Medicine

Excipients will play a vital role in personalized medicine by enabling the development of tailored formulations for specific patient populations.

2. Nanotechnology

Excipients will be used to develop nanocarriers for targeted drug delivery, improving the efficacy and reducing the side effects of pharmaceuticals.

3. 3D Printing

Excipients will be used in 3D printing to create complex dosage forms with customized release profiles.

4. Biodegradable Excipients

There will be an increasing demand for biodegradable excipients, such as plant-based polymers, to reduce the environmental impact of pharmaceutical products.

5. Co-Processed Excipients

Co-processed excipients, which combine multiple functions in a single excipient, will become more prevalent to simplify formulations and improve manufacturing efficiency.

6. Excipient Design for Pediatric and Geriatric Populations

Excipients will be designed to meet the specific needs of pediatric and geriatric populations, such as taste-masking agents for pediatric formulations.

7. Excipient Development for Biologics and Biosimilars

Excipients will be developed to stabilize and deliver biologics and biosimilars, which require specialized formulations to maintain their integrity.

8. Regulatory Frameworks and Harmonization

Regulatory frameworks will evolve to address the increasing complexity of excipient development, and international harmonization efforts will continue to facilitate the global supply of excipients[5].

Excipients might need to be evaluated experimentally for genotoxicity, general toxicity, carcinogenicity, or other specific toxicity endpoints; they should additionally be evaluated for their source, quantity, purity, degradation profiles, and possible interactions with APIs and other ingredients in the dosage form. The safety profiles of new excipients must be systematically assessed for potential risk in humans, depending on data available and precedent.

Natural Excipients

Natural excipients are compounds used in formulations that come from a variety of natural sources, such as plants, animals and minerals. Natural excipients are frequently employed due to their low cost, reduced toxicity and less adverse effects. Natural excipients have attracted a lot of attention lately because of their many medicinal uses[6].6

Since ancient times, natural substances have been utilized. The traditional medical system in India is called Ayurveda and it uses medicinal plant parts and extracts to treat a wide range of illnesses. They only employed a small spectrum of synthetic substances under specific guidelines because of their significant toxicity or adverse effects. They only employed a small spectrum of synthetic substances under specific guidelines because of their significant toxicity or adverse effects. Additionally, scientists favor using natural excipients whenever feasible or semi-synthetic chemicals to increase a compound's potency. The formulation of solid dosage forms makes considerable use of natural polysaccharides. Mono-saccharide polymers, such as sugars are cheap and come in a range of forms and characteristics. Changes to this can be utilized to create new delivery system designs.

Polymer was first used in the design and development of drug delivery systems as a result of the fusion of pharmaceutical sciences and polymer science. Hydrophilic matrices made of natural polymers, such as polysaccharides, continue to be widely used biomaterials for controlled-release dosage forms[7].

Cellulose:

It is the most prevalent biopolymer found in nature. Cellulose is the primary component of many natural fibers, including cotton and higher plants. Cellulose is composed of lengthy sequences of anhydro-D-glucopyranose units (AGU), with each molecule containing three hydroxyl groups per AGU, except at the terminal ends. It is characterized by its insolubility in water and most common solvents, a property largely due to the robust intramolecular and intermolecular

hydrogen bonding that occurs between the individual chains. Despite its limited solubility, cellulose is widely utilized in various applications, including paper production, packaging, composite materials, netting, upholstery and coatings. Cellulose is chemically modified to enhance its processing capabilities and create cellulosics, or cellulose derivatives, that can be customized for certain industrial uses. Cellulosics are utilized in many biomedical applications because they are generally robust, repeatable, recyclable and biocompatible. At the hydroxyl groups of cellulose, esterification and etherification are the most common changes. These substitution processes produce the majority of water-soluble and organic solvent-soluble cellulose derivatives and they typically result in significant alterations to the cellulose's original characteristics.

