M. Sc Chemistry

DEVELOPMENT AND CHARACTERIZATION OF pH – RESPONSIVE CMTKG/PAM/PEG HYDROGEL FOR ORAL ADMINISTRATION OF ETOPHYLLINE

A PROJECT WORK

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS
FOR THE AWARD OF THE DEGREE
OF

MASTER OF SCIENCE
IN
CHEMISTRY

Submitted by:

KAJAL
(2K21/MSCCHE/24)

RAMENDER KUMAR
(2K21/MSCCHE/36)

Under the supervision of
Prof. S.G.Warkar

DEPARTMENT OF APPLIED CHEMISTRY
DELHI TECHNOLOGICAL UNIVERSITY
(Formerly Delhi College Of Engineering)
Bawana Road, Delhi-110042
MAY,2023

Kajal, Ramender Kumar 2023

CONTENTS

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Bawana Road, Delhi -110042

CANDIDATE'S DECLARATION

We (Kajal, Ramender Kumar), 2K21/MSCCHE/24, 2K21/MSCCHE/36 students of M.Sc. (Department of Applied Chemistry), hereby declare that the project Dissertation Titled " Development And Characterization of pH-responsive CMTKG/PAM/PEG hydrogel for oral administration of Etophylline" which is submitted by us to the Department of Applied Chemistry, Delhi Technological University, Delhi in partial fulfillment of the requirement for the award of the degree of Master of Science, is original and not copied from any source without proper citation. This work has not previously formed the basis for the award of any degree, Diploma Associateship, Fellowship or other similar title or recognition.

Place: Delhi Kajal, Ramender Kumar

Date:

DEPARTMENT OF APPLIED CHEMISTRY

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College Of Engineering)

Bawana Road, Delhi -110042

CERTIFICATE

I hereby certify that the project Dissertation titled "Development and Characterization of pH – responsive CMTKG/PAM/PEG hydrogel for oral administration of Etophylline". Which is submitted by Kajal, Ramender kumar 2K21/MSCCHE/24, 2K21/MSCCHE/36 Department of Applied Chemistry, Delhi Technological University, Delhi in partial fulfillment of the requirement for the award of the degree of Master of Chemistry is a record of the project work carried out by the students under my supervision. To the best of my knowledge this work has not been submitted in part or full for any Degree or Diploma to this University or elsewhere.

Place: Delhi

Date:

Prof. S.G.Warkar

SUPERVISOR

ACKNOWLEDGEMENT

Thanks to the gracious Lord for all the immeasurable blessings you have bestowed upon us as well as to our family and friends for their love and support.

We would like to thank our project supervisor **Prof. S.G.Warkar Department of Applied Chemistry, Delhi Technological University, Delhi** who provide us the opportunity to work under their guidance.

We would like to thank the honourable **Prof. Yogesh Singh, Vice Chancellor of the Delhi Technological University** and **Prof. Anil kumar, Head of the Department of Applied Chemistry** for their kind permission to work in the Department of Applied Chemistry and providing utilities for the completion of this project.

We would like to thank also all the faculty members of the Department of Applied Chemistry for their constant support, encouragement and suggestions which help us to complete our project on time.

We also convey our ardent gratitude to the **Ph.D. scholar Miss Priyanka Meena** & non-teaching staffs of the Department of Applied Chemistry, Delhi Technological University for their valuable help over the span of the project work.

Kajal, Ramender kumar

ABSTRACT

The present research aims to create a pH-responsive hydrogel based on carboxymethyl tamarind kernel gum (CMTKG), polyacrylamide (PAM), and polyethylene glycol (PEG) for the regulated delivery of etophylline using the free radical mechanism. To achieve the expected characteristics of the hydrogels, the formulation was customised by varying the quantity of the biopolymer, initiator, and crosslinker. The consequence of these changes on the swelling ratio was also examined. Attenuated Total Reflection-Fourier Transform Infrared Spectroscopy (ATR-FTIR), Powder X-ray Diffraction (PXRD), Scanning Electron Microscopy (SEM), and Thermogravimetry Analysis (TGA) were used to analyse the produced hydrogel. Studies on drug release and swelling were carried out at pH 1.2 and 7.4 to examine the hydrogel's sensitivity to pH. Results of swelling and drug release show that increased swelling and drug release were seen at pH 7.4 in contrast to pH 1.2, which suggests that the formed polymeric network behaves differently depending on the pH. The regulated release profile of etophylline was validated by drug release kinetics, with the best-fitting model being the Korsmeyer-Peppas model with Fickian diffusion serving as the drug release mechanism at pH 1.2 and non-fickian diffusion at pH 7.4. So, CMTKG/PAM/PEG hydrogel can be used as a potentially effective material for the regulated pH-dependent release of etophylline.

LIST OF FIGURES

LIST OF TABLES

LIST OF SYMBOLS & ABBREVATIONS

1. Potassium persulphate	KPS
2. N,N- Methylenebisacrylamide	MBA
3. Carboxymethylene Tamarind kernel gum	CMTKG
4. Poly(ethylene glycol)	PEG
5. Poly(Acryl amide)	PAM

CHAPTER 1

INTRODUCTION

1.1 Hydrogel

The hydrogel, which is a three-dimensional polymeric network of synthetic and natural polymers, is capable of retaining huge amounts of water instead of dissolving in water[1] . A pore-forming polymer structure, consisting of hydrophilic functional groups like amines, carboxylic acids, hydroxyls, sulphonyls, etc., which are connected to the polymeric backbone, provides a very good capacity for the absorption of water and biological fluids[2] . It is well known that hydrogels possess excellent biocompatibility, biodegradability, porosity, mechanical strength, and soft rubbery nature, making it a promising choice for the administration of a number of therapeutic agents as compared to other carrier systems[3] .

There are two basic categories of hydrogels, smart and stimuli-responsive. Smart and stimuli-responsive hydrogels are defined as hydrogels that respond to a variety of physical or chemical stimuli, including electric fields, ionic strength, temperature, pH, light, etc[2][4].. In order to deliver targeted drugs in the most efficient manner, pH-responsive hydrogels are the most commonly used hydrogels. By changing in the amount or shape of the pH-responsive hydrogels with slight changes in external stimuli, the pH-responsive hydrogels can adapt to the pH values of bio-fluids, making them suitable for biomedical applications that require them to adjust to a variety of pH values. Through the use of pH-sensitive hydrogels, we have been able to develop an oral release system that can be used for delivery of drugs under controlled conditions[5].

1.2 Poly(acrylamide) (PAM)

A polymer known as polyacrylamide (PAM) is a pH-sensitive, non-toxic polymer that has brilliant biocompatibility and is widely explored in drug delivery applications. It contains CONH2 group, which has the property of promoting swelling under pH-dependent conditions and also enable the release of the drug. A PAM is composed of a basic amide group that forms the hydrogen bond with the drug. As a result, the loading and release capacity of the drug is enhanced. In addition to

maintaining their shape and mechanical strength, PAM-based hydrogels also have a high mechanical strength[6]. A wide range of natural and synthetic polymers have been used to make composite hydrogels with excellent properties. Such polymers include carboxymethyl cellulose, polyethylene glycol, carboxymethyl tamarind kernel gum, etc[7].

1.3 Poly(ethylene glycol) PEG

Another type of nontoxic, water-soluble, and pH-sensitive polymer is poly(ethylene glycol), also known as PEG. Among the many applications of PEG hydrogels, there have been numerous studies demonstrating their compatibility with other polymers due to the fact that they contain a functional hydroxyl group at the end of their polymeric chains. The excellent ligand binding properties of PEG-based hydrogels have made them more and more popular as drug carriers. Due to its enhanced solubility and biocompatibility, PEG can be used as an effective drug delivery material for a wide variety of applications, such as drug delivery systems. Its property of fast clearance from the body makes it a good applicant in drug delivery systems[8]. It also has the advantage of not having significant side effects, making it a safe choice for medical treatments. Furthermore, its low cost makes it an attractive option for medical applications[9].

1.4 CarboxyMethyl Tamarind Kernel gum (CMTKG)

The Tamarindus indica L. plant is one of the cheapest sources of biopolymers on earth, and it also produces quite a bit of plastic. A number of biopolymers are derived from the sap of the Tamarind tree, including Tamarind kernel gum (TKG). There are chemical modifications carried out on tamarind kernel gum (TKG), leading to carboxymethyl tamarind kernel gum (CMTKG), which is characterized by many properties that include high drug loading capacity, hydrophilicity, stability, as well as wide pH tolerance[10]. The polymer is pH-sensitive, meaning it swells or deswells based on the pH of the medium, making it an excellent candidate for site-specific drug delivery, due to its ability to swell or deswell as pH changes[11].

