DESIGN AND DEVELOPMENT OF NOVEL GELATIN/POLYACRYLAMIDE/CMTKG HYDROGEL FOR DELIVERY OF AMPICILLIN SODIUM

Dissertation Submitted

In Partial Fulfillment of the Requirements for the

Degree of

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CHEMISTRY

by

TANUJA KUMARI

(2k22/MSCCHE/43)

&

NITIN

(2k22/MSCCHE/23)

Under the Supervision of

Prof. S.G. WARKAR

Department of Applied Chemistry

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daulatpur, Main Bawana Road, Delhi-110042, India

May 2024

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daulatpur, Main Bawana Road, Delhi-110042, India

CANDIDATE'S DELCLARATION

We Tanuja Kumari (2k22/MSCCHE/43) and Nitin (2k22/MSCCHE/23) hereby certify that the work which is being presented in the dissertation entitled **"Design and development of novel Gelatin/Polyacrylamide/Carboxymethyl Tamarind Kernel Gum Hydrogel for delivery of Ampicillin Sodium"** in partial fulfillment 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 my/our own work carried out during the period from August 2023 to March 2024 under the supervision of Prof. S.G. Warkar.

The matter presented in the dissertation has not been submitted by us for the award of any other degree of this or any other Institute.

Tanuja Kumari (2k22/MSCCHE/43)

Nitin (2k22/MSCCHE/23)

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daulatpur, Main Bawana Road, Delhi-110042, India

CERTIFICATE

Certified that Tanuja Kumari (2k22/MSCCHE/43) and Nitin (2k22/MSCCHE/23) has carried out their search work presented in this dissertation entitled **"Design and development of novel Gelatin/Polyacrylamide/Carboxymethyl Tamarind Kernel Gum Hydrogel for delivery of Ampicillin Sodium"** 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

DATE -

Prof. S.G. Warkar

(SUPERVISOR)

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Tanuja Kumari & Nitin

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daulatpur, Main Bawana Road, Delhi-110042, India

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ABSTRACT

This research focuses on developing pH-responsive hydrogels using a blend of gelatin, polyacrylamide (PAM), and carboxymethyl tamarind kernel gum (CMTKG), loaded with the hydrophilic drug ampicillin sodium. Optimization of the formulation involved varying cross-linker, and initiator amounts, aiming to enhance the swelling ratio. Characterization techniques including Scanning Electron Microscopy (SEM), Powder X-ray Diffraction (PXRD), and Attenuated Total Reflection-Fourier Transform Infrared Spectroscopy (ATR-FTIR) were employed to analyze the fabricated hydrogel. Swelling behavior and drug release profiles were assessed in both acidic (pH 1.2) and alkaline (pH 7.4) buffer solutions. The synthesized hydrogels demonstrated more favorable outcomes under alkaline conditions. Kinetic modeling data supported the suitability of the Korsmeyer-Peppas model in explaining the drug release mechanism, with observations suggesting Fickian diffusion at pH 1.2 and non-Fickian diffusion at pH 7.4. Hence, the Gelatin/PAM/CMTKG hydrogel exhibits promising potential for tailored release of ampicillin sodium in response to pH variations, holding significant implications for targeted drug delivery applications.

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AM- Acrylamide

ATR-FTIR- Attenuated Total Reflection- Fourier Transform Infrared Spectroscopy

CMTKG- Carboxymethyl Tamarind Kernel Gum

COO^- - Carboxylate ion

$-CONH_2$ - Amide group

Cukα- Copper kα

DEE- Drug Entrapment Efficiency

DL- Drug Loading

g- Gram

IPN- Interpenetrating Polymeric Hydrogel

KPS- Potassium per sulphate

MBA- N', N' -methylene-bis (acrylamide)

mg- Milligram

N_2 - Dinitrogen

NH_2- Amine group

OH- Hydroxyl group

PAM- Polyacrylamide

PXRD- Powder X-ray diffraction

R^2- Regression coefficient

$-SO_3H$- Sulfonic group

SEM- Scanning Electron Microscopy

SR- Swelling Ratio

TGA- Thermal Gravimetric Analysis

TKG- Tamarind Kernel Gum

UV-Vis- Ultraviolet- visible spectroscopy

W_d- Weight of dried hydrogel

W_s- Weight of swollen hydrogel

%- Percentage

λ_{max}- Maximum Wavelength

CHAPTER 1

INTRODUCTION AND LITERATURE SURVEY

INTRODUCTION

The term "hydrogel" was first introduced in 1894 to describe a colloidal gel . The initial documented application of hydrogels in the biomedical field was reported by Wichterle and Lim in 1960.[1] Since then, the use of hydrogels has expanded to a wide range of biomedical and pharmaceutical applications. Hydrogels are soft, rubbery and have high water absorbing capacity. By its definition, hydrogels are polymeric networks possessing a three-dimensional structure capable of absorbing significant quantities of water or biological fluids.[2] The hydrophilic groups such as -COOH, $-SO_3H$, -OH and $–CONH_2$ etc., facilitate the absorption of fluid into the matrix.[3]

Hydrogels are well-known for their biocompatibility and eco-friendliness, making them extensively utilized in industrial, pharmaceutical, and agricultural sectors.[4] Hydrogels display exceptional biodegradability, biocompatibility, porosity, mechanical strength, and a soft, rubbery texture, making them a promising option for delivering various therapeutic agents compared to alternative carrier systems.[5]A variety of synthetic and natural polymers, such as polyacrylamide, collagen, gelatin, hyaluronic acid, among others, have been utilized in constructing hydrogel networks.[6, 7] The cross-linking within the structure of hydrogels plays a vital role in preventing their dissolution in aqueous environments.

Hydrogels can be classified in various ways, such as by their method of preparation, polymer type, biodegradability. However, the most common classification of hydrogels is based on their responsiveness to the surrounding environment.[8]

1.1 Classification of Hydrogel :

Hydrogel can be classified according to various factors, as outlined below.

- **Classification based on source**
  Hydrogels can be categorized into two groups according to their origins, either natural or synthetic[9].
- **Classification according to polymeric composition**
  The preparation method results in the formation of several significant classes of hydrogels. These can be exemplified as follows:
  (a) Homopolymeric hydrogels denote polymer networks derived from a single species of monomer, which serves as the fundamental structural unit within any polymer network. Depending on the nature of the monomer and the polymerization technique employed, homopolymers may exhibit a cross-linked skeletal structure[10].
  (b) Copolymeric hydrogels consist of two or more distinct monomer species, with at least one component being hydrophilic. These monomers are arranged in a random, block, or alternating configuration along the polymer network chain[11].
  (c) Multipolymer Interpenetrating polymeric hydrogels (IPN), a significant class within hydrogels, consist of two distinct cross-linked synthetic and/or natural polymer components, contained within a network structure. In semi-IPN hydrogels, one component is cross-linked while the other component remains non-cross-linked[12].
- **Classification based on configuration**
  The classification of hydrogels, based on their physical structure and chemical composition, can be outlined as follows:
  (a) Crystalline.

(b) Semicrystalline: A blend of amorphous and crystalline phases.
(c) Amorphous (non-crystalline).

- **Classification based on the type of cross-linking**
  Hydrogels can be categorized into two groups depending on the chemical or physical nature of the cross-link junctions. Chemically cross-linked networks possess permanent junctions, whereas physical networks feature transient junctions that result from polymer chain entanglements or physical interactions like ionic interactions, hydrogen bonds, or hydrophobic interactions[10].
- **Classification based on network electrical charge:**

  Hydrogels can be classified into four groups depending on the presence or absence of electrical charge situated on the cross-linked chains:
  (a) Ionic (comprising anionic or cationic charges).
  (b) Nonionic (neutral).
  (c) Amphoteric electrolyte (ampholytic), containing both acidic and basic groups.
  (d) Zwitterionic (polybetaines), containing both anionic and cationic groups within each structural repeating unit[13].

Hydrogels, also known as intelligent gels or smart hydrogels, are a class of materials that can exhibit responsiveness to various chemical and physical stimuli. These stimuli include changes in pH, ionic strength and magnetic fields, which cause hydrogels to undergo significant changes in their properties, such as swelling, shrinking, or altering their mechanical strength.[8] Among these stimuli, pH is particularly significant in biomedical applications. pH-responsive hydrogels can be categorized as either anionic or cationic. Anionic hydrogels display increased swelling in alkaline pH environments due to ionization occurring at a pH exceeding their pKa. Conversely, cationic hydrogels exhibit greater swelling in acidic pH conditions, as they undergo ionization at a pH lower than their pKa.[14]

Polyacrylamide (PAM) is a non-toxic polymer, that is sensitive to pH, renowned for its outstanding biocompatibility. It contains the $–CONH_2$ group, which facilitates versatile swelling and drug release behaviors under different pH conditions.[15] PAM-based hydrogels demonstrate the capacity to retain their form and mechanical resilience. They have been extensively combined with natural and synthetic polymers such as carboxymethyl cellulose, polyethylene glycol, and carboxymethyl tamarind kernel gum to yield composite hydrogels with outstanding properties.[16]

Tamarind kernel gum (TKG) is among the most cost-effective biopolymers, derived from the Tamarindus indica L. tree[17]. However, rapid biodegradability, low solubility in cold water, and dull color are among the various drawbacks associated with TKG. Thus, there is a need to modify TKG in order to enhance and alter its pharmaceutical and physicochemical characteristics for improved efficacy and broader applicability. Chemical modification of TKG produces its derivative carboxymethyl tamarind kernel gum (CMTKG), with molar proportions of xylose, glucose and galactose as 2:3:1, resulting from carboxymethylation of TKG[18]. It is an anionic polymer with high swelling capacity and pH-dependent release properties, making it suitable for drug delivery application[19]. Introducing a carboxylic group to TKG imparts anionic properties to CMTKG due to the presence of carboxylate ions. Carboxymethylation of tamarind kernel gum leads to enhanced stability, solubility, and biodegradability of the biopolymer[20].