Oxidation:

A reaction involving cellulose and an oxidizing agent, such as gaseous chlorine, hydrogen peroxide, peracetic acid, chlorine dioxide, nitrogen dioxide (dinitrogen tetraoxide), persulfates, permanganate, dichromate in sulfuric acid, hypochlorous acid, hypohalites, or periodates, leads to the formation of oxidized celluloses, commonly referred to as oxycelluloses. These substances are characterized by their water insolubility. In addition to the presence of hydroxyl groups, these oxidized celluloses may also exhibit carboxylic, aldehyde, and/or ketone functional groups, which are influenced by the specific oxidant used and the conditions of the reaction.

Micro Crystallization:

The partially depolymerized cellulose known as purified microcrystalline cellulose is made by applying mineral acids to α-cellulose, which is derived from fibrous plant pulp. Usually, the degree of polymerization is below 400. The degree of polymerization reduces when cellulose reacts with acid because the acetal linkage is broken and the β (1–4) glycoside bond is attacked, causing the chain to hydrolyze. However, the hydroxyl groups react to generate carbonyl and carboxyl groups when oxidizing substances affect the cellulose chain. Thus, the average length of the cellulose chain is shortened by the oxidation reaction, and cellulose is transferred into MCC with carboxyl groups by the use of HNO3 or N2O4.

Etherification:

It is incorrect to think of the water-soluble cellulose ethers as only water-soluble cellulose. Although the ethers are cellulose derivatives, their molecular makeup only partially retains the original cellulose structure. In the esterification process, an alcohol and an acid combine to generate an ester and water, which is a common equilibrium reaction. Certain acids, including phosphoric, sulfuric, nitric and acetic acids are used to esterify cellulose. Schonbein's method, which involves reacting an aqueous cellulose slurry with nitric acid in the presence of sulfuric acid, is used to prepare all cellulose nitrates. Since the reaction occurs in equilibrium, the loss of water during the reaction drives it to finish, and the final DS is determined by the relative concentrations of the reacting species. Cellulose derivatives with lower flammability than cellulose nitrate were created as a result of the discovery that cellulose esters may be made with organic substituents. Cellulose acetate, the most significant organic ester, is created when cellulose and acetic anhydride combine with sulfuric acid. To obtain the triester (defined as having a DS larger than 2.75), the reaction is conducted for approximately 8 hours using acetic acid as the solvent. The triester is hydrolyzed by hydrochloric acid to produce the required substitution, which results in the derivatives with lower DS values[8].

Microcrystalline Cellulose:

Although MCC has been very beneficial in the creation of solid dosage forms, certain features have restricted its use, including sensitivity to lubricants, moderate flowability, loss of compatibility following wet granulation, and very low bulk density. MCC's silicification enhances its functioning by improving its density, low moisture content, flowability, lubricity, compatibility, compressibility and larger particle size. A suspension of MCC particles and colloidal silicon dioxide is codered to produce silicified MCC (SMCC), which has a final product that is 2% colloidal silicon dioxide after drying[9].

Lepidium sativum

L. sativum Linn. (Family Brassicaceae) is an annual, glabrous, erect, and fast-growing, herb commonly known as “common cress,” “garden cress,” “garden pepperweed,” “chandrashoor,” “raktbija”, “aseliyo” and many more. L. sativum is native of Egypt and

Southwest Asia that as a naturalised or more frequently casual plant is widespread in Asia, Afghanistan, Bhutan, China, India, Japan, Kazakhstan, Kyrgyzstan, Nepal, Pakistan, Russia, Tajikistan, Turkmenistan, Uzbekistan, Vietnam; Africa; Europe; North America; South American Seeds are ovate, oblong, 3 lobed, 3 mm long, 1 mm broad, brownish-red in colour as shown in Fig. 1

Fig. 1: *Lepidium sativum* seed (Garden Cress seed)