1.5 Etophylline as a model drug

There are several types of bronchodilators, such as etophylline, which are used to treat diseases such as chronic obstructive pulmonary disease, asthma, and chronic bronchitis. In order to make sure that the lungs are relaxing and making the bronchioles wider, it relaxes the muscles of the

lungs. To avoid this complication, it is necessary to develop a new formulation strategy that will ensure superior oral bioavailability as well as reduce patient discomfort due to unpleasant injection delivery because of the poor oral absorption of etophylline[12].

The purpose of this study is to synthesize CMTKG/PAM/PEG hydrogel and Etophylline loaded CMTKG/PAM/PEG hydrogel for the pH-dependent release of Etophylline from these hydrogels. It has been determined that the developed hydrogels have been characterized using ATR-FTIR, SEM, PXRD, and TGA. In addition, the impact of biopolymers, crosslinkers, and initiators on the swelling behavior of Etophylline loaded hydrogels was analyzed in pH 1.2 and pH 7.4. We used a variety of models to assess drug release kinetics, including Hixson-Crowell, Higuchi, Zero-Order, Korsmeyer-Peppas, and First-Order.

CHAPTER 2

EXPERIMENTAL

2.1 Materials

It is our great pleasure to receive CMTKG which has been generously provided by Hindustan Gum and Chemicals Ltd. in Bhiwani, Haryana. There were five types of compounds used in this experiment: N,N'-methylenebis(acrylamide) (MBA, Merck, Germany), Acrylamide (AM, CDH, Delhi), Polyethyleneglycol (PEG, High Purity Laboratory Chemicals Pvt. Ltd., Mumbai, India), Potassium persulphate (KPS, Fischer Scientific, Bombay) and Etophylline (UnicurePvt. Ltd., U.P., India) which were all used exactly as received.

2.2 Synthesis of CMTKG/PAM/PEG and Etophylline loaded CMTKG/PAM/PEG hydrogels

CMTKG/PAM/PEG hydrogels were prepared by mixing an aqueous solution of PEG, AM, and CMTKG separately, followed by the addition of PEG, AM, and CMTKG to the solution. A continuous stirring was carried out during the addition of KPS, and MBA. A test tube was filled with the resultant solution, which was then placed in a water bath at a temperature of 60°C. Hydrogel pellets were extracted, cut into pellets, and dried at 50°C for 48 hours in an oven after they had been extracted[6].

As far as the synthesis of CMTKG/PAM/PEG hydrogel loaded with etophylline is concerned, the same procedure was followed with the exception of a few minor alterations. It should be noted that as MBA and KPS were being introduced into the process, Etophylline was also being added, with the rest of the procedures remaining the same as shown in Fig.1. Different Etophylline-loaded hydrogel formulations were synthesized by using varying percentages of CMTKG, KPS, and MBA to produce different formulations using the same process[13].

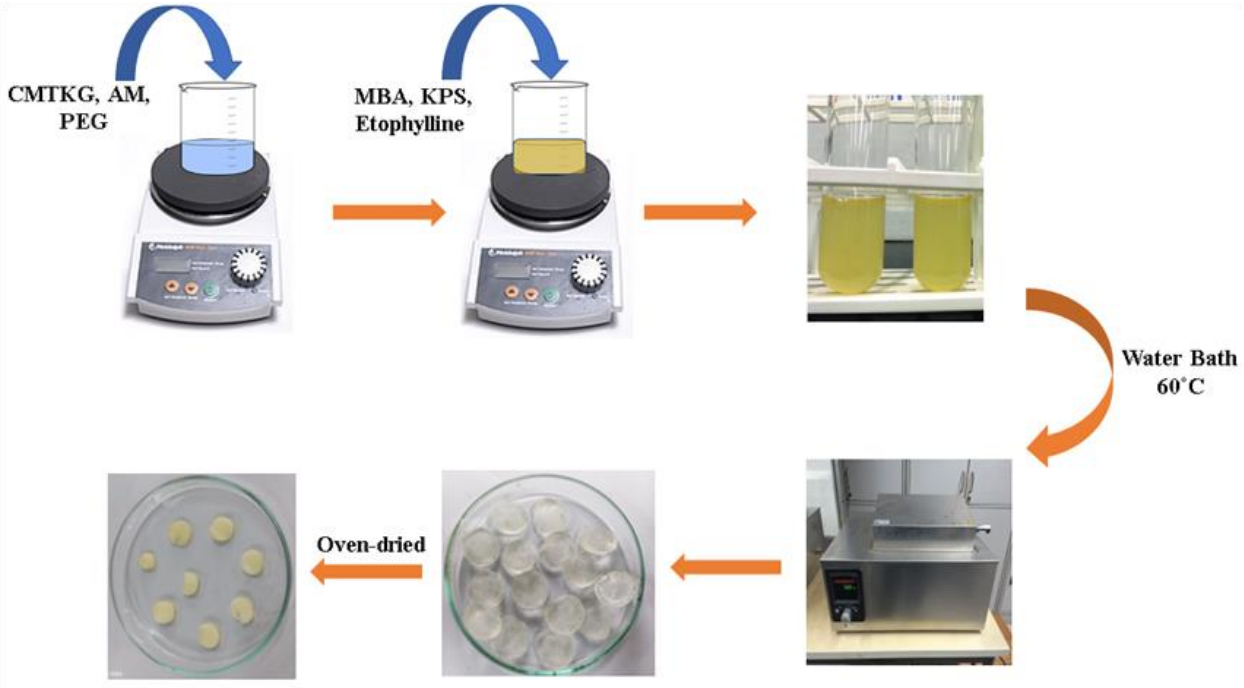


Fig 1 Schematic diagram for the synthesis of Etophylline-loaded hydrogels

Table 1 Amount of reactants required in the formulation of hydrogels and their swelling ratio

Sample	CMTKG (g)	AM (g)	PEG (g)	KPS (mg)	MBA (mg)	Etophylline (mg)	Swelling Ratio (%)	
							pH 7.4	pH 1.2
P-1	0.1	2.4	0.1	32	20	50	740	734
P-2	0.2	2.4	0.1	32	20	50	777	773
P-3	0.3	2.4	0.1	32	20	50	805	778
P-4	0.4	2.4	0.1	32	20	50	820	799
P-5	**0.5**	**2.4**	**0.1**	**32**	**20**	**50**	**864**	**849**
P-6	0.5	2.4	0.1	22	20	50	714	703
P-7	0.5	2.4	0.1	27	20	50	755	750
P-8	0.5	2.4	0.1	37	20	50	782	768
P-9	0.5	2.4	0.1	42	20	50	779	768
P-10	0.5	2.4	0.1	32	25	50	804	771
P-11	0.5	2.4	0.1	32	30	50	727	622
P-12	0.5	2.4	0.1	32	35	50	650	569
P-13	0.5	2.4	0.1	32	40	50	648	538
P-14	0.5	2.4	0.1	32	20	00	855	842

2.3 Swelling analysis

As a result of the gravimetric method, we performed swelling studies on all synthesized hydrogels. In this study, completely dry hydrogel discs were soaked in pH 1.2 and pH 7.4 solutions for a short time and extra fluid was blotted with filter paper after each dip. After being soaked in the solutions for a short time, swollen hydrogel discs were removed, weighed, and then heated again. The equation was used to measure the amount of swelling that occurred when a sample of material was submerged in a solution[3]. This measure was used to calculate the absorption capacity of the material, which is an important factor in determining its suitability for a particular application.

$$\text{Swelling ratio}(\%) = (W_f - W_i)/Wi \qquad (1)$$

Where, W_f and W_i are the final and initial weight of the hydrogel respectively.