Gelatin is regarded as one of the most efficient bio-based polymers due to its numerous benefits, including non-toxicity, high water absorption, biodegradability, and

biocompatibility[21]. Gelatin is derived through the thermal denaturation of collagen sourced from animal skin, bones, and occasionally fish scales. Its structure primarily comprises of three amino acids—glycine (occurring every third residue), proline, and 4-hydroxyproline[22]. Gelatin readily dissolves in water at 37°C, is non-immunogenic, and demonstrates amphoteric behavior. These characteristics make gelatin-based hydrogels suitable for manufacturing contact lenses, tissue engineering matrices, and drug delivery systems[7].

Ampicillin sodium is indeed a versatile β-lactum antibiotic employed in the treatment of various infections in both humans and animals. It is a part of the broader penicillin class, known for its effectiveness against a wide array of bacterial infections due to its ability to interfere with the synthesis of peptidoglycan, a key component of bacterial cell walls. This disruption is particularly effective against rapidly growing bacteria, making ampicillin a go-to antibiotic for infections like those affecting the gastrointestinal tract, the urinary system, and the meninges, among others. Its ability to target both gram- positive bacteria and gram-negative bacteria enhances its utility in a clinical setting, through resistance patterns must be considered to maintain its efficacy.

Polyacrylamide and Gelatin are being employed with carboxymethyl tamarind kernel gum (CMTKG) for the first time, to the best of our knowledge. This work involves synthesis, characterization, swelling studies, drug release experiment, and the kinetic modeling of drug released from Ampicillin sodium-loaded PAM, CMTKG and Gelatin -based hydrogels.

This research delves into the fabrication of Gelatin/PAM/CMTKG hydrogel and Ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel, aiming for a pH-dependent release of the drug ampicillin sodium. Characterization techniques such as PXRD, ATR-FTIR, SEM were employed to assess the hydrogels. Additionally, the study investigates how varying amounts of biopolymer, cross-linker and initiator affect the swelling behavior of the hydrogels under pH 1.2 and 7.4 buffer conditions. Furthermore, the drug release kinetics were evaluated using different mathematical models, including Hixson-Crowell, Korsmeyer-Peppas, Higuchi, Zero order and First order.

LITERATURE SURVEY

Hydrogel	Crosslinker	Drug	Reference
Sodium Alginate/HPMC K100	$CaCl_2$	Ampicillin sodium	[23]
CMTKG/PSA/XG	MBA	Metformin HCL	[14]
PVP/AMPS	Ethylene glycol dimethacrylate	Release of acyclovir HCL	[8]
CMC/PAM/PVP	MBA	Release of theophylline	[24]
Gelatin/PMAA	MBA	salbutamol sulphate	[25]
PAM/MMT/CMC/MgO	MBA	Release of acyclovir	[26]

CHAPTER 2

MATERIAL AND SYNTHESIS

2.1 Materials

Hindustan Gum and Chemicals Ltd., Bhiwani, Haryana, kindly offered CMTKG, which has 0.20 degree of substitution. Potassium per sulphate (KPS, Fischer scientific, Mumbai, India), Gelatin (Fischer Scientific, Mumbai, India), Acrylamide (CDH, New Delhi India) and N', N'-methylene-bis(acrylamide) (MBA, Merck, Germany) were utilized as provided. For the preparation of the solutions, distilled water was used.

2.2 Synthesis of Gelatin/PAM/CMTKG hydrogel

The hydrogels based on PAM, CMTKG and Gelatin were fabricated by free radical polymerization with MBA as the cross-linker and KPS as initiator, as represented in Fig.2.1. The reaction procedure was carried out by dissolving required amount of CMTKG, AM and Gelatin in distilled water while being stirred continuously for 1 hour. Following this, KPS and MBA were added to the solution and magnetically stirred for an additional hour. The mixture was then poured into test tubes and heated for 1 hour in water bath at 60 ^{0}C. Afterward, the synthesized hydrogels were removed from the test tubes, sliced and over-dried at 50 ^{0}C.

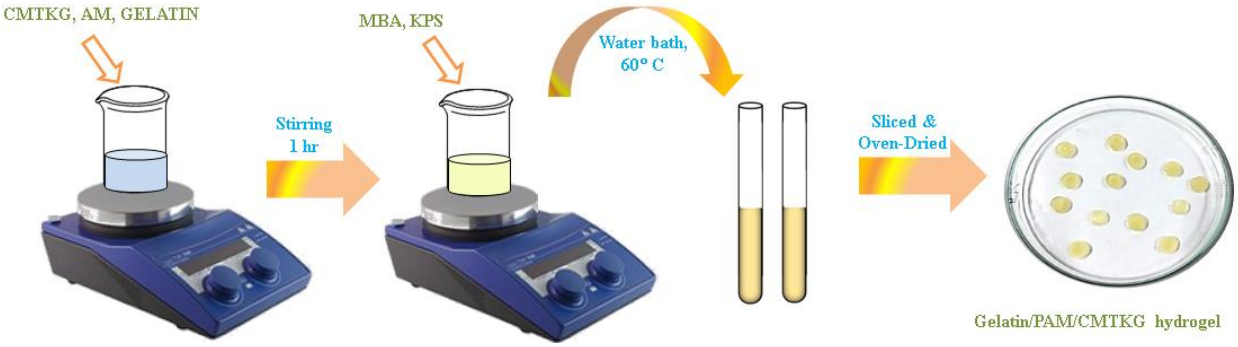


Fig. 2.1 Synthesis of Gelatin/PAM/CMTKG hydrogel.

Table 1 Composition of Gelatin/PAM/CMTKG hydrogels and their swelling ratio

Sample code	CMTKG (g)	AM (g)	MBA (mg)	KPS (mg)	Gelatin (g)	Swelling Ratio (%)	
						pH 7.4	pH 1.2
A-1	0.2	0.5	10	32	0.5	1371	1218
A-2	0.2	0.5	15	32	0.5	1295	1130
A-3	0.2	0.5	20	32	0.5	1274	1010
A-4	0.2	0.5	25	32	0.5	897	812
A-5	0.2	0.5	30	32	0.5	823	711
A-6	0.2	0.5	10	14	0.5	1226	1047
A-7	0.2	0.5	10	20	0.5	1259	1108
A-8	0.2	0.5	10	26	0.5	1334	1115
A-9	0.2	0.5	10	38	0.5	1063	1031

CHAPTER 3

EXPERIMENTAL SECTIONS

3. EXPERIMENTAL SECTIONS

3.1 Swelling Studies

The swelling behavior of the fabricated hydrogels was evaluated in pH 7.4 and pH 1.2 buffer solutions over a 24-hour period. The dried hydrogel discs were precisely weighed before being immersed in the respective solutions. After 1 hour, each hydrogel disc was taken out, gently blotted with filter paper to remove excess liquid, and then reweighed. The discs were then placed back into the solutions. This procedure was performed in triplicate to ensure accuracy.

W_S = swollen hydrogel's weight

$W_{d=}$ dry hydrogel initial weight

$$\text{Swelling Ratio } (\%) = \frac{(W_s - W_d)}{W_d} \times 100 \quad (1)$$

3.2 Ampicillin Sodium loading and entrapment efficiency

Drug loading for formulation (A-1) was carried out because it exhibited the highest swelling among all the hydrogels. The ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel was prepared according to section 2.2, with the modification of adding ampicillin sodium (the drug) during the addition of MBA and KPS to the solution; the rest of the process remained same. During loading (DL %) and drug entrapment efficiency (DEE %) calculations were performed for the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel. A hydrogel sample was immersed in a pH 7.4 buffer for 24 hours, and its absorbance at λ_{max} 203 nm was measured using UV-Vis spectrophotometer (Model: Cary UV-Vis). The amount of drug loaded was calculated using Beer-Lambert law[27, 28].

The formula used for calculation of drug loading

$$DL(\%) = \frac{Amount\ of\ drug\ loaded\ in\ hydrogel}{Weight\ of\ hydrogel} \times 100 \quad (2)$$

Formula used for calculation of encapsulation efficiency: -

$$DEE(\%) = \frac{Amount\ of\ drug\ laoded\ in\ hydrogel}{Initial\ amount\ of\ drug\ added\ in\ hydrogel} \times 100 \quad (3)$$

3.3 In vitro Ampicillin Sodium release

The in vitro study of the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel was conducted in an orbital incubator shaker using pH 1.2 and pH 7.4 buffer solutions. A precise amount of ampicillin sodium-loaded hydrogel disc was immersed in 100 ml of each buffer solution at 37 ^{0}C. After 1 hour, 3ml of the solution was withdrawn and replaced with an equal volume of fresh buffer. The absorbance of the solution was then measured using a UV spectrophotometer (Model: Cary 300 UV-Vis) at λ_{max} 203nm. The amount of drug was determined from calibration curve and entire process repeated three times.