L. sativum has been traditionally used for many diseases such as cold infusion of seed is used in treatment of high cough, spleen and liver chronic enlargement, flatulence, diarrhea, dysentery, indigestion, rheumatic pain, inflammation, viscous humors, tenesmus, secondary syphilis, abortion, anemia and weakness. For sprains, dysentery, leprosy, skin diseases they are mostly used as poultices. The seeds are also recommended as depurative, tonic, aphrodisiac, hemogenic, galactagogue and diuretic. Moreover, seeds are useful in preventing the hair loss and stimulating the appetite. Seeds serve a source of mucilage that is used as pharmaceutical excipients. Seed cake of L. sativum is also used as a water purifying bed. Also seeds have the capacity to absorb heavy metals from the soil and so are used as soil purifier for their removal of from the soil.

Table 1: Taxonomic classification

Kingdom	Plantae
Subkingdom	Tracheobionta
Super division	Spermatophyta
Division	Magnoliophyta
Class	Magnoliopsida
Subclass	Dilleniidae
Order	Capparales
Family	Brassicaceae
Genus	Lepidium
Species	Lepidium sativum

Table 2: Physical characteristics of garden cress seed

Parameters	
Colour	Reddish-brown
Shape	Oval
Taste	Peppery, pungent taste
Length	2.60mm
Width	1.20mm
Thickness	0.94mm
Sphericity	54.59%
Bulk density	729.74 Kg/m^3
True Density	1230Kg/m^3
Porosity	40.67%
Angle of repose	20.59^0
Weight of 1000 seeds	1.86g

II. MATERIAL AND METHODS

Methods of extraction of cress seed mucilage:

Method under optimal conditions: At 35°C, pH of 10, and a water-to-seed ratio (wt./vol) of 30:1, the method performed optimally. After 30 minutes, the mixture was blended to separate the slurry from the cress seeds. The solution was passed through a muslin cloth to obtain slurry. Before being used, the mucilage was freeze-dried and stored in a dry, cool environment.

Extraction of mucilage by using various Drying Method

Initially, cress seeds were immersed in a 30:1 ratio of distilled water for a duration of 30 minutes at a temperature of 35 °C and a pH of 7. Following this soaking period, the seeds were extracted using an extractor model 700P from Rasht, and the surface of the gum layer was scratched with the extractor. Subsequently, the mucilage obtained from the cress seeds was filtered and collected. In the next phase, the collected mucilage underwent drying through various techniques. The drying methods employed for the cress seed mucilage included vacuum drying, freeze drying, and microwave drying. For the freeze-drying process, samples were initially frozen at -20 degrees Celsius before being transferred to a freeze dryer chamber set at 40 degrees Celsius. In the vacuum dryer, the samples were subjected to drying at temperatures of 40, 60, and 80 degrees Celsius. Additionally, the samples were microwaved for a duration of 50 minutes at a power setting of 360 watts.

Precipitation of soaked and blended seed in acetone

Wet and mixed seed acetone precipitation A total of 100g of seed was soaked in 800ml of distilled water for 12 hours. After that, pure the soaked seeds for 15 minutes at 2000 rpm with a Phillips HR 1453 hand

blender. The contents of this mixer were filtered using a muslin cloth. To achieve optimal yield, an additional 200ml of water was incorporated, mixed thoroughly, and subsequently filtered using muslin fabric. The resulting filtrate, which contains an equal volume of acetone, facilitates the precipitation of mucilage. The white mass of the supernatant should be separated using muslin cloth or through filtration. The mucilage was then precipitated and collected on a glass slab, where it was dried for 16 hours in a tray drier set at 60°C. The mucilage can be easily transformed into flakes by applying acetone onto a glass surface. These flakes were then subjected to further drying for an additional 5 minutes at 60°C.

Precipitation of soaked seed mucilage in alcohol

Seeds weighing 100 grams were immersed in 1000 mL of distilled water along with 5 mL of chloroform for a duration of 24 hours. The resulting viscous solution was then filtered through a muslin cloth. To precipitate the mucilage, 95 percent ethanol was added to the 1 liter of the mucilaginous mixture. The precipitated mucilage was subsequently collected and dried at temperatures between 40 °F and 45 °C until fully dehydrated. The yield was then determined.