2.4 Etophylline Efficiency and Encapsulation Efficiency

As a result of this study, the composition P-5 was chosen as an example of the composition that might be used to test the loading and encapsulation efficiency of Etophylline, since it shows the highest swelling in both buffer solutions. In order to determine the pH of the buffer solution, 100 ml of buffer solution with a pH of 7.4 was dipped with the hydrogel pellet (0.1g) for 24 hours. In

order to determine the absorbance of the sample after it had been drawn, a UV-visible spectrophotometer was used (Model: Cary 300 UV-Vis) at λmax273 nm[14]. Etophylline loading and entrapment efficiency (%) was determined by the given equations.

$$\text{Drug Loading (\%)} = \frac{Drug\ content\ in\ hydrogel\ discs \times 1oo}{weight\ of\ hydrogel\ discs} \qquad (2)$$

$$\text{Drug Encapsulation efficiency(\%)} = \frac{Drug\ content\ in\ hydrogel\ Discs \times 100}{Theoretical\ drug\ content\ in\ hydrogel\ disc} \qquad (3)$$

2.5 *In-vitro* Release Analysis of Etophylline

It was determined that formulation P-5 exhibits the greatest swelling among all the formulations that were studied in terms of drug release by HCl-KCl buffer and phosphate buffer saline. For the determination of the release pattern of Etophylline in a 100 ml solution of pH 1.2 and 7.4 at 37°C, weighted hydrogel discs (0.1g) were soaked in 100 ml of pH 1.2 and 7.4 and after a certain interval of time, 3 ml of the solution was collected and a fresh buffer of equivalent volume was added to the solution to maintain volume. Using a UV-visible spectrophotometer (Model: Cary 300 UV-Vis), Etophylline release was measured at a wavelength of 252 nanometers (max 273). The procedure was conducted in triplicate and a calibration curve was drawn to determine drug content .This procedure was done because the pH levels and temperature of the buffer can affect the rate of release of Etophylline from the hydrogel discs. By testing the drug release in different pH levels, we can get a better understanding of how the release is affected. The calibration curve is used to compare the drug content in the hydrogel discs at different pH levels. This helps to determine how much of the drug is being released at each pH level and if the release rate is affected by changes in pH. By testing the drug release in different pH levels, we can get a better understanding of how the release rate is affected and optimize the drug delivery system accordingly[15].

2.6 Kinetic Modeling of Etophylline

Various models were used to analyze the kinetics of Etophylline's release from a synthesized hydrogel based on the release data. Here the Hixson-Crowell, Higuchi, Zero-Order, Korsmeyer-Peppas, and First-Order models were used to analyze how Etophylline releases from a synthesized hydrogel. In order to determine which model is the best fit, the coefficients of determination (R^2) values from each model were compared, and the model that has the highest R^2 value closer to 1

was assumed to be the best fit model. This is because the coefficient of determination (R^2) value is a measure of how well the model fits the data. The closer the R^2 value is to 1, the better the model fits the data[16].

2.7 Characterization

A Bruker D8 diffractometer was used at 2Θ to perform PXRD measurements between 5 and 70°C using CuKα radiation. An ATR-FTIR spectrophotometer, Nicolet iS50 FTIR, was used to obtain the ATR-FTIR spectra. An N2 atmosphere was used during the thermogravimetric analysis with a heating rate of 10°C/min from 30°C to 850°C, using Perkin Elmer TGA 4000. With the aid of SEM (JEOL Japan mode: JSM 6610LV) the surface morphology was observed. The Bruker D8 diffractometer was used to measure the phase transformation behavior of the sample at different temperatures by obtaining the PXRD spectra. The ATR-FTIR spectrophotometer was used to analyze the chemical composition of the sample. Finally, the SEM was used to observe the surface morphology of the sample.

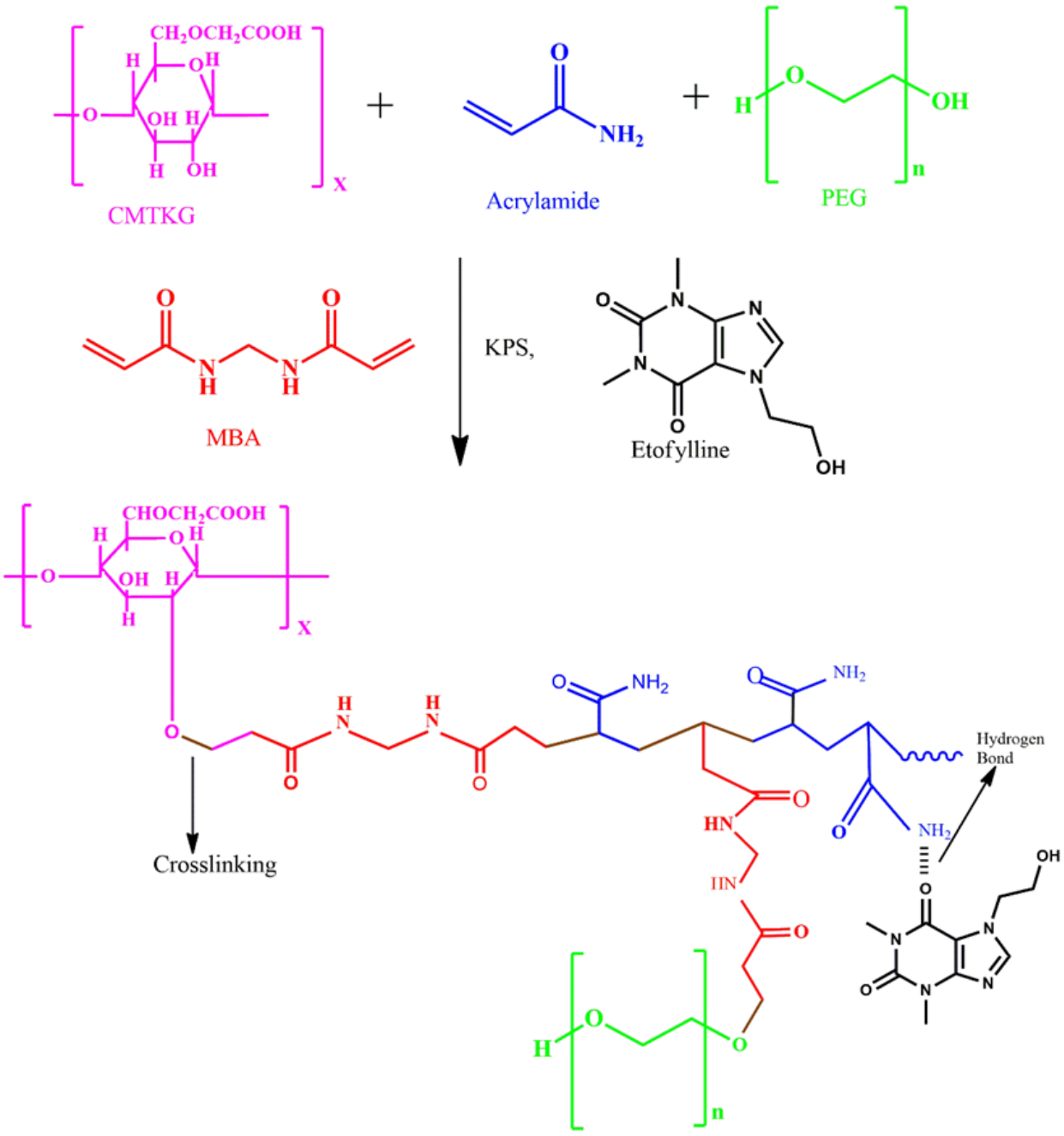
CHAPTER 3

RESULT AND DISCUSSION

3.1 Proposed Mechanism of synthesis of Etofylline loaded CMTKG/PAM/PEG hydrogel

At 60°C, there is first a breakdown of the initiator KPS, which leads to the production of the sulfate radicals. During the process of cleavage, a sulphate radical was introduced into the reaction system, which subsequently caused the release of acrylamide radicals as a result. In Scheme 1e, this acrylamide molecule attacks the other acrylamide molecule which is loaded on the CMTKG/PAM/PEG hydrogel, and subsequently the chain propagation step occurs which leads to the formation of polyacrylamide. The CMTKG and PEG molecules provide a hydrophilic and porous structure that enables the Etophylline molecules to form hydrogen bonds with the PAM molecules, creating a stable hydrogel matrix that holds the Etophylline molecules in place. The MBA radical provides a cross-linking agent to strengthen the hydrogel matrix, further increasing its stability and efficacy[21]. The CMTKG and PEG molecules form a hydrophilic network which allows the entrapment of water molecules between them. This creates a porous matrix that allows for the Etophylline molecules to form hydrogen bonds with the PAM molecules. The MBA radical

then enhances this network and serves as a connecting agent between the different molecules, creating a more robust matrix that helps to keep the Etophylline molecules in place, resulting in a more effective and stable hydrogel.



Scheme 1 Proposed mechanism of Etophylline loaded CMTKG/PAM/PEG hydrogel

3.2 Swelling Studies

According to Table 1, the swelling ratios of all the developed hydrogels were measured for pH 1.2 and pH 7.4 at both pH 1.2 and pH 7.4 at the time of development. In Figure 2, it is shown that the presence of biopolymer, initiator, and cross-linker content of the hydrogel has a significantly positive impact on its swelling[25]. This indicates that the higher the biopolymer, initiator, and cross-linker content of the hydrogel, the greater the swelling ratio at both pH 1.2 and pH 7.4. This suggests that these components play a key role in determining the hydrogel's swelling properties.

