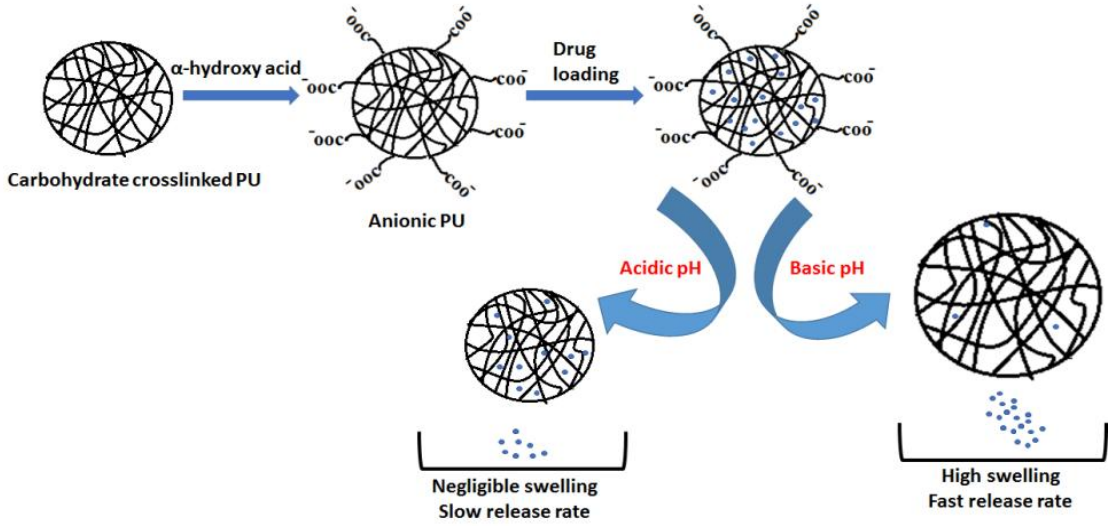
Chapter 4

Cellulose crosslinked pH-responsive waterborne polyurethanes for drug delivery: α-hydroxy acids as drug release modifiers



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4.1 INTRODUCTION

After reaching this stage, we could understand role of stoichiometry of the PUs to command the rate of drug release. In order to move towards synthesis of waterborne PUs and aiding stimuli responsive characteristics to the PUs, we carried our research in the direction as shall be discussed in this chapter.

4.1.1 WATERBORNE PUS: A BRIEF INTRODUCTION

PUs are one of the most adaptable polymeric materials with regard to both processing methods and mechanical properties [1]. However, the PU products synthesized by conventional process usually contain a significant amount of hazardous air pollutants (HOPs) and sometimes even free isocyanate monomers [2]. Besides this, conventional PU resin systems usually contain a high proportion of volatile organic components (VOC), leading to serious environmental problems. Therefore, looking to the increased demand of environmental legislation for raw materials with low VOCs, solvent-based PUs have been gradually replaced by the waterborne polyurethanes (WPUs). The increased use of WPUs nowadays is attributed to their exceptional set of properties such as [3–5]:

- Resistance to chemicals, solvents and water
- Flexibility and toughness
- Abrasion resistance
- Adhesion to various substrates

WPUs are especially suitable for applications including painting plastics and wood, and for coating both metallic and mineral substrates. The advantages of WPUs are illustrated in table 4.1.

The fundamental aspect to the employment of polymers in aqueous media is the fact that certain polar functional groups are capable of conferring water solubility or water dispersibility to PUs which are otherwise water-insoluble. The most commonly adopted functional groups are carboxylic acid groups, sulphonic acid groups and tertiary amine groups [3]. The concentration of such functional groups plays a vital role in determining the solubility or dispersibility in an aqueous environment. Usually, higher concentration of such functional groups leads to water soluble polymer, whereas low concentration results in water-dispersible polymer. This is possible provided its molecular weight/viscosity is not excessive. At even lower concentration, the polymer can obtain charge stabilization or steric stabilization impacted due to the presence of polar groups.

