# Grafting of Poly [(2-Diethylamino)Ethyl Methacrylate] onto Chitosan

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# ABSTRACT

In this study, chitosan was grafted with poly[(2-diethyl amino)ethyl methacrylate] in homogeneous and heterogeneous systems by using potassium persulphate as the initiator. The effect of temperature, time, (2-diethyl amino)ethyl methacrylate concentration and the concentration of the initiator on the grafting yield was examined in aqueous acetic solution under nitrogen atmosphere. The maximum grafting percentage of poly[(2-diethyl amino)ethyl methacrylate] on to chitosan was found to be 180% under homogeneous conditions using 0.5 mL of monomer concentration with 1.00 g chitosan dissolved in 1.0mL of 0.10% (w/v) acetic acid solution, temperature at 70°C and 4 hour reaction time. The products were precipitated in ethanol in the powder form. They were found to be water soluble which is an improvement over the dissolution properties of the parent molecule, namely chitosan. Chitosan tripolyphosphate gel beads which were chemically crosslinked by glutaraldehyde or ethylene glycol digylcidyl ether were used as solid substrates for grafting of poly[(2-diethyl amino)ethyl methacrylate] PDEAEM in a heterogeneous system at the optimum conditions determined for the homogeneous system. The maximum grafting yield was found to be in the range 40-50%. The swelling kinetics of the grafted beads was followed in acidic, neutral and basic buffer solutions. The experiments demonstrated 166-4811% swelling within 72 hours. The synthesized copolymers were characterized by using Fourier Transform Infrared spectroscopy (FTIR), thermo gravimetric analysis (TGA), and scanning electron microscopy (SEM).

**Keywords**: graft copolymerization, natural polymer, chitosan, chitosan tripolyphosphate bead, (2-diethyl amino)ethyl methacrylate (DEAEM), water soluble polymer.

# ÖZ

Bu çalışmada kitosanın potasyum per sülfat başlatıcı kullanarak homojen ve heterojen ortamlarda poli[(2-dietil amino)]etil metakrilat ile aşılanması incelenmiştir. Derişimi 1.0% (w/v) olan sulu asetik asit çözeltisi içinde ve azot ortamında sıcaklık, zaman, monomer ve başlatıcı konsantrasyonlarının aşılanma yüzdesi üzerindeki etkisi çalışılmıştır. Homojen ortamda 1.00 g kitosan ve 0.5 mL (2-dietil amino)etil metakrilat örneğinin 1mL çözelti içinde 70°C sıcaklıkta 4 saat sonunda %180 aşılanma oranı elde edilmiştir. Poli[(2-dietil amino)etil metakrilat] aşılanmış kitosan etanol çöktürücü ile toz halde çözeltiden ayrılmıştır. Ürünlerin saf suda tamamen çözündükleri ve dolasıyla kitosanla karşılaştırıldığında önemli bir avantaja sahip oldukları gözlemlenmiştir. Sistem, homojen ortamda belirlenen en iyi koşullar sağlanarak heterojen ortamda da test edilmiştir. Glutareldehit veya etilen glikol diglisidil eter kullanılarak kimyasal çapraz bağlanmaya uğratılmış kitosan tripolifosfat jel boncukların, poli[(2-dietil amino)]etil metakrilat ile aşılanmasıyla %40-50 arasında değişen aşılanma oranlarına ulaşılmıştır. Aşılanmış jel boncukların şişme kinetiği de izlenmiş ve asit, nötral ve baz tampon çözeltilerde 72 saat içinde 166-4811% şişme kapasitesine sahip oldukları bulunmuştur.Ürünler FTIR spektroskopisi, termal gravimetric analiz (TGA) ve tarayıcı electron mikroskopisi SEM yöntemleri ile karakterize edilmişlerdir.

**Anahtar kelimeler:** aşı kopolimerizasyonu, doğal polimerler, suda çözünen polimerler (2-dietil amino)etil metakrilat, DEAEM, kitosani kitosan tripolifosfat.

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# LIST OF SYMBOLS ABBREVIATIONS

| | |
|---|---|
| DEAEM | (2-Diethylamino)ethyl Methacrylate |
| Chi-TPP | Chitosan Tripolyphosphate |
| KPS | Potassium Persulfate |
| FT-IR | Fourier Transform Infrared |
| Rpm | Rounds per Minute |
| SEM | Scanning Electron Microscopy |
| TGA | Thermograviemtric Analysis |
| TPP | Tripolyphosphate |
| Glu | Glutaraldehyde |
| EGDE | Ethylene Glycol Diglycidyl Ether |

# Chapter 1

# INTRODUCTION

In this study, chitosan was grafted with poly[(2-(diethylamino)ethyl methacrylate]. This copolymer is a pH responsive system which may find applications in drug delivery, gene delivery (Saranya, 2011), metal recovery (Ricardo, 2003) as well as an antimicrobial agent (Ramya, 2012).

The grafting system consisted of an aqueous solution containing the monomer, (2-diethylamino)ethyl methacrylate, the initiator potassium persulfate and the substrate, chitosan. The system was optimized with respect to the reaction time, the amount of initiator, the amount of the monomer and the temperature by calculating the grafting yield.

A similar approach was taken for surface modification of chitosan tripolyphosphate (Chi-TPP) gel beads. Chi-TPP beads were prepared by coagulating chitosan in acetic acid solution in aqueous solution of penta sodium tripolyphosphate (Hennink, 2002). Then the beads were grafted by poly[(2-diethylamino)ethyl methacrylate] by redox initiation. These beads have the potential to serve as adsorbents for heavy metals or dyes in water treatment, or as drug carriers in drug delivery application.

The products were characterized gravimetrically to find out the grafting yield. The products were analyzed by FTIR spectroscopy and TGA analysis. They were also

tested to determine the swelling and solubility properties. The bead surface before and after grafting was characterized by SEM analysis.

## 1.1 Polysaccharides

Polysaccharides are polymers with a high molecular weight made up of monosaccharides as the repeat units joined together with glycosidic bonds. Polysaccharides exist in nature and have the ability to act as energy reservoirs. A large number of polysaccharides are non-toxic and benign to mammalian tissues. As a result of these properties they receive growing interest scientifically and commercially on the ways to fully utilize them for different applications. Since polysaccharides are obtained from renewable (plant and animal origin), there is a rising interest in them as non-petroleum based polymer feed stock. The sources biodegradable nature of these polymers is another asset. Examples include structural polysaccharides like chitin and cellulose and storage polysaccharides like glycogen and starch.

### 1.1.1 Chitosan

Chitosan is a polyaminosaccharide which is synthesized by the deacetylation of chitin with the help of a strong alkali usually sodium hydroxide. It can be found naturally in the outer shells of insects, cell walls of bacteria and shells of crustaceans (Majeti, 2000).

Nowadays, chitosan has received a lot of interest as compared to other polysaccharides in applied chemistry, because of the presence of the amino group (Figure1-1) as an important functional group. Amino group allows easy modification to realize the required properties. It provides special biological functions and solubility in aqueous acidic medium. The molecular weight of chitosan usually

ranges from $5 \times 10^4$ Da to $2 \times 10^6$ Da and the deacetylation degree from (40% - 98%) . Molecular weight, particle size, viscosity, density and degree of deacetylation are important features of chitosan which affect its physical behaviour and applications (Majeti, 2000).

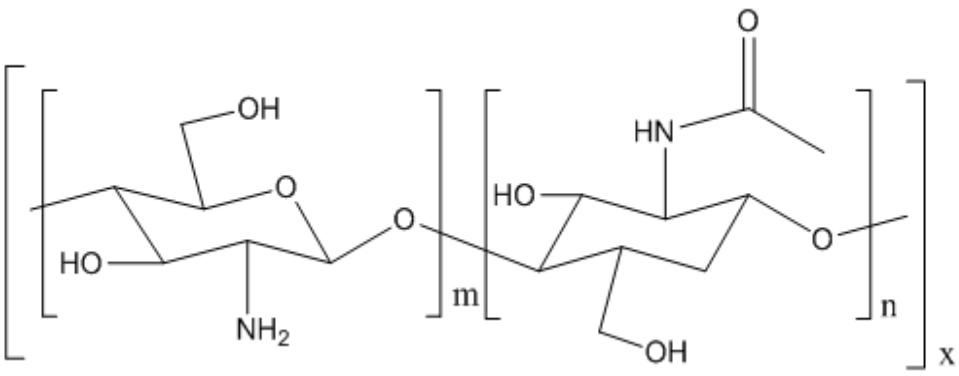
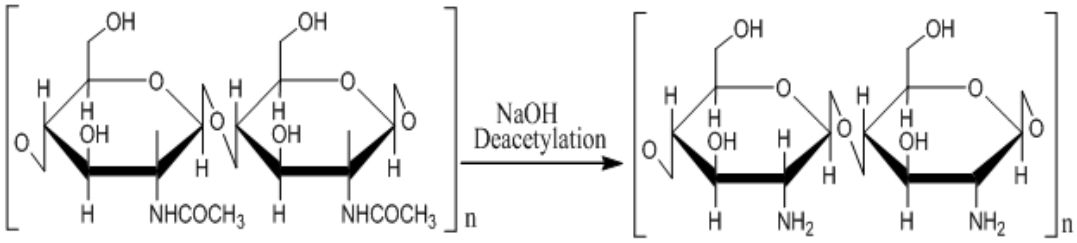



Figure 1-1: Chitosan Structure

Chitosan can be produced by removing the acetyl group from chitin. This involves a rough treatment with a concentrated aqueous solution of NaOH with care taken to prevent the reaction mixture from coming in contact with oxygen. This is done by the use of a nitrogen purge or adding sodium borohydride to guard against any unwanted reactions, for instance, depolymerization and production of reactive radicals (Mourya, 2008). The process is shown in Scheme 1.




Scheme 1. Producing Chitosan from Complete Deacetylation of Chitin

#### 1.1.1.1 Properties and Applications of Chitosan

Chitosan has amino and hydroxyl groups as the functional groups on its structure giving it a broad area of applications which include biomedical applications, in cosmetics, food and nutrition, paper and textile and also water treatment (Skjak, 1985). Chitosan is insoluble in organic solvents and water; however it is soluble in acidic solvents such as acetic, oxalic, hydrochloric and lactic acid. It forms a polycation i.e. positively charged polymer with a high charge density in solution. The solubility of chitosan in acidic solvents is as a result of the protonation of the amino groups present in the chain structure. Therefore it displays a pH responsive behaviour because of the amino groups on its structure. It bears useful biological properties, like biocompatibility, biodegradability, and antimicrobial activity. Chitosan, when degraded, produces non-carcinogenic and non-toxic by products (Majeti, 2000).

A large number of chitosan and its derivatives have been synthesized only with the aim of adsorption of metals. Presence of amino and hydroxyl groups make chitosan structure more effective for removing heavy metals and dyes (Wan, 2011). In acidic medium amino groups of chitosan have the ability to be protonated, therefore, they adsorb anionic dyes robustly via electrostatic attraction. Crosslinkers such as glutaraldehyde, ethylene glycon diglycidyl ether, epichlorohydrin, have been used to improve chitosans derivative performance as an adsorbent. Crosslinkers stabilize

chitosan in acidic medium and improve its mechanical properties (Wan, 2011). Another common application of chitosan derivative is in drug delivery. Chitosan and its derivatives act as a carrier for different drugs such as chlorohexidine buccal tablets in oral drug delivery or lidocaine hydrochloride in transdermal drug delivery (Tapan, 2012). Having amino groups enable it to react with negatively charged polymers and polyanions found in aqueous media. Furthermore, chitosan and its derivatives can be used in gene delivery, and this is a promising process for treating different genetic diseases and cancers (Saranya, 2011). A number of grafting studies have carried out on chitosan with the aim of producing natural/ synthetic hybrid materials for various applications as mentioned above (Majeti, 2000).

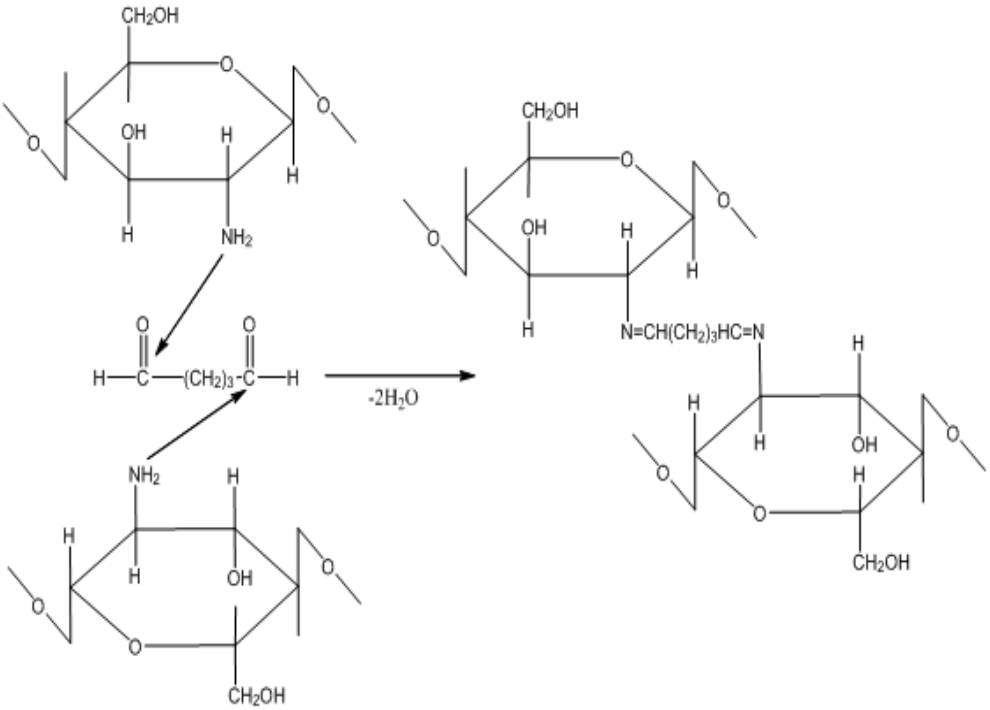
#### 1.1.1.2 Crosslinking of Chitosan

Some of the disadvantages of chitosan powder (e.g., poor resistance to heat, insufficient mechanical properties, and solubility in acidic media) limits its utilization as an insoluble absorbent. Crosslinking is one method that can be used to overcome these disadvantages of chitosan powder. The crosslinked raw polymer can be shaped into beads, membranes or microparticles with more useful properties. After crosslinking, the solubility in aqueous acetic media is limited (Mourya, 2008). Furthermore the swelling capacity can be controlled by adjusting the degree of crosslinking. Hence, crosslinked chitosan based matrices can be applied as bioadsorbents for water treatment. They usually find applications as drug carriers in drug delivery systems. It is possible to form chemically or physically crosslinked chitosan. Difunctional reagents or multivalent anions are used for chemical or physical crosslinking respectively. Each method has its advantages and disadvantages (Hennink, 2002). In the recent years, physically crosslinking method has more interesting since the use of toxic crosslinking reagents is avoided.

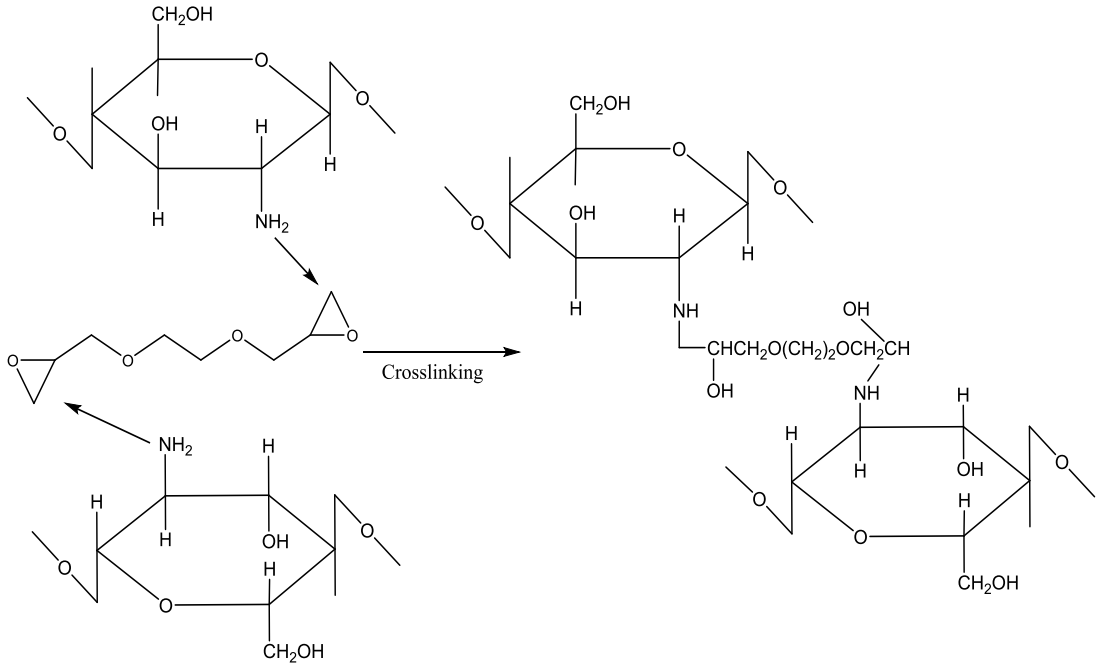
Chemically crosslinking reagents for example glutaraldehyde usually is toxic and produce toxic compounds which have to be separated or extracted from the gels before they can be applied (Hennink, 2002).

**1.1.1.2.1 Chemical Crosslinking of Chitosan Beads**

Multifunctional molecules are required for covalent crosslinking to form bridges among polymeric chains. Reagents like glutaraldehyde (GA) or ethylene glycol diglycidyl ether (EGDE) are widely used for crosslinking chitosan. Thermal crosslinking is also possible. Scheme 2 and 3 show chemical crosslinking of chitosan using GA and EGDE respectively (Kumbar, 2002).