Extraction of cress seed mucilage

A seed was acquired from a local market, thoroughly washed, and subsequently verified by a botanist. The mucilage surrounding the seeds was eliminated, as it expands upon contact with distilled water. Consequently, the seeds were submerged in water for several hours. Specifically, 300 grams of seeds were soaked in 300 milliliters of distilled water for a duration of 24 hours, resulting in the formation of a clear, watery capsule around each seed. Following this, the seeds were agitated at a speed of 1,000 revolutions per minute using an overhead stirrer to produce a viscous fluid from the capsule. This viscous fluid was then subjected to vacuum filtration with a pore size of 200 micrometers, allowing the seeds to separate from the thick liquid. Ethyl alcohol, methanol, and acetone were employed as precipitants for the mucilage extraction from the viscous fluid solvents, with these solvents being utilized at a concentration ten times greater than standard practice. The resulting precipitate was mucilage, which was subsequently collected and allowed to dry at room temperature.[10]

Fig 2: Extracted Cellulose

Seeds of *Lepidium sativum* belong to family Brassicaceae was collected from market of 100gm pack. Acetone and oxalic acid dihydrate were procured from local supplier Vijay chemicals.

Method of extraction:

Lepidium sativum seed was collected. 100 gm of seed was soaked in 800 ml of distilled water for 12 hours. After that, the soaked seeds were poured in beaker for blending for 10 min at 1500 rpm using Remi lab stirrer. Content of mixer was filtered through muslin cloth. To produce best result, 200 ml extra water was added, stirred, and filtered through muslin cloth. Equivalent amount of acetone was added for mucilage precipitation. Allowed to precipitate. The precipitated mucilage was separated by using muslin cloth. Mucilage was dried in oven at 65° C for 6 hours. Dried mucilage was reduced using mortar and pestle and passed through sieve no 30. Powder of seed mucilage was obtained.

Modification of Seed Mucilage/ Cellulose:

By Sulphuric acid Hydrolysis:

Microcrystalline Cellulose (MC) was produced by sulfuric acid hydrolysis of extracted cellulose. The hydrolysis was done by using sulfuric acid (H_2SO_4) solution [58% v/v]. Mix the cellulose and H_2SO_4 in ratio of 1:20 g/ml (MC/ dil H_2SO_4). Heat at temperature of 45°C for 30 min. Formed suspension was cooled into cold distilled water by ratio of suspension to cold distilled water of 1:20 to stop hydrolysis. Centrifugation was conducted at 4000rpm for 15min to abolish the acid solution. The obtained precipitate was collected by filtration and washed with water to neutralize the condition. Precipitate was dried in room temperature.

Preparation of cellulose oxalate:

Cellulose oxalate has been alternative to other method to prepare cellulose crystals. It is reaction between extracted cellulose and oxalic acid dihydrate to produce cellulose oxalate. The cellulose oxalate may

be low-cost method and may open up new applications areas for cellulose nanomaterials.

4 gm of cellulose and 15.40gm of oxalic acid dihydrate was taken in beaker. It was heated to 110° C on oil bath for 35 min. After time was reached, mixture lifted from oil bath and allowed to cool room temperature. After cooling and solidifying, acetone wad added to dissolve excess oxalic acid dihydrate. Solution was allowed to mix overnight. Added solvent was removed by vaccum filtration. Remaining solid was washed by acetone. Collected and dried at room temperature.