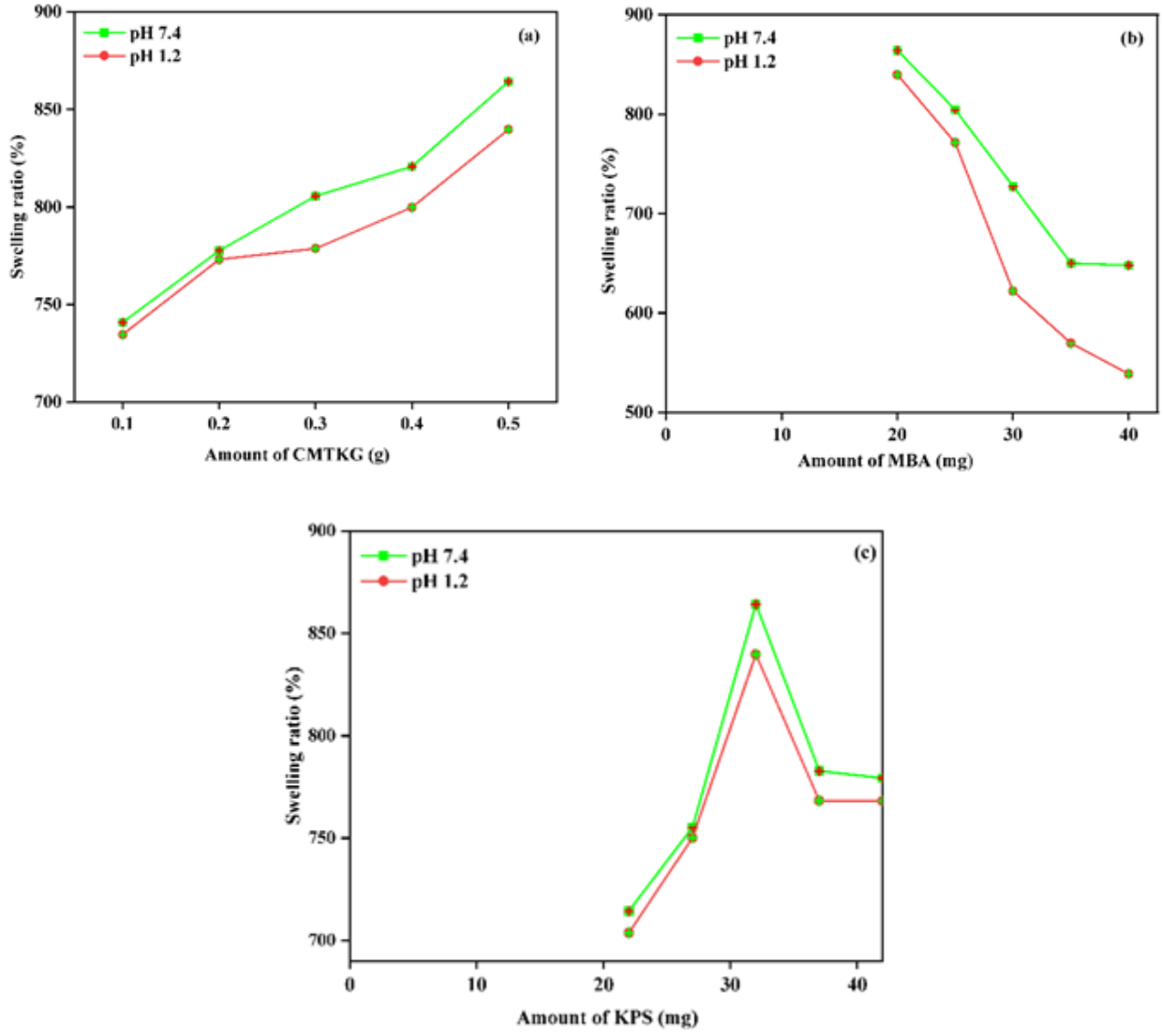


Fig 2 Impact of (a) biopolymer, (b) cross-linker, and (c) initiator content on the swelling ratio of developed hydrogels

3.2.1 Impact of Biopolymer

The impact of CMTKG content on the swelling ratio is depicted in Fig. 2. The swelling of hydrogel increases as the CMTKG content increased from 0.1 to 0.5 gm, due to the deprotonated COO- across the polymeric network[24]. These like charged COO- groups repel one another, which promotes chain relaxation and enhances free space in the network, leading to an increase in swelling. This indicates that there is an optimal concentration of biopolymer needed for the hydrogel to be able to swell properly and not become too thick and difficult to work with[17]. Beyond that optimal concentration, the hydrogel is unable to swell properly due to the increased repulsion between the negatively charged COO- molecules. This repulsion between like-charged molecules is especially strong due to the electrostatic force, and at higher concentrations, this force causes the biopolymer chains to be pushed apart, resulting in less free space in the network and consequently, a decrease in swelling[36].

A comparison of the swelling properties of formulation P-5 against the swelling properties of other hydrogel samples is shown in figure 3. The swelling evidence for the P-5 formulation was greater in pH 7.4 due to the deprotonation of carboxylic acid, which then causes the formation of COO- which repeal each other, resulting in a relaxation of the polymer network and the consequent increase in swelling[18]. While, at pH1.2 hydrogen bonds between PAM, CMTKG, and PEG exist, which decreases its swelling ability. This is because the hydrogen bonding leads to the formation of crosslinks between the polymers, which makes the network more rigid and reduces its ability to swell[17]..

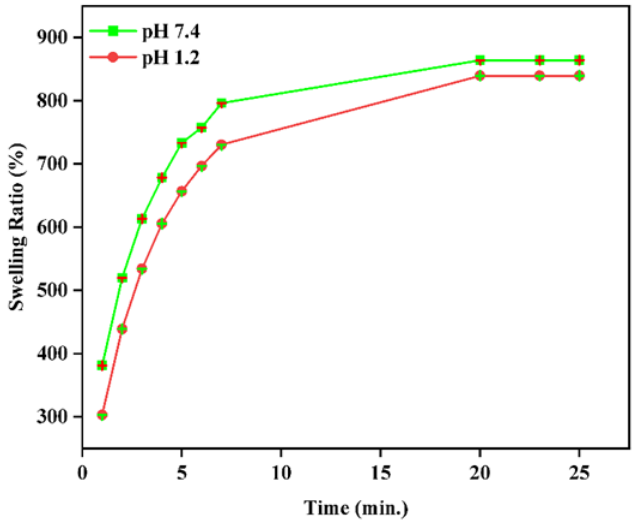


Fig 3 Swelling curve of Etophyline loaded CMTKG/PAM/PEG hydrogel (P-5) in pH 1.2 and pH 7.4

3. 2.2 Impact of Crosslinker

A decrease in the swelling ratio was observed with the increase in MBA content as shown in Figure 2. It was found that when the MBA concentration of a hydrogel is exceeded by 20 mg, the crosslinking density of the hydrogel reduces, due to reduction in the mesh size, resulting in a decrease in swelling of the hydrogel. A hydrogel with a water content of less than 20 mg tended to form jelly when MBA levels decreased below that threshold[17].

3.2.4 Impact of Initiator

As illustrated in Figure 2, the cross-linker concentration was kept at 20 mg, the KPS content was changed, and swelling results were then analyzed. When the initiator content is increased, there is an increase in swelling ratio at first, but then a decrease in swelling ratio shortly afterwards[22]. The highest swelling was detected at 32mg of the initiator's content, but the swelling decreased significantly below 32 mg. The reason for this phenomenon is that the soluble monomer is unreacted, resulting in a loose polymer network, which subsequently results in swelling declining. When the initiator content is above 32 mg, the swelling will decrease because colliding free radicals will form between monomers, causing oligomers to be generated as a result of the collision[23]. Therefore, oligomeric components became more soluble as a result of the

solubilization process and swelling of the polymeric network was greatly diminished as a result[17]

CHAPTER 4

CHARACTERIZATION

This figure illustrates the PXRD analysis of Etophylline, CMTKG/PAM/PEG hydrogel, and Etophylline-loaded CMTKG/PAM/PEG hydrogel, obtained as described earlier. It was found that Etophylline has a crystalline structure based on its PXRD curve, which showed two sharp peaks at 10.12, 12.32, 14.04, 17.32, 21.48, 24.76, and 29.4 which indicates that Etophylline has a crystalline structure[12]. Using a PXRD curve of the CMTKG/PAM/PEG hydrogel, we observed a broad band, which indicates that the hydrogel is an amorphous one. On the other hand, in a PXRD curve corresponding to Etophylline-loaded CMTKG/PAM/PEG hydrogel, the etophylline is not detected anywhere in the crosslinked polymer network, which indicates that the etophylline has completely distributed itself throughout the crosslinked polymer network[3].