Scheme 2. Chitosan Crosslinked with Glutaraldehyde (GA)

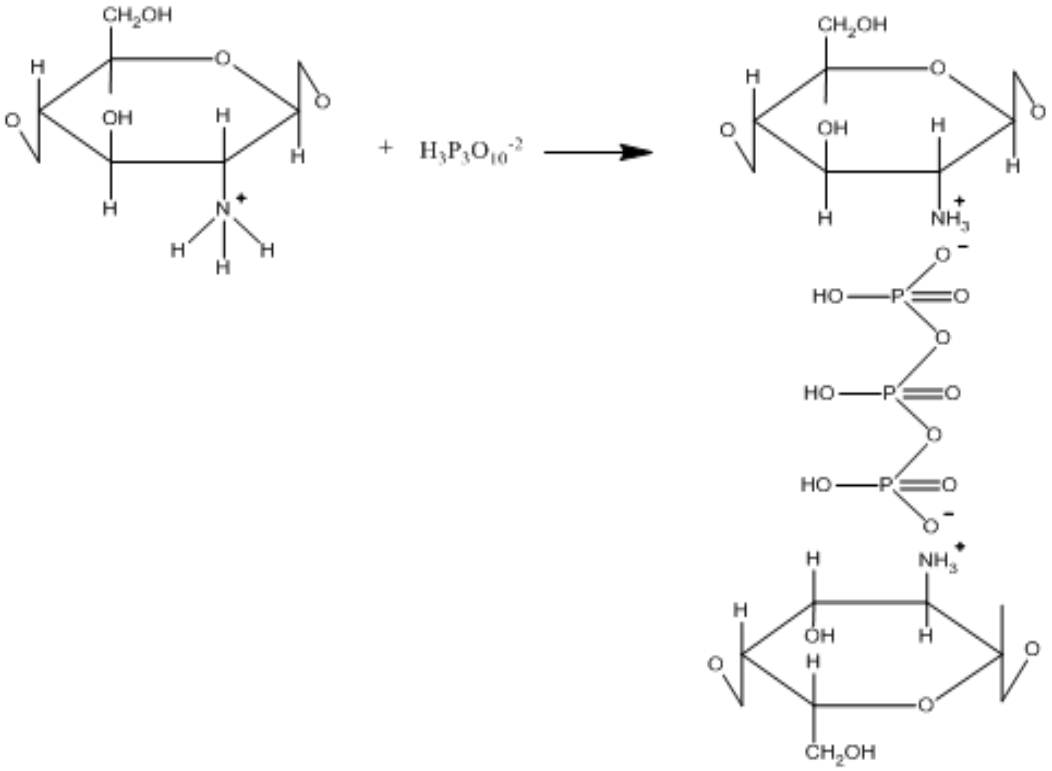



Scheme 3. Chitosan Crosslinked with Ethylene Glycol Diglycidyl Ether (EGDE)

**1.1.1.2.2 Physical Crosslinked Chitosan Beads**

A physically crosslinked network is formed due to the ionic interactions between positively charged chitosan repeat units and negatively charged polyanion (Berger, 2004). In some cases, chitosan or chitin derivatives with negative charge can be crosslinked ionically with cations, such as iron (III) (Shu, 2002).

TPP, is a polyanion and non-toxic, reacts with chitosan through electrostatic forces to create ionic crosslinked networks. Chitosan beads and microsphere can be prepared using tripolyphosphate due to fast gelling capability (Sung, 2001). A small amount of crosslinker leads to faster crosslinking reaction because it spreads easily. Chitosan ionic crosslinking with TPP solution of PH= 8.6 is shown in Scheme 4. (Sung, 2001).



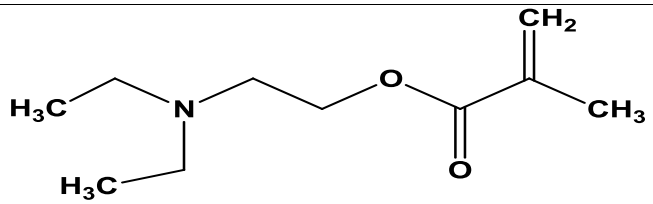

Scheme 4. Chitosan Ionic Cross Linking in Aqueous TPP Solution

Citrate, sulfate with three and negative charges are two other useful polyanion for chitosan crosslinking. The crosslinking method for sulfate and citrate was observed to be faster than that of tripolyphosphate; however once totally crosslinked, chitosan-TPP beads possess excellent mechanical strength because of strong interaction between chitosan and TPP (Shu, 2002).

## 1.2 (2-Diethylamino)Ethyl Methacrylate (DEAEM)

In this research, graft copolymerization of (2-diethylamino)ethyl methacrylate (DEAEM) on to chitosan was carried out in acidic medium by using redox initiation method. New grafted chitosan powder and grafted chitosan-TPP beads were obtained. The chemical and physical properties of the monomer are listed in Table 1-1(Sigma). DEAEM is an interesting monomer to study, since this compound and its analogue bearing moiety have been shown to be useful in the synthesis of functional polymers aimed for use in biomedical applications. Some very recent examples carried out with DEAEM are as follows. Molecularly imprinted polymers of DEAEM are selective for recognition of borine serum albumin (BSA). These systems were shown to recognize BSA 1,6 -2.5 times better than control samples (David, 2012). Photo responsive and pH responsive star copolymers containing DEAEM were synthesized in another study, for biological diagnosis, cell imaging and detection (Ying, 2012). Dendritic copolymers of DEAEM with methyl methacrylate and PEG were prepared by ATRP method. The copolymer films demonstrated antimicrobial activity (Giovanni, 2012). PDEAEM based gene delivery materials; gene transfection is the transfer of genetic materials like DNA into cells. Cationic polymers which form nanocomplexes with DNA, so called non- viral gene vectors, are a highly promising platform for efficient gene transfection (Agrawal, 2012).

Table 1-1: Chemical and Physical Properties of (2-Diethylamino) Ethyl Methacrylate

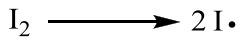
| 2-(Diethylamino) ethyl methacrylate | |
|---|---|
| Chemical structure |  |
| Chemical formula | $C_{10}H_{19}NO_2$ |
| Acute toxicity | LD50 Oral – rat-4.696 mg/kg |
| Boiling point | 239.327 ˚C at 760 mmHg |
| Relative density | 0.922 g/cm3 at 25 ˚C |
| Flash point | 77 ˚C closed cup |
| Molecular weight | 185.26 g/mole |

## 1.3 Graft Copolymerization

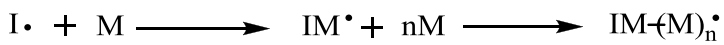
Graft copolymerization is a well- known method for modifying the physical and chemical characteristics of polymers. Graft copolymerization can be initiated by various means including chemical treatment, photo-irradiation, high energy radiation technique and enzymatic degradation techniques (Bhattacharya, 2004).

Radicals are created on the polymer backbone by the action of the initiator on the substrate polymer. These radicals further initiate polymerization of the monomer from the active site on the substrate polymer. Several different grafting mechanisms have been proposed including ring opening via oxidation and grafting from amine nitrogen and /or oxygen on C-3 and /or C-6 (Abduel, 2000). Homopolymerization of the monomer is also initiated. A general mechanism can be written as given bellow (Bhattacharya, 2004).

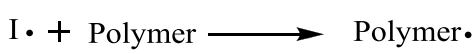
Dissociation of the initiator

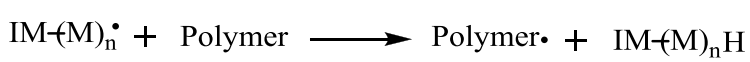


Homopolymerization of the monomer:

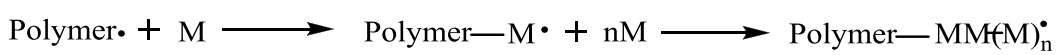


Creation of active sites on the polymer by H- abstraction:

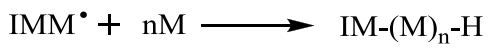


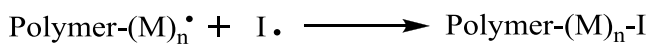


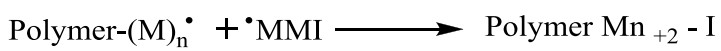
Graft copolymerization:



Termination:







Grafting yield is affected by the concentration of the monomer, the initiator and the concentration or the amount of substrate. Time and temperature are two other reaction conditions that influence the grafting yield (Hatice, 2005). Hence grafting systems need to be optimized with respect to these factors.

Several grafting studies onto chitosan have been reported in the literature (Hamit, 2007). Vinyl monomers have been copolymerized onto chitosan under homogenous or heterogeneous conditions to produce hybrid materials with improved properties. Some examples to grafting work carried out in our laboratory are synthesis, characterization and antibacterial activity of poly(N-vinylimidazole) (Hamit, 2007), preparation and characterization of maleic acid grafted Chitosan (Hatice, 2005),

synthesis, characterization and biocompatibility studies on chitosan-graft-poly (EGDMA) (Adali, 2009).

# Chapter 2

# EXPERIMENTAL

## 2.1 Materials

All materials used are listed in Table 2-1. They were all used as received.

Table 2-1: The Chemicals and Their Manufacturers

| No | Chemicals | Company |
|---|---|---|
| 1 | Acetic Acid | Riedel-deHäen-Germany |
| 2 | Acetone | Kemiteks Kimyevi Maddeler Tic.Ltd.Sti.-Turkey |
| 3 | Ammonia | Aldrich-Germany |
| 4 | Hydrochloric acid | AnalaR-UK |
| 5 | Sodium hydrogen carbonate | AnalaR-UK |
| 6 | Chitosan (medium molecular weight) | Aldrich-Germany |
| 7 | 2-(Diethylamino)ethyl methacrylate | Aldrich-Germany |
| 8 | Sodium tripolyphosphate pentabasic | Aldrich-Germany |
| 9 | EGDE (ethylene glycol diglycidyl ether) | Aldrich-Germany |
| 10 | Glutaraldehyde | Aldrich-Germany |
| 11 | Potassium chloride | Aldrich-Germany |
| 12 | Sodium hydroxide | Aldrich-Germany |
| 13 | Iron (III) chloride | Aldrich-Germany |
| 14 | Potassium persulfate | Aldrich-Germany |

## 2.2 Methods

### 2.2.1 Preparation of Chitosan-graft-Poly[(2-Diethylamino)Ethyl Methacrylate] Powders

Chitosan was grafted with poly[(2-diethylamino)ethyl methacrylate], PDEAEM, in the presence of potassium persulfate, KPS, initiator. A 25 mL sample of chitosan solution of concentration 1% (w/v) prepared in 1% (v/v) acetic acid solution was placed in a two-neck reaction vessel. A required amount of KPS, and monomer (2-diethylamino)ethyl methacrylate, DEAEM, were then added into chitosan solution respectively under nitrogen atmosphere and at constant temperature (60℃, 70℃,80℃). The reaction was carried out for a various period of time (1h-12h) under vigorous magnetic stirring at 1200 rpm. Then, it was precipitated in acetone and was dried at 50 ℃ overnight. Preparation conditions of all samples are given in detail in Table 2-2.

The powder formation process has been illustrated in Figure 2-1.

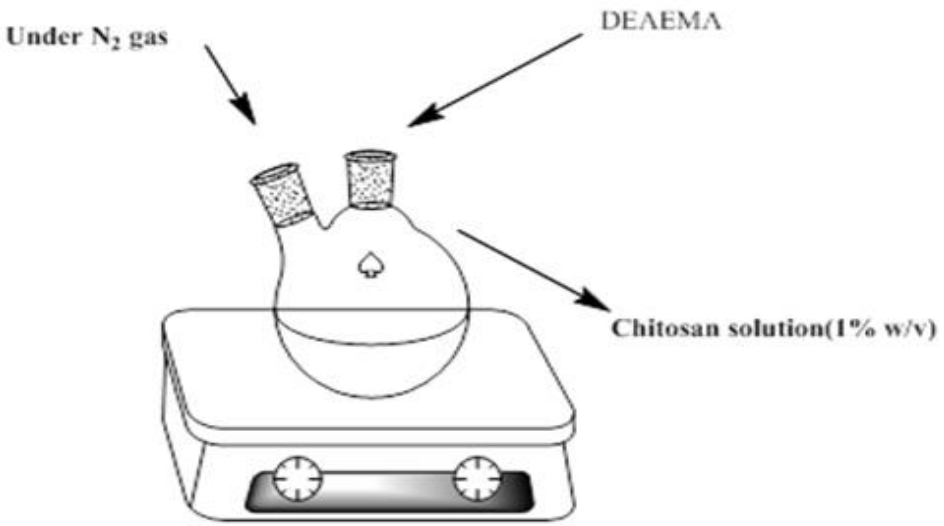



Figure 2-1: The Preparation of Chitosan-graft-Poly[(2-Diethylamino)Ethyl Methacrylate] powders

Table 2-2: Preparation Conditions of All Powders.

| Sample ID | DEAEM (mL) | T (°C) | Time (hr) | KPS (g) |
|---|---|---|---|---|
| P1 | 0.50 | 60 | 1h | 0.1250 |
| P2 | 0.50 | 60 | 2h | 0.1250 |
| P3 | 0.50 | 60 | 3h | 0.1250 |
| P4 | 0.50 | 60 | 4h | 0.1250 |
| P5 | 0.50 | 60 | 6h | 0.1250 |
| P6 | 0.50 | 60 | 9h | 0.1250 |
| P7 | 0.50 | 60 | 12h | 0.1250 |
| P8 | 0.50 | 70 | 1h | 0.1250 |
| P9 | 0.50 | 70 | 2h | 0.1250 |
| P10 | 0.50 | 70 | 3h | 0.1250 |
| P11 | 0.50 | 70 | 4h | 0.1250 |
| P12 | 0.50 | 70 | 6h | 0.1250 |
| P13 | 0.50 | 70 | 9h | 0.1250 |
| P14 | 0.50 | 70 | 12h | 0.1250 |
| P15 | 0.50 | 80 | 1h | 0.1250 |
| P16 | 0.50 | 80 | 2h | 0.1250 |
| P17 | 0.50 | 80 | 3h | 0.1250 |
| P18 | 0.50 | 80 | 4h | 0.1250 |
| P19 | 0.50 | 80 | 6h | 0.1250 |
| P20 | 0.50 | 80 | 9h | 0.1250 |
| P21 | 0.50 | 80 | 12h | 0.1250 |
| P22 | 0.25 | 70 | 4h | 0.1250 |
| P23 | 0.75 | 70 | 4h | 0.1250 |
| P24 | 1.00 | 70 | 4h | 0.1250 |
| P25 | 0.5 | 70 | 4h | 0.0600 |
| P25 | 0.5 | 70 | 4h | 0.1800 |
| P26 | 0.5 | 70 | 4h | 0.2400 |

### 2.2.2 Preparation of Chitosan Tripolyphosphate (Chi-TPP) Beads, Crosslinked Chitosan Tripolyphosphate (Chi-TPP) Beads and Chitosan-TPP-graft-Poly[(2-Diethylamino)Ethyl Methacrylate]

A chitosan solution of concentration 2% (w/v) was prepared in 1% (v/v) acetic acid. The solution was added dropwise into 5% (w/v) pentasodium tripolyphosphate (TPP) solution prepared in distilled water. The pH of this solution was measured with a pH-meter to be 8.6. Chi-TPP beads formed instantaneously upon coagulation at room temperature under magnetic stirring of 20 rpm. They were dried in the oven at 50 °C overnight. Crosslinked beads were prepared by adding EGDE or GA into the TPP solution. Preparation conditions of Chi-TPP beads are given in Table 2-3.

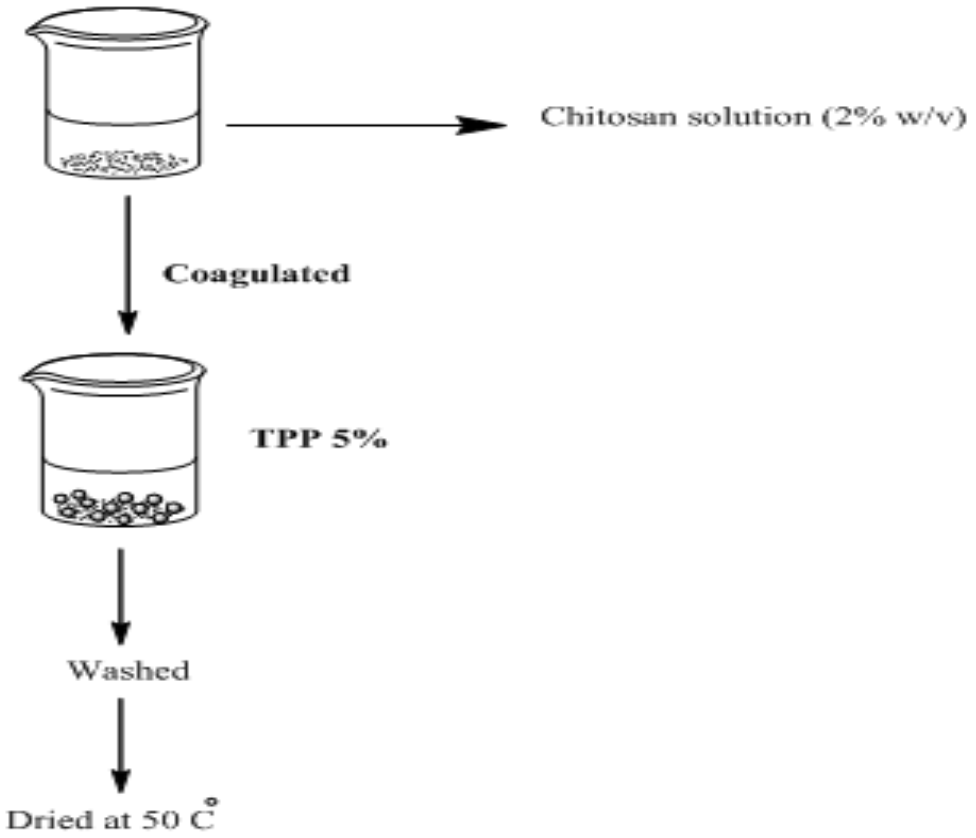
Table 2-3: Preparation Conditions of Chi-TPP Beads and Chi- TPP Beads Crosslinked with GA and EGDE in Distilled Water.

| Sample ID | GLU conc. | EGDE conc. | Temperature (C°) | Drying Temp. over night |
|---|---|---|---|---|
| b1 | --------- | ---------- | 25 | 50C° |
| b2 | 0.1 | | 25 | 50 C° |
| b3 | 0.3 | | 25 | 50 C° |
| b4 | 0.5 | | 25 | 50 C° |
| b5 | 1.0 | | 25 | 50 C° |
| b6 | | 2.5 | 25 | 50 C° |
| b7 | | 8 | 25 | 50 C° |
| b8 | | 16 | 25 | 50 C° |

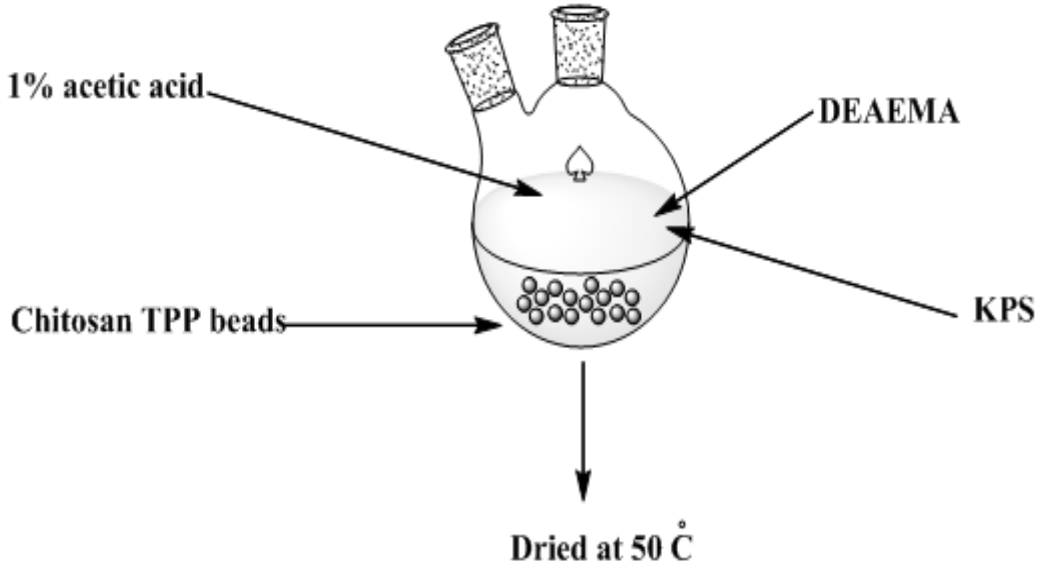
b refers to chi-TPP beads in distilled water.