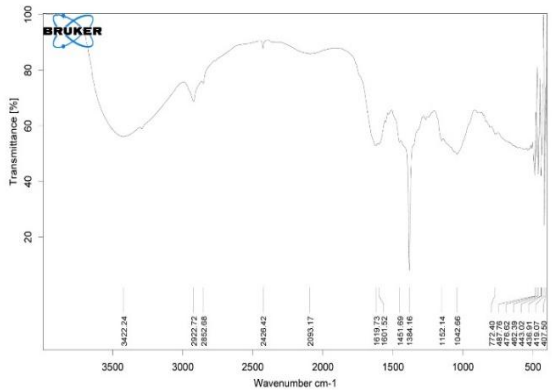
Fig No 3: Microcrystalline Cellulose

Fig No 4: Cellulose Oxalate

III. RESULT AND DISCUSSION

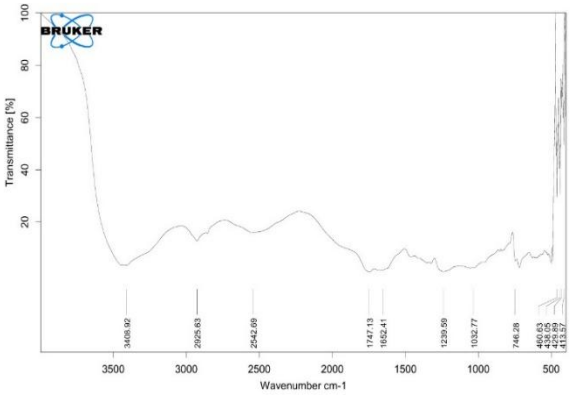
1. Fourier transform infrared (FT-IR) spectroscopy:

Crude Cellulose IR



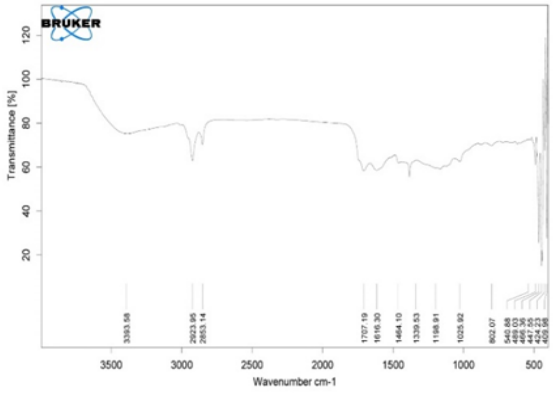
Sr No	Functional Grp	Peak Wavelength
1	Stretching & bending of OH	3422.24
2	CH Stretching	2922.72
3	COOH	2093.17
4	Stretching & bending vibration of OH	1601.52
5	Symmetric bending of CH2	1451.69
6	Bending vibration of C-H & C-O	1384.16
7	C-O-C pyranose ring Stretching Vibration	1042.66
8	C-H rock vibration of cellulose	722.40

Cellulose Hydrolysis IR



Sr No	Functional Grp	Peak Wavelength
1	Stretching & bending of OH	3399.66
2	CH Stretching	2925.18
3	COOH	1710.19
4	Stretching & bending vibration of OH	1622.00
5	Symmetric bending of CH2	1437.20
6	Bending vibration of C-H & C-O	1318.66
7	C-O-C pyranose ring Stretching Vibration	1009.97
8	C-H rock vibration of cellulose	873.52
9	C-O-S grp vibration due to sulphate ester during hydrolysis	850.46