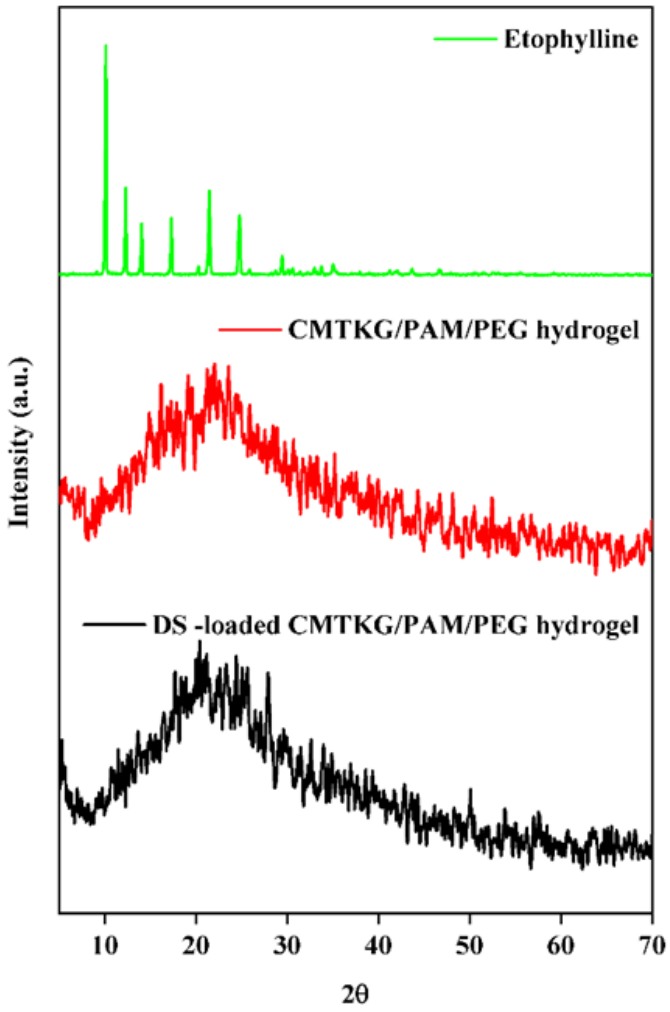


Fig 4 PXRD pattern of Etophylline, CMTKG/PAM/PEG hydrogel (P-14), andEtophylline-loaded CMTKG/PAM/PEG hydrogel (P-5)

In this figure, we are illustrating the spectra of the ATR-FTIR spectra of the developed CMTKG/PAM/PEG hydrogel, the hydrogel compound that was processed with etophylline, and the hydrogel compound that was prepared with MBA. 5. The band at 3324cm^{-1} and 3181cm^{-1} in CMTKG/PAM/PEG hydrogel, and the band at 3330cm^{-1} and 3183cm^{-1}, respectively, has been assigned to the overlapping stretch of the -OH and -NH molecules in CMTKG/PAM/PEG hydrogel loaded with Etophylline. As far as COO-symmetric stretching is concerned, the peaks are located at 1318 and 1324cm^{-1} for the CMTKG/PAM/PEG hydrogel and the Etophylline-loaded CMTKG/PAM/PEG hydrogel, respectively, while asymmetric stretching is located at 1598 and 1600 cm^{-1} for these materials. CMTKG/PAM/PEG hydrogel and Etophylline-loaded

CMTKG/PAM/PEG hydrogel exhibit symmetric C-H stretch peaks at 2938cm^{-1} and 2932cm^{-1}, respectively, indicating C-H stretch peaks of CMTKG/PAM/PEG hydrogel and Etophylline-loaded CMTKG/PAM/PEG hydrogel, respectively. In contrast, the COO- asymmetric vibration bands correspond to peaks at 1567cm^{-1} and 1569cm^{-1}, respectively . Moreover, as a result of the C-O-C group peak attribute being observed on both the CMTKG/PAM/PEG hydrogel, as well as the Etophylline-loaded CMTKG/PAM/PEG hydrogel, it has been found that 1106cm^{-1} is positive for both groups[18] .

As shown in the ATR-FTIR spectrum of MBA, a peak attribute to C-N occurred at 1311cm^{-1}, which shifted to 1318cm^{-1} and 1324cm^{-1} in the CMTKG/PAM/PEG hydrogel and Etophylline-loaded CMTKG/PAM/PEG hydrogel, respectively, indicating a decrease in conjugation as a result of the cross-linking. This shift in frequency is due to the increase in conjugation as the hydrogel is cross-linked. The peak attribute to C-N in the ATR-FTIR spectrum of MBA indicates the presence of an amide bond, which is broken as the hydrogel is cross-linked, leading to a decrease in conjugation and a shift in the peak frequency[19].

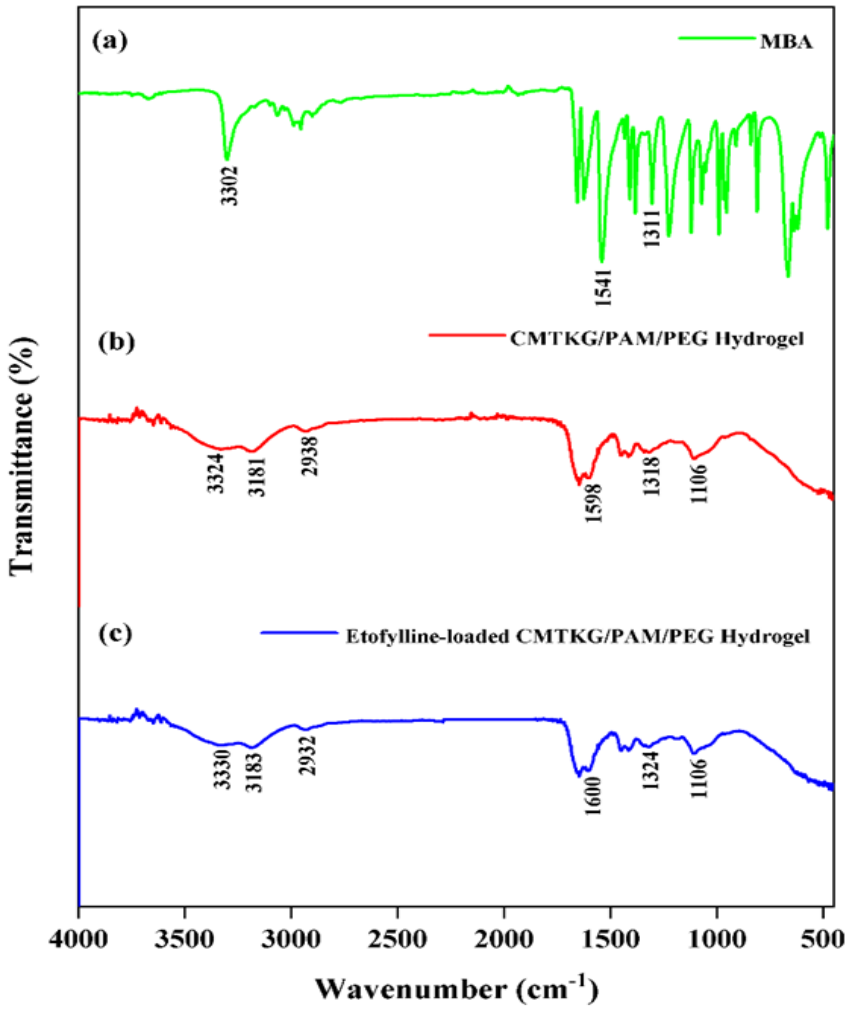


Fig 5 ATR-FTIR spectrum of CMTKG/PAM/PEG hydrogel, Etophylline-loaded CMTKG/PAM/PEG and MBA

It was as a result of this that cross-linked CMTKG/PAM/PEG hydrogels were developed. A CMTKG/PAM/PEG hydrogel containing etophylline showed no additional peak as compared to a hydrogel without etophylline, suggesting only a physical or weak interaction between the drug and the hydrogel (hydrogen bonding), this means that the drug is not chemically bound to the hydrogel, and is likely to be released in an environment-dependent manner. The weak interaction between the drug and the hydrogel also means that the drug can be released from the hydrogel in a controlled manner, allowing for greater control over the rate of drug release.

This figure illustrates the four stages of decomposition that are followed during the thermal degradation of CMTKG/PAM/PEG hydrogel and Etophylline-loaded CMTKG/PAM/PEG

hydrogel as shown in Fig.6. Since the first stage is between 30-260°C, weight loss ranging between 14.9% and 19.1% indicates that water is vaporizing in CMTKG/PAM/PEG hydrogel and Etophylline-loaded hydrogel respectively. The four stages of decomposition are indicative of the chemical degradation that occurs in the hydrogel when exposed to elevated temperatures[35]. The vaporization of water in the first stage is due to the hydrolysis of the polymers and the removal of the associated water molecules[33].