Grafting of PDEAEM onto Chi-TPP Beads was carried out as follows. A sample of Chi-TPP beads weighing 0.25 g was placed and 25 mL water. The monomer, 0.50 mL, mixed with 1.0 mL ethanol was added into the flask containing 0.25g Chi-TPP beads and 0.1250 g KPS initiator in 25 mL acetic acid and Grafting was carried under nitrogen atmosphere. Preparation condition for the grafted beads is given in Table 2-4.

The formation process for the preparation of chitosan tripolyphosphate (Chi-TPP) Beads and chitosan tripolyphosphate (Chi-TPP)-graft-poly[(2-diethylamino)ethyl methacrylate] has been illustrated in Scheme 5 (a) and 5 (b) respectively.




Scheme 5. (a) Preparation of Chitosan Tripolyphosphate (Chi-TPP) Beads




Scheme 5. (b). Preparation of Chitosan Tripolyphosphate (Chi-TPP)-graft-Poly[(2-Diethylamino)Ethyl Methacrylate] at 70 °C and for 4 Hours

Table 2-4: Preparation Conditions for the Grafted Chi-TPP Beads Crosslinked with GA and EGDE with PDEAEM in Acetic Acid

| Sample ID | GLU conc. | EGDE conc. | T ( C°) | KPS (g) | DEAEM conc, | Time (hr) |
|---|---|---|---|---|---|---|
| B1 | ------ | -------- | 70 | 0.125 | 0.5mL | 4 |
| B2 | 0.1 | | 70 | 0.125 | 0.5mL | 4 |
| B3 | 0.3 | | 70 | 0.125 | 0.5mL | 4 |
| B4 | 0.5 | | 70 | 0.125 | 0.5mL | 4 |
| B5 | 1.0 | | 70 | 0.125 | 0.5mL | 4 |
| B6 | | 2.5 | 70 | 0.125 | 0.5mL | 4 |
| B7 | | 8 | 70 | 0.125 | 0.5mL | 4 |
| B8 | | 16 | 70 | 0.125 | 0.5mL | 4 |

B refers to chi-TPP grafted with poly[(2-diethyl amino)ethyl methacrylate].

## 2.3 Characterizations

### 2.3.1 FTIR Analysis

The FTIR spectra of synthesized samples were recorded on a Perkin Elmer Spectrum-65 FTIR spectrometer, using KBr pellets of the samples.

### 2.3.2 Gravimetric Analysis

% grafting yield was calculated by the following equation.

$$Grafting\ (\%) = \frac{m\ grafted\ chitosan - m\ chitosan}{m\ chitosan} x\ 100 \quad \ldots\ldots (1)$$

### 2.3.3 Dissolution Properties of Products

The swelling kinetics of the beads were studied in aqueous buffer solutions with pH values of 1.2, 7 and 11 respectively. Buffer solutions used in these experiments were prepared using potassium chloride and hydrochloric acid as shown in Table 2-5. The swelling % was calculated as follows:

$$Swelling\ (\%) = \frac{\text{m swollen} - \text{m dried}}{\text{m dried}}\ x100 \quad \ldots\ldots (2)$$

Table 2-5 : Preparation Conditions of Buffer Solutions.

| PH | Components | Total Volume |
|---|---|---|
| 1 | 25mL of 0.2M KCl+ 67.5mL of 0.2M HCl | 100mL |
| 1.2 | 25mL of 0.2m KCl+ 42.5mL of 0.2M HCl | 100mL |
| 7 | 0.681g of potassium dihydrogen phosphate + 29.1mL of 0.10M NaOH | 100mL |
| 11 | 0.210g of sodium bicarbonate and 22.7 mL of 0.10M NaOH | 100mL |

### 2.3.4 SEM Analysis

Morphological properties of products were analyzed by SEM at TUBITAK-MAM.

### 2.3.5 TGA Analysis

Thermogravimetric properties of products were analyzed by TGA at TUBITAK-MAM.

### 2.3.6 Bead Size

Bead size was measured by using sieves of mesh size 710µm, 500µm, 212µm.

# Chapter3

# RESULTS AND DISCUSSION

Chitosan-graft-poly[(2-diethylamino)ethyl methacrylate], was prepared under homogeneous conditions using potassium persulphate (KPS) as redox initiator. The grafting reaction was carried out in solution and the product was precipitated from solution as powder. The effect of monomer concentration, temperature, and initiator amount on the extent of grafting (% G) was investigated by the gravimetric method. Chi-TPP beads were also prepared and grafted with (2-diethylamino)ethyl methacrylate and the sample were characterized by SEM, FTIR and TGA analysis.

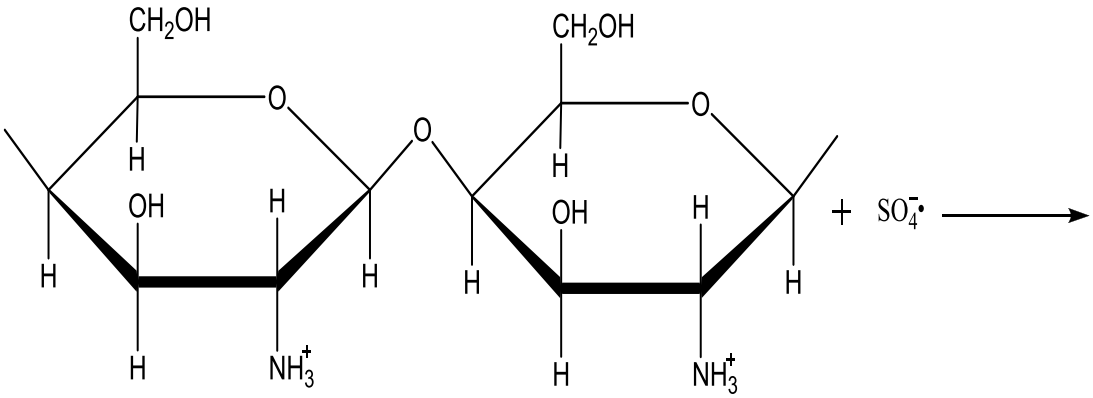
## 3.1 Preparation of Chitosan-graft-Poly[(2-Diethylamino)Ethyl Methacrylate] in Solution

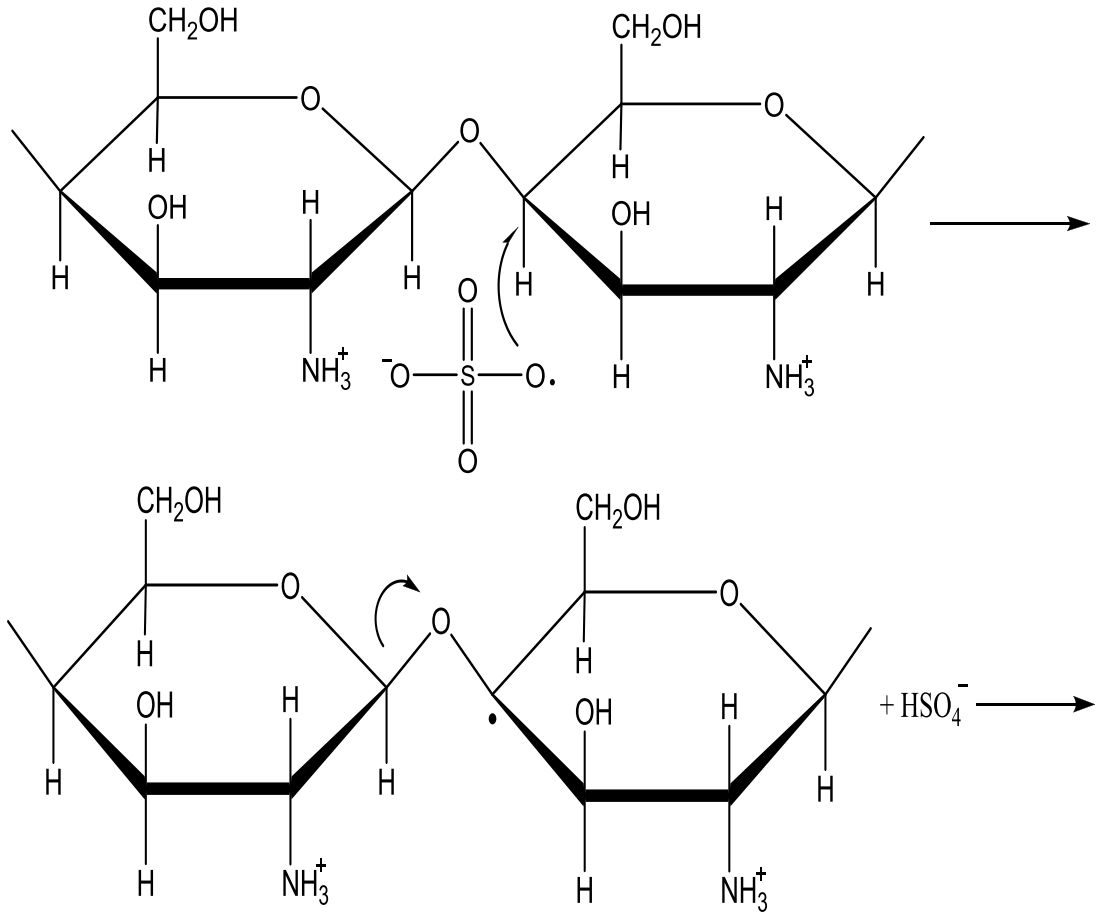
Redox initiating systems are suitable for initiating grafting of vinyl monomers onto polysaccharides as these agents can be applied in aqueous media, at ambient temperatures. Even though redox initiators have been shown to be successful in grafting of synthetic polymers onto polysaccharides in numerous systems, they do have their drawbacks as well. Two very frequently used redox initiators for grafting vinyl polymers onto polysaccharides are cerium ammonium nitrate (CAN) and potassium persulfate (KPS). One disadvantage of CAN is that it may form insoluble complexes that remain as impurities in the system (Abduel, 2000). Another disadvantage of these initiators is that during redox initiation, degradation of the polymer also occurs. One proposed mechanism by Shih-Chang Hsu (Shih, 2002) is shown in Scheme 6.

Another possible mechanism shown in Scheme 7 which proposed ring opening via formation of aldehyde and/or imine groups (Abduel, 2000). Then grafting of the monomer and propagation of polymerization should occur either form C-2 or C-3 (Scheme 7). It is equally probable that a radical forms on the O-atom on C-6 and propagation occurs via addition of the monomer to the chitosan backbone from O-6. Grafting onto Chi-TPP beads are shown in Scheme 8 and 9 using a similar approach to that explained for Scheme 7 and 8 above. It should be noted that FTIR analysis only is not sufficient to prove the chemical structure proposed. Further detailed analysis could be needed to support the mechanisms proposed.

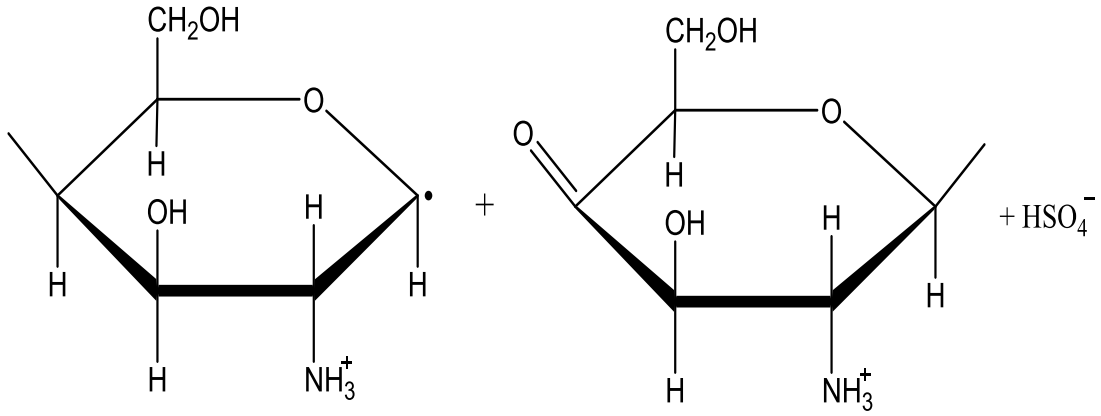
1- Thermal dissociation of KPS.

2- Persulfate ion is attracted to the cationic amino group and free radical is transferred to the C-4 carbon.

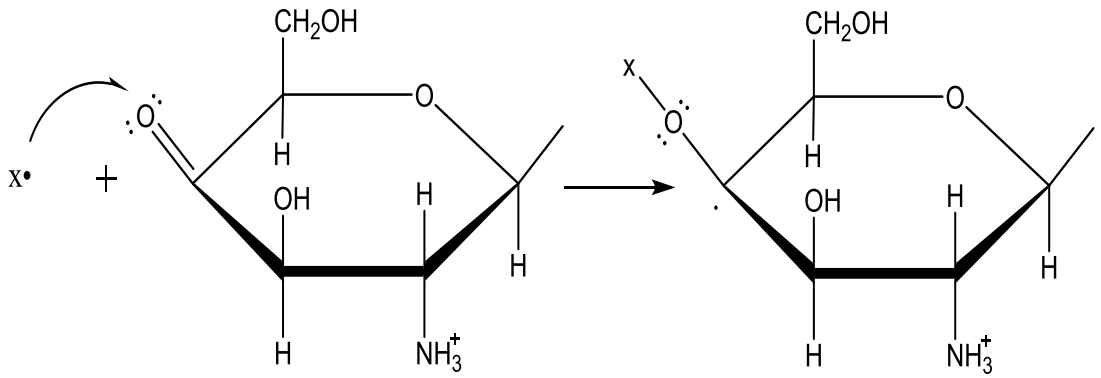




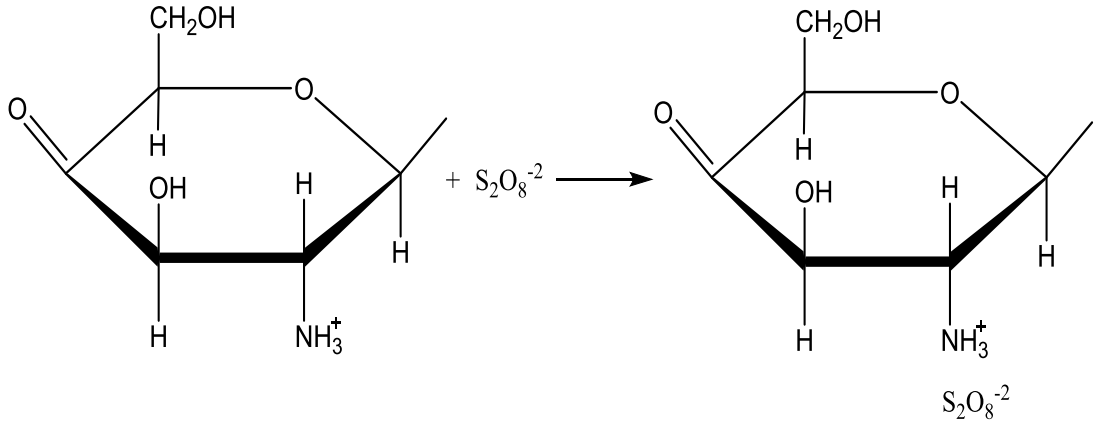
3- Chain scission at the C-O-C bond.



