PECTIN-EGGSHELL BASED HYDROGEL FOR ANTHOCYANIN RELEASE

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CANDIDATE'S DECLARATION

I Ashu (2k24/MSCCHE/54) hereby certify that the work which is being presented in the dissertation entitled **"Pectin-Eggshell Based Hydrogel Incorporated by Anthocyanin"** in partial fulfilment of the requirements for the award of Degree of Master of Science, submitted in the Department of Applied Chemistry, Delhi Technological University is an authentic record of our own work carried out during the period from August 2025 to May 2026 under the supervision of Prof. Sudhir. G. Warkar.

We have not submitted the matter presented in the dissertation for the award of any other degree of this or any other Institute.

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Certified that Ashu (2k24/MSCCHE/54) has carried out their search work presented in this dissertation entitled **"Pectin-Eggshell Based Hydrogel for Anthocyanin Release"** for the award of Master of Science from Department of Applied Chemistry, Delhi Technological University, Delhi, under my supervision. The dissertation embodies results of original work, and studies are carried out by the students themselves, and the contents of the dissertation do not form the basis for the award of any other degree to the candidate or to anybody else from this or any other University/ Institution.

PLACE – DELHI

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ABSTRACT

In this study, we focus on addressing the need for sustainable, bio-based materials that connect the domains of green chemistry and agriculturally based research has increased due to the world's population rise. This work used a moderate acid-assisted extraction method with citric acid to successfully extract pectin from jackfruit peel (Artocarpus heterophyllus), a common agricultural waste. Through an in-situ crosslinking mechanism, calcium oxide (CaO), which is derived from calcined eggshell waste, was used as an environmentally friendly crosslinker to create a Pectin-Calcium (Pectin-Ca^{2+}) based hydrogel that formed a distinctive egg-box conformations stabilized by ionic and hydrogen bonding interactions.

The pigment (anthocyanin) was extracted from the petals of Alcea rosea (Gul Khaira) by using dilute ethanoic acid in distilled water. This gave a stable purple colored pigment. The anthocyanin was incorporated to the Pectin-Ca 2+ hydrogel matrix, in order to study its effect on hydrogel formation, its structure and release properties. The hydrogel formed was characterized using FESEM-EDS, DSC, Zeta Potential and Rheology Test to identify the formation of a well-crosslinked material, surface morphology and temperature stability respectively. The swelling investigation were gravimetrically carried out in distilled water. The effect of the amount of crosslinker and initiator on equilibrium swelling ratio were all studied extensively. The anthocyanin release from the hydrogel was investigated in aqueous media, and release kinetics were predicted to understand the diffusion process. The results reveal that the Pectin-Ca^{2+} hydrogel including anthocyanin displays better structural stability, pH-responsive activity, and a prolonged release profile, making it a suitable option for food packaging, biomedical, and agrochemical applications. This work develops a straightforward, economical, and ecological friendly method for turning agricultural waste into useful biopolymer materials.

Keywords: Pectin, Jackfruit Peel, Gul-Khaira Anthocyanin, Eggshell crosslinking, Controlled release, Alcea rosea, Biopolymer, Sustainable chemistry.

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LIST OF ABBREVIATIONS AND SYMBOLS

JPP	Jack fruit Peel Pectin
CH_3COOH	Ethanoic Acid
DSC	Differential Scanning Colorimetry
ATR-FTIR	Attenuated Total Reflection- Fourier Transform Infrared Spectroscopy
COO^-	Carboxylate ion
COOH	Carboxylic Acid Group
cm	Centimeter
Ca	Calcium
N_2	Dinitrogen
g	Gram
R^2	Regression coefficient
OH	Hydroxyl group
EA	Ethanoic acid
ml	Milliliter
mg	Milligram
FESEM	Field Emission Scanning Electron Microscopy
EDX/EDS	Energy dispersive X-ray spectroscopy
Capacity Λ_{max}	Maximum Wavelength

Ca^{2+}	Calcium ion
COO^-	Carboxyl group
SR	Swelling Ratio
C-H	Alkyl group
DSC	Differential Scanning Colorimetry
UV-vis	Ultraviolet-Visible spectroscopy
H_2O	Water
O-H	Hydroxyl group

Chapter 1: INTRODUCTION

1.1 Introduction

The Population is growing at an exponentially pace, and the world need for food supplies is growing rapidly. In response to this issue, food production has expanded by more than three times during the last fifty years [1]. The incineration of the waste generates a substantial amount of greenhouse gases, pollutants and particulate matter [1]. Because of the burning of the wastes greenhouse gas, pollutant & particulate matter have been produced in large amount [2]. In preservation of both human health and plant's ecosystem, adopting the sustainable practice in order to manage agriculture and food waste are vital. Because agriculture and food wastes possess enriched qualities, these beneficial materials must be removed and valorized [3]. Among them, Pectin is a one of the valuable biowaste and biopolymer which is comprised of α-1,4-linked D-galacturonic acid, which is partly methyl esterized, and the side chain comprises various neutral sugars, such as L-rhamnose, L-arabinose, and D-galactose [4]. The chemical significance across all domains like food, pharmaceutical, cosmetics etc. Can be explained by its natural, safe, easy and moldable nature [5, 6].

Jackfruit (Artocarpus heterophyllus) is a common and ubiquitous fruit in the Indian market, especially throughout the summer months. It is therefore generated in large quantities. Agricultural waste has therefore been produced. The peel represents about 59 per cent of the weight of a ripe fruit and has been discarding so far. The peel contains vital substances such dietary fiber, Polysaccharides, and polyphenolic chemicals [7, 8]. Using this process is possible to extract the pectin from jackfruit peel, which is one of the most heteropolysaccharides components (7.5-15% on dry weight basis) in the peel. If the process is optimized, the yield may

be higher, to about 28-38%, depending on the acids and optimization method used [16, 17]. As the pectin molecule was plentiful in carboxyl groups, so the carboxyl groups combined or coordinated with the Ca^{2+} when using the in-situ Ca^{2+} crosslinking method. Then the egg-box conformation was created. And this structure of Pectin-Ca^{2+} was stable due to ionic and hydrogen bonding [18]. The shell calcination to obtain this crosslinking. Too much money (and thus resources) in terms of the minerals: Protein, calcium carbonate (limestone), magnesium, etc. was wasted due to careless disposal. Calcium is recovered from the egg shell as CaO, $CaCO_3$, $Ca(OH)_2$, calcium phosphate, or hydroxyl apatite, in contrast to the other natural calcium sources. Egg shell calcium oxide (CaO) was over calcined, Efficient waste management preserve natural resources, lowers global warming, and promotes the development of environmentally friendly products [15].