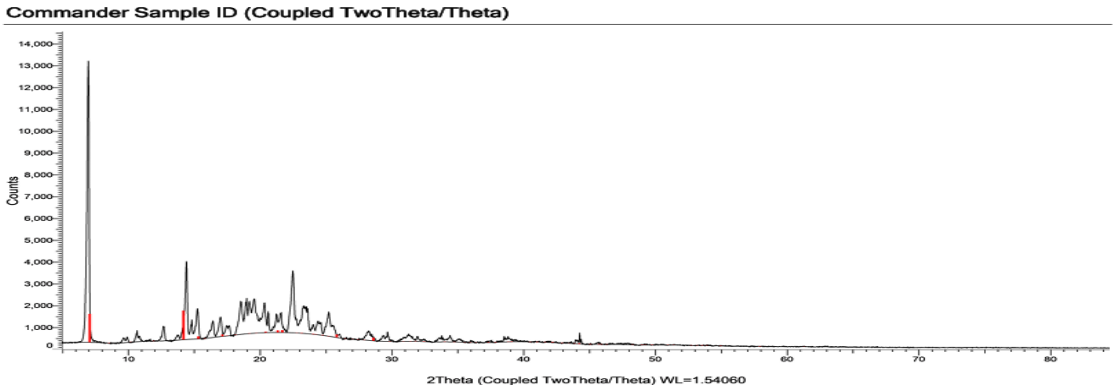
Cellulose Oxalate IR



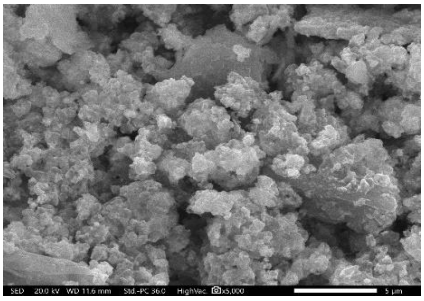
Sr No	Functional Grp	Peak Wavelength
1	Stretching & bending of OH	3408.92
2	CH Stretching	2925.63
3	C=O stretching of carbonyl grp	1747.13
4	Stretching & bending vibration of OH	1652.41
5	Symmetric bending of CH2	
6	Bending vibration of C-H & C-O	1239.59
7	C-O-C pyranose ring Stretching Vibration	1032.77
8	C-H rock vibration of cellulose	746.28

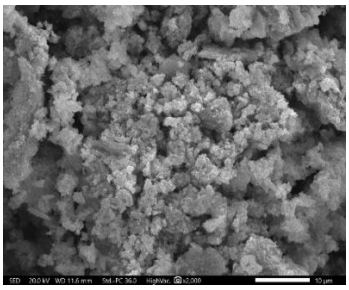
X-Ray Diffraction Spectroscopy:

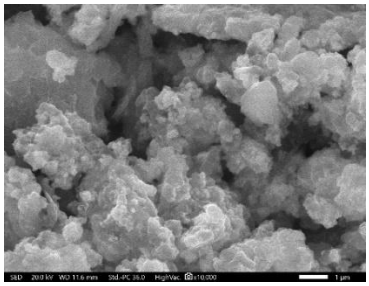
x ray diffraction pattern of hydroliswed cellouse at room temprature from 2-80 °C as show in fig the xrd pattern of hydrolised cellouse demonstrated stromg peak at 12.475,10.321,8.682,7.424,6.236,4.492 indicates crystalline nature of modified cellouse



2. Scanning Electron Microscopy (SEM):







The modification process has resulted in fragmentation of cellulose fibers, converting them into aggregated particles. The increased porosity and surface roughness suggest improved reactivity, making it suitable for adsorption, filtration, or biomedical applications. Modified cellulose is showing clusters into small particles due to chemical modifications, reducing individual fiber visibility. Higher magnifications show nano-scale voids and interconnected networks**,** **s**uggesting enhanced porosity and surface area**.**

IV. CONCLUSION

This study highlights the potential of Lepidium sativum (garden cress) seed mucilage as a valuable natural excipient in pharmaceutical formulations. The extraction and modification of cress seed mucilage demonstrate promising characteristics such as biocompatibility, biodegradability and favourable rheological properties, making it a suitable alternative to synthetic excipients. The process of cellulose extraction and its subsequent modifications, including hydrolysis and oxalate formation, further enhance its functional properties for various pharmaceutical applications. The analytical techniques, such as FT-IR, X-ray diffraction, and SEM, confirm the structural and morphological changes that improve the material's properties for potential use in drug delivery systems and other biomedical applications. The research emphasizes the growing significance of natural excipients in reducing toxicity and environmental impact while improving the overall quality and effectiveness of pharmaceutical products.

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