4- Inhibition of free radical by degraded chitosan chain with carbonyl group at the end

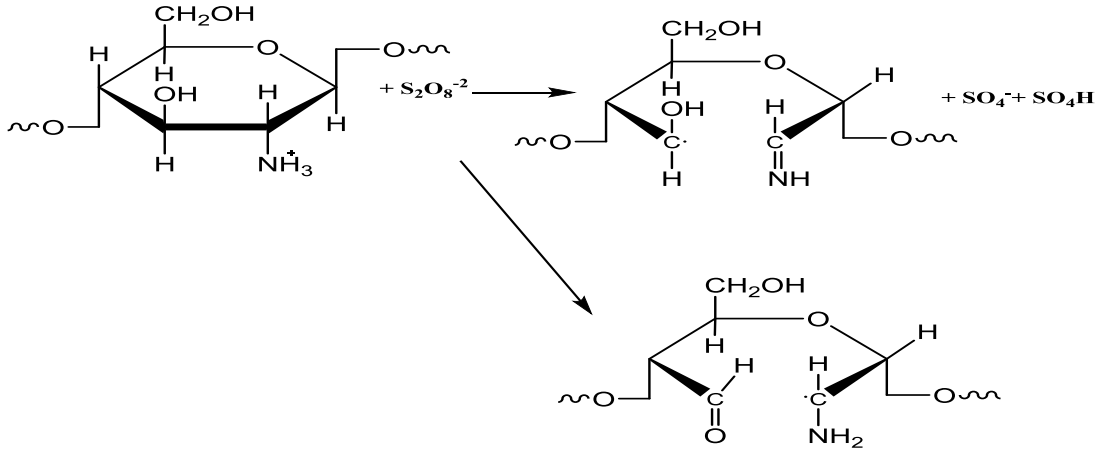



5- Deactivation of persulfate ions

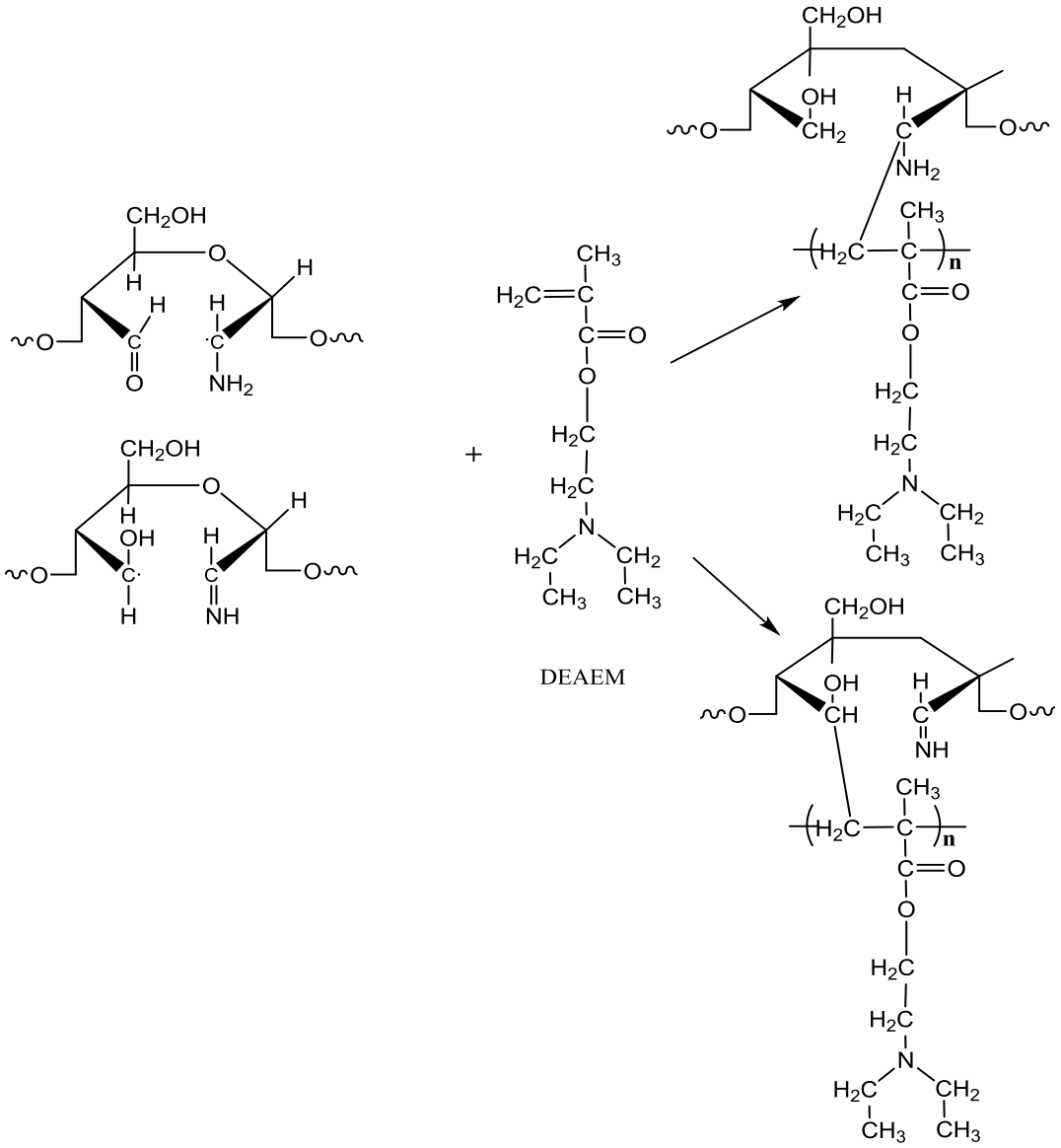



Scheme 6. Suggested Degradation Mechanism of Chitosan by Potassium Persulfate Free Radical

**Initiation**

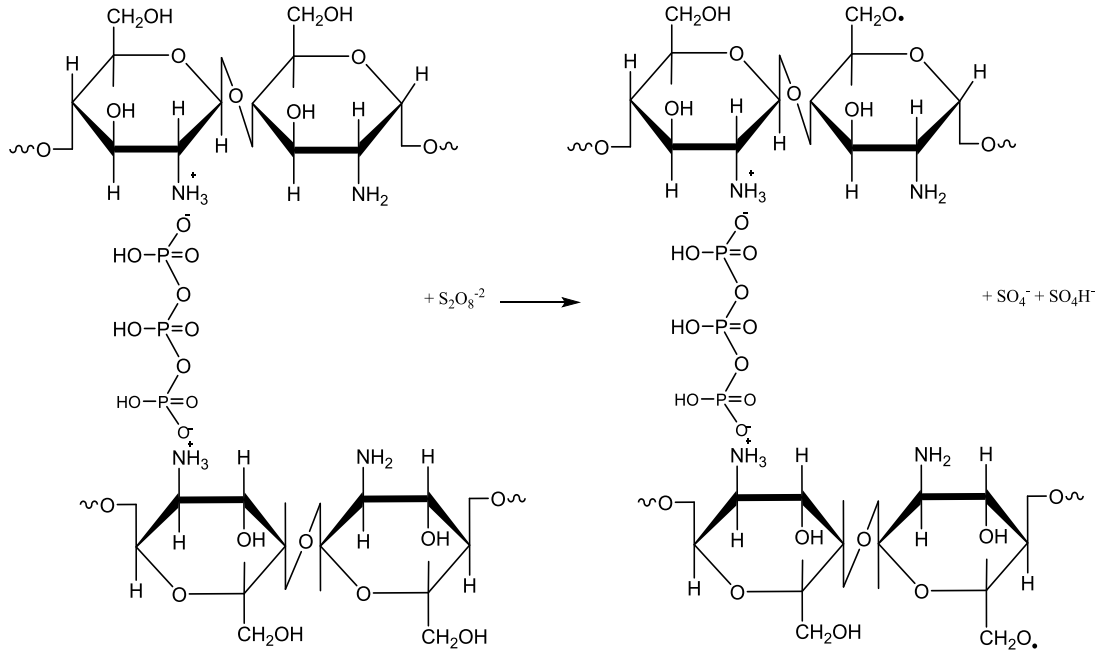



**Propagation**

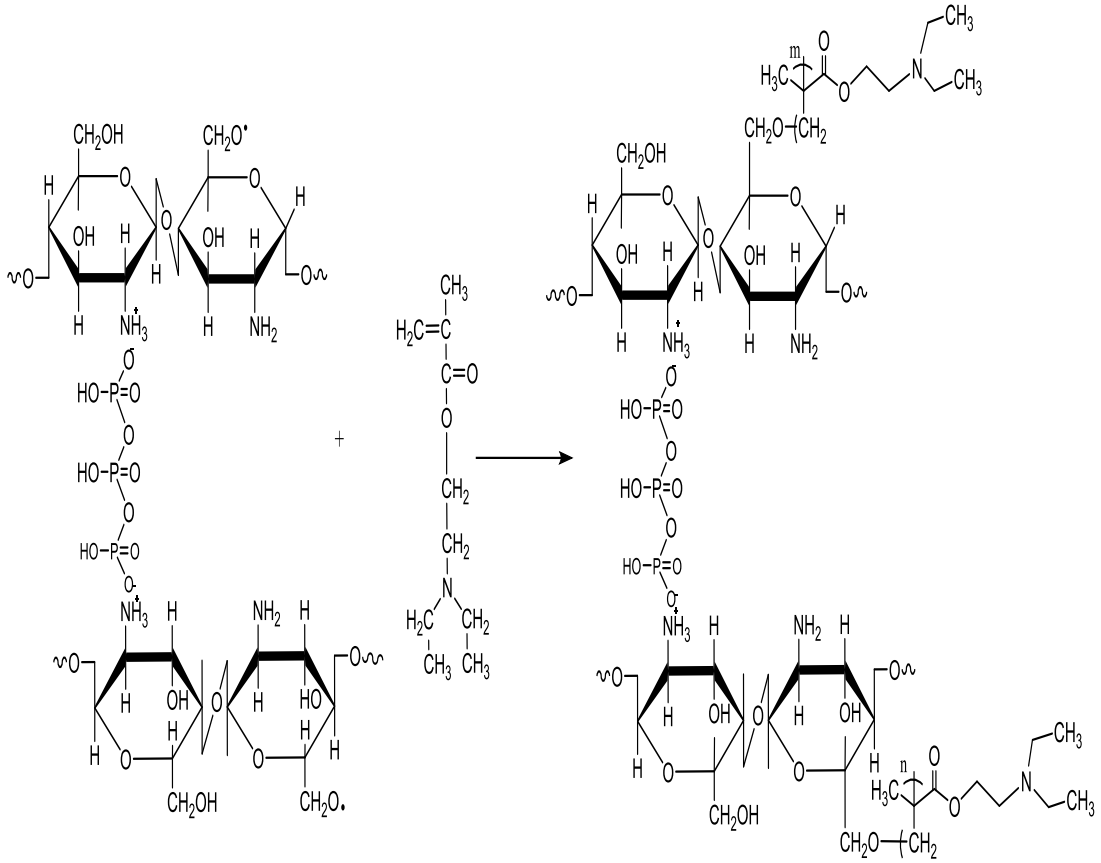



Scheme7. Proposed Structure Mechanism for Grafting Chitosan Powder with (DEAEM) on C-2 &C-3

**Initiation**

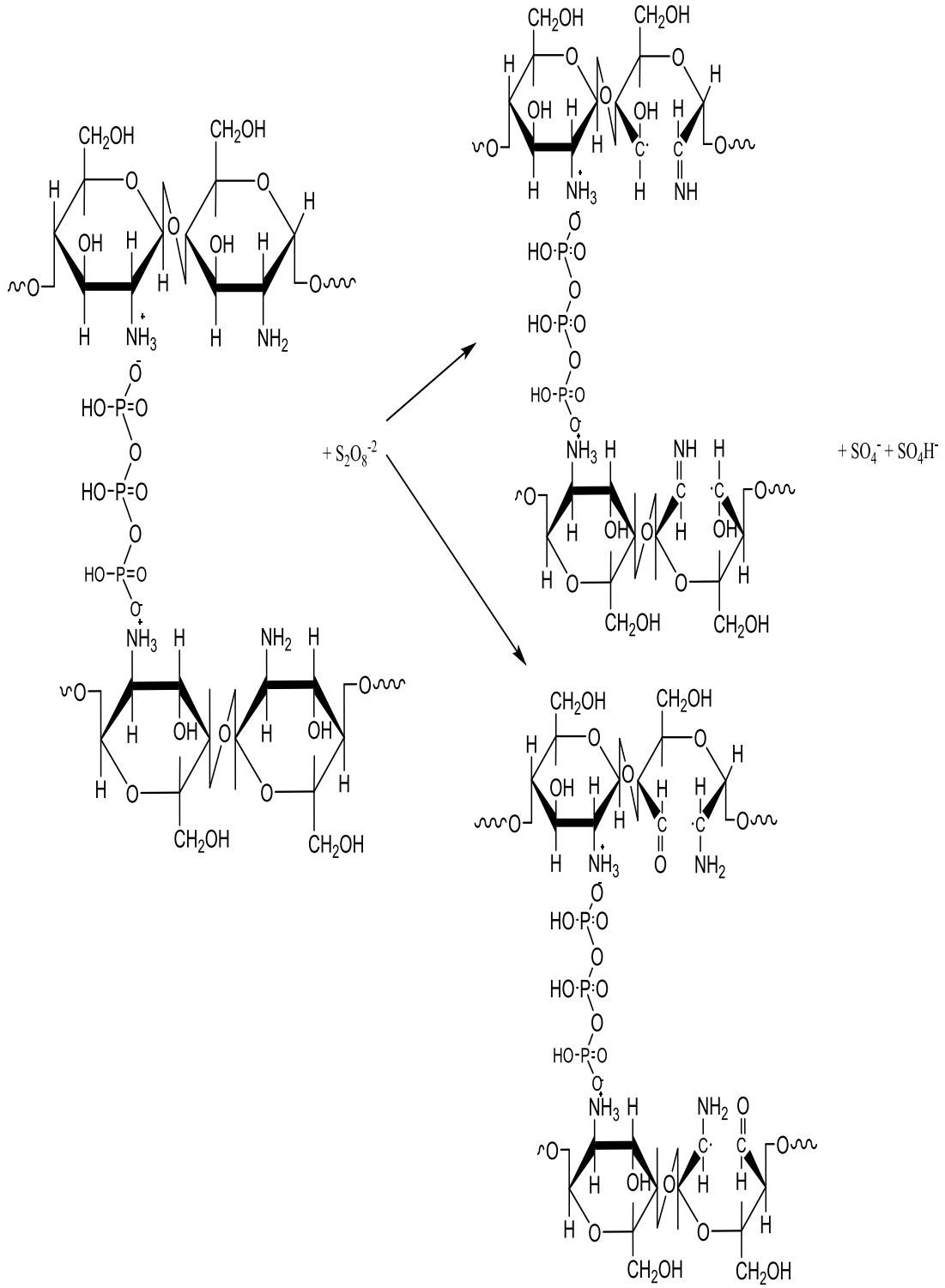


**Propagation**

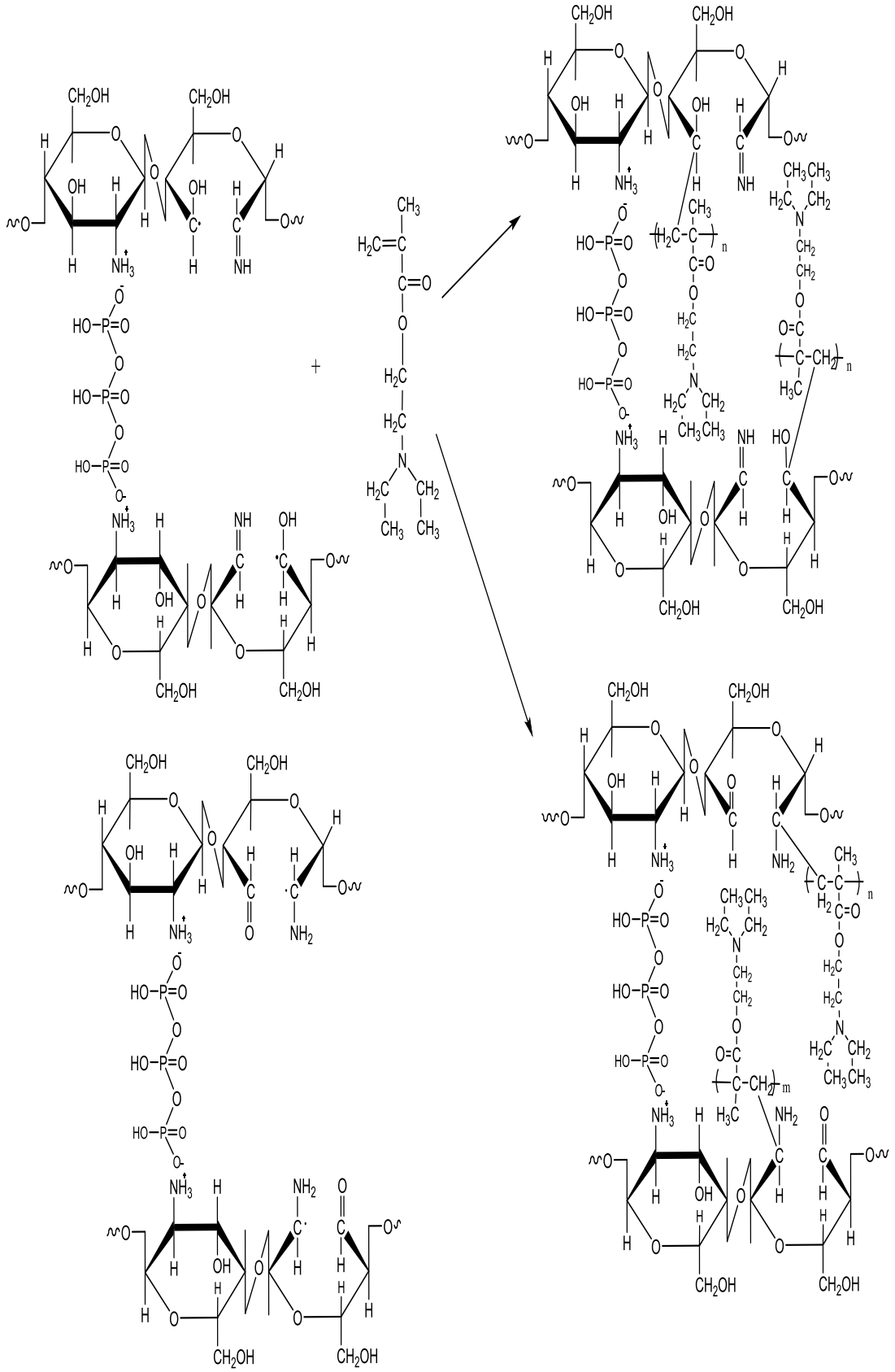


Scheme 8. Proposed Structure Mechanism for Grafting Chi-TPP Bead with (DEAEM) on C-6