Table 4.1 Waterborne Polyurethane Advantages

Quick dry
Outstanding flexibility and impact resistance
Excellent abrasion resistance
Non-flammable
Easy water clean-up
Low volatile organic content = less pollution

The commercially adopted process for the preparation of WPUs can be described as follows. The first step is the formation of a medium-molecular weight isocyanate-terminated prepolymer by the reaction of suitable polyol with stoichiometric excess of isocyanate. After this stage, the different procedures utilize different ways for incorporation of chain extender.

Aqueous PU dispersions may be divided into two classes. One class consists of polymers stabilized by external emulsifiers, and the other of those in which stabilization is achieved by the inclusion of hydrophilic centers in the polymer. Such hydrophilic centers, in principle, may be one of three types:

- Anionic groups: e.g. carboxylate or sulphonate groups
- Cationic groups: e.g. alkylated or protonated tertiary amines
- Nonionic groups: e.g. polyethylene oxide chains

Such hydrophilic groups act as internal emulsifiers, making it consentient to produce stable aqueous emulsions. In the solution process, the isocyanate-terminated polyurethane prepolymer is chain extended in solution to prevent an excessive viscosity from being attained. The preferred solvent is acetone, and hence this process is frequently referred as the *acetone process* [6]. This process, however, is naturally limited to the requirements that the polymer be uncross-linked and acetone soluble. The need for removal of solvent is another obvious disadvantage.

Sometimes, the polymer to be dispersed in water is functionalized with water solubilizing/dispersing groups which are introduced either into the prepolymer prior to chain extension or are introduced as a part of chain extension agent. Thus, without the use of an externally added surfactant, small-particle stable dispersions can be obtained.

4.1.2 STIMULI-RESPONSIVE POLYMERS- AN OVERVIEW

In recent years, considerable attention is focused on the controlled drug delivery systems capable of releasing their drug payload in response to external stimuli. Such systems find increasing applications in tissue targeting, achieving specific intracellular locations, or promoting controlled drug release. Currently there is an increasing demand of stimuli-sensitive drug devices having a

modulated drug release in response to external factors such as pH [7,8], temperature [9,10], magnetic field [11,12] etc. The release of drug from the polymer can be controlled by external stimuli such as temperature, pH, ionic strength, an electric field, magnetic field or metal, etc (Fig. 4.1(a)). Polymers that respond to these external stimuli can be used as controlled-release devices. Such type of polymers is called stimuli-responsive polymers or "smart polymers". For a brief overview of such systems, the examples of temperature and pH-responsive polymers are considered as follows.

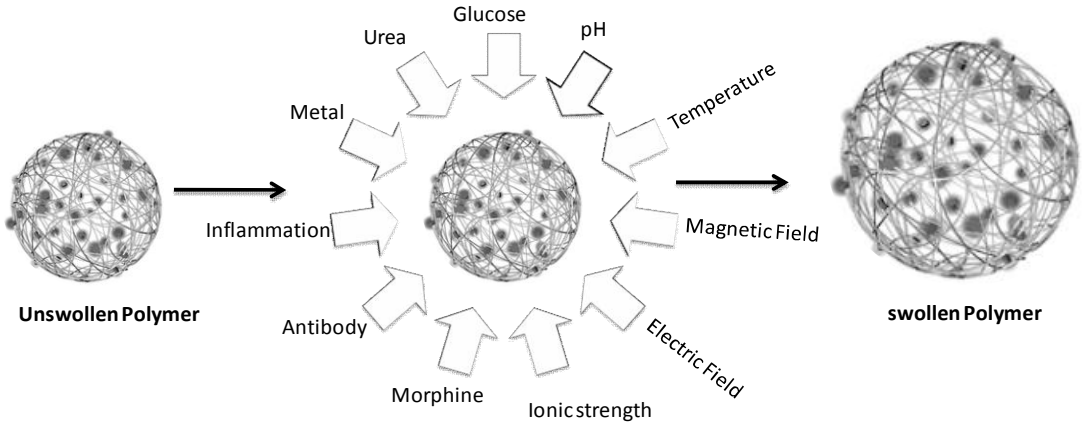


Figure 4.1(a) A schematic diagram showing stimuli responsive swelling of polymers