In the second stage of hydrogel formation, the carboxymethyl and hydroxyl groups in the polymeric backbone are lost, with weight loss of 13.25% and 13.02%, respectively, between 260 and 334°C for CMTKG/PAM/PEG hydrogels and Etophylline-loaded CMTKG/PAM/PEG hydrogels, indicating the loss of these groups in the polymeric backbone. This is due to the thermal decomposition of the polymeric backbone, which is brought about by the breaking of the carbon-oxygen bonds that hold the carboxymethyl and hydroxyl groups in place. This results in a decrease in the overall weight of the hydrogel. In the third step of the process, there was a weight loss of 37.04% and 38.35% in CMTKG/PAM/PEG as well as the Etophylline-loaded CMTKG/PAM/PEG hydrogel after 334°C and 455°C, respectively, since the crosslinking present in CMTKG/PAM/PEG has disappeared. At 455-750°C, the weight loss was observed to be about 26.46 % and 23.84 %, respectively, due to the complete degradation of CMTKG/PAM/PEG and Etophylline-loaded CMTKG/PAM/PEG hydrogels respectively. A study was conducted to determine whether Etophylline-loaded CMTKG/PAM/PEG hydrogel exhibited comparable thermal stability to unloaded CMTKG/PAM/PEG hydrogel, indicating that physical interaction between Etophylline and the polymer network was the only factor responsible for thermal stability[13].

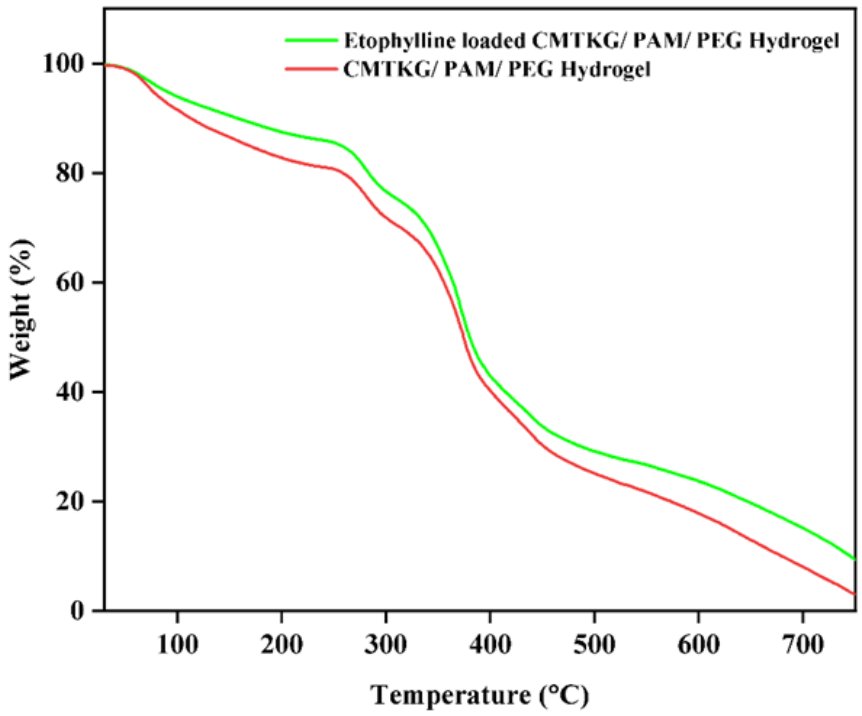


Fig 6 TGA curve of the CMTKG/PAM/PEG, Etophylline- loaded CMTKG/PAM/PEG hydrogel (P-5)

SEM micrographs of the hydrogels comprised of CMTKG/PAM/PEG and Etophylline-loaded CMTKG/PAM/PEG are shown in the following Fig. 7. It is important to note that the surface of the CMTKG/PAM/PEG hydrogel is very rough and porous, which enables it to effectively load the Etophylline molecule into a matrix. SEM micrographs of Etophylline-loaded CMTKG/PAM/PEG hydrogel, on the other hand, show a more smooth and less porous surface as compared to the loaded hydrogel, presumably because the introduced drug occupies the porous

hydrogel matrix, rather than filling it with molecules[20].

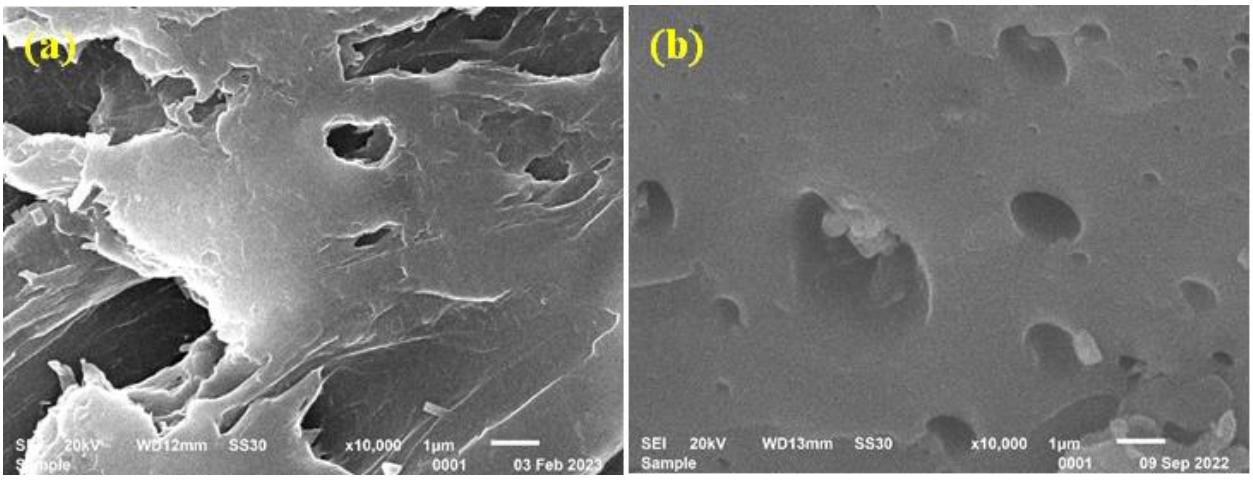


Fig 7-SEM images of CMTKG/PAM/PEG hydrogel and Etophylline-loaded CMTKG/PAM/PEG hydrogel

4.1 Loading of Drug and Encapsulation Efficiency

A CMTKG/PAM/PEG hydrogel (P-5) containing Etophylline was used to determine the drug encapsulation efficiency and loading percentage. According to our analysis, the drug encapsulation efficiency (%) is observed to be 62.96%, while the drug loading efficiency (%) is 16.23%. This suggests that the hydrogel was able to effectively encapsulate the drug, and that there is a good balance between the total amount of drug encapsulated and the amount of drug that is actually released [32].

4.2 *In-vitro* release studies of Etophylline

A 24-hour release study of Etophylline-loaded CMTKG/PAM/PEG hydrogel (P-5) at 37°C was carried out *in-vitro* in pH 1.2 and pH 7.4 buffer solutions . The results of this study are shown in Fig 8. The amount of drug released was higher at pH 7.4 as compared to pH 1.2 due to the deprotonated COO-ions that were present there. There is a strong repelling tendency between deprotonated COO-groups at pH 7.4, which leads to an expansion of the polymeric chain and, as a result, swelling increases, which leads to increased drug release as well[28]. It is thought that this carboxylated ion protonates at pH 1.2, which leads to the formation of a hydrogen bond between CMTKG, PEG, and PAM. Consequently, a shrinking polymeric network causes another

increase in swelling and drug release due to a reduction in the polymeric network. Moreover, the study indicates that there is a pH-dependent channel for releasing Etophylline from CMTKG/PAM/PEG hydrogels that contain Etophylline as a drug. The protonation of the carboxylated ion at a pH of 1.2 causes a hydrogen bond to form between CMTKG, PEG, and PAM. This bond weakens the polymeric network, causing it to shrink and resulting in an increase in swelling as well as drug release[27]. The pH-dependent channel provides a controlled release of Etophylline from the hydrogels, allowing for a more efficient delivery of the drug[6].