The need for sustainable, edible, or biodegradable materials has grown as a result of the sharp shift towards agricultural and food waste-associated biopolymers due to concerns about the effects of synthetic plastic on the environment and human health. The anionic polysaccharide pectin follows the capacity of polysaccharides, a class of carbohydrates, to bond with water molecules to create an inexpensive, biodegradable film and offer improved capabilities once they were homogenized in water as gel. Building on this Ca^{2+}-crosslinked matrix, the design allows for control over polyphenol-matrix interactions and polysaccharide network fine-tuning while maintaining bioactivity. This allows the crosslinked network and polyphenols to cooperate and stabilizes sensitive anthocyanins. Plant anthocyanins facilitate the development of the stable and intelligent polymer [18,19]. This flower of Alcea rosea (holly hock flower, Gul Khaira) is an upright, relatively short-lived biennial or perennial herb, capable of reaching a height of 3m and it possesses anthocyanins. All 5 of the papery crinkled petals, ranging from a pink to a strong crimson, spread out from the central column of stamens [20]. Water soluble flavonoids give the

flower a purple or deep red color pigment in an acidic solution, it is made up of glycosylated polyphenolic chemicals. The pigmented anthocyanins are derived from the glycosylides of flavylium ions. There are six forms of flavylium ions with a sugar molecule attached. The six types are, cyanidin, dephinidin, malvidin, peonidin, pelargonidin and petunidin. Cyanidin and dephinidin among these ensure anti-inflammatory effects of plants [19, 23]. Because of its superior biological activity and environmental safety, anthocyanin is a common natural pigment. Its stability is jeopardized since it will deteriorate over time due to its susceptibility to light, heat, and oxygen, Divalent metals species such as calcium (Ca^{2+}) have been investigated for use as cross-linkers to improve the stability or strength of the resultant polymer matrix [21, 22].

The objective of this study is to extract pectin from agriculture or food waste through a simple environment friendly method and synthesize their hydrogel/bio film to understand structure property relationship. In this study, pectin was isolated from jackfruit peel by using mild acidic environment. The effect on the synthesis of hydrogel when anthocyanin is present, as well as release characteristics, will also be investigated. This method develops the neutral hydrogel [9]. The possible formation of hydrogel by using chemical crosslinker egg shell, study the function of anthocyanin incorporation to the hydrogel/bio film formation. Gul Khaira (Alcea rosea) produces the purple color pigment by the reaction of ethanoic acid into distilled water. Extracting purple color Anthocyanin in the presence of ethanoic acid, blocking blue-green and UV, caring cells from excessive light stress (Photoinhibition) [9]. Another is to explore ability of anthocyanin to coordinate with metal ion such as Ca^{2+} (divalent) and form stable complexes, if there is possibility then check its swelling and structural modifications [1313].

1.2 Literature Survey

1.2.1 Pectin: Sources, Extraction, and Properties

Pectin has been long researched as a natural polysaccharide as of the advantage such as a biocompatible, biodegradable, tunable gelling property. It has been used in food, drug, biomedical applications [5]. The amount and quality of the pectin obtained is highly dependent on the extraction conditions such as the concentration of the acid, temperature and pH of the solution [4, 6]. Under such circumstances, the readily available jack fruit (Artocarpus heterophyllus) peel waste is considered as the waste material that is mostly wasted waste [10]. Yield from the range of 7.5 to 38% has been obtained on extraction and purity of biopolymers obtained is enough for using for the direct formulation of hydrogel [7, 8].

1.2.2 Hydrogel Synthesis and Crosslinking Mechanisms

Hydrogels are three-dimensional crosslinked polymeric networks capable of absorbing large volumes of water without structural disintegration, making them suitable for controlled release, wound healing, and agricultural applications [24]. Ionic crosslinking with Ca^{2+} is particularly favored for polysaccharide-based hydrogels due to its simplicity and low toxicity. A Ca^{2+} cross links through the ‘egg-box mechanism’ between the carboxylate groups of adjacent pectin chains (Figure3a), a complex gel networks [25]. That Ca^{2+} -crosslinked pectin matrices effectively stabilize anthocyanin pigments, which is directly relevant to the present study [18]. Studies concerning carboxymethyl tamarind kernel gum hydrogels [26, 27, 28] set up a great milestone in biopolymer hydrogels on controlled release of nutrients for agricultural uses.

1.2.3 Anthocyanin: Extraction, stability, and Encapsulation

Anthocyanins are pigments derived from plants and are accountable for red, purple and blue shades of plants and fruits. These contain confirmed antioxidative and anti-inflammatory

outcomes [20]. Their practical application is constrained by susceptibility to heat, oxidation, and pH fluctuations. Coordination with divalent metal ions such as Ca^{2+} has been shown to significantly improve anthocyanin stability and color retention [13]. Encapsulation within pectin-based matrices has proven equally effective, as demonstrated by who successfully incorporated anthocyanins into bioactive films for food freshness monitoring [19, 21]. These results further support our current hypothesis and approach regarding the pectin-Ca^{2+} complex formation method for stabilization and controlled release of anthocyanin.

1.2.4 Calcium Oxide from Eggshell Waste

Eggshells are mostly Calcium carbonate (~94% of composition and were considered for this purpose a high-volume product for Food Industry Waste having a considerable amount of calcium is available to recover. Calcination of eggshell at high temperatures results in the formation of highly reactive CaO that can function as an ionic crosslinking agent within the polymer [15]. By utilizing eggshell-derived CaO as a Ca^{2+} source in this study for in-situ pectin crosslinking, this work accomplishes both the independent use of non-synthetic agents such as $CaCl_2$, and the two stream wastes from agriculture-jackfruit peel and eggshell-are valorise in this green framework in one time.

1.2.5 Swelling Behavior and Release Kinetics of Hydrogels

Understanding how a hydrogel swells is key to knowing how it behaves, especially regarding how it releases the molecules trapped inside. We looked into this by establishing a clear way to measure the swelling properties of superabsorbent hydrogels. Our work showed that for these polysaccharide systems, the release of molecules is primarily driven by the swelling process itself [29, 30]. To analyze these release patterns, the Korsmeyer-Peppas equation is frequently used to distinguish between simple diffusion and other types of release. As Siepmann and Peppas have noted, choosing the right model depends on the shape of the material and how much it

swells [31]. In the present work, the Korsmeyer-Peppas model will be applied to elucidate the anthocyanin release mechanism from the Pectin-Ca^{2+} hydrogel Matrix.