**Initiation**



**Propagation**



Scheme 9. Proposed Structure Mechanism for Grafting Chi-TPP Bead with (DEAEM) on C-2 &C-3

## 3.2 Preparation of Chitosan powder-graft-Poly[(2-Diethylamino)Ethyl Methacrylate] Under Homogeneous Conditions

### 3.2.1 FT-IR Analysis for Chitosan-graft-PDEAEM Powder Products

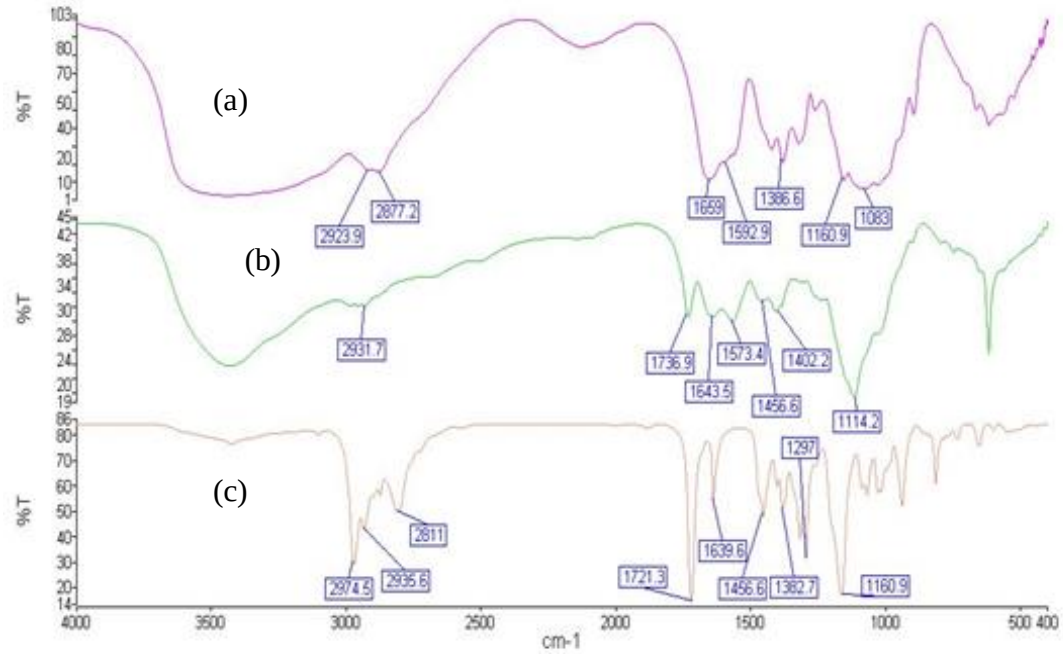



Figure 3-1: FTIR Spectrum of (a) Chitosan (b) Chitosan-graft-PDEAEM Powder (c) DEAEM

Samples were analyzed by FTIR spectroscopy to test the achievement of grafting reaction carried out. The FTIR spectrum of chitosan shows absorption bands in the range 2900-2800 at 2875cm$^{-1}$ which belong to C-H stretching. Chitosan shows special band at 1659 cm$^{-1}$ which belong to amide absorption. The band at 1386.6 cm$^{-1}$ refers to C-H rock and 1160 cm$^{-1}$, 1083 cm$^{-1}$ belong to C-O-C stretching.

DEAEM shows adsorption band at 2974 cm$^{-1}$ and 2935.6 cm$^{-1}$ band which belong to C-H stretching . The band at 2811 cm$^{-1}$ belong to H-C=O band, and 1721.3 cm$^{-1}$ belong to C=O stretching. The band at 1639 cm$^{-1}$ belong to C=C stretching, whereas

1456.6 $cm^{-1}$ belong to C-H bending and 1382.7 $cm^{-1}$ refer to C-H rocking .The peak at 1297 $cm^{-1}$ belong to C-H wag.

In the spectrum of the grafted powder the peak 1736.9 $cm^{-1}$ refer to C=O stretching and the peak at 1643.5 $cm^{-1}$ belong to the amide C=O of chitosan. The shift from 1659 $cm^{-1}$ to 1643.5 $cm^{-1}$ is an indication of grafting of the monomer onto chitosan from the amide nitrogen. The band at 1456.6 $cm^{-1}$ and 1402.2 $cm^{-1}$ refers to C-H bending, and the peak at 1115 $cm^{-1}$ refer to C-O-C of chitosan.

### 3.2.2 Gravimetric Analysis for Chitosan-graft-PDEAEM Powder Products

The effects of reaction conditions on the grafting yield are summarized in Table 3-1. The grafting reaction was carried out at 60℃, 70℃ and 80℃ as can be followed from Table 3-1 and Figure 3-3, all temperatures studied maximum grafting yield was obtained at 4h reaction time. At 60℃, 70℃ and 80℃ the grafting yield values are 170%, 180%.and 149% respectively. Hence, 70℃ and 4h are the optimum temperature and time values to obtain maximum grafting yield in this system. In these experiments, the amount of chitosan (1.00gm), DEAEM (0.50mL) and the amount of KPS (0.125g) were kept constant in 25mL solution. The effect of amount of KPS and monomer on grafting% was examined at 70 ℃ and it was found that with 0.125g KPS and 0.5 mL monomer yield highest % grafting. It was observed maximum grafting percentage was obtained when the amount of KPS concentration being (0.125g) , amount of DEAEM (0.5mL) , temperature 70℃ and time 4 hours.

Figure 3-2: Chitosan-graft-PDEAEM Powder Product

Table 3-1: The Effect of Reaction Duration, Temperature, KPS Concentration, and DEAEM Concentrations on Grafting % of Chitosan-graft-PDEAEM Powder Carried out at 70℃, 60℃ and 80℃

| Sample ID | DEAEM(mL) | T (℃) | Time (hr) | KPS (g) | G% |
|---|---|---|---|---|---|
| P1 | 0.50 | 60 | 1 | 0.1250 | 73.5 |
| P2 | 0.50 | 60 | 2 | 0.1250 | 109 |
| P3 | 0.50 | 60 | 3 | 0.1250 | 130 |
| P4 | 0.50 | 60 | 4 | 0.1250 | 170 |
| P5 | 0.50 | 60 | 6 | 0.1250 | 173.5 |
| P6 | 0.50 | 60 | 9 | 0.1250 | 133 |
| P7 | 0.50 | 70 | 12 | 0.1250 | 127 |
| P8 | 0.50 | 70 | 1 | 0.1250 | 111 |
| P9 | 0.50 | 70 | 2 | 0.1250 | 128 |
| P10 | 0.50 | 70 | 3 | 0.1250 | 147 |
| P11 | 0.50 | 70 | 4 | 0.1250 | 180 |
| P12 | 0.50 | 70 | 6 | 0.1250 | 181 |
| P13 | 0.50 | 70 | 9 | 0.1250 | 145 |
| P14 | 0.50 | 70 | 12 | 0.1250 | 131 |
| P15 | 0.50 | 80 | 1 | 0.1250 | 65 |
| P16 | 0.50 | 80 | 2 | 0.1250 | 99.5 |
| P17 | 0.50 | 80 | 3 | 0.1250 | 127 |
| P18 | 0.50 | 80 | 4 | 0.1250 | 149 |
| P19 | 0.50 | 80 | 6 | 0.1250 | 151 |
| P20 | 0.50 | 80 | 9 | 0.1250 | 125 |
| P21 | 0.50 | 80 | 12 | 0.1250 | 84.5 |
| P22 | 0.25 | 70 | 4 | 0.1250 | 88 |
| P23 | 0.75 | 70 | 4 | 0.1250 | 108 |
| P24 | 1.00 | 70 | 4 | 0.1250 | 61 |
| P25 | 0.50 | 70 | 4 | 0.0600 | 85 |
| P26 | 0.50 | 70 | 4 | 0.1800 | 143 |
| P27 | 0.50 | 70 | 4 | 0.2400 | 137 |

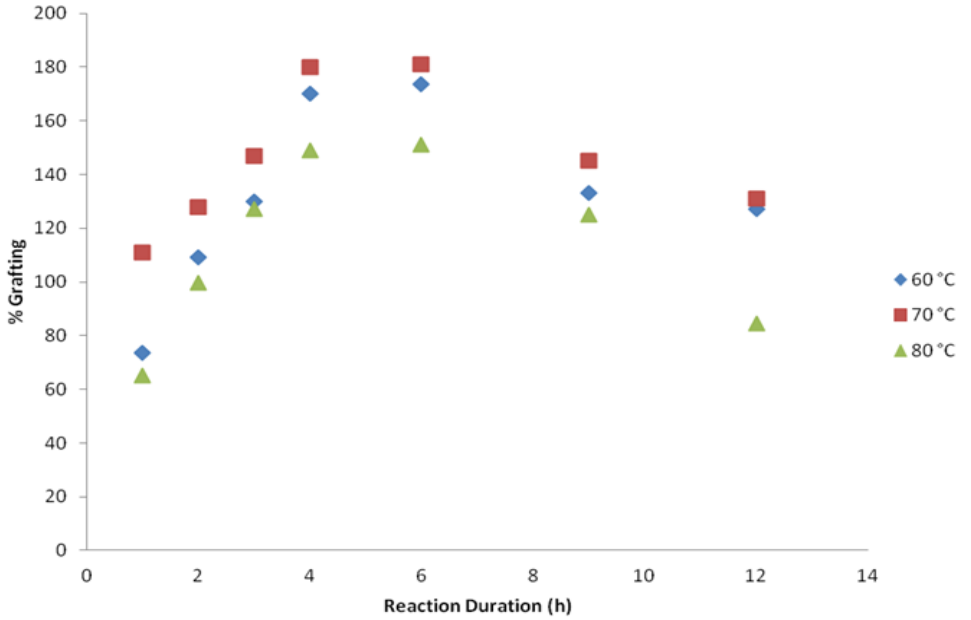



Figure 3-3: The Comparison of Temperature on Grafting % of Chitosan-graft-PDEAEM Powder Carried Out at 60, 70 and 80 °C

The grafting reactions were carried on with different temperature at 60, 70 and 80 °C. Maximum grafting% was obtained with increasing reaction duration. The maximum grafting which is 180 % occurs at 70°C within 4 hours, a further increase in temperature or time leads to decreased grafting% since grafting site is reduced and formation of homopolymer occurred.

The effect of the amount of the initiator and the monomer are shown in Table 3-1 and trends observed are illustrated in Figure 3-4 and Figure 3-5.

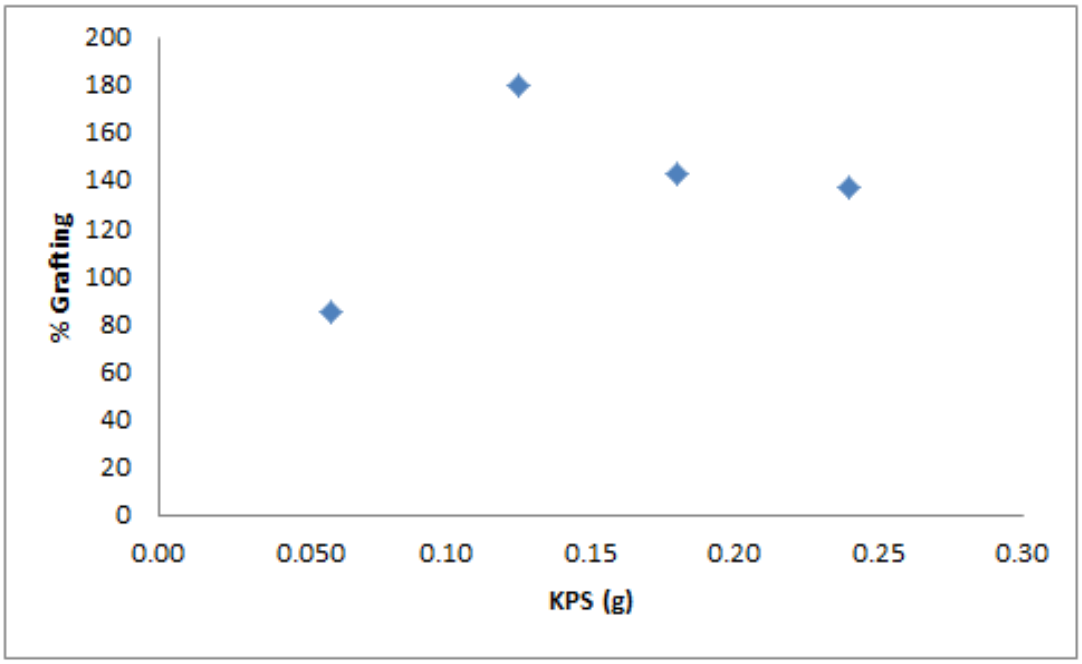



Figure 3-4: The Effect of Amount of KPS on Grafting % of Chitosan-graft-PDEAEM Powder Carried Out at 70 °C, 4h

The optimum amount of initiator, KPS, was 0.125g. Increasing the KPS amount lead to decrease the grafting% as shown in Figure 3-4. This may be explained by the formation of homopolymer which compete with grafting reaction for the available monomer.

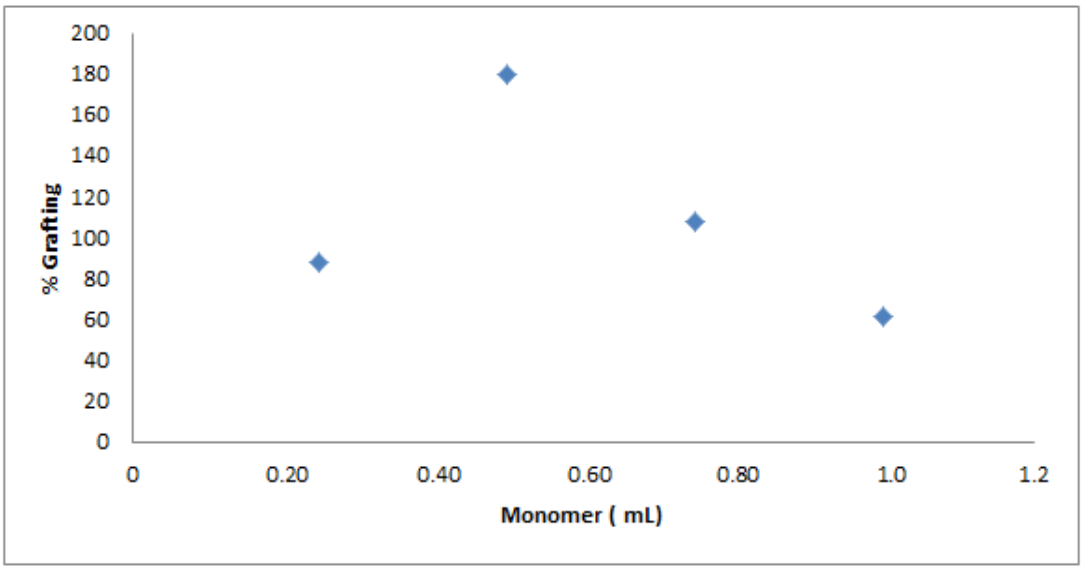



Figure 3-5: The Effect of Amount of Monomer on Grafting % of Chitosan-graft-PDEAEM Powder Carried Out At 70 ℃, 4h

The maximum grafting 180% obtained when DEAEM concentration was (0.5mL). With increasing DEAEM concentration, the grafting% were reduced. Since homopolymer of DEAEM lead to increase the viscosity of reaction media thereby the mobility of the growing polymer chains to the active site is limited.

### 3.2.3 Dissolution of Grafted Chitosan Powder

The dissolution of chitosan grafted PDEAEM powder was studied for P11 180%, P3 150% and P15 65% at pH=1.2, pH=7 and pH=11. All those powders were completely soluble at pH=7 after 24 hours. The amount of weight loss of the P3 and P11 was twice (approximately 55%) in acidic and alkaline media. On the other hand, P15 has complete weight loss in neutral and alkaline media.

## 3.3 Preparation of Chitosan TPP-graft-Poly[(2-Diethylamino)Ethyl Methacrylate] Under Heterogeneous Condition

### 3.3.1 Gravimetric Analysis of Chi-TPP-graft-PDEAEM

Crosslinked chi-TPP beads prepared either by using GA or EGDE were grafted with PDEAEM under heterogeneous conditions. The experimental conditions, grafting yield values and bead sizes before grafting are given in Table 3-2.

Table 3-2: % Grafting yield (%G), and Size of the Chitosan-TPP-graft-PDEAEM Beads DEAEM = 0.50mL, time = 4h, T = 70 ℃ and Medium 2% w/v Acetic Acid and Sizes of EGDE and GA Crosslinked Chitosan-TPP-graft-PDEAEM Beads in Acidic and Aqueous Media

| Sample ID | **GA, mL** | **EGDE, mL** | **%G** | **Size ( µm)** |
|---|---|---|---|---|
| B1 | - | - | 31.5 | 710 |
| B2 | 0.1 | - | 49.5 | 710 |
| B3 | 0.3 | - | 43 | 710 |
| B4 | 0.5 | - | 47.9 | 500 |
| B5 | 1 | - | 54 | 212 |
| B6 | - | 2.5 | 47.5 | 710 |
| B7 | - | 8 | 32 | 500 |
| B8 | - | 16 | 50.6 | 500 |
| B9 | 0.1 | - | 80.3 | 710 |

B9 refer to GA crosslinked chitosan-TPP-graft-DEAEM bead in distilled water.