3.4 Kinetic modeling of Ampicillin Sodium

Various mathematical models were used to determine drug release mechanisms, including Korsmeyer-Peppas, Higuchi, Hixson-Crowell, Zero-order, and First-order models. The regression coefficient (R^2) values were compared, and the model with R^2 value closest to 1 was deemed the best fit.

CHAPTER 4

FABRICATION METHOD AND PROCESS

4.1 UV-Visible Spectroscopy (UV-Vis)
Ultraviolet-Visible spectroscopy is widely used to measure how much a chemical substance absorbs light. Here is the detailed explanation of UV-visible principle:-

Principle-

Ultraviolet (UV) absorption occurs when a molecule or ion absorbs UV radiation, leading to the promotion of an electron from a lower energy (ground state) to a higher energy level (excited state). This transition usually involves moving an electron from a bonding or non-bonding orbital to an anti-bonding orbital. The particular wavelengths of UV light that are absorbed correspond to the energy difference between those states[29].

All the possible electronic transition are as follows:

1. σ to σ*

2. n to σ*

3. n to π*

4. π to π*

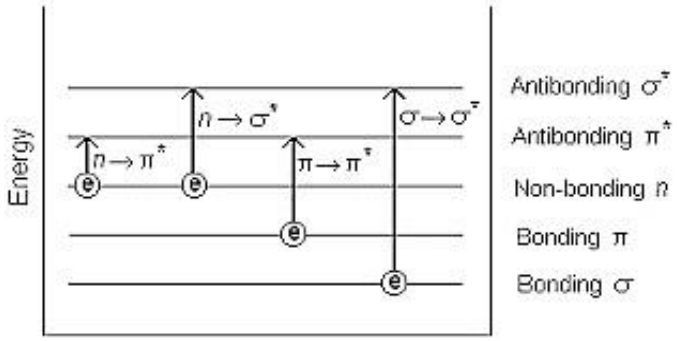


Fig 4.1 Electronic Transition

4.1.1 Beer Lambert's Law-
The absorbance of a sample is directly proportional to the concentration of the absorbing species and its path length.

$A = \varepsilon cl$ (4)

A=absorbance
C=concentration
l= path length, ε= molar extinction coefficient

4.1.2 Instrumentation:

Two types of absorbance:
a. Single beam UV-Vis spectrophotometer
b. Double beam UV-Vis spectrophotometer

Single beam spectrometer has a monochromator in between source and analyze at a time.

Double beam spectrometer has a single source and monochromator and there is a splitter and mirrors to split the light into two parts[29].

Components:

1. Light source
2. Monochromator
3. Sample cell
4. Detector

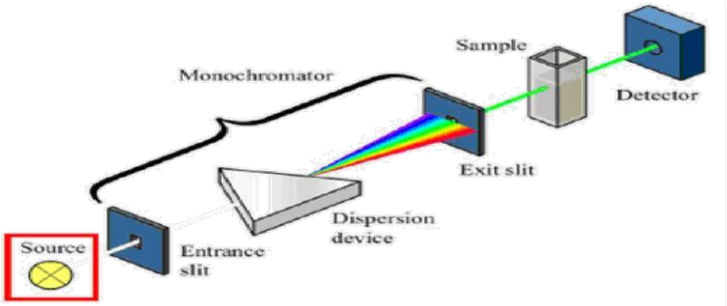


Fig 4.2 Single Beam UV-Vis spectrophotometer

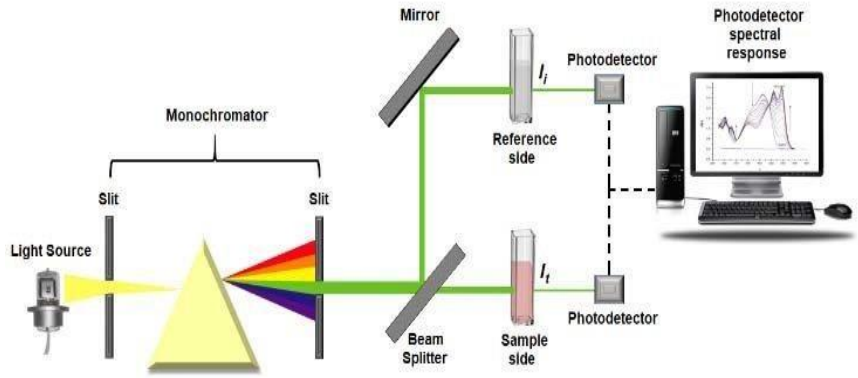


Fig 4.3 Double Beam UV-Vis spectrophotometer

The basic instrumentation (as shown in Figure 4) of a UV-Vis spectrophotometer includes:

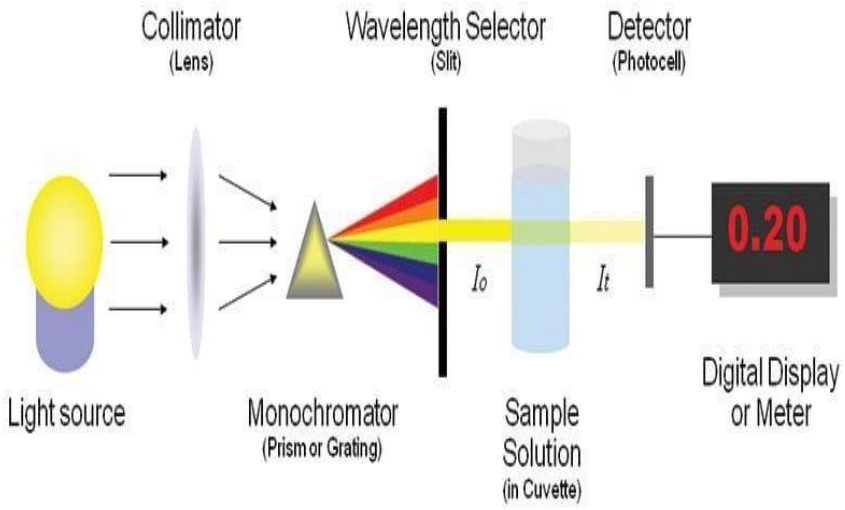


Fig 4.4 Instrumentation of UV-Vis Spectrophotometer

4.2 PXRD

Powder X-ray diffraction (PXRD) is an analytical technique used to analyze composition, atomic spacing, structure and properties of crystalline material. PXRD works on the following principle-

Principle:

Interaction of incident radiations with constructive interference [30]

Bragg's Law-

$n\lambda = 2d\sin\theta$ (5)

n=integer

λ=wavelength

d= interplanar spacing.

Instrumentation:

It was performed using a high resolution Bruker D8 diffractor at 2θ range of 10-70 ^{0}C with Cukα radiation (λ=1.5406 ^{0}A) to investigate the crystalline phases present in the sample.

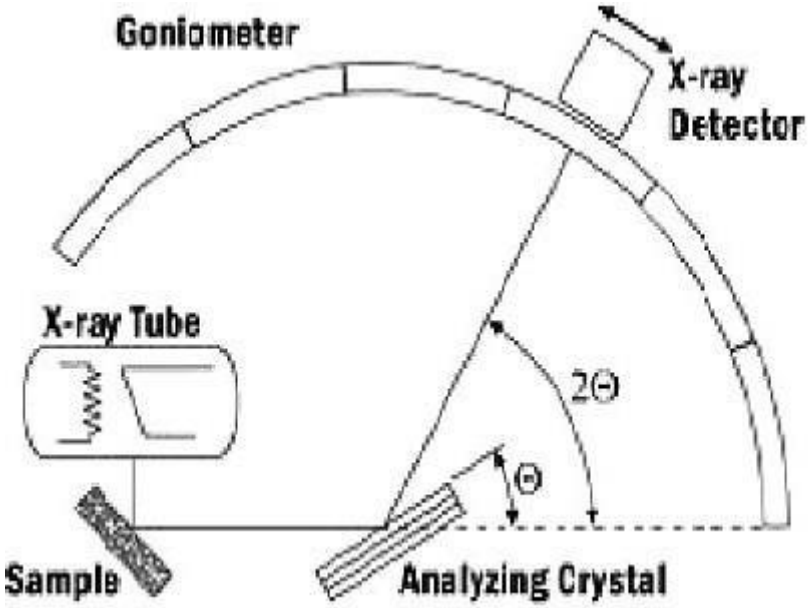


Fig 4.5 Schematic representation of a diffractometer system

4.3 Attenuated Total Reflection-Fourier Transform Infrared Spectroscopy (ATR-FTIR)
Fourier-transform infrared spectroscopy is used to determine the functional groups, chemical bonds and molecular structure of inorganic and organic compounds. FTIR principle works on the following process:

Principle:

In FTIR spectroscopy, IR radiation is passed through the sample and intensity of transmitted light is measured in wavelength.

For non-linear molecule degree of freedom= 3N-6

For linear molecule degree of freedom= 3N-5

There are two types of molecular vibrations that is: -

a. Stretching vibrations
- Symmetric
- Asymmetric

b. Bending vibrations
- Rocking
- Scissoring
- Wagging
- Twisting [31].

Instrumentation:
FTIR spectrometer is made up of various components, a broadband IR source, a sample compartment, an interferometer and detector.