Chapter 2

MATERIAL AND SYNTHESIS

2.1 Materials

Jackfruit Peel (JP) and Eggshell (ES) were collected from the vegetable market and kitchen waste, respectively. Gul Khaira flower petals (Alcea rosea) were collected from the campus of Delhi Technological University, Delhi, India. Citric acid (10%), sodium hydroxide (NaOH, 98%), and hydrochloric acid (HCl, 36%) were purchased from Thermo Fisher Scientific (Delhi, India). Ethanoic acid (glacial acetic acid) was used as the extraction solvent for anthocyanin. All chemicals were used as received without further purification. All solutions were prepared using deionized (DI) water.

2.2 Extraction of Pectin from Jackfruit Peel

The jackfruit peel was collected fresh, washed thoroughly with deionized water to remove surface impurities, and chopped into small pieces. The peel was first blanched in hot distilled water at 80°C for 15 minutes to inactivate pectinase enzymes and then dried in a hot air oven at 60°C for 24 hours. The dried peel was ground into a fine powder using a mechanical grinder and passed through a 60-mesh sieve to obtain fine and uniform particle size.

For pectin extraction, five grams of purified bio-waste were treated with 6% citric acid for five to six hours at 60°C. After cooling to room temperature and beginning the separation process using Whatman filter paper, the collected supernatant liquid was heated to 60°C for to 15 to 20 minutes to make it more concentrated, then cooled to room temperature, after that 90% ethanol were added. The mixture were then kept into dark for 24 hr. the precipitated pectin were filtered, washed with ethanol to remove impurities and dried at 50°C for 24 hr. The dried pectin was

grounded by motar and pestle and stored in an airtight polythene at room temperature for further use. According to estimates, the amount of pectin extracted from jackfruit peel was roughly 13%, 12% and 10% of their dry weights, respectively. The fig.1. shows extraction process.

sFigure 1*:* Extraction mechanism of Jackfruit Peel Pectin by environmentally friendly method

2.3 Preparation of Calcium Oxide (CaO) from Eggshell

Eggshells were collected from kitchen waste, washed with deionized water to remove egg white residues and other biological matter, and air dried for about 24 hours. The cleaned and dried eggshells were crushed into small fragments and then calcined in a muffle furnace at 600°C for 2 hours which convert calcium carbonate ($CaCO_3$) of eggshell to more reactive form i.e. calcium oxide (CaO) according to the thermal decomposition reaction:

$$CaCO_3 \rightarrow CaO + CO_2$$

$$CaO + H_2O \rightarrow Ca(OH)_2 \quad \text{and} \quad Ca(OH)_2 \rightarrow Ca^{2+} + 2OH^-$$

The resulting white color eggshell powder (CaO) were obtained and cooled in a desiccator and stored in an airtight container to prevent from moisture. The calcined eggshell was used as the crosslinking agent for the hydrogel synthesis.

2.4 Extraction of Anthocyanin from Alcea rosea (Gul Khaira)

Flower of Alcea rosea (Gul Khaira) were collected from the DTU campus then their petal were washed gently with distilled water. From this 5 g of fresh petals were macerated in 100 mL of ethanoic acid solution (1% v/v in distilled water) for 3 hours at 50°C-60°C in the beaker. The resulting petals imparted their deep purple-colored pigment into the solution were this extract were filtered through Whatman No. 1 filter paper to remove plant debris. The filtrate, containing the anthocyanin pigment in the ethanoic acid medium, was collected and stored at room temperature in dark place to prevent from photoreduction. The ethanoic acid medium maintains an acidic pH that stabilizes the flavylium cation form of anthocyanin, which exhibits the characteristic purple-to-wine red coloration. The maximum absorbance (λ_{max}) of the anthocyanin extract was measured by UV-Vis spectroscopy to confirm the pigment identity and estimate concentration. Can be understand by the below equation:

Figure 2: Extraction of anthocyanin

2.5 Fabrication of Pectin-Eggshell (ES) Hydrogel

The Pectin-ES hydrogel was prepared by dissolving 40 mg of the extracted jackfruit peel pectin (JPP) in 10 mL of deionized water under continuous stirring at 60°C until a clear, viscous solution was obtained. To this pectin solution, 2 mL of the eggshell solution (40% w/v, prepared were added dropwise with constant stirring. The immediate formation of viscous hydrogel due to

the eggshell (CaO) cross linking with the free carboxylate ($–COO^-$) groups in the pectin chain to form ionic crosslinks (Pectin-Ca^{2+}), producing a characteristic egg-box conformation. The final hydrogel obtained was placed in a small beaker to obtain gel discs which were then air dried for 24 hours at room temperature. Then it was dried for 48 hours at 50°C and the dried hydrogel disks collected and stored in sealed polythene until further characterization and swelling experiment was carried out.

2.6 Fabrication of Anthocyanin-Incorporated Pectin-ES Hydrogel

The procedure for the preparation of anthocyanin-loaded Pectin-Eggshell (ES) hydrogel: A solution of jackfruit peel pectin (40mg) was prepared by dissolving the pectin in 10mL deionized water at room temperature. Then add in 2 ml (2g) the fresh and dried Alcea rosea anthocyanin extracted powder into make a consistent pectin-anthocyanin solution. After adding the anthocyanin, the solution's colour is a dark purple solution. After that, the Eggshell (ES) solution (40%w/v) (2mL) was added dropwise into the mixture solution and continued stirred for in-situ ionic crosslinking.

In addition to the Ca^{2+} ions (from Eggshell, CaO) which cross-linked neighbouring pectin chains, also served to coordinate with the OH and carbonyl group of the anthocyanin to stabilize them into the hydrogel matrix.

Chapter 3

EXPERIMENTAL ANALYSIS

3.1 Swelling Studies

The swelling behavior of all synthesized Pectin hydrogels was examined based on gravimetric analysis. The accurately weighed dried hydrogel (W_D) were immersed in distilled water. At certain time intervals, the swollen hydrogels were removed from the distilled water, and the extra water on the hydrogel disc was blotted off with the help of filter paper and weighed again (W_S) [22]. The equilibrium swelling ratio was determined for all synthesized hydrogels at equilibrium (maximum swelling) by using the given formula [23].

$$\text{Equilibrium Swelling Ratio} = \frac{W_{SG} - W_{DG}}{W_{DG}} \quad \text{.....(1)}$$