Figure 3-6: GA Crosslinked Chitosan-TPP-graft-PDEAEM Bead

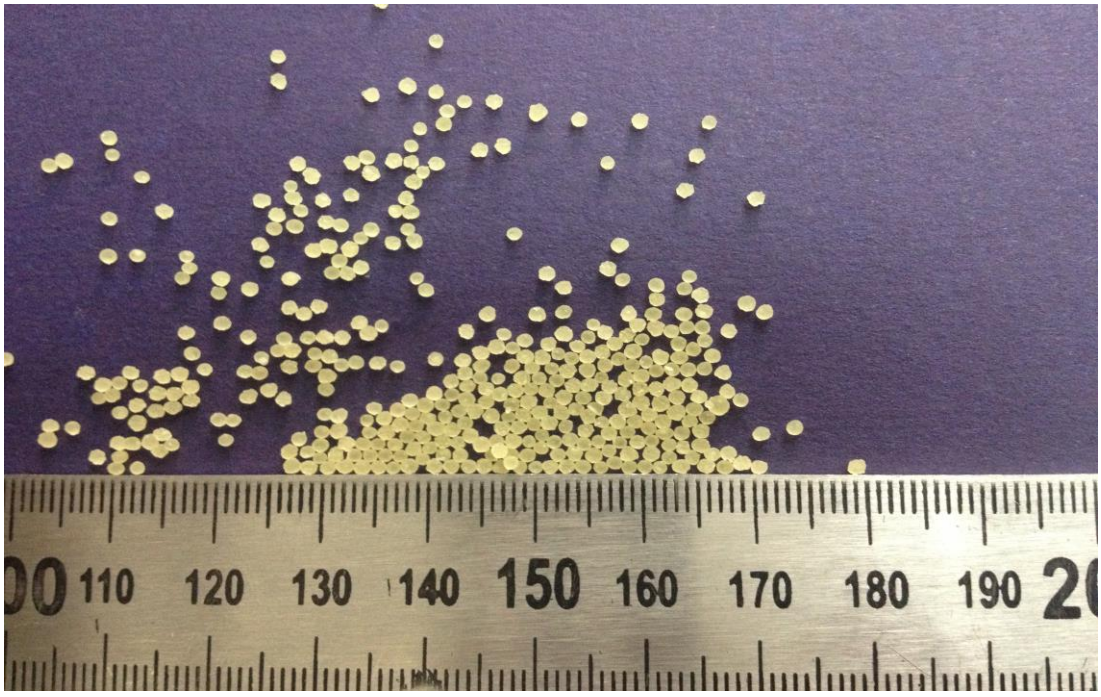


Figure 3-7: EGDE Crosslinked Chitosan-TPP-graft-PDEAEM Bead

It should be noted that increasing amount of crosslinker is expected to consume more of the grafting sites hence leaving behind a lower fraction of free $–NH_2$ or –OH groups available for grafting. Another factor to be considered is the degree of swelling of the beads during grafting reaction. As grafting proceeds degree of swelling changes. However, using an oversimplification and assuming that diffusion ability of the monomer onto the outer surface of the beads would not be affected by the degree of swelling of the bead to a significant effect it is possible compare the %G results with the respect to bead size. When %G values are examined for various beads, it can be observed that %G values range in between 43% - 51% which may be considered as invariant within experimental error. Hence, it can be concluded that several factors such as the bead size, degree of crosslinking and degree of swelling compensate for each other under the experimental conditions studied. The only significant different on %G is observed when grafting medium is changed from aqueous acetic acid to distilled water. Sample B9 grafted in distilled water has %G value of 80.3% while its counterpart B2 grafted in aqueous acetic acid has a %G value 49.5%. This difference can be attributed to higher reactivity of both the monomer and chitosan substrate under neutral conditions. All grafting experiments except for B9 were carried out in of acetic acid solution to study under parallel conditions in homogeneous and heterogeneous grafting sets. Similar size of B2 and B9 was prepared with using same amount of GA. The grafting % of those bead form samples are quite different from each other. The B9 which is prepared in aqueous medium less soluble compared to B2 which is prepared in acidic medium. EGDE crosslinked B7 and B8 beads which possess 500 µm size showed 32% and 51% grafting yield, respectively. Increasing EGDE crosslinker enhance the stability of beads against to dissolution, therefore increase grafting %.

### 3.3.2 FTIR Analysis of Chi-TPP-graft-PDEAEM

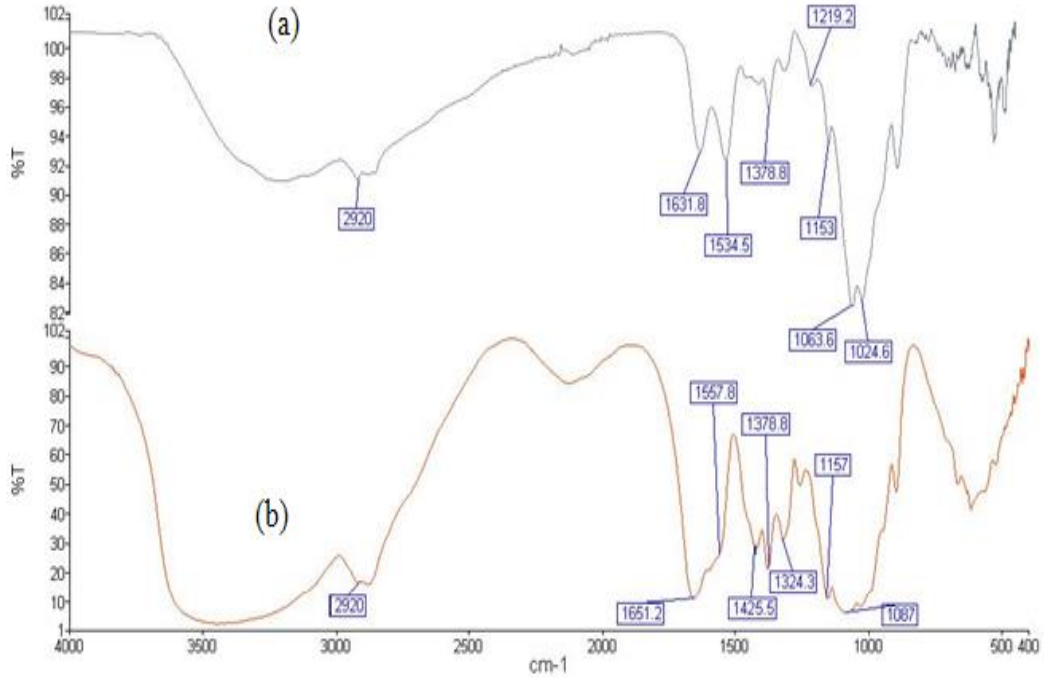



Figure 3-8: FTIR Spectrum of (a) Chitosan TPP Beads (b) Chitosan

The FTIR spectrum of chitosan-TPP and chitosan are compared in Figure 3-8(a) and (b). In Figure 3-8(a) the band at 2920 $cm^{-1}$ belongs to C-H stretching for (Chi-TPP) aliphatic compound. The band at 1631.8 $cm^{-1}$ belongs to C=O stretching. The band at 1534.5 $cm^{-1}$ peak belongs to N-H bending for $NH_3^+$. The band at 1378.8 $cm^{-1}$ refer to C-H rock. The peak at 1063.6 $cm^{-1}$ belong to C-N stretching. The bands in the range 1024.6-1063.6 $cm^{-1}$ belongs to $C_6$-O of chitosan TPP and the band at 1216 $cm^{-1}$ belongs to the P-O.

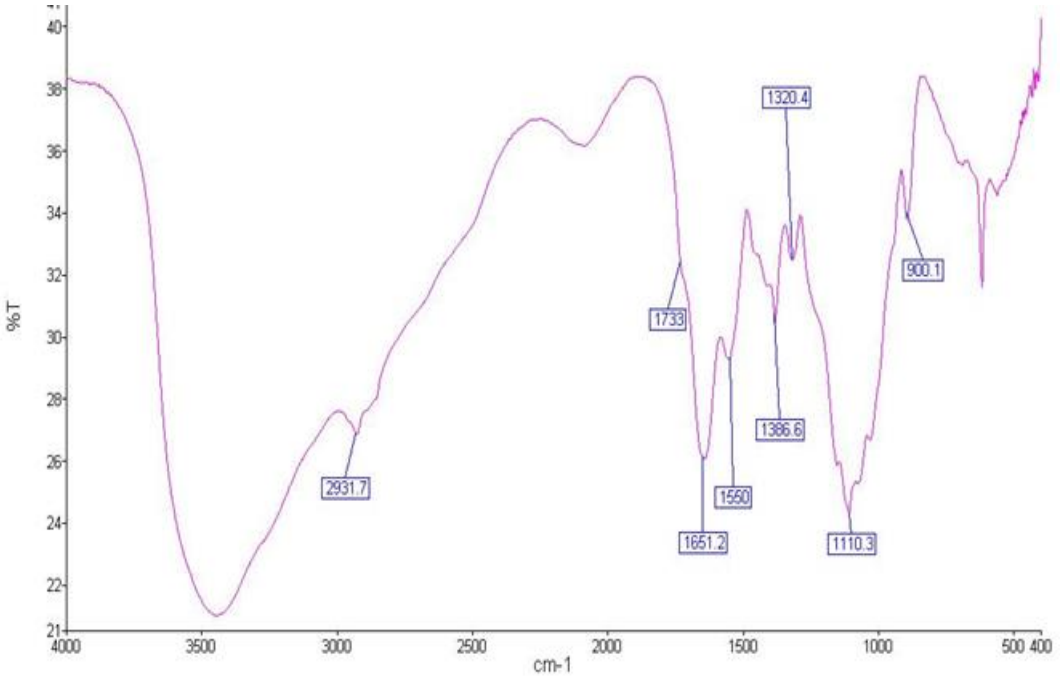



Figure 3-9: FTIR Spectrum of GA Crosslinked Chitosan-TPP-graft-PDEAEM Bead

The FTIR spectrum of the GA crosslinked and grafted beads is shown in Figure 3-9.The band at 2931.7 $cm^{-1}$ belongs to C-H stretching. The peak at 1733$cm^{-1}$ belong to C=O stretching of the polymer grafted. The band at 1651.2 $cm^{-1}$ belong to amide absorption of chitosan. The band at 1550 $cm^{-1}$ peak belongs to C=N of chemical crosslinking glutaraldehyde with chitosan , which take place of ionic interaction Chi-TPP. The band at 1461 $cm^{-1}$ belongs to C-C stretching. The band at Peak 1383 $cm^{-1}$ refer to C-H rock. The band at 1317 $cm^{-1}$ peak belong to C-O stretching and the band at 1115 $cm^{-1}$ peak belong to C-O-C of chitosan pyranose ring. It can be interpreted from the FTIR spectrum of the grafted and crosslinked product that during chemical crosslinking most of the TPP is dissolved away as it is not possible to see a peak characterizing P-O band.

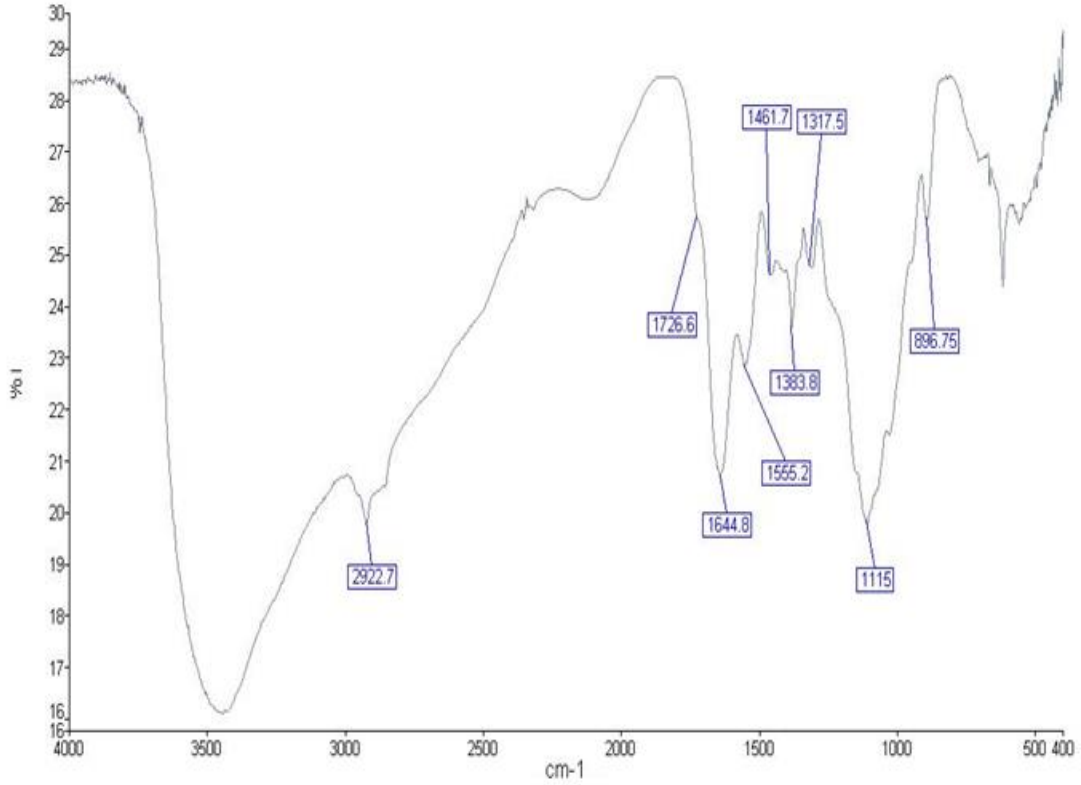



Figure 3-10: FTIR Spectrum of EGDE Crosslinked Chitosan-TPP-graft-PDEAEM Bead

The FTIR spectrum of the EGDE crosslinked and grafted beads is shown in Figure 3-10. The band at 2922.7 $cm^{-1}$ belong to C-H stretching. The peak at 1726.6 $cm^{-1}$ belong to C=O stretching of the polymer grafted. The band at 1644.8 $cm^{-1}$ belong to amide of chitosan. The band at 1555.2 $cm^{-1}$ peak belong to C=N of chemical crosslinking which take place of ionic interaction Chi-TPP. The band at 1461 $cm^{-1}$ belong to C-C stretching. The peak at 1383 $cm^{-1}$ refer to C-H rocking. The band at 1317 $cm^{-1}$ peak belong to C-O stretching and the band at 1115 $cm^{-1}$ peak belong to C-O-C of chitosan pyranose ring.

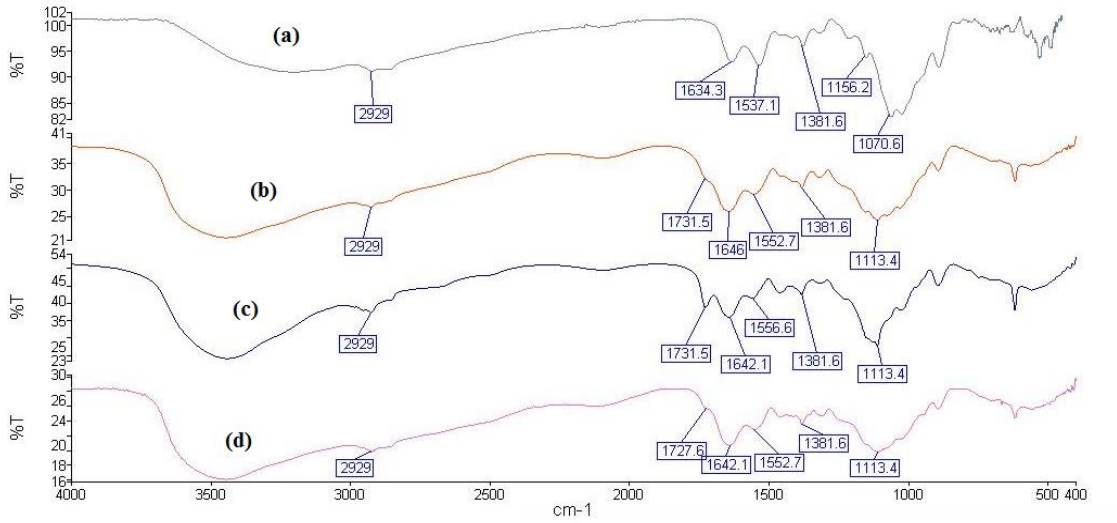



Figure 3-11: FTIR Spectrum of (a) b1 (b) B3 (c) B9 (d) B7

The FTIR spectra as shown in Figure3-11 above shows that GA crosslinked chitosan-TPP-grafted-PDEAEM bead in acid medium (B3), GA crosslinked chitosan-TPP-grafted-PDEAEM bead in water medium (B9) and EGDE crosslinked chitosan-TPP-grafted-PDEAEM bead (B7) spectrum have some desorption different from that Chi-TPP bead (b1)spectrum. It can be observed that absorption bands at 1727.6 $cm^{-1}$ or 1731.5 $cm^{-1}$ which belongs to C=O stretching that are due to grafted DEAEM which is not present in chitosan TPP. Chemical crosslinking via formation C=N (imine) bonds at 1552.7 $cm^{-1}$ , 1556.6 $cm^{-1}$ and 1552,7 $cm^{-1}$ peaks of GA crosslinked chitosan-TPP-grafted-PDEAEM bead ( acidic and water medium) and EGDE crosslinked chitosan-TPP-grafted-PDEAEM bead in acid medium which take place at the expense of ionic interaction between chitosan and TPP.

### 3.3.3 SEM Analysis

SEM micrographs of the b1(ungrafted uncrosslinked Chi-TPP bead), B1 (grafted but uncrosslinking), B3 (grafted and crosslinked with GA), B7 grafted and crosslinked with EGDE are given in Figure 3-12and 3-13.

In Figure 3-7 and 3-12, it is illustrated that Chi-TPP beads have uniform spherical shapes with rough surfaces. The presence of some cracks was observed in Figure 3-12, which can be caused by drying process. The smoother surface of Chi-TPP beads was obtained after crosslinking with both crosslinker. Also some porosity observable, as shown in Figure 3-13.