4.4 Scanning Electron Microscopy (SEM)

Scanning Electron Microscopy (SEM) is a technique utilized to visualize structure of material and surface morphology at spatial resolution and magnification. SEM principle works on the following principle:

Principle of SEM:
Sample is subjected to high-energy electron beam in SEM, it reveals crucial details about material's morphology, topography, composition, orientation, crystallography and chemistry [32].

Instrumentation:
SEM components: electromagnetic lenses, specimen chamber, detectors, electron gun and computer system for image processing.
Electron guns develop a finely focused beam of electrons, to accelerate energies ranging from kiloelectron volt to tens of kiloelectron volt.
Electromagnetic lenses scan and focus electron beams across the sample with high precision.
Detector collects different type of signals emitted by sample.
Computer system processes collected signals to develop high resolution images and sample's surface.

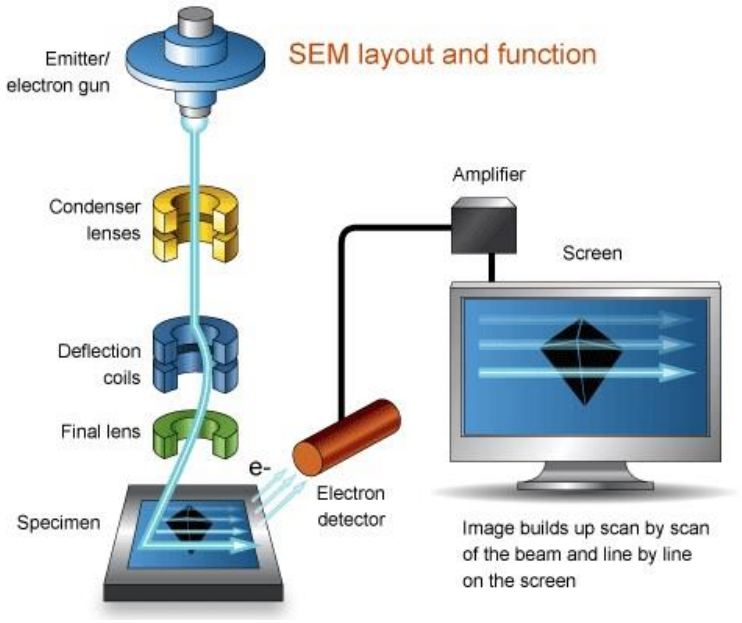


Fig 4.6 Schematic diagram of SEM

CHAPTER 5

CHARACTERIZATION TECHNIQUES

5.1 Powder X-RAY Diffraction (PXRD)

PXRD analysis was performed on ampicillin sodium, Gelatin/PAM/CMTKG hydrogel, and ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel to investigate their crystalline characteristics as illustrated in fig.2. Sharp crystalline peaks for ampicillin sodium were observed at 2θ values of 12.7, 15.5, 17.3, 18, 19.3, 19.8, 21.8, 23.9, 25.8, 26.3 and 26.7 [33]. Conversely, the PXRD spectra of both the Gelatin/PAM/CMTKG hydrogel and the ampicillin sodium -loaded Gelatin/PAM/CMTKG hydrogel exhibited broad bands, indicative of their amorphous nature. Additionally, the absence of discernible peaks for ampicillin sodium in the PXRD pattern of the drug-loaded hydrogel implies its uniform distribution within the polymers cross-linked network[34].

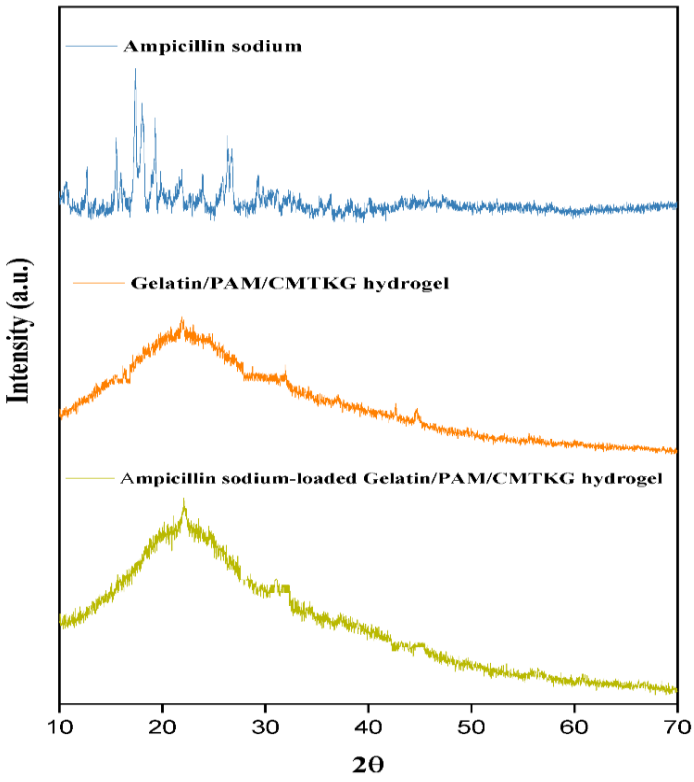


Fig 5.1 PXRD pattern of ampicillin sodium, Gelatin/PAM/CMTKG hydrogel and Ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel.

5.2 ATR-FTIR Spectroscopy

ATR-FTIR spectra for Gelatin/PAM/CMTKG hydrogel, ampicillin-loaded Gelatin/PAM/CMTKG hydrogel, and MBA are depicted in fig. 5.2. In both Gelatin/PAM/CMTKG and ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogels, bands at 3317 cm^{-1} and 3194 cm^{-1} and 3321 cm^{-1} and 3196 cm^{-1} correspond to overlapping -NH and -OH stretching vibrations. Peaks at 1323 cm^{-1} and 1321 cm^{-1} are attributed to COO^- symmetric stretch, while peaks at 1649 cm^{-1} and 1647 cm^{-1} indicate the C=O stretch in

Gelatin/PAM/CMTKG hydrogels, respectively[35]. Peaks at 2926 cm^{-1} and 2931 cm^{-1} relate to C-H stretch in the Gelatin/PAM/CMTKG hydrogels. Additionally, peaks assigned to the C-O-C group appear at 1094 cm^{-1} and 1106 cm^{-1} in Gelatin/PAM/CMTKG and ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogels, respectively[19].

In the ATR-FTIR spectrum of MBA, the peaks at 1303 cm^{-1}, ascribed to C-N stretch , shifts to 1321 cm^{-1} and 1323 cm^{-1} in the Gelatin/PAM/CMTKG and ampicillin sodium loaded-Gelatin/PAM/CMTKG hydrogels, respectively. This shift indicates a reduction in conjugation due to cross-linking and confirms the successful formation of the cross-linked hydrogel network[18]. Additionally, the absence of any new peaks in the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel, suggests that there is likely only a weak or physical interaction between the drug and hydrogel.

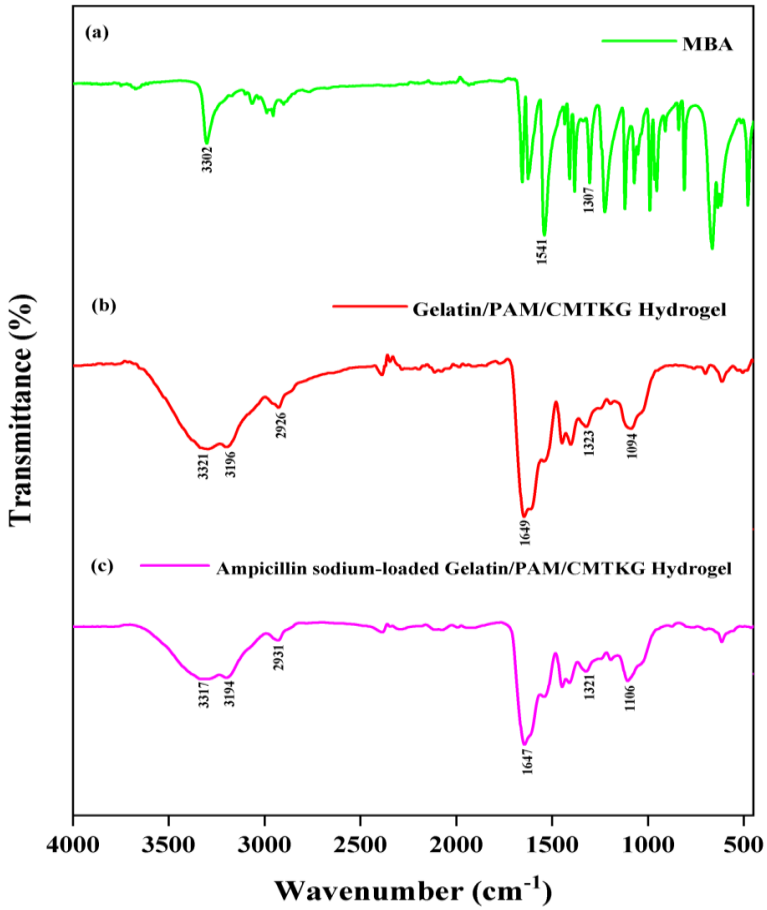


Fig 5.2 FTIR spectra of MBA, Gelatin/PAM/CMTKG hydrogel and Ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel

5.4 Scanning Electron Microscopy (SEM)

SEM images of the Gelatin/PAM/CMTKG hydrogel and the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel are displayed in fig. 5.3. The Gelatin/PAM/CMTKG hydrogel shows a highly uneven and porous surface, facilitating the effective encapsulation of ampicillin sodium crystals within the polymer matrix. However, the SEM micrograph of the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel reveals a smoother and less porous surface, as the entrapped drug fills the pores of the hydrogel matrix[8].