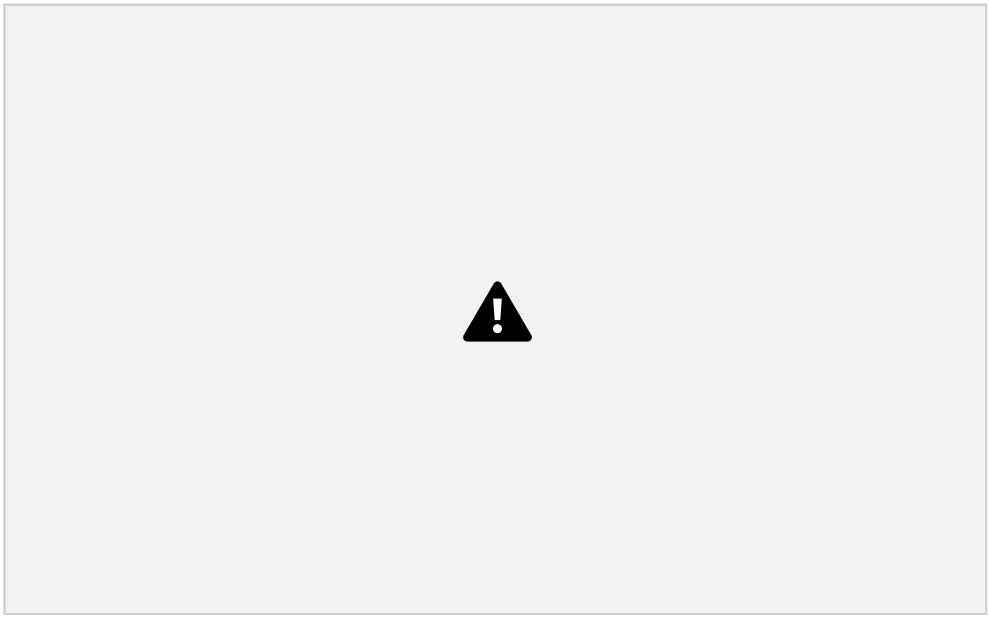
Similarly, the swelling analysis for the Ca^{2+} incorporated Pectin-Anthocyanin hydrogel was carried out in distilled water [5].

3.2 Release analysis of anthocyanin in water

The in-vitro release of anthocyanin from the Pectin-ES-Anthocyanin hydrogel was studied in distilled water at room temperature (25 ± 1°C). A pre-weighed, dried anthocyanin-loaded hydrogel disc was immersed in 10 mL of distilled water in a small beaker. At specified time intervals (0.5, 1, 2, 4, 6, 8, 12, 24, 48, and 72 hours), an aliquot of 3 mL was withdrawn from the release medium and to ensure that volume remained steady throughout the experiment, an equal volume of fresh distilled water was poured into the beaker.

The anthocyanin concentration in the withdrawn aliquots was determined by UV-Vis spectrophotometry at the characteristic maximum wavelength (λmax) of the anthocyanin extract

(approximately 200–500 nm) for cyanidin-based anthocyanins in acidic conditions). A calibration curve was constructed using known concentrations of the anthocyanin extract to quantify the cumulative anthocyanin released over time. The cumulative percentage release was calculated and plotted as a function of time. Experiments were performed in triplicate and mean values with standard errors are reported.



3.4 Characterization

The FTIR Spectra of Jackfruit Peel Pectin (JPP), Eggshell (ES), Gulphaira anthocyanin (GA), JPP-ES hydrogel and Anthocyanin-loaded JPP-ES hydrogel were obtained using the ATR-FTIR

Nicolet iS50 FTIR Tri detector spectrometer within a wavenumber range of 4000-450 cm^{-1}. The DSC (Differential Scanning Colorimetry) was taken for Jackfruit Peel Pectin (JPP), Eggshell (ES), Gulphaira anthocyanin (GA), JPP-ES hydrogel and Anthocyanin-loaded JPP-ES hydrogel using Hitachi NEXTA DSC 200/600 within a temperature range of 30-300°C at a uniform heating rate of 10°C/min. FESEM EDS was performed for Jackfruit Peel Pectin (JPP), Eggshell (ES), Gulphaira anthocyanin (GA), JPP-ES hydrogel and Anthocyanin-loaded JPP-ES hydrogel using FESEM: Gemini SEM 500 mode with Energy-Dispersive X-ray Spectroscopy (EDS)detector for elemental analysis and mapping. UV-Vis spectroscopy was carried out for anthocyanin released data.