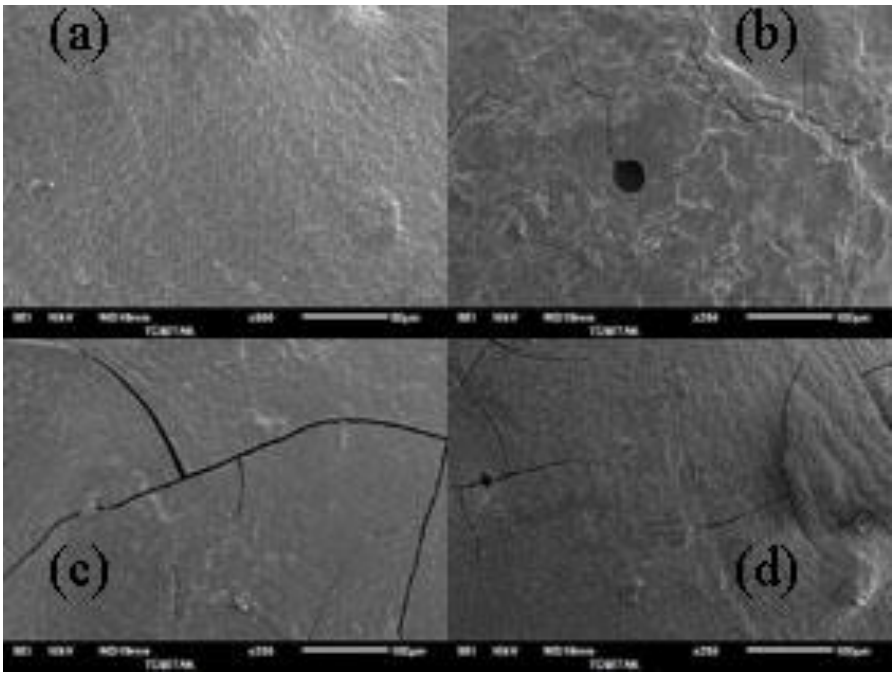


Figure 3-12: SEM Micrograph of b1, B1, B3, B7 Magnified by 500x

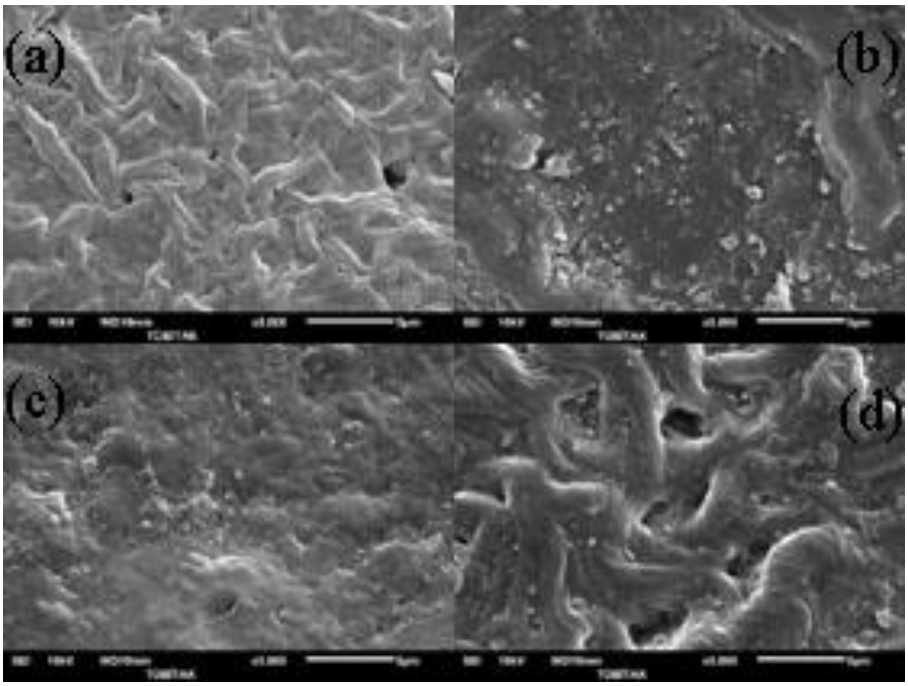


Figure 3-13: SEM Micrograph of b1, B1, B3, B7 Magnified by 5000x

### 3.3.4 Dissolution and Swelling Properties of Products

Swelling is affected by both the hydrophilicity and hydrophobicity of the polymer backbone. Swelling can be varied by the type of crosslinker and degree of crosslinking. The swelling behavior of B7, B6, B3, B2, B1 products is shown in Figure 3-14,3-15and 3-16 in PH= 1.2, 7.0 and 11.0 respectively. High crosslinking density leads to reduction in swelling. B3 has lower swelling in comparison to B2 as shown in Table 3-3, 3-4 and 3-5. Non crosslinked Chi-TPP (B) showed highest swelling in all pH=1.2 and pH=7.0 at 3 rd hour compared to the others. In alkaline media (pH=11.0) non crosslinked Chi-TPP (B) had the highest swelling at 24th hours. Then it started to dissolve with time. The swelling properties of B1 and b1 were investigated. b1 showed 4800 % in pH=1.2, 195% in neutral pH and 248% in alkaline buffer. B1 had 166 % swelling in acidic solution and 255% in alkaline, whereas dissolved in neutral buffer as shown in Figures 3-14, 3-15 and 3-16.

Table 3-3: Swelling % in pH=1.2

| Time (h) | B7 | B6 | B3 | B2 | B1 |
|---|---|---|---|---|---|
| 1 | 96 | 147 | 160 | 204 | 328 |
| 2 | 123 | 139 | 173 | 261 | 432 |
| 3 | 153 | 120 | 172 | 285 | 472 |
| 24 | 163 | 139 | 200 | 351 | 166 |
| 48 | 174 | 118 | 201 | 345 | dissolved |
| 72 | 180 | 129 | 206 | 367 | dissolved |

Table 3-4: Swelling % in pH=7

| Time (h) | B7 | B6 | B3 | B2 | B1 |
|---|---|---|---|---|---|
| 1 | 67 | 88 | 86 | 80 | 126 |
| 2 | 62 | 82 | 98 | 89 | 127 |
| 3 | 56 | 88 | 98 | 90 | 132 |
| 24 | 64 | 98 | 109 | 115 | dissolved |
| 48 | 70 | 94 | 112 | 127 | dissolved |
| 72 | 96 | 97 | 120 | 128 | dissolved |

Table 3-5: Swelling % in pH=11

| Time (h) | B7 | B6 | B3 | B2 | B1 |
|---|---|---|---|---|---|
| 1 | 75 | 97 | 101 | 115 | 146 |
| 2 | 67 | 106 | 105 | 125 | 176 |
| 3 | 73 | 119 | 108 | 127 | 214 |
| 24 | 132 | 137 | 122 | 214 | 255 |
| 48 | 89 | 144 | 113 | 220 | dissolved |
| 72 | 50 | 145 | 116 | 234 | dissolved |

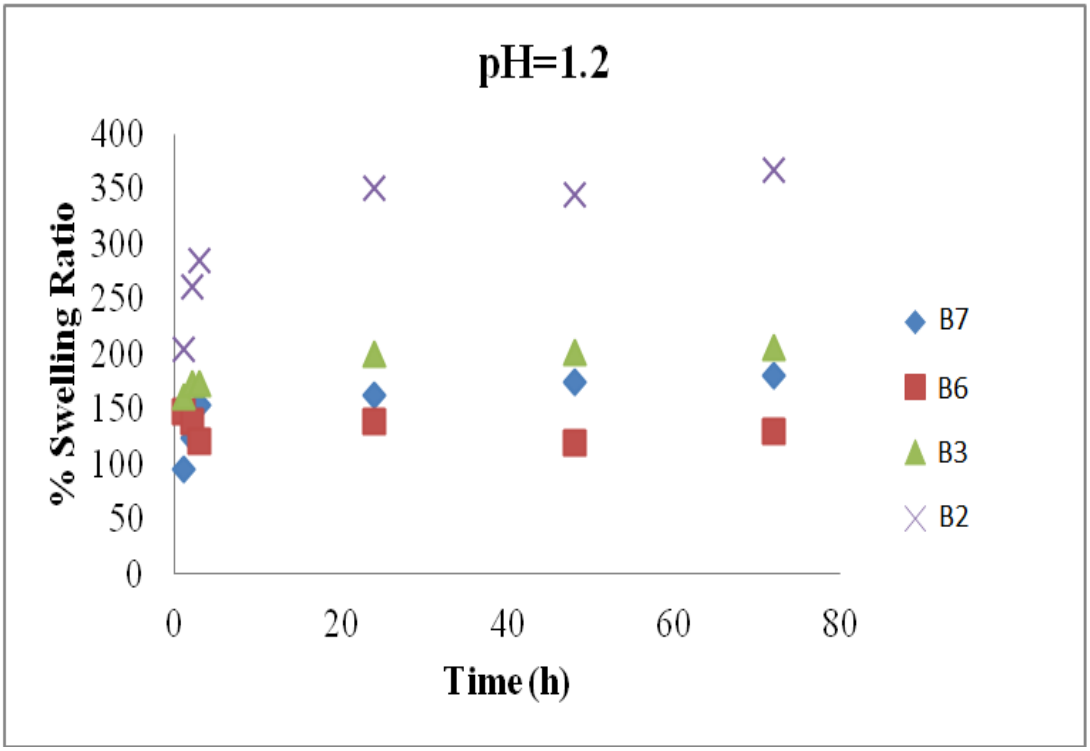


Figure 3-14: Swelling % of Chitosan-TPP-graft-PDEAEM in pH=1.2

GA crosslinked chitosan-TPP-graft-PDEAEM swell more than EGDE crosslinked chitosan-TPP-graft-PDEAEM. EGDE crosslinker leads to less swelling with avoiding dissolution. This can be explained by chemical modification occurred on the bead surface. Both Crosslinker EGDE and GA improved both resistance to dissolution and swelling.

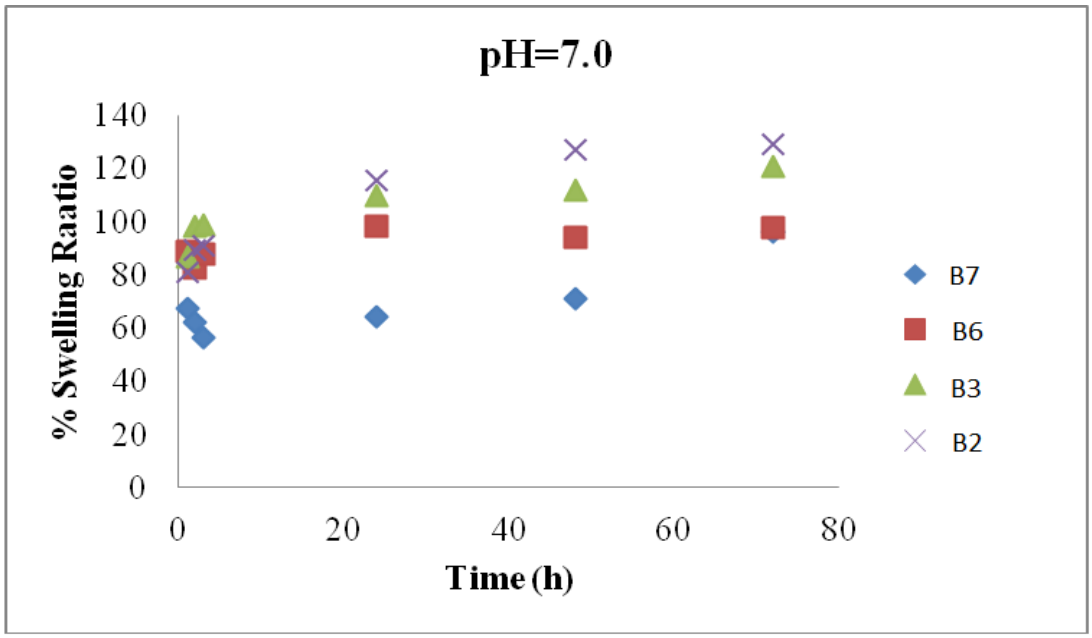



Figure 3-15: Swelling % of Chitosan-TPP-graft-PDEAEM Beads in pH=7

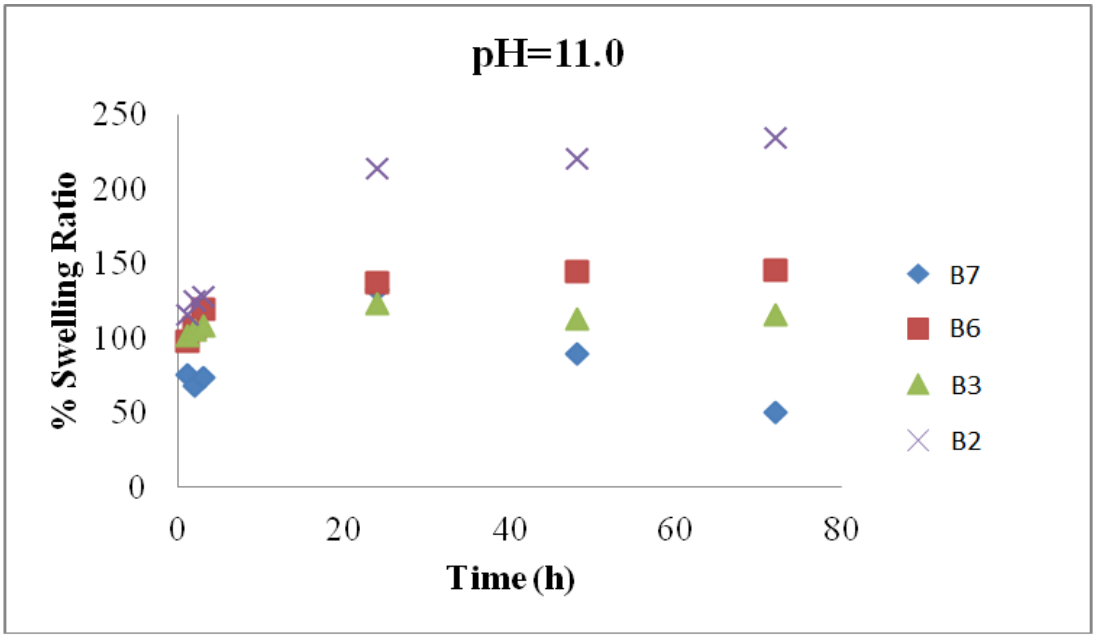



Figure 3-16: Swelling % of Chitosan-TPP-graft-PDEAEM in pH=11

Table 3-6: Swelling % of Chitosan-TPP-graft-PDEAEM Beads and Chitosan-TPP Beads in pH=1.2, pH=7.0 and pH=11.0

| %Swelling | pH=1.2 | pH=7 | pH=11 |
|---|---|---|---|
| b1 | 4811 | 195 | 247 |
| B1 | 166 | Dissolves | 255 |

### 3.3.5 Thermal Gravimetric Analysis (TGA)

TGA analysis for Chi- TPP, Chi- TPP bead grafted with PDEAEM, Chi-TPP beads crosslinked with GA and grafted with PDEAEM and Chi-TPP beads crosslinked with EGDE and grafted with PDEAEM is shown below in Figure 3-17, 3-18, 3-19 and 3-20 respectively. Figure (3-17) Chi- TPP bead decompose and show the first step weight loss at 50°C (9%) which is related to releasing water molecules. The onset of weight loss of Chi- TPP start at 220°C with two steps: first at 220°C (43%) and the second step at 600°C (6%), due to the degradation of the polysaccharide. 40% remaining is not decomposed at 900°C.

Figure (3-18) Chi- TPP grafted bead with PDEAEM has three steps of weight loss first start at 220◦C (12%), the second step at 280°C (23 %) and the third step at 600°C (6%) due to the degradation. 36% is remaining at 900°C.

Figure (3-19) Chi-TPP beads crosslinked with GA and grafted with PDEAEM has three steps for weight loss, first start at 220C (10%), second step start at289°C (25%) and the third step start at 600°C (6 %). 36% remain at 900 °C. the second peak (289°C) should be due to the decomposition of the crosslinked chains.

Figure (3-20) show TGA Thermal gravimetric for Chi-TPP beads crosslinked with EGDE and grafted with PDEAEM. The only difference compared to Figure 3-18 and 3-19 is an additional decomposition peak observed at 330 ˚C. this is an indication of other type of crosslinking reactions which may have occurred when this sample was formed. With limited number of samples studied it is not possible to make further detailed discussion.