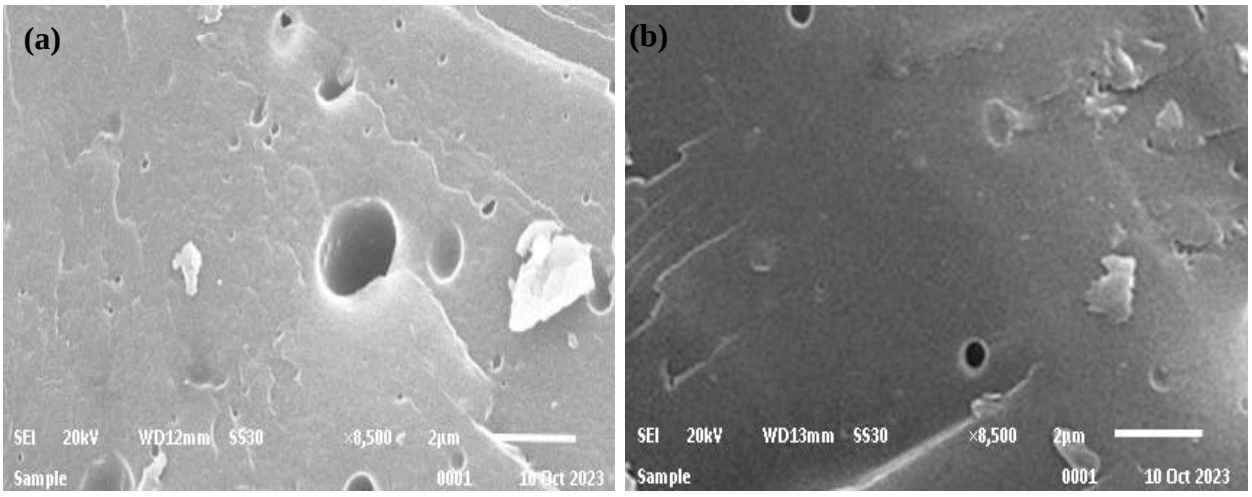


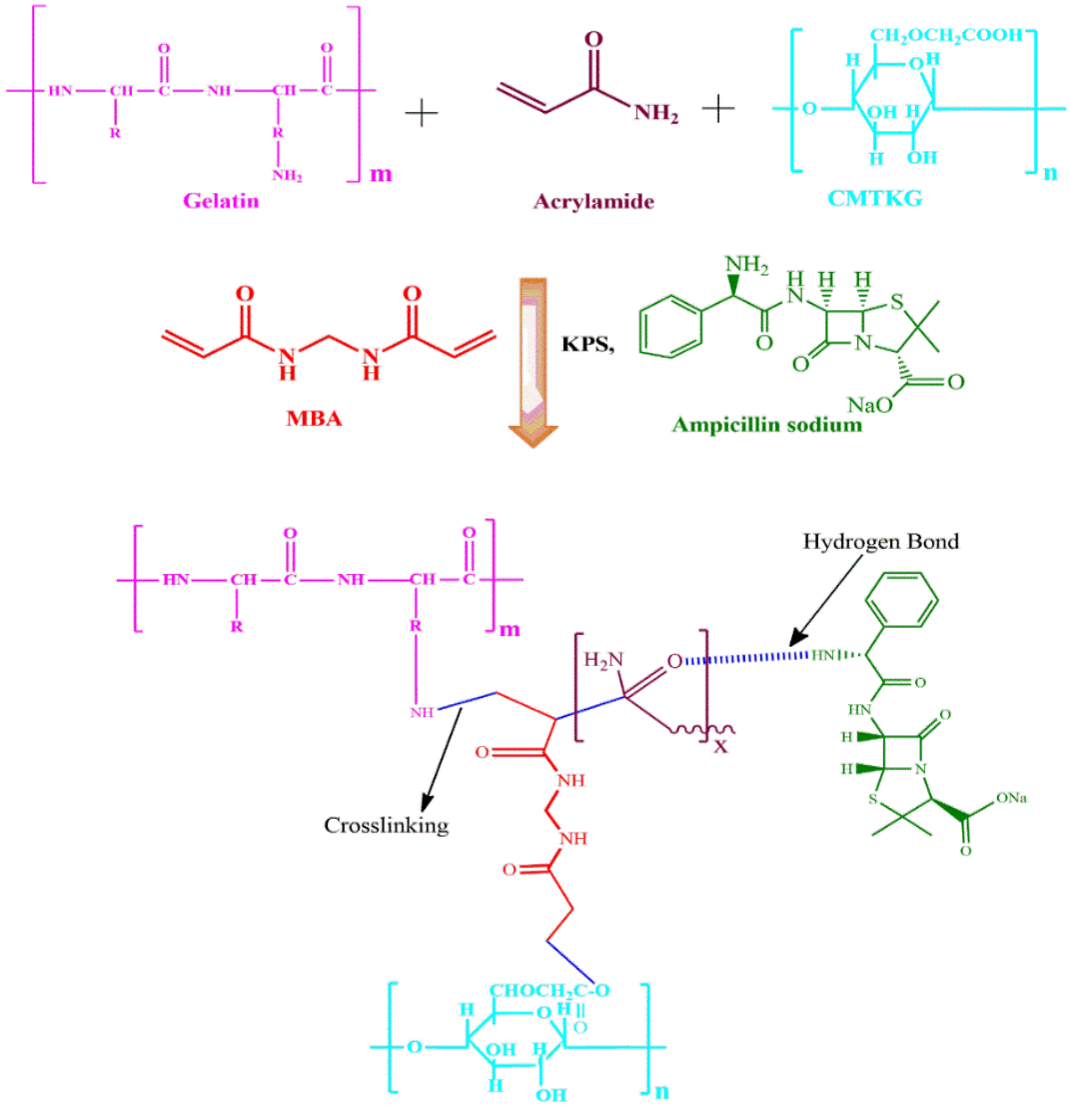
Fig 5.3 SEM images of (a) Gelatin/PAM/CMTKG hydrogel and (b) Ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel.

CHAPTER 6

RESULT AND DISCUSSION

6.1 Overview of Gelatin/PAM/CMTKG hydrogels and ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel formation

Multiple hydrogel formulations are created by varying the quantities of KPS and MBA through free radical polymerization, as shown in Table 1. The initiator KPS decomposes to yield sulfate radicals at 60°C. In the presence of sulfate radicals, the vinyl bond of AM undergoes decomposition, resulting in the formation of acrylamide radicals. These radicals initiated chain formation, propagated, and terminated, ultimately resulting in the formation of polyacrylamide. The sulfate radical additionally removes hydrogen atoms from the -COOH group of CMTKG and the $–NH_2$ group of Gelatin, thereby producing radicals on the polymeric chains. The MBA served as a cross-linker, linking the radicals present on the GELATIN, CMTKG, and PAM chains, which results in the formation of a three-dimensional polymeric hydrogel network[26]. Furthermore, in the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel, the hydrogen bonding between ampicillin sodium (drug) and PAM played a role in the formation of the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel, as illustrated in Scheme 1.



Scheme 1: Mechanism of synthesis of Ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel

6.2 Swelling Studies

Swelling analysis was performed in pH 7.4 and pH 1.2 solutions for all synthesized hydrogels, as detailed in Table 1. Figure 6.1 depicts how the quantities of the initiator (KPS) and crosslinker (MBA) affect the swelling ratio of the hydrogel.

6.2.1 Impact of cross-linker

MBA concentration significantly affected the swelling of hydrogels (Fig. 6.1 (a)). The synthesized hydrogel (A-1) displays a higher swelling compared to all other samples, as shown in Table 1. As the MBA concentration increased from 10 to 30 mg, the swelling capacity decreased, as higher concentrations of the cross-linker minimized the free volume within the polymeric network. Furthermore, reducing the MBA by 10 mg led to the formation of a gel-like structure due to inadequate cross-linking within the polymer network[36].

6.2.2 Impact of initiator

The MBA concentration remains constant at 10 mg, while variations in initiator and KPS content (14-38 mg) are investigated for their impact on swelling, as depicted in Fig. 6.1 (b). Initially, an increase in initiator content leads to a rise in the swelling ratio, with a maximum of 32 mg and afterwards decreases. However, a decrease is observed below 32 mg, attributed to unreacted monomers, which leads to a less compact polymer network and consequently reduces swelling. Moreover, initiator content exceeding 32 mg leads to a decrease in swelling because of the formation of a soluble oligomer chain instead of a polymeric chain, resulting in reduced swelling[18]. In all formulations, greater swelling was noted under alkaline pH conditions compared to acidic environments. Under alkaline pH, deprotonation of the COOH groups in the polymeric chains occurs, causing electrostatic repulsion among the COO^- ions, which in turn facilitates relaxation of the hydrogel's polymeric chains and their adoption to an elongated conformation. This elongation creates additional space, enabling biofluid to infiltrate the hydrogel matrix and enhance swelling. Additionally, at pH 1.2, the hydrogen bonding between CMTKG, PAM, and gelatin contracts the hydrogel network, diminishing the swelling efficiency of the hydrogel[26].