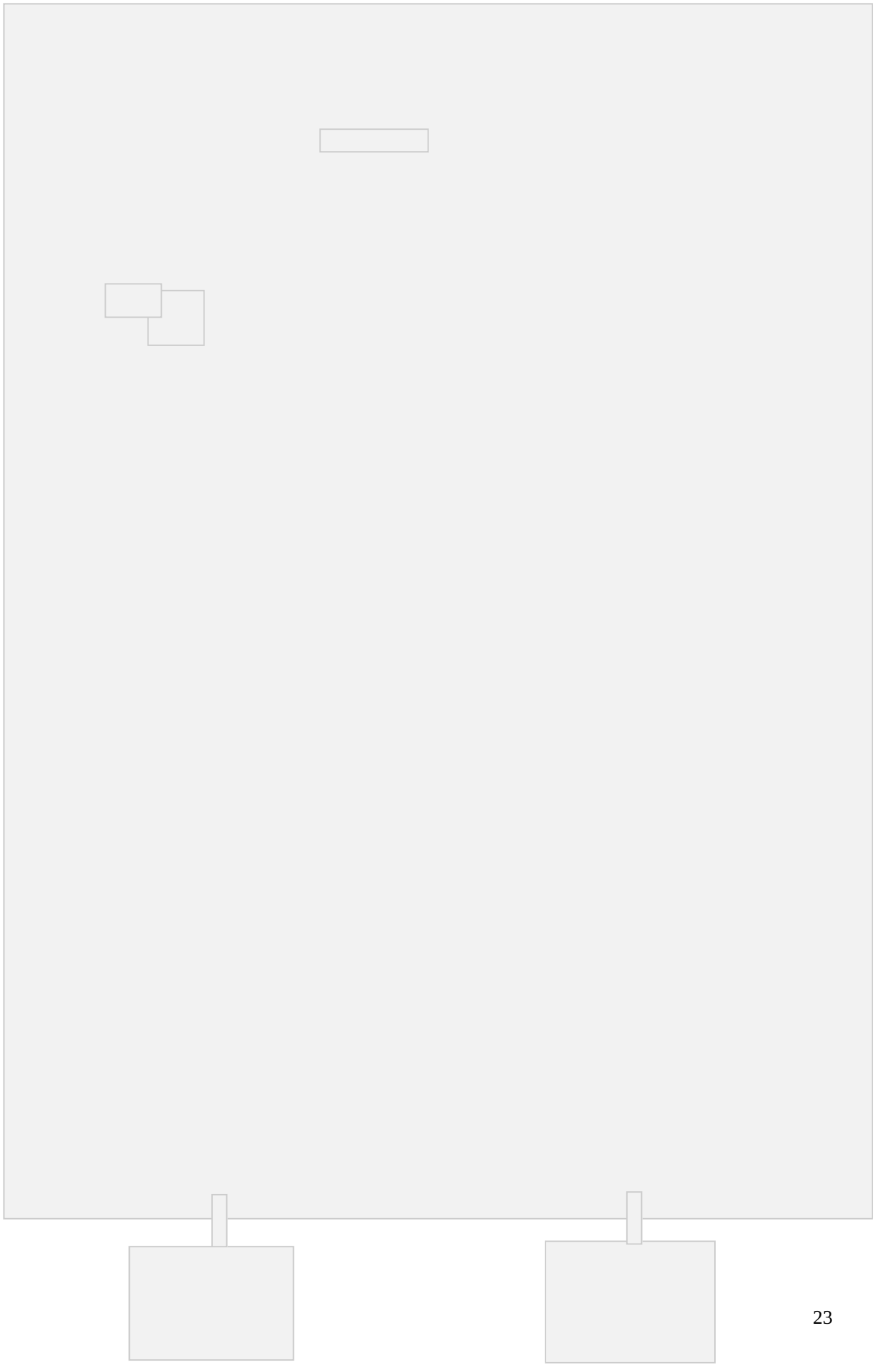
Chapter 4

RESULT AND DISCUSSION

4.1 Mechanism of synthesis of Jackfruit Peel Pectin and eggshell hydrogel

Prior to this section we understand about the structural and chemical properties of pectin, now in this section we will study about the presence of esters and other molecules in the pectin matrix which tell us its gelation and structural properties. Through its degree of esterification, we will understand the mechanism of pectin gelation. Through Ionic gelation mechanism the multivalent species, such as divalent Ca^{2+} from the activated CaO major component of Eggshell (ES) takes part in crosslinking with JPP and the stabilization of the Pectin-crosslinked ES hydrogel through this Ionotropic mechanism. Gul-Khaira Anthocyanin (GA) plays critical role in stabilization of gel formation between Pectin of jackfruit peel and ions of eggshell providing the additional influence of gel formation. Additionally, anthocyanin form hydrogen bonding with water molecule resulting the restricted motion of free water thus influencing the concentration of polymeric environment

by leading gelation.



The gel formation is majorly influenced by three factors:

1. Degree of acetylation (DA)
2. Degree of methoxylation (DM)
3. Degree of esterification (DE)

The constituents of Eggshell help in creation of “egg-box” structure by the presence of free carboxylate ions(-COO^-) and sulfate ion in the heteropolymer, significantly strengthen the polymeric chain formation or intercalation. The acetyl group typically attach to galacturanic acid by the -OH group in pectin chain. The degree of acetylation alters the gel formation, high DA make it difficult to form gel while at low DA it influencing the gelling it also influencing gel formation at low pH with the high degree of acetylation as carboxylate ions more actively participated in crosslinking to form hydrogel, with this neutral sugar will also take part and help in the formation of Pectin hydrogel [25].

4.2. Swelling studies

The swelling characteristics of the hydrogels fabricated were tested in distilled water for its water absorption capacity and the induced mechanical stress on the polymer network [18].

Figure 4.2 The time-dependent percentage swelling ratio shows the comparison between the eggshell crosslinker and addition of Gulphaira anthocyanin pigment.

Fig. 4.2. Swelling ratio (%) of synthesized hydrogel by (a)JPP with ES and (b) by the incorporation of Gulphaira anthocyanin with Jackfruit Peel Pectin (JPP) crosslinked by Eggshell (ES).

From the swelling curves, the initial rate of water absorption for both pure (a) JPP + ES hydrogel

and anthocyanin-contained (b) JPP + GA + ES composite showed the fast rate of water absorption. This initial phase is characteristic of hydrophilic polymeric networks, driven by the rapid hydration of the matrix via hydrogen bonding. The swelling ratio of JPP-crosslinked ES hydrogel and anthocyanin composite hydrogel showed the highest to 839.12 ± 156% and 1360 ± 52% after immersing 5 hrs and 6 hrs into distilled water respectively. Beyond this peak, the swelling ratio stabilized and exhibited a slight decline, marking the onset of initial polymeric chain degradation and partial network erosion.

Mechanistically, these swelling and stabilization processes are strongly dependent on the ionic interactions in the matrix. Activated CaO, in which calcined eggshell is a valuable resource, readily gives up its divalent Ca2+ ions to the aqueous system. Such ions facilitate excessive crosslinking of the polymer network because they can coordinate with plenty functional group of carboxylate (-COO-) and hydroxyl (-OH) existed on pectin chain. This targeted chelation are the ones which build up the characteristic "egg-box" junction zones, which form the basic structural unit of the hydrogel network [18, 25].

Also, the use of anthocyanin affects significantly the swelling kinetics because it has extra phenolic hydroxyl binding sites which can bind with the free Ca2+ ions available from eggshell. The introduction of this additional chelation resulted in a very stable and highly linked semi-interpenetrating network structure. Consequently, the anthocyanin not only modulates the water uptake capacity—allowing the matrix to accommodate a significantly larger volume of solvent before reaching osmotic equilibrium—but also enhances the overall mechanical integrity of the hydrogel matrix, delaying premature structural degradation [19].

4.3 FTIR

The Fig. 4.3 (a) to (e) illustrates the FTIR spectra of Jackfruit Peel Pectin (JPP), Eggshell (ES), Gulphaira anthocyanin (GA), JPP-ES hydrogel and Anthocyanin-loaded JPP-ES hydrogel respectively. In the FTIR spectrum of JPP (in Fig. 4.3.a), the peaks observed at 3224 cm^{-1}, 2920 cm^{-1}, 1730 cm^{-1},1614 cm^{-1}, 1100 cm^{-1} and 1014 cm^{-1} correspond to O-H stretching vibration, C-H

stretching vibration, C=O stretching vibration, and peaks COO^- stretching vibration and other two corresponding peaks shows presence of polysaccharides groups such C-O-H and C-O-C respectively [32].