The temperature responsive polymers can be utilized in the field of controlled drug delivery due to the fact that the actual body temperature often deviates from the physiological value (37°C) in the presence of pathogens or pyrogens. This type of temperature variations is an advantageous stimulus to activate the release of drugs from various polymers that are responsive to such temperature, especially for diseases accompanied by fever. The variety of properties of polymers such as thermally reversible transition, swelling properties and glass transition are used for construction of temperature responsive polymers [13]. The example of such system includes poly(N-isopropyl acryl amide) (NIPAAm) which has been extensively employed as a negative thermosensitive hydrogel [14–16].

4.1.3 pH RESPONSIVE POLYMERS

Amongst all the stimuli listed above, the one having pH as an external stimulus is much interesting for the fact that a gradient of changes in the pH are found physiologically. Such gradient can be used to drive the targeted drug delivery using pH of the relevant body part. For instance, gradient between the cellular compartments (pH 4.5 to 6.5), and extracellular environment (pH 7.4) [17], pathological and normal tissues and along gastrointestinal (GI) track are well exemplified. Strategies to prevent GI degradation and/or to promote absorption in the intestine by making use of the pH gradient found along the GI tract appear promising. Besides this, pH of normal blood (7.4) differs greatly form pH of cancerous tissues (5.7) [18], which offers advantage for preparation of polymers which release anticancer drug in acidic conditions to target cancer cells distinctively.

Examples of pH responsive drug delivery system include use of poly(acrylic acid) (PAA) with carboxylic groups and poly(N, N-dimethylaminoethyl methacrylate) with amino groups for controlling drug release in alkaline and acidic pH environments, respectively [19].

The pH responsive polymers are synthesized by incorporation of acidic or basic pendant groups that either accept or donate protons in response to the environmental pH. Swelling of polymer is directly proportional to pH of external media in case of polymers with weakly acidic (anionic) groups, but inversely proportional in case of polymers with weakly basic (cationic) groups [20] (Fig. 4.1(b)).

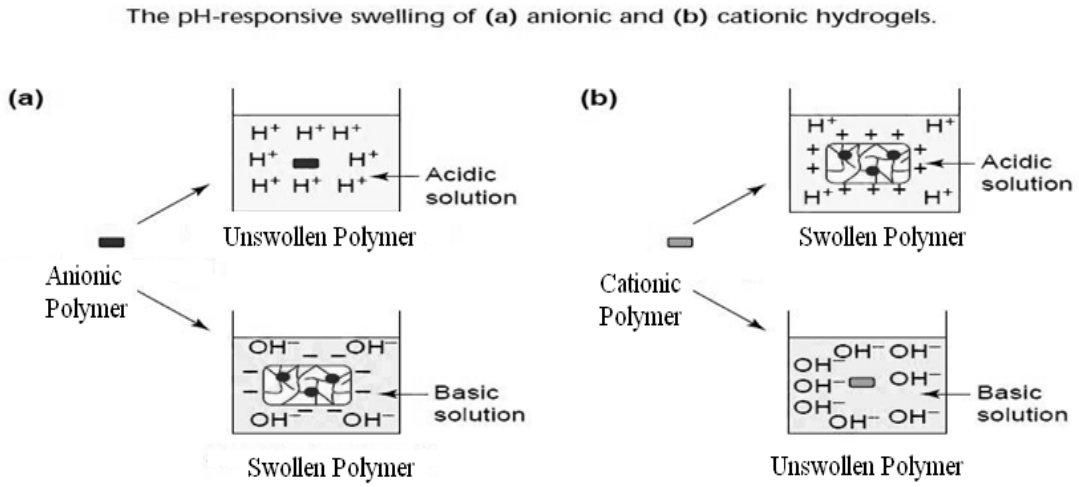


Figure 4.1(b) Effect of pH of external media on the swelling properties of polymer