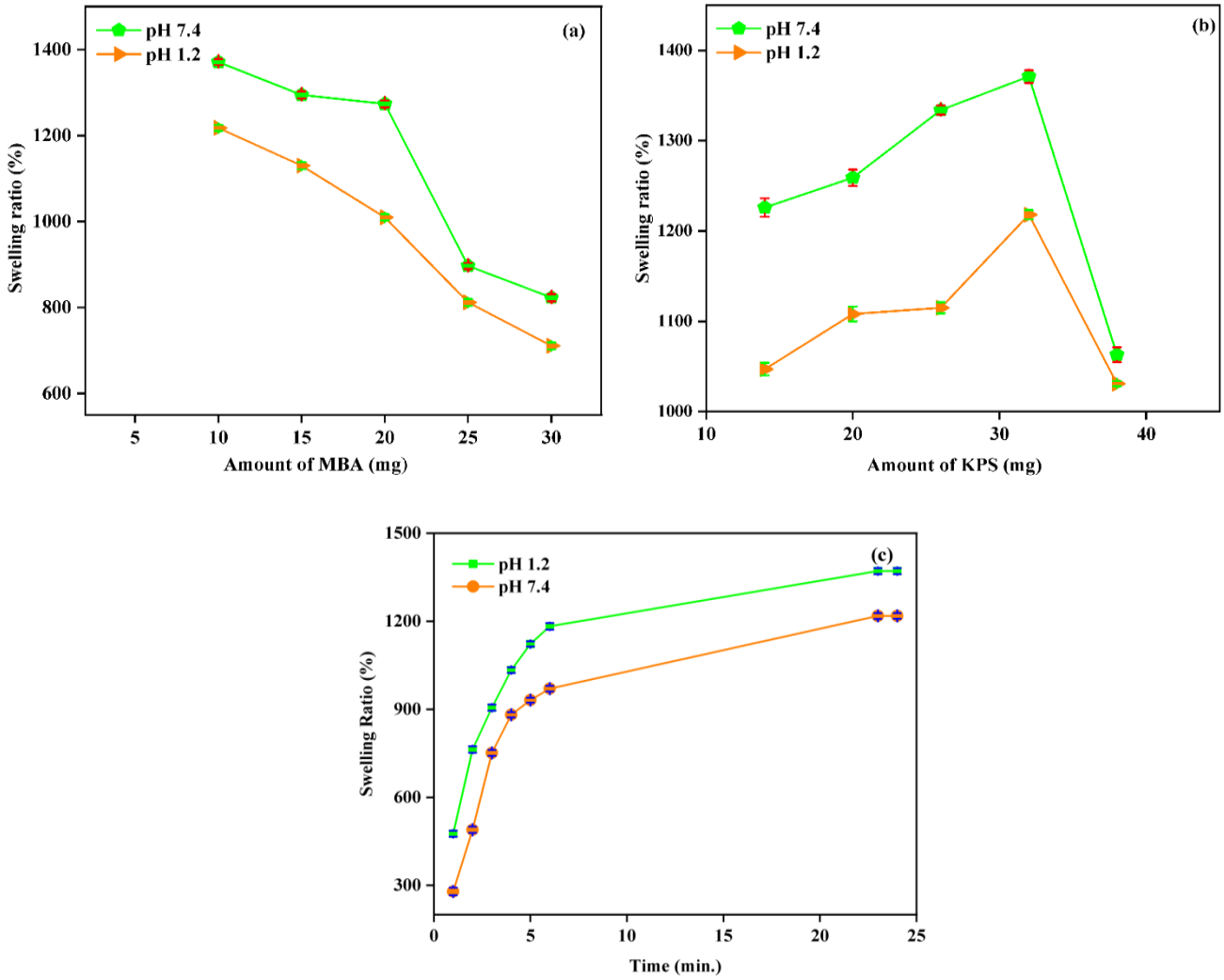


Fig 6.1 Impact of (a) cross-linker, (b) initiator on swelling ratio and (c) swelling ratio v/s time plot for A-1 formulation.

6.3 Loading of Drug and Encapsulation Efficiency

The drug loading and drug encapsulation efficiency for the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel were examined. The drug encapsulation efficiency (%) was determined to be 61.16% and drug loading (%) was 19.54%.

6.4 In vitro release of Ampicillin sodium

An in vitro investigation was carried out on ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel, as illustrated in Fig. 6.2. A significant proportion of drug release was observed under pH 7.4 in contrast to pH 1.2, attributed to the presence of deprotonated COO^- ions in CMTKG. Under pH 7.4 conditions, these COO^- ions demonstrate repulsive forces, inducing expansion of the polymer chain. As a result, this expansion leads to enhanced swelling, thereby facilitating increased diffusion of the drug from the hydrogel matrix. At pH 1.2, the COOH group undergoes protonation, promoting the establishment of hydrogen bonds among the CMTKG, Gelatin, and PAM chains. This interaction induces contraction of the polymeric network, resulting in reduced swelling and, consequently, diminished drug release from the hydrogel. This observation indicates the pH-dependent influence on drug release from the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel[34].

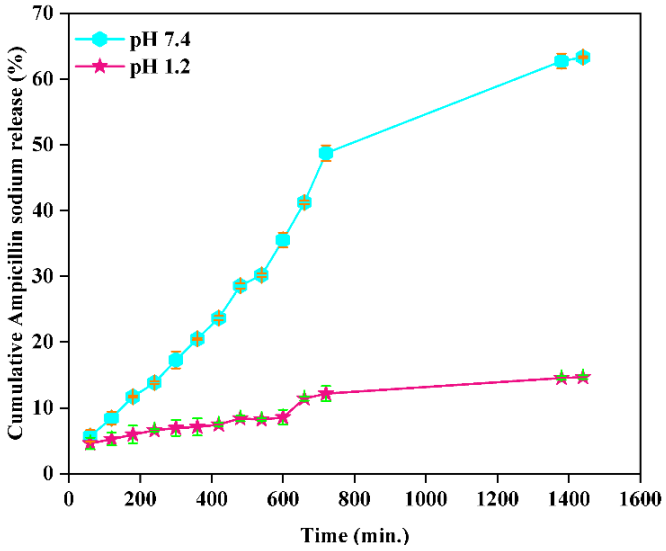


Fig 6.2 Ampicillin sodium release from Gelatin/PAM/CMTKG hydrogel (A-1) in pH 7.4 and 1.2.

6.5 Kinetic modeling of ampicillin sodium

The drug release mechanism from the ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel was evaluated employing various kinetic models, as outlined in Table 2. The findings indicate that the data closely adhered to the Korsmeyer-Peppas model for both pH conditions, as depicted in Fig. 8. At pH 7.4, with an R^2 value of 0.9871 and an n value of 0.7847, it suggested a non-Fickian diffusion mechanism, indicating both drug diffusion and polymeric chain relaxation. In contrast, at pH 1.2, the R^2 value was observed as 0.9892, with an n of 0.2557, indicating the Fickian diffusion mechanism of release, suggesting that only drug diffusion occurred[37].

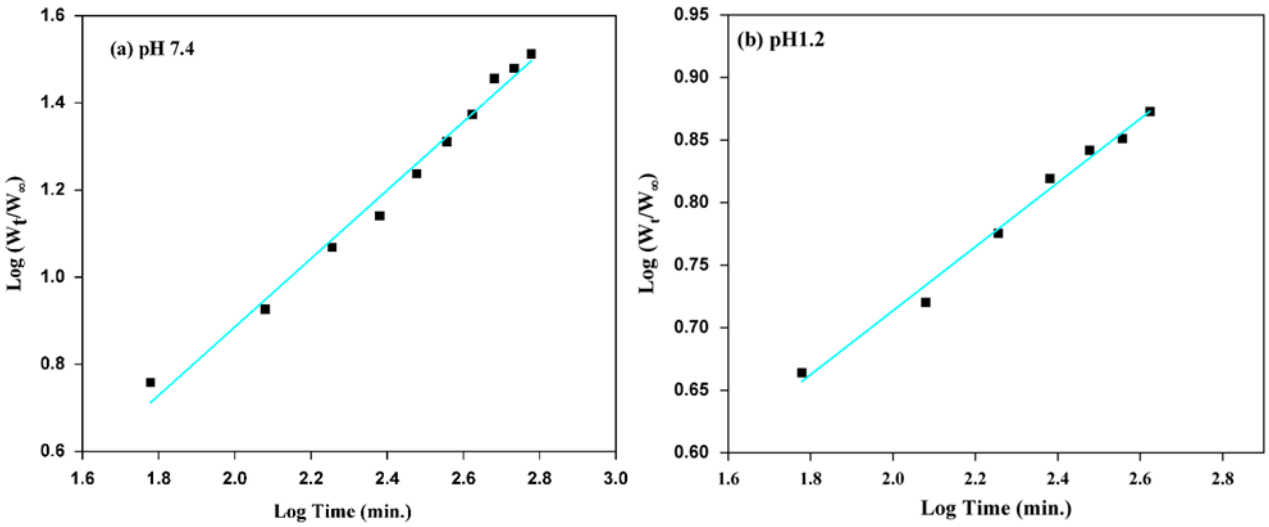


Fig 6.3 Kinetic modeling plot of ampicillin sodium as per Korsmeyer-Peppas model in (a) pH 7.4, and (b) 1.2.