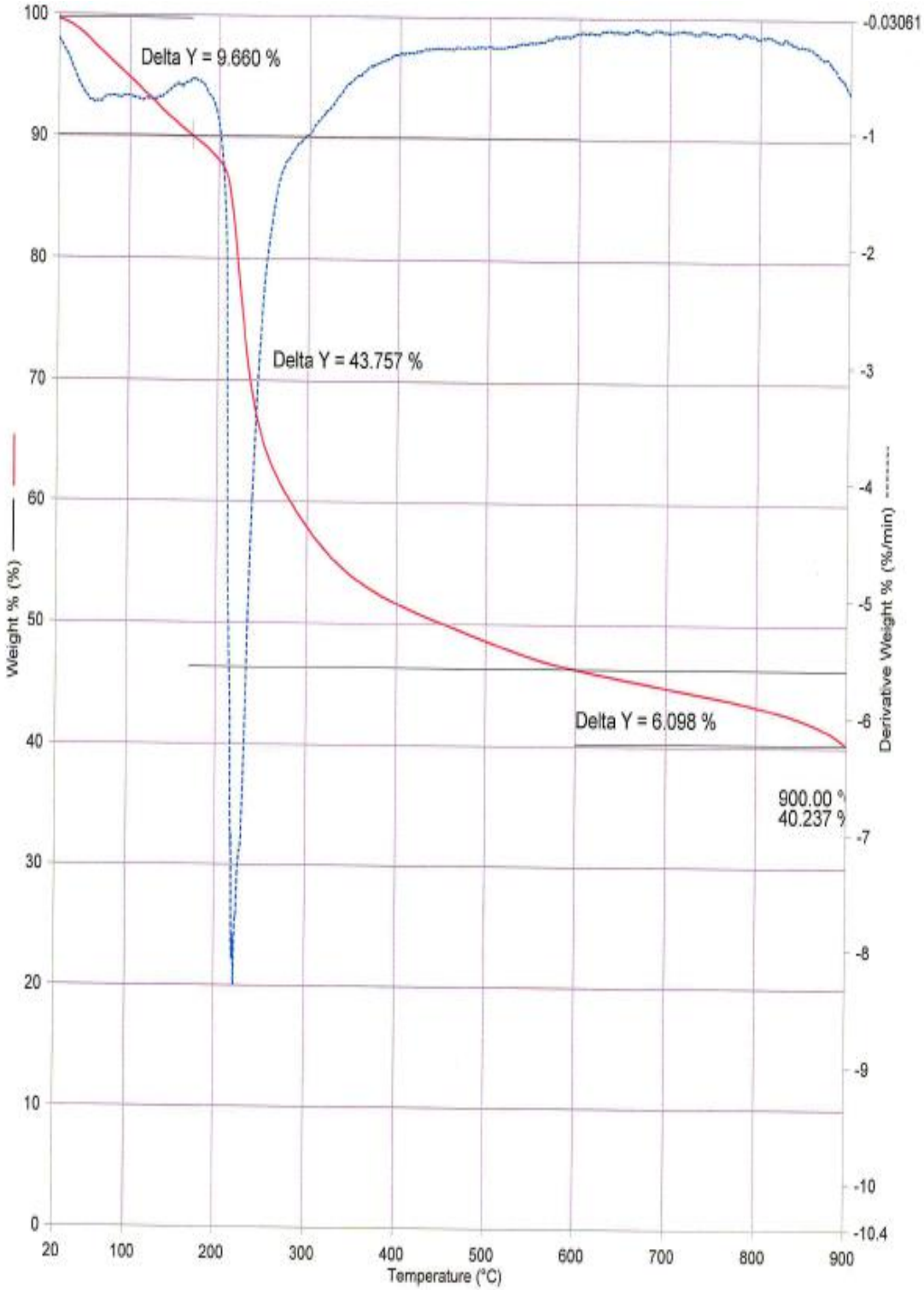



Figure 3-17: TGA Spectrum of Chi-TPP Bead

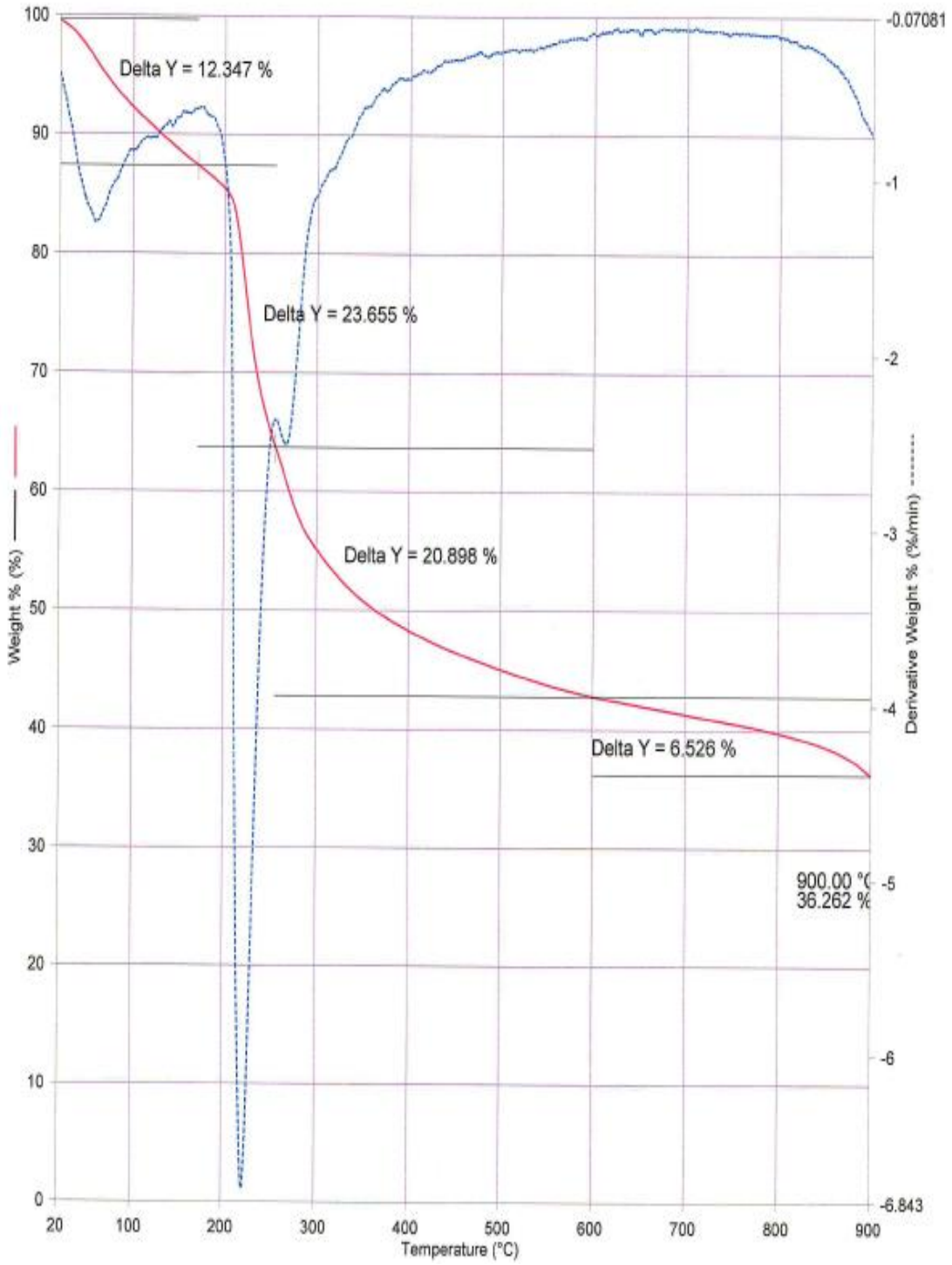



Figure 3-18: TGA Spectrum of Chi- TPP Bead Grafted with PDEAEM

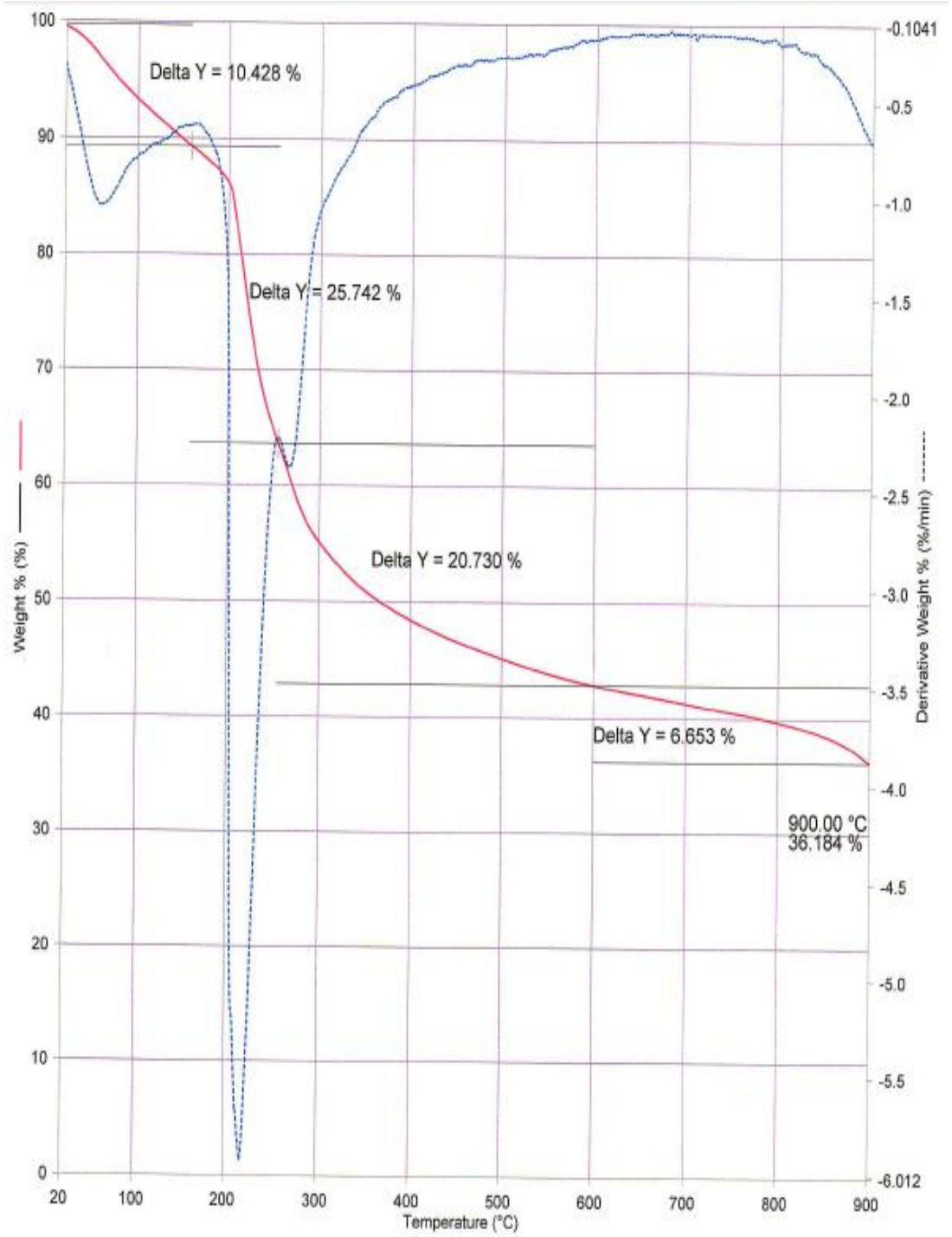



Figure 3-19: TGA Spectrum Chi-TPP beads Crosslinked with GA and Grafted with PDEAEM

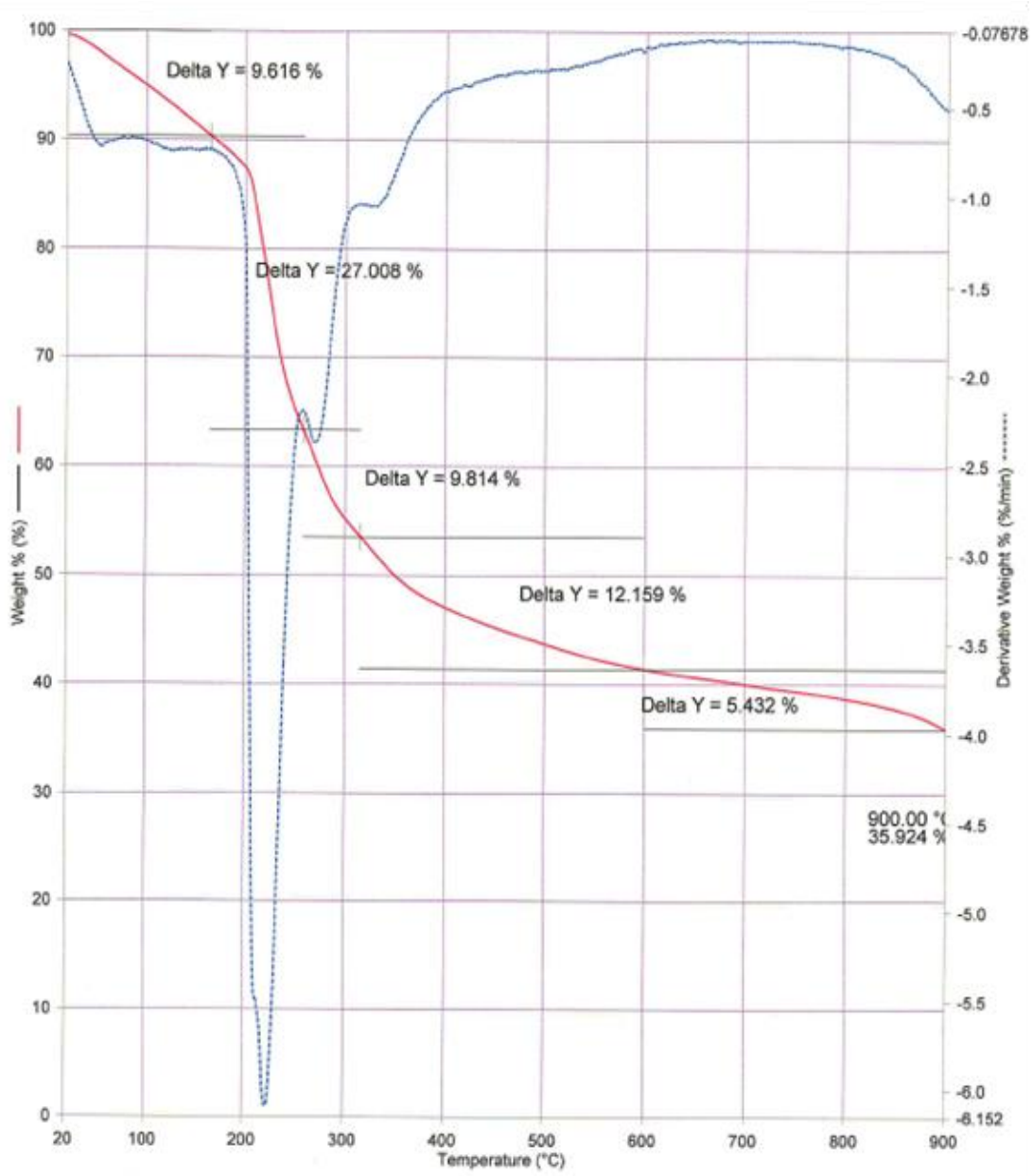



Figure 3-20: TGA Spectrum Chi-TPP Beads Crosslinked with EGDE and Grafted with PDEAEM

# Chapter 4

# CONCLUSIONS

PDEAEM can be grafted on to chitosan in solution, under homogeneous conditions to produce water soluble chitosan-grafted –PDEAEM powder products. The grafting values range between 61% to 181%. The optimum grafting condition are temperature 70 ℃ , time 4h ,the amount of initiator (0.1250gm) and DEAEM concentration (0.5mL). Giving rise 181% grafting. PDEAEM can be grafted onto chitosan –TPP beads to create surface modification of chitosan-TPP beads. %G values of 43% - 51% can be achieved using glutaraldehyde or EGDE crosslinker chitosan –TPP beads, %G is not affected significantly with the amount of chemical crosslinker used. PDEAEM grafted beads are capable of swelling under acidic, neutral or basic medium. They are thermally stable up to 300℃ and have rough bead surface becoming more homogenous with grafting. Bead morphology bears some porosity as well. The water soluble chitosan-graft-PDEAEM powders are anticipated to be useful in gene delivery applications and as antibacterial polymers. The surface grafted beads have a potential to find a place in drug delivery as well as biobased adsorbent.

# REFERENCES


Bhattacharya, B. (2004). Grafting: A versatile means to modify polymers techniques, factors and applications. *Progress in Polymer Science*, 767–814.

Abduel majid k. Najjarm Wan Md Zin Wan Yunus, M. B. (2000). Preparation and characterization of poly (2-acrylamido-2-methylamido-2-methylpropane-sulfonic acid) grafted using potassium persulfate as redox initiator. *Polymer Science,*, 2314–2318.

Agrawal, S., & Yi Zhang, S. M. (2012). PDMAEMA based gene delivery materials. *Materials Today*, 389- 393.

David R Kruscio, N. A. (2012). Surface imprinted thin polymer film systems with selective recognition for bovine serum albumin. *Analytica Chimica Acta*, 109-115.

Skjak-Braeck, T. P. (1985). Chitosan: commercial uses and potential applications. In chitin and chitosan: sources, chemistry, biochemistry, physical properties and applications. *Elsevier Applied Science*, 51-69.

Giovanni G Vigliotta, M. M. (2012). Modulating antimicrobial activity by synthesis: dendritic copolymers based on nonquaternized 2-(dimethyl amino)ethyl methacrylate by Cu- mediated ATRP. *Biomacromolecules*, 833-41.

Hamit Caner, E. Y. (2007). Synthesis, characterization and antibacterial activity of poly (N-vinylimidazole) grafted chitosan. *Carbohydrate Polymers*, 318–325.

Hatice Nilay Hasipoglu, E. Y. (2005). Preparation and characterization of maleic acid grafted Chitosan. *International Journal of Polymer Analysis and Characterization*, 313-327.

Berger, M. R. (2004). Structure and interactions in covalently and ionically crosslinked chitosan hydrogels for biomedical applications. *European Journal of Pharmaceutics and Biopharmaceutics*, 19–34.

Majeti, N. R. (2000). A Review of Chitin and Chitosan Applications. *Reactive & Functional Polymers*, 1-27.

Saranya, A. M. (2011). review chitosan and its derivatives for gene delivery . *Biological Macromolecules*, 234-238.

Ramya.R, V. ,. (2012). Biomedical application of chitosan : An overview. *Biomaterials and Tissue Engineering*, 100-110.

Ricardo Navarro, J. G. (2003). Recovery of metal ions by chitosan: Sorption mechanisms and influence of metal speciation. *Macromolecular Bioscience*,, 552-561.

Kumbar, A. a. (2002). Crosslinked chitosan microspheres for encapsulation of diclofenac sodium: effect of crosslinking agent. *Microencapsulation*, 173-180.

Sigma, A (n.d.). http://www.sigmaaldrich.com/catalog/product/aldrich/408980?lang=en&region=TR.

Sung- Tao Lee, F.-L. M.-J. (2001). Equlibrium and kinetic studies of copper (II) ion uptake by chitosan- tripolyphosphate chelating resin. *Polymer*, 1879-1892.

Tapan Kumar Girin, A. T. (2012). Review modified chitosan hydrogels as drug delivery tissue engineering systems: present status and applications. *Acta Pharmaceutica Sinica*, 439-449.

Terin Adali, E. Y. (2009). synthesis, characterization and biocompatibility studies on chitosan-graft-poly (EGDMA). *Carbohydrate Polymers*, 136–141.

Mourya, N. N. (2008). Review Chitosan-modifications and applications: Opportunities galore. *Reactive & Functional Polymers,*, 1013–1051.

Hennink, C. v. (2012). Novel crosslinking methods to design hydrogels. *Advanced Drug Delivery Reviews*, 223–236.

Hennink, C. N. (2002). Novel crosslinking methods to design hydrogels. *Advanced Drug Delivery Reviews*, 13–36.

Wan Ngaha, L. T. (2011). Adsorption of dyes and heavy metal ions by chitosan composites: A review. *Carbohydrate Polymers*, 1446-1456.

Shu, K. Z. (2002). Controlled drug release properties of ionically cross- linked chitosan beads: the influence of anion structure International. *Pharmaceutics*, 217–225.

Ying Y Wang, C.-Y. C.-Y. (2012). Spiropyran- based hyperbranched star copolymer: synthesis, phototropy, FRET, and bioapplication. *Polymer*, 95-3703.