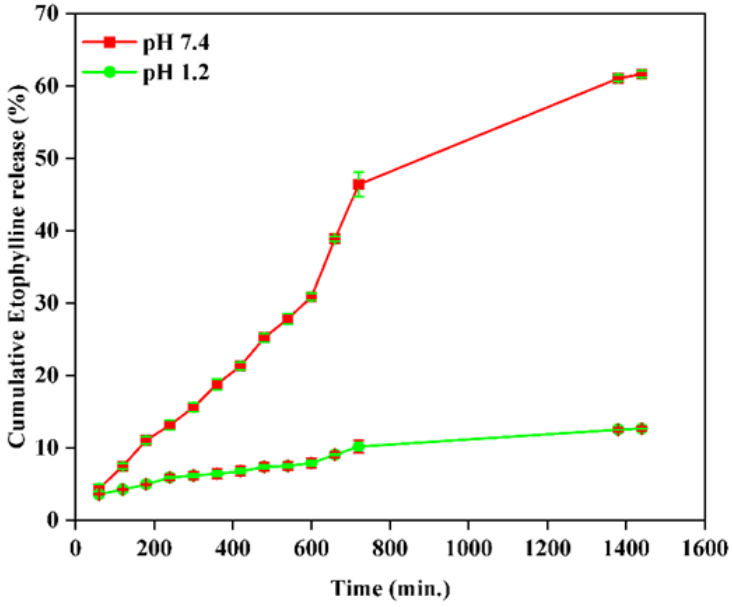


Fig 8 Etophylline release profile from CMTKG/PAM/PEG hydrogel (P-5) in pH 1.2 and pH 7.4

4.3 Kinetic modeling of Etophylline

The release kinetics of Etophylline loaded CMTKG/PAM/PEG (P-5) hydrogel have been investigated using a number of different kinetic models, such as Zero-order, Hixson-Crowell, First-order, Higuchi, and Korsmeyer-Peppas as shown in Table 2. According to the results of this study, the Korsmeyer-Peppa model of release data fits very well to the release data at both pH. A value of R^2 of 0.9966 with an n value of 0.8440 was observed for pH 7.4, indicating that hydrogels followed non-Fickian diffusion, which implies that drug diffusion, as well as polymeric chain relaxation, both occurred, as reflected by Figure 9. At pH 1.2, however, we found that the R^2 value

was 0.9857, and the n value was 0.3487, which suggests that hydrogels follow the Fickian diffusion principle, which means that only drug diffusion is observed in the hydrogels[13]

.

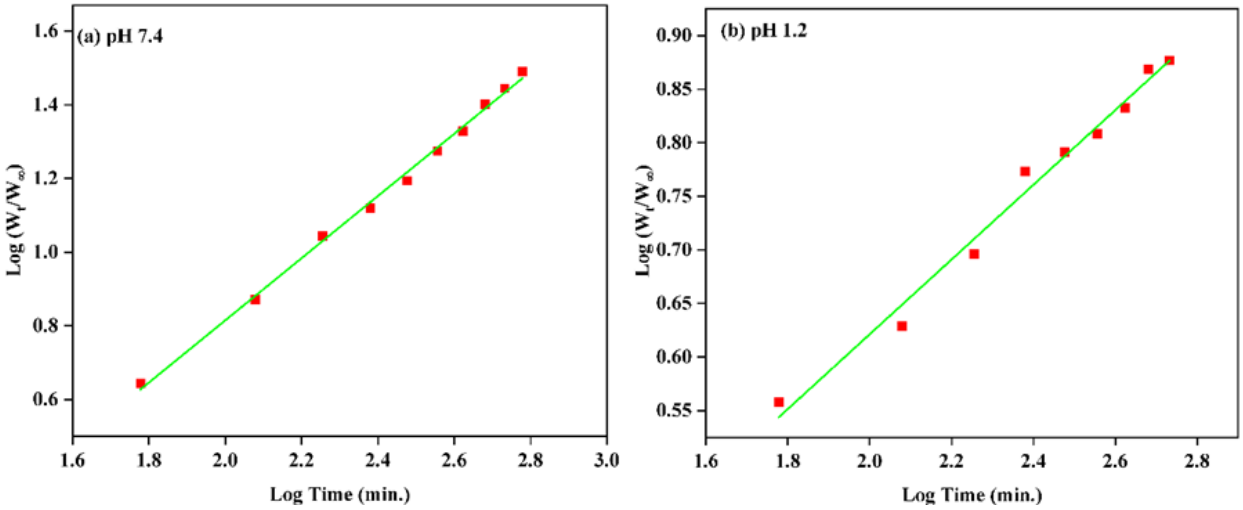


Fig 9 Kinetic release modelling plot of Etophylline as per Korsmeyer-Pepppas model in pH 1.2 and pH 7.4

Table 2 Kinetic modeling data of Etophylline employing various models

Model	Mechanism	Equation	pH 7.4		pH 1.2		Ref.
			n	R^2	n	R^2	
Zero Order	Rate is independent of drug concentration	$W_t = W_\infty + W_0 t$	-	0.9906	-	0.9532	[29]
Higuchi	Fickian diffusion followed	$F = W_t/W_\infty = k_H t^{½}$ k_H = kinetic constant	-	0.9612	-	0.9780	[20]
First Order	The amount of the drug and its release rate is directly proportional.	$LogW_t = LogW_\infty + \frac{kt}{2 \cdot 303}$ k = rate constant	-	0.7792	-	0.8615	[30]
Hixson-Crowell	Erosion	$(W_0)^{1/3} - (W_t)^{1/3} = k_{HC}.t$ k_{HC} = Hixson Crowell constant	-	0.8951	-	0.8641	[30]
Korsmeyer –Peppas	1.Fickian Diffusion ($n < 0.5$) 2. Non-Fickian Diffusion ($0.89 \geq n \geq 0.5$) 3. Case II transport ($n > 0.89$)	$F = W_t/W_\infty = kt^n$ k = kinetic constant n = diffusion exponent	0.8440	0.9966	0.3487	0.9857	[30]

4.4 Conclusion

In this study, we synthesized a pH-responsive CMTKG/PAM/PEG hydrogel containing Etophylline[34]. MBA (crosslinking agent) and KPS (initiator)[31]. ATR-FTIR, PXRD, SEM, and TGA have been used to characterize the hydrogel formula, as well as ways in which it can be improved[29]. Based on the kinetic modeling of the Etophylline released data, the Korsmeyer-Peppas model fit well in both pH and followed the fickian diffusion mechanism in pH 1.2 as well as the non-fickian diffusion mechanism in pH 7.4 and followed the non-fickian diffusion mechanism[30]. The Korsmeyer-Peppas model is a mathematical expression which describes the phenomena of drug release from a drug delivery system[26]. The model takes into account various parameters, such as the amount of drug released, the diffusion coefficient, and the porosity of the drug delivery system. By using the Korsmeyer-Peppas model to analyze the Etophylline released data, it was found that the synthesized CMTKG/PAM/PEG hydrogel could be used to effectively control the release of Etophylline in the colon and thus achieve the desired results.

REFERENCE

1. Rafique N; Ahmad M; Minhas M et al, Designing gelatin-based swellable hydrogels system for controlled delivery of salbutamol sulphate: characterization and toxicity evaluation. Polym Bull**2022**,794535–4561. https://doi.org/10.1007/s00289-021-03629-6.

2. Rizwan M; Yahya R; Hassan A; et al, pH sensitive hydrogels in drug delivery: Brief history, properties, swelling, and release mechanism, material selection and applications. Polymers**2017**, 9. https://doi.org/10.3390/polym9040137.

3. Suhail M Kha; Rosenholm JM; et al, Fabrication and characterization of diclofenac sodium loaded hydrogels of sodium alginate as sustained release carrier. Gels**2021**, 7, 1–16. https://doi.org/10.3390/gels701001.0

4. Kulkarni R V; Sa B, Evaluation of pH-sensitivity and drug release characteristics of (Polyacrylamide-Grafted-Xanthan)-carboxymethyl cellulose-based pH-sensitive interpenetrating network hydrogel beads. Drug Dev Ind Pharm **2008**, 34, 1406–1414. https://doi.org/10.1080/03639040802130079.

5. Raza MA; Park SH, Irradiated Ch/GG/PVP-based stimuli-responsive hydrogels for controlled drug release. J Appl Polym Sci **2020**, 137, 49041.

https://doi.org/10.1002/app.49041.