Table 2 Kinetic modeling data of ampicillin sodium loaded Gelatin/PAM/CMTKG hydrogel

Model	Equation	pH 7.4		pH 1.2		Ref.
		n	R^2	n	R^2	
Zero Order	$W_t = W_\infty + k_0 t$ k_0 = zero-order constant	-	0.9012	-	0.9327	[38]
Higuchi	$W_t/W_\infty = k_{HG} t^{1/2}$ k_{HG} = Higuchi constant	-	0.9762	-	0.9809	[39]
First Order	$\text{Log } W_t = \text{Log } W_\infty + \frac{kt}{2.303}$ k = first order constant	-	0.7268	-	0.7831	[40]
Hixson-Crowell	$(W_t)^{1/3} - (W_\infty)^{1/3} = k_{HX} t$ k_{HX} = Hixson Crowell constant	-	0.8172	-	0.8239	[40]
Korsmeyer–Peppas	$W_t/W_\infty = kt^n$ k = kinetic constant n = diffusion exponent	0.7847	0.9871	0.2557	0.9892	[25]

CHAPTER 7

CONCLUSION

Conclusion

The synthesis of pH-responsive ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel was successfully achieved utilizing MBA as a crosslinker and KPS as the initiator. Comprehensive analysis of the fabricated hydrogel was performed employing ATR-FTIR, PXRD, and SEM techniques. Additionally, an investigation was carried out to understand the influence of MBA and KPS quantities on the swelling behavior of the hydrogels. The findings revealed that increasing amounts of MBA and KPS resulted in enhanced swelling; however, a further increase in KPS led to a decrease in swelling values. Swelling and drug release analyses were conducted in pH 1.2 and 7.4 buffer solutions, highlighting higher swelling and drug release at pH 7.4. Kinetic modeling of drug release aligned well with the Korsmeyer–Peppas model at both pH levels, suggesting a Fickian diffusion mechanism at pH 1.2 and a non-Fickian diffusion mechanism at pH 7.4. Consequently, the synthesized ampicillin sodium-loaded Gelatin/PAM/CMTKG hydrogel exhibits promise for achieving pH-dependent release of ampicillin sodium.

CHAPTER 8

REFERENCES

REFERENCES

1. Thakur S, Thakur VK, Arotiba OA (2018) History, Classification, Properties and Application of Hydrogels: An Overview. Springer Singapore

2. Nagam SP, Naga Jyothi A, Poojitha J, et al (2016) A Comprehensive review on hydrogels. Int J Curr Pharm Rev Res 8:19–23.

3. Ghauri ZH, Islam A, Qadir MA, et al (2021) Development and evaluation of pH-sensitive biodegradable ternary blended hydrogel films (chitosan/guar gum/PVP) for drug delivery application. Sci Rep 11:1–10.

4. Zainal SH, Mohd NH, Suhaili N, et al (2021) Preparation of cellulose-based hydrogel: A review. J Mater Res Technol 10:935–952..

5. Kajal, Kumar R, Meena P, Warkar SG (2023) Development and characterization of pH-responsive CMTKG/PAM/PEG hydrogel for oral administration of etophylline. Colloid Polym Sci.

6. Varghese SA, Rangappa SM, Siengchin S, Parameswaranpillai J (2019) Natural polymers and the hydrogels prepared from them. Elsevier Inc.

7. Jaipan P, Nguyen A, Narayan RJ (2017) Gelatin-based hydrogels for biomedical applications. MRS Commun 7:416–426.

8. Shoukat H, Pervaiz F, Noreen S, et al (2020) Fabrication and evaluation studies of novel polyvinylpyrrolidone and 2-acrylamido-2-methylpropane sulphonic acid-based crosslinked matrices for controlled release of acyclovir. Polym Bull 77:1869–1891.

9. Zhao W, Jin X, Cong Y, et al (2013) Degradable natural polymer hydrogels for articular cartilage tissue engineering. J Chem Technol Biotechnol 88:327–339.

10. Ahmed EM (2015) Hydrogel: Preparation, characterization, and applications: A review. J Adv Res 6:105–121.

11. Yang L, Chu JS, Fix JA (2002) Colon-specific drug delivery: New approaches and in vitro/in vivo evaluation. Int J Pharm 235:1–15.

12. Maolin Z, Jun L, Min Y, Hongfei H (2000) The swelling behavior of radiation prepared semi-interpenetrating polymer networks composed of polyNIPAAm and hydrophilic polymers. Radiat Phys Chem 58:397–400.

13. Garg S, Garg A (2016) HydrogelClassificationPropertiesPreparationandTechnicalFeatures. Hydrogel Classif Prop Prep Tech Featur 2:163–170

14. Tushar, Saraswat Y, Meena P, Warkar SG (2023) Synthesis and characterization of novel xanthan gum-based pH-sensitive hydrogel for metformin hydrochloride release. Colloid Polym Sci 301:1147–1158.

15. Zhou C, Wu Q (2011) A novel polyacrylamide nanocomposite hydrogel reinforced with natural chitosan nanofibers. Colloids Surfaces B Biointerfaces 84:155–162.

16. Mandal BB, Kapoor S, Kundu SC (2009) Biomaterials Silk fibroin / polyacrylamide semi-interpenetrating network hydrogels for controlled drug release. Biomaterials 30:2826–2836.

17. Khushbu, Warkar SG (2020) Potential applications and various aspects of polyfunctional macromolecule- carboxymethyl tamarind kernel gum. Eur Polym J 140:110042.

18. Meena P, Singh P, Warkar SG (2023) Development and assessment of carboxymethyl tamarind kernel gum-based pH-responsive hydrogel for release of diclofenac sodium. Eur Polym J 197:112340.

19. Mali KK, Dhawale SC, Dias RJ (2017) Synthesis and characterization of hydrogel films of carboxymethyl tamarind gum using citric acid. Int J Biol Macromol 105:463–470.

20. Kaur G, Jain S, Tiwary AK (2010) Chitosan-carboxymethyl tamarind kernel powder interpolymer complexation: Investigations for colon drug delivery. Sci Pharm 78:57–78.

21. Treesuppharat W, Rojanapanthu P, Siangsanoh C, et al (2017) Synthesis and characterization of bacterial cellulose and gelatin-based hydrogel composites for drug-delivery systems. Biotechnol Reports 15:84–91.

22. Pal K, Banthia AK, Majumdar DK (2007) Preparation and Characterization of Polyvinyl Alcohol – Gelatin Hydrogel Membranes for Biomedical Applications. 8:

23. Sevinç Özakar R, Özakar E (2021) The effect of polymer amount and crosslinker ratio in polymeric hydrogel beads on characterization. J Res Pharm 25:653–666.

24. Wei QB, Fu F, Zhang YQ, et al (2014) PH-responsive CMC/PAM/PVP semi-IPN hydrogels for theophylline drug release. J Polym Res 21:.

25. Rafique N, Ahmad M, Minhas MU, et al (2022) Designing gelatin-based swellable hydrogels system for controlled delivery of salbutamol sulphate: characterization and toxicity evaluation. Polym Bull 79:4535–4561.

26. Sabbagh F, Muhamad II (2017) Acrylamide-based hydrogel drug delivery systems: Release of Acyclovir from MgO nanocomposite hydrogel. J Taiwan Inst Chem Eng 72:182–193.

27. Khan R, Zaman M, Salawi A, et al (2022) Synthesis of Chemically Cross-Linked pH-Sensitive Hydrogels for the Sustained Delivery of Ezetimibe. Gels 8:1–17.

28. Shahid N, Erum A, Zaman M, et al (2021) Ph-responsive nanocomposite based hydrogels for the controlled delivery of ticagrelor; in vitro and in

29. Alekhya M, Jyothi M, Ramya M (2023) R EVIEW A RTICLE A Review of Hydrocephalus. Rev Lit Arts Am 18:216–220. 8

30. Bunaciu AA, Udriştioiu E gabriela, Aboul-Enein HY (2015) X-Ray

Diffraction: Instrumentation and Applications. Crit Rev Anal Chem 45:289–299.

31. Ismail AA, van de Voort FR, Sedman J (1997) Chapter 4 Fourier transform infrared spectroscopy: Principles and applications. Tech Instrum Anal Chem 18:93–139.

32. Abdullah A, Mohammed A (2019) Scanning Electron Microscopy (SEM): A Review. Proc 2018 Int Conf Hydraul Pneum - HERVEX 77–85

33. Profile SEE (2021) The effect of polymer amount and crosslinker ratio in polymeric hydrogel beads on characterization.

34. Ashames A, Ullah K, Al-Tabakha M, et al (2022) Development, characterization and In-vitro evaluation of guar gum based new polymeric matrices for controlled delivery using metformin HCl as model drug. PLoS One 17:1–20

35. Trivedi JH (2013) Synthesis, characterization, and swelling behavior of superabsorbent hydrogel from sodium salt of partially carboxymethylated tamarind kernel powder-g-PAN. J Appl Polym Sci 129:1992–2003.