FTIR spectrum of eggshell (ES) has been shown in the figure 4.3(b). The O-H stretching vibration was associated with a peak in FTIR spectrum of ES at 3735 cm^{-1} which represents water content or surface hydroxyl groups. Additionally, the absorbance peaks at 2982 cm^{-1} and 1200 cm^{-1} can be attributed to the C-H stretching vibration and the C-O stretching vibration of organic components of eggshell like remaining proteins and lipids [15] respectively. Also, the sharp and broad peak found at 1400 cm^{-1} is the result of the asymmetric C-O stretching vibration of carbonate ion, Although the 2545 cm^{-1} or so band is an overtone to the main carbonate vibrations. In addition to, sharp absorptions peaks at 872 cm^{-1}, and 713 cm^{-1}, the characteristic frequencies of the out-of-plane bending mode and in-plane bending mode for carbonate ion, which confirms more particularly the dominance of a mineral nature in the composition of the eggshell with the calcium carbonate calcite allotrope [16].

FTIR spectrum of the hydrogel consisting of jackfruit peel pectin and eggshell (JPP+ES) can be observed in the fig.4.3(c). In JPP+ES spectrum, the broad peak centered at 3328 cm^{-1} is the O-H stretching vibration involved with intermolecular hydrogen bonding, the peak at 2980 cm^{-1} is attributed to the C-H stretching vibration of galacturonic acid ring. Additional peak at 1740 cm^{-1} indicates the C=O stretching vibration of the esterified carboxyl groups which is present in pectin, with 1570 cm^{-1} assigned to the asymmetric stretching of the carboxylate (COO^-)ion [18]. The big absorption peak at 1400cm also represents the symmetric stretching of COO^- group. Simultaneously overlapping with the strong carbonate ion (CO) stretching band originating from the eggshell moiety [16]. The intense peak around 1068 cm^{-1} is the stretching vibration of C-O-C of pyranose ring from the main backbone of pectin polysaccharide [18].

Fig. 4.3(d) below represents the FTIR spectrum of the anthocyanin extract. Among the bands in the FTIR spectrum, the absorption peak in 3300 cm^{-1} is a strong and broad absorption peak

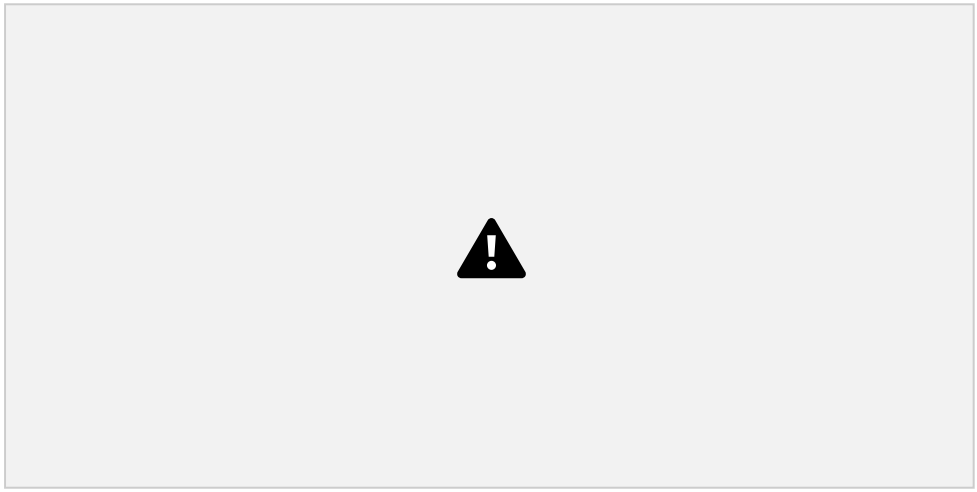
which represents the O-H stretching vibration, shows presence of phenolic hydroxyl groups and inter-molecular H-bonding. In addition, the bands at 2982 cm^{-1} and 2888 cm^{-1} can be assigned as C-H stretching of aliphatic and aromatic group. Next, peak at 2127 cm^{-1} is CC stretching. The intensity of the peak is quite strong at 1617 cm^{-1}, peak corresponds to C=C stretching vibration, the aromatic systems which constitute the anthocyanin structure are very characteristic.

Another unique peak that occurs at 1270 cm^{-1} may be attributed to the stretching vibration of the C-O bond in phenols or flavonoid ethers. Last the following bands around 1385 cm^{-1}, 1035 cm^{-1} and 840 cm^{-1} are due to different C-H bending, stretching and out-of-plane vibration of skeleton of anthocyanins molecule [14].

FTIR spectrum of JPP+Antho+ES composite hydrogel loaded with anthocyanins is shown in the fig.4.3(e). O-H stretching vibration of pectin that existed in spectrum of composite hydrogel produced broad peak at 3280 cm^{-1} was attributed to the presence of a strong intermolecular hydrogen bonding between pectin, anthocyanins and the matrix of eggshell. Peak at around 2982 cm^{-1} arose from C-H stretching vibrations.

Secondly, the individual absorption at 1743 cm^{-1} corresponds to the C=O stretching of the esterified carboxyl groups belonging to the backbone of pectin [18]. The peak at 1565 cm^{-1} arises from overlapping asymmetric stretching of the carboxylate (COO -) ion of pectin and C=C aromatic ring stretching of added anthocyanins [23]. Finally, the wideband in 1393 cm^{-1} is interpreted as the symmetric COO^- stretching, overlapped with strong stretching of carbonate ion (CO) from eggshell [16]. The cm^{-1} vibration band is a stretching vibration band for the C-O linkage characteristic of phenolic compounds in anthocyanin compounds [14] and the steep vibration band at 870 cm^{-1} indicates the bending mode of out of the plane for the carbonate ion of calcite [16]. Thus, the all three species were incorporated into the hydrogel matrix.

4.4 Anthocyanin Release Study



The Gul Khaira anthocyanin's release kinetics and structural stability from the pectin-ES hydrogel in deionized water was analysed with the time-resolved UV-Visible spectroscopy. A gradual increase in the value of the maximum intensity of the typical visible absorption band (max = 272 and 320 nm) during the measurement process has shown the diffusion of the pigment from polymer matrix during all the time, and no hypochromic or bathochromic effects prove that the structural integrity of the flavylium cation was preserved during transferring into aqueous solution [35, 36]. A steady progression of the increase in the typical visible absorption band during the study indicated that diffusion of pigment from the polymer remained throughout the experimental time; furthermore, no hypochromic or bathochromic shifts observed shows no change in the integrity of the flavylium cation in passing into aqueous solution [35, 36]. The primary observed UV range, at 272 nm, was attributed to the aromatic ring system of the aglycone moiety the basic flavanoid structural element. Meanwhile, unique shoulder at 320 nm corresponds to aromatic acylation in glycosidic unit that reflects esterification by such as hydroxyl cinnamic acid derivatives [35, 37]. This stability for the specific UV-Vis spectral profiles of the release suggests that these chemical features remained protected in the pectin-ES matrix and that hydrolysis of acylated anthocyanin complexes was largely prevented during release.