6. Wei QB; Fu F; Zhang YQ, et al, PH-responsive CMC/PAM/PVP semi-IPN hydrogels for theophylline drug release. J Polym Res**2014**, 21. https://doi.org/10.1007/s10965-014-0453-0
7. D'souza AA; Shegokar R, Polyethylene glycol (PEG): a versatile polymer for pharmaceutical applications. Expert Opin Drug Deliv **2016**, 13, 1257–1275. https://doi.org/10.1080/17425247.2016.1182485.
8. Mandal BB; Kapoor S; Kundu SC, Silk fibroin/polyacrylamide semi-interpenetrating network hydrogels for controlled drug release. Biomaterials**2009**, 30, 2826–2836. https://doi.org/10.1016/j.biomaterials.2009.01.040
9. Peppas NA; Keys KB; Torres-lugo M; Lowman AM, Poly (ethylene glycol) -containing hydrogels in drug delivery**1999**, 62, 81–87..
10. Khushbu; Warkar SG, Potential applications and various aspects of polyfunctional macromolecule- carboxymethyl tamarind kernel gum. Eur Polym J**2020**, 140, 110042. https://doi.org/10.1016/j.eurpolymj.2020.110042.
11. Mali K; Dhawale S; Dias R; Ghorpade V, Delivery of drugs using tamarind gum and modified tamarind gum: A review. Bull Fac Pharmacy Cairo Univ**2019**, 57, 1–24. https://doi.org/10.21608/BFPC.2019.47260.
12. Pardeshi C V; Agnihotri V V; Patil KY; et al, Mannose-anchored N,N,N-trimethyl chitosan nanoparticles for pulmonary administration of etofylline. Int J Biol Macromol**2020**, 165, 445–459. https://doi.org/10.1016/j.ijbiomac.2020.09.163
13. Khushbu; Warkar SG; Thombare N, Controlled release and release kinetics studies of boron through the functional formulation of carboxymethyl tamarind kernel gum-based superabsorbent hydrogel. Polym Bull**2022**, 79, 2287–2303. https://doi.org/10.1007/s00289-021-03634-9.

14. Niu B; Jia J; Wang H; et al, In vitro and in vivo release of diclofenac sodium-loaded sodium alginate/carboxymethyl chitosan-ZnO hydrogel beads. Int J Biol Macromol**2019**, 141, 1191–1198. https://doi.org/10.1016/j.ijbiomac.2019.09.059.

15. Jana S; Sharma R; Maiti S; Sen KK, Interpenetrating hydrogels of O-carboxymethyl Tamarind gum and alginate for monitoring delivery of acyclovir. Int J Biol Macromol, **2016**, 92, 1034–1039. https://doi.org/10.1016/j.ijbiomac.2016.08.017.

16. Varahala Setti ML; Ratna JV; Preparation and evaluation of controlled release tablets of carvedilol. Asian J Pharm **2009**, 3, 252–256. https://doi.org/10.4103/0973-8398.56307.

17. Khushbu; Warkar SG; Kumar A, Synthesis and assessment of carboxymethyl tamarind kernel gum based novel superabsorbent hydrogels for agricultural applications. Polymer **2019**, 182,121823. https://doi.org/10.1016/j.polymer.2019.121823.

18. Mali KK; Dhawale SC; Dias RJ, Synthesis and characterization of hydrogel films of carboxymethyl tamarind gum using citric acid. Int J Biol Macromol**2017**, 105, 463–470. https://doi.org/10.1016/j.ijbiomac.2017.07.058.

19. Roy; Brahma C; Nandi; Parida K, Formulation and design of sustained release matrix tablets of metformin hydrochloride: Influence of hypromellose and polyacrylate polymers. Int J Appl Basic Med Res **2013**, 3, 55. https://doi.org/10.4103/2229-516x.112242

20. . Shoukat; Pervaiz F; Noreen S; et al, Fabrication and evaluation studies of novel polyvinylpyrrolidone and 2-acrylamido-2-methylpropane sulphonic acid-based crosslinked matrices for controlled release of acyclovir. Polym Bull**2020**, 77, 1869–1891. https://doi.org/10.1007/s00289-019-02837-5.

21. Malik R; Warkar SG; Saxena R, Carboxy-methyl tamarind kernel gum based bio-hydrogel for sustainable agronomy. Mater Today Commun **2023**, 35, 105473.

https://doi.org/10.1016/j.mtcomm.2023.105473

22. Goonoo N; Bhaw-Luximon A; Ujoodha R; et al, Naltrexone: A review of existing sustained drug delivery systems and emerging nano-based systems. J Control Release**2014**, 183, 154–166. https://doi.org/10.1016/j.jconrel.2014.03.04.6

23. Das D; Das R; Mandal J; et al, Dextrin crosslinked with poly(lactic acid): A novel hydrogel for controlled drug release application. J Appl Polym Sci**2014**, 13, 1–12. https://doi.org/10.1002/app.40039.
24. Thirupathi; Phan TTV; Santhamoorthy M; et al, pH and Thermoresponsive PNIPAm-co-Polyacrylamide Hydrogel for Dual Stimuli-Responsive Controlled Drug Delivery. Polymers **2022**, 15, 167. https://doi.org/10.3390/polym1501016.7.
25. Amtul Jamil Sami*; Madeeha Khalid; Sitara Nasar HAM; YNB, Sustained Release of Ibuprofen by a Novel Formulated Hydrogel Containing Molecular Pharmaceutics & Organic Process Research Sustained Release of Ibuprofen by a Novel Formulated Hydrogel Containing Graphene Oxide. Mol Pharm Org Process Res**2018**, 6, 1000143. https://doi.org/10.4172/2329-9053.100014.3.
26. Mathematical models of drug release. (2015). *Strategies to Modify the Drug Release from Pharmaceutical Systems*, *63–86.* doi:10.1016/b978-0-08-100092-2.00005-9
27. Li J, Mooney DJ. Designing hydrogels for controlled drug delivery. Nat Rev Mater. 2016 Dec;1(12):16071. doi: 10.1038/natrevmats.2016.71. Epub 2016 Oct 18. PMID: 29657852; PMCID: PMC5898614.
28. Rizwan M, Yahya R, Hassan A, Yar M, Azzahari AD, Selvanathan V, Sonsudin F, Abouloula CN. pH Sensitive Hydrogels in Drug Delivery: Brief History, Properties, Swelling, and Release Mechanism, Material Selection and Applications. Polymers (Basel). 2017 Apr 12;9(4):137. doi: 10.3390/polym9040137. Erratum in: Polymers (Basel). 2017 Jun 14;9(6): PMID: 30970818; PMCID: PMC6432076.
29. Croitoru, Catalin & Pop, Mihai Alin & Bedo, Tibor & Cosnita, Mihaela & Roata, Ionut & Hulka, Iosif. (2020). Physically Crosslinked Poly (Vinyl Alcohol)/Kappa- Carrageenan Hydrogels: Structure and Applications. Polymers. 2020. 560. 10.3390/polym12030560.
30. Lisik A, Musiał W. Conductomeric Evaluation of the Release Kinetics of Active

Substances from Pharmaceutical Preparations Containing Iron Ions. Materials (Basel). 2019 Mar 3;12(5):730. doi: 10.3390/ma12050730. PMID: 30832401; PMCID: PMC6427111.

31. Singhal, Reena & Tomar, Rajiv & Nagpal, A.. (2009). Effect of cross-linker and initiator concentration on the swelling behaviour and network parameters of superabsorbent hydrogels based on acrylamide and acrylic acid. International Journal of Plastics Technology. 13. 22-37. 10.1007/s12588-009-0004-4.
32. Wu YJ, Tsai TW, Huang SJ, Mou Y, Lin CJ, Chan JC. Hydrogen bond formation between citrate and phosphate ions in spherulites of fluorapatite. Langmuir. 2013 Sep 17;29(37):11681-6. doi: 10.1021/la402392b. Epub 2013 Sep 5. PMID: 24007413.
33. Enrica Caló, Vitaliy V. Khutoryanskiy, Biomedical applications of hydrogels: A review of patents and commercial products,European Polymer Journal,Volume 65, 2015, Pages 252-267, ISSN 0014-3057, https://doi.org/10.1016/j.eurpolymj.2014.11.024.
34. Ghosh G, Barman R, Sarkar J, Ghosh S. pH-Responsive Biocompatible Supramolecular Peptide Hydrogel. J Phys Chem B. 2019 Jul 11;123(27):5909-5915. doi: 10.1021/acs.jpcb.9b02999. Epub 2019 Jun 27. PMID: 31246033.
35. Szymańska E, Winnicka K. Stability of chitosan-a challenge for pharmaceutical and biomedical applications. Mar Drugs. 2015 Apr 1;13(4):1819-46. doi: 10.3390/md13041819. PMID: 25837983; PMCID: PMC4413189.
36. Bashir S, Hina M, Iqbal J, Rajpar AH, Mujtaba MA, Alghamdi NA, Wageh S, Ramesh K, Ramesh S. Fundamental Concepts of Hydrogels: Synthesis, Properties, and Their Applications. Polymers (Basel). 2020 Nov 16;12(11):2702. doi: 10.3390/polym12112702. PMID: 33207715; PMCID: PMC7697203.