36. Ghumman SA, Noreen S, Hameed H, et al (2022) Synthesis of pH-Sensitive Cross-Linked Basil Seed Gum/Acrylic Acid Hydrogels by Free Radical Copolymerization Technique for Sustained Delivery of Captopril. Gels 8:.

37. Suhail M, Hsieh YH, Khan A, et al (2021) Preparation and in vitro evaluation of aspartic/alginic acid based semi-interpenetrating network hydrogels for controlled release of ibuprofen. Gels 7:.

38. Goonoo N, Bhaw-Luximon A, Ujoodha R, et al (2014) Naltrexone: A review of existing sustained drug delivery systems and emerging nano-based systems. J Control Release 183:154–166.

39. Rani I, Warkar SG, Kumar A (2023) Nano ZnO embedded poly (ethylene glycol) diacrylate cross-linked carboxymethyl tamarind kernel gum (CMTKG)/poly (sodium acrylate) composite hydrogels for oral delivery

of ciprofloxacin drug and their antibacterial properties. Mater Today Commun 35:105635.

40. Das D, Das R, Mandal J, et al (2014) Dextrin crosslinked with poly(lactic acid): A novel hydrogel for controlled drug release application. J Appl Polym Sci 131:1–12.

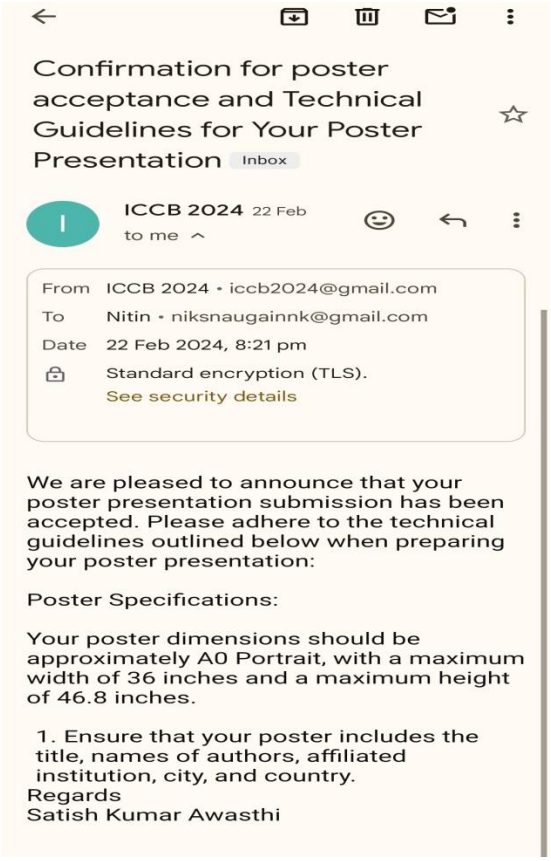
CHAPTER 9

LIST OF CONFERENCE ATTENDED

9.1 Conference details

1. Presented our research work at the ***"International Conference on Crossroads of Chemistry, Biology & Atmospheric Environment: A Modern Perspective"*** organized by the **University of Delhi, Indian Science Congress Association (ISCA), Delhi Chapter and The Indian Society of Analytical Scientist-Delhi Chapter**, held at Department of Chemistry, University of Delhi on 26,27 & 28 February 2024.

9.2 Registration details



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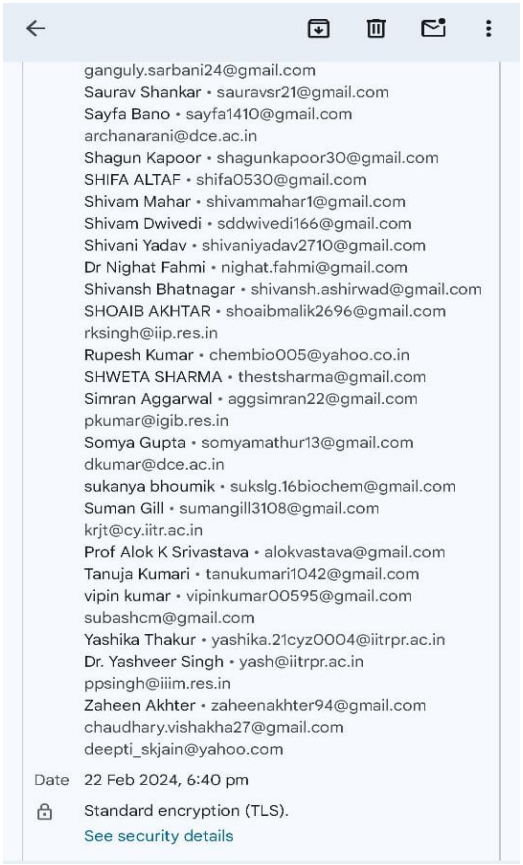

ganguly.sarbani24@gmail.com
Saurav Shankar • sauravsr21@gmail.com
Sayfa Bano • sayfa1410@gmail.com
archanarani@dce.ac.in
Shagun Kapoor • shagunkapoor30@gmail.com
SHIFA ALTAF • shifa0530@gmail.com
Shivam Mahar • shivammahar1@gmail.com
Shivam Dwivedi • sddwivedi166@gmail.com
Shivani Yadav • shivaniyadav2710@gmail.com
Dr Nighat Fahmi • nighat.fahmi@gmail.com
Shivansh Bhatnagar • shivansh.ashirwad@gmail.com
SHOAIB AKHTAR • shoaibmalik2696@gmail.com
rksingh@iip.res.in
Rupesh Kumar • chembio005@yahoo.co.in
SHWETA SHARMA • thestsharma@gmail.com
Simran Aggarwal • aggsimran22@gmail.com
pkumar@igib.res.in
Somya Gupta • somyamathur13@gmail.com
dkumar@dce.ac.in
sukanya bhoumik • sukslg.16biochem@gmail.com
Suman Gill • sumangill3108@gmail.com
krjt@cy.iitr.ac.in
Prof Alok K Srivastava • alokvastava@gmail.com
Tanuja Kumari • tanukumari1042@gmail.com
vipin kumar • vipinkumar00595@gmail.com
subashcm@gmail.com
Yashika Thakur • yashika.21cyz0004@iitrpr.ac.in
Dr. Yashveer Singh • yash@iitrpr.ac.in
ppsingh@iiim.res.in
Zaheen Akhter • zaheenakhter94@gmail.com
chaudhary.vishakha27@gmail.com
deepti_skjain@yahoo.com

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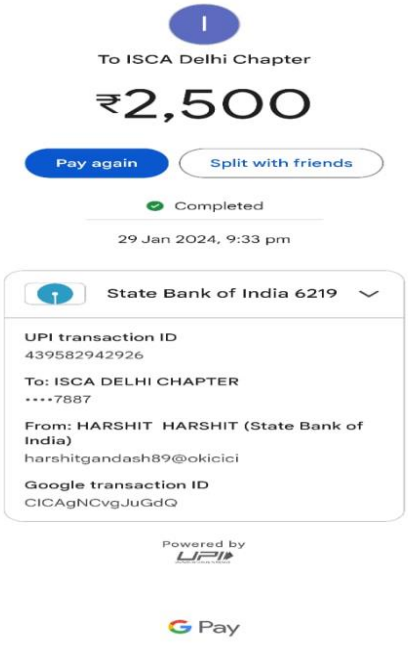
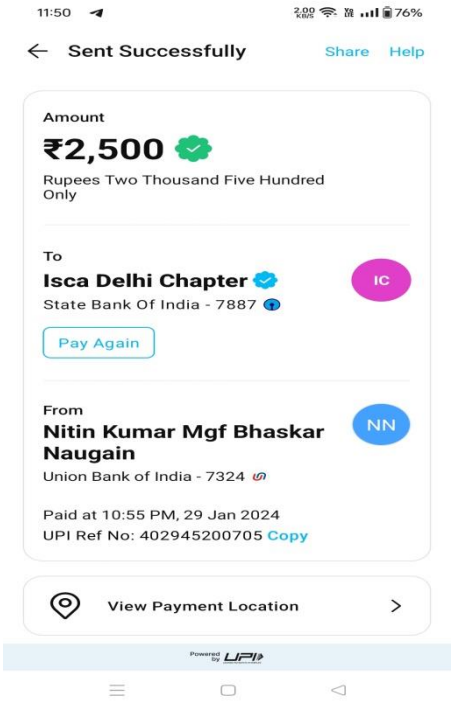
9.3 Payment details





9.4 Certificates

ICCB-2024

International Conference
on
CROSSROADS OF CHEMISTRY, BIOLOGY & ATMOSPHERIC ENVIRONMENT:
A MODERN PROSPECTIVE

February 26 to 28, 2024

CERTIFICATE OF PARTICIPATION

This is to certify that Prof./Dr./Ms./Mr. Janya Kumari from Delhi Technological University has participated as Invited Speaker/Oral/Poster/Participant in the "International Conference on Crossroads of Chemistry, Biology & Atmospheric Environment: A Modern Prospective" jointly organized by University of Delhi, Indian Science Congress Association (ISCA), Delhi Chapter and The Indian Society of Analytical Scientists-Delhi Chapter, held at Department of Chemistry, University of Delhi.

Prof. Satish K. Awasthi
Convenor, ICCB-2024
Department of Chemistry, University of Delhi

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Prof. Satish K. Awasthi
Convenor, ICCB-2024
Department of Chemistry, University of Delhi