4.5 Thermal Analysis

DSC (Differential Scanning Calorimetry):

Fig. 4.5 (i) to (v) shows the DSC thermograms for the thermal stability studies of JPP, ES, JPP+ES, GA, and JPP+GA+ES composite hydrogel. DSC thermogram of raw JPP presented a pronounced endothermic peak at about 205°C that attributed to the thermal degradation and depolymerization of the backbone of galacturonic acid of pectin [17]. The thermogram of ES shows a very stable thermal profile up to 300 °C without significant degradation peaks, which is typical for the strong inorganic calcium carbonate matrix [15]. For the extracted GA, a strong endothermic peak is found at around 145–150, corresponds to the melting point and structural disintegration of the anthocyanin molecules [13]. In JPP+ES, the pectin degradation sharp peak vanishes and the broad endothermic band appear around 115 °C after ionic crosslinking, confirming that the thermal stability is improved due to the successful coordination between the free carboxylate groups of pectin and the calcium ions from the eggshell. Finally, the different thermal degradation peaks of the separate components are entirely veiled in the JPP+GA+ES composite hydrogel. Instead, a broad endothermic peak is detected at around 120 °C which corresponds to the evaporation of highly hydrogen-bonded and physically entrapped water molecules. This significant shift indicates that anthocyanin loaded hydrogel network has been well entrapped and formed good crosslinking structures with sound thermal stability [19, 22].

4.6 Zeta potential and Particle size

The properties of the particle size and surface charge of jackfruit peel pectin (JPP) were analyzed by DLS and zeta potential. The intensity size distribution indicates that a narrow monodisperse colloidal system is formed in water as is shown by the very low PDI of 0.065. The zeta potential is also shown to be unimodal with an average value of 12.5 mV. The overall Z-average size is 1645 nm. In addition, the zeta potential analysis shows a unimodal distribution with an average value of −12.5 mV. This negative surface charge is largely

believed to result from the deprotonation of the free carboxylate groups of the galacturonic acid (-COO) in the extracted pectin polymer chains [18]. Zeta potential of -12.5 mV shows



4.7 Rheological Measurements

Viscoelastic properties of the developed hydrogel were studied by oscillatory rheological tests. Amplitude sweep measurement as a function of shear strain (Fig. 4.7a) shows that both the storage modulus (G′) and loss modulus (G″) have been maintained at plateau stability during LVR for small strain values (around 0.01-3%) with G′ (~200 Pa) having higher value compared to G″ (~50 Pa). This predominance of the elastic property over the viscous one is indicative of a good cross-linking process of the jelly-like network, indicating the formation of a stable three-dimensional hydrogel matrix through ionotropic Ca^{2+}-egg box cross-links in the pectin gel matrix [25, 26]. In addition to the critical strain (~5–10%), there was an evident

crossover of G' and G" that reflects network breakdown and gel-to-sol transformation due to large deformation, which is analogous to the strain-dependent yield behaviour of physically cross-linked pectin gels [12, 25]. Amplitude sweep data versus shear stress (4.7b) again shows results compatible with the other techniques since an elastic Plateau (at the very minimum shear stresses) declines, before suddenly diving under a certain shear stress limit (typically 2 - 5 Pa).

The shear stress at which G′ = G″ corresponds to the yield stress of the hydrogel, beyond which irreversible structural breakdown initiates [34]. This well-defined yield point is consistent with the cohesive network architecture arising from Ca^{2+} coordination across galacturonate residues of pectin chains, forming the characteristic egg-box supramolecular assembly [18, 25].

The frequency sweep (Fig.4.7c) demonstrated a monotonic increase in both G′ and G″ over the angular frequency range of 0.1–100 rad/s, with G′ maintaining predominance over G″ across the entire frequency window. The absence of any crossover or terminal flow behaviour within this frequency range confirms the predominantly elastic, solid-like character of the hydrogel network and suggests a stable, entangled crosslink density without significant chain relaxation at the timescales probed [12, 26]. Such frequency-dependent stiffening is a hallmark of Ca^{2+}-crosslinked pectin hydrogels and reflects restricted chain mobility imposed by the multivalent ionic coordination junctions [25].

The complex viscosity(η*) plot (Fig.4.7d) displayed a standard power-law drop in with increase in angular frequency with linear log-log response - denoting shear-thinning viscoelastic behaviour. This decline in η* with frequency, combined with the maintained gel-like moduli, reflects the frequency-dependent disentanglement of the crosslinked network under oscillatory shear, consistent with a loss tangent ($\tan \delta = G''/G'$) less than unity across the frequency range [34]. The shear-thinning behaviour proves useful for potential

injectability or flow processing approaches and post-shear structural recovery indicated the ability for the network to be reformed due to the physical crosslinking [26, 30]. In conjunction, rheological characterization confirms that the pectin-anthocyanin-eggshell composite hydrogel had a well-structured and elastic-predominating viscoelastic network with distinct yield stress and shear-thinning behaviour of flowing which should originate from the ionic egg-box mechanism of crosslinking and it also in agreement with our earlier works on ionotropically crosslinked pectin-based hydrogel system [12, 18, 25].

4.8 FESEM

FESEM images were taken to investigate the surface morphology of the eggshell particles, jackfruit peel pectin and its hydrogel (Fig.4.8).

Fig.4.8. FESEM images of Eggshell (ES), Pectin derived from jackfruit peel (JPP), and there synthesized hydrogel.

Eggshell (ES) consists of irregular, angular, and blocky granular aggregates (Fig.4.8a, c). Particles display distinctly rigid, fractured geometric features that are highly characteristic of crystalline mineral phases (typically calcium carbonate). At 1µm (Fig.4.8b), the surface topography reveals a rough, heavily textured, and compact structure. This hard, granular micro structure together with its gradient distribution of particle size can maintain structural integrity, having adequate interface area for use as a bio-reinforcing filler or active adsorbent. Whereas that pectin particles are aggregated into irregular, heterogeneous clumps, suggests

that lack uniform crystalline geometry, asymmetrical agglomerates (Fig.4.8d, f). The diameter ranging from 50 to 80 µm are highly heterogeneous. At 2 µm highly corrugated, wrinkled, and porous topologies are visible (Fig.4.8e). These surfaces are deeply folded, indented, and completely amorphous in shape. Because of this rugged and discontinuous surface microstructure, the specific surface area of the pectin is maximized, which allows for convenient structural gaps and surface area for rapid hydration and effective matrix entrapment during gelation.

FESEM images of pectin-ES composite hydrogel shows that the hydrogel particles are distributed in widely dispersed, irregular clusters at 20 µm (fig.4.8i) . Implies that hydrogel formation and subsequently drying caused fragmentation and separation of the composite to separate, broken down agglomerates rather than a complete monolith.The diameter ranging from approximately 10 to 80 µm, a dense granular composites and flake-like shapes are visible in the pectin-ES hydrogel matrix (Fig.4.8g, h). These composite blocks are highly textured, rigid, and distinctly uneven in shape. Due to incorporation of the eggshell (ES) into the hydrogel, the smooth amorphous structures of the base pectin were completely disrupted, and heavily coated, granular structures were observed across the composite surfaces. Furthermore, significant discontinuities, fractured planes, and microscopic gaps between the granular structures were observed throughout the pectin-ES hydrogel, presenting a highly heterogeneous, interlocking, and rough surface morphology.

Elemental analysis

Energy dispersive X-ray spectroscopy (EDX) was used for elemental analysis and pure determination of eggshell, JPP and the produced hydrogels. The analysis demonstrated that carbon and oxygen were the primary constituents across the mapped samples. Traces of structural and trace elements such as calcium, potassium, aluminium and phosphorus, were found in varying amounts. The separation that these additional components (in particular, Ca

and P) present themselves as, are also a feature to be anticipated from a precursor like eggshells, within an inorganic mineral matrix. List the atomic percent composition (as calculated by Sum Spectrum): is given in Table 1.

Table 1: Elemental composition of eggshell, pectin, and its hydrogel.

Atomic (%)	C	O	Ca	P	K	Al	F	Zn	Fe	Cu	Mg
Eggshell (ES)	50.86	30.17	1.35	6.87	-	4.03	-	0.70	0.40	0.29	-
Jackfruit peel pectin (JPP)	50.83	48.21	0.38	-	0.51	-	-	-	0.04	-	0.02
JPP-ES hydrogel	40.01	47.66	5.13	0.77	3.76	0.17	1.27	0.26	0.21	0.32	0.24

Chapter 5

CONCLUSION, FUTURE SCOPE, AND SOCIAL IMPACT

5.1 CONCLUSION

This investigation successfully establishes a comprehensive, green-chemistry framework for the fabrication of a fully bio-derived, functionalized hydrogel matrix. This approach of extracting pectin using mild conditions and using a citric acid catalyst from abundant agricultural waste material – Artocarpus

heterophyllus peel to a value-added biopolymer – appears a very practical method. Moreover, we demonstrated the calcium oxide obtained from calcinating waste eggshells works as an effective green ionic crosslinking agent. This approach helped the establishment of a sturdy three-dimensional polymeric matrix exhibiting typical ‘egg-box’ structural configuration resulting into a dual-waste valorization paradigm. Anthocyanin derived from Alcea rosea (Gul Khaira) petals through ethanoic acid was successfully incorporated in hydrogel to obtain a very stable photophysical active pigment. Spectroscopic examination such as UV-Vis, gave a range of highest absorbance at between 520-540 nm range. The structural integrity and chemical coordination of this composite were extensively corroborated through FTIR and FESEM analyses. The FTIR spectra showed that the interaction between the carboxylate groups of the pectin and the calcium cations had occurred and that the anthocyanin molecules had been incorporated into the system without disturbing the bulk material. Morphologically, the composite exhibited a highly porous, interconnected architecture, which was subtly modified by the presence of the botanical pigment. Thermal and kinetic studies have further confirmed the material's stability and viability. Thermal Analysis (DSC) revealed that Eggshell-mediated crosslinking dramatically elevated the thermal stability of the matrix. In comparison to the incorporation of anthocyanin, the introduction of pigment resulted in particular degradation effects specifically tied to the anthocyanin pigment. Overall, the thermal stability of the hydrogel was not negatively

affected. Swelling kinetics demonstrated a highly tunable water-absorption capacity, dictated directly by the crosslinker concentration, allowing for precise control over network density. Ultimately, this work delivers a cost effective, easily scalable biopolymeric system with substantial potential for active compound delivery.

5.2 FUTURE SCOPE

The foundational methodologies and empirical outcomes established in this study provide a versatile platform for several advanced technological and industrial applications. A primary avenue for immediate exploration lies in the realm of intelligent packaging systems. As anthocyanins have clear predictable colorimetric transitions in response to pH changes, the prepared hydrogel can be used to construct a visual indicator smart film. Films like that might display real-time shelf life of things like milk, poultry and seafood through some visual indication of the change in pH caused by bacterial spoilage. Other than packaging, there is high potential to utilize matrix in the field of agro-chemicals, and biomedical. The developed pectin-calcium based matrix is biocompatible and can be functionalized with a range of plant derived antimicrobials, essential oil or targeted nutraceuticals. From an agronomic point of view, such expansion dynamics of the hydrogel could be interesting to conceive superabsorbent and controlled-release matrixial materials for agriculture. Adding important trace minerals like iron, copper, or zinc to the polymer, or growth hormones, would

make it a vehicle that releases nutrients over time to promote fertile soil conditions and also provide a useful tool in areas of the world where drought conditions are most challenging.

5.3 SOCIAL IMPACT

The impact of this research goes beyond that, contributing immensely to the fields of environment and community betterment. Such endeavours are crucial as they facilitate the creation of a bio-circular economy through the elimination of not just one, but two waste streams-jackfruit peel (which creates a seasonal disposal problem in India) and eggshells. Provides a practical, scalable approach to the municipal solid waste problem that is currently being addressed by this project, and produces state-of-the-art materials. This fits the development agenda strongly into creating a more remote, circular, economy around the rural areas which represent the core of agricultural waste production. This synthesis of a biodegradable and plant-derived option will, additionally combat the growing issue of petro-chemical plastic contamination, and thus offer a non-toxic, alternative to plastic wrap which could pose long-term risks. Used as the cheapest water conservation and soil-conditioning water storing devices on farms this could help resource-poor farming operations. The technology can help to strengthen regional food security by promoting water conservation and enhancing the resistance of crops to drought and climate stress. Finally, research on the topic really calls for to leave poisonous man-made colors as well as

transition to well-being natural colors as well as is in line with public outcry to “clean” up honor mark that implies non-toxic food. From an educational standpoint, affordable, locally sourced plant waste and basic laboratory procedures also present this technology as an excellent candidate for the classroom: an easily replicated low-cost experiment in hands of tomorrow’s community volunteers or the next generation of researchers can really foster the environment around green chemistry and scientific literacy.

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