Most anionic pH-sensitive polymers are based on polyacrylic acid (PAA) or its derivatives. In addition to this,, polymethacrylic acid (PMMA), poly(ethylene imine) and poly(l-lysine) have also been explored for use in drug delivery [21]. On the other hand, cationic pH-sensitive polymers are based on chitosan. Glycine, chitosan, and glutaraldehyde were used to prepare spherical crosslinked beads for a pH-dependent swelling behavior which makes them appropriate for delivery of drugs in an acidic environment [22,23]. The drug release in pH responsive systems is generally triggered by protonation (or deprotonation) of free carboxylic acid groups present in a polymer. Depending on the route of drug administration and the physical barriers to overcome, three potentially Inter-related pH-responsive drug release approaches/strategies can be envisaged (Fig. 4.1(c)), as follows [24].

1. Destabilization (via collapse or swelling) of drug delivery systems upon changes in pH
2. Dissociation or
3. pH-dependent changes in partition coefficient between the drug and the delivery vehicle.

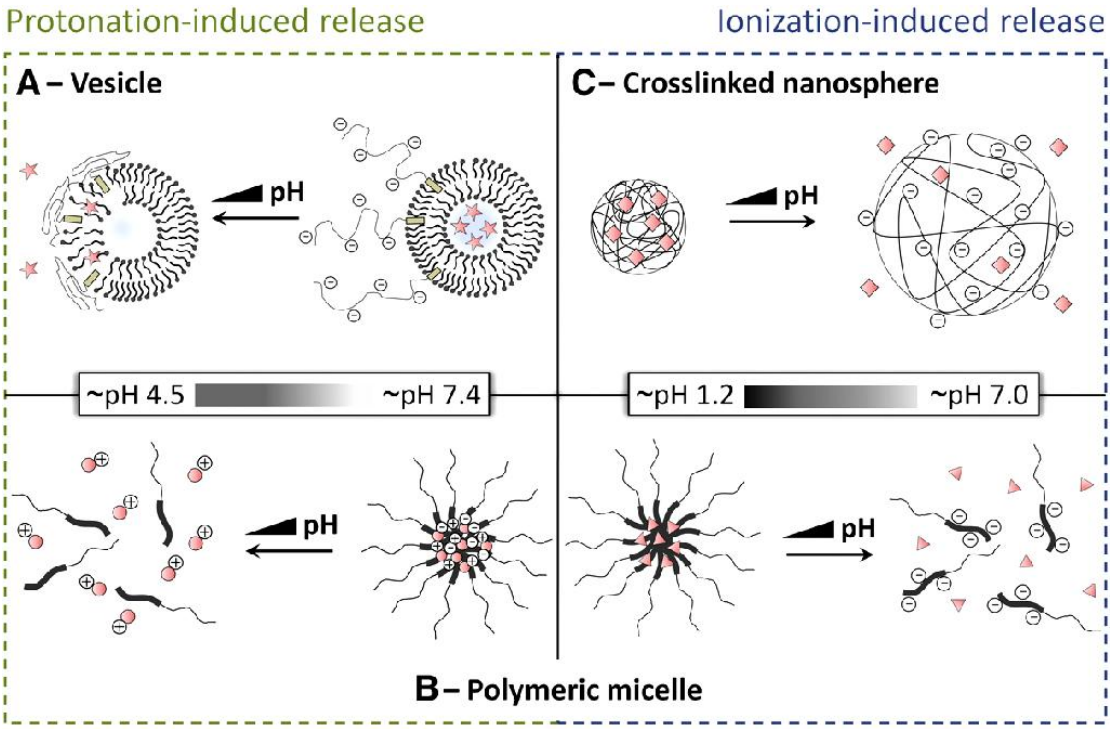


Figure 4.1(c) schematic representations of drug release mechanisms of pH responsive polymers (A) Collapse of the polyanion makes the liposomal membrane leaky and promotes efflux of the drug from the liposomes. (B) Protonation-induced (left) and ionization-induced (right) destabilization of multimolecular PMs. (C) Ionization-induced swelling leads to drug release from cross-linked polymeric nanospheres *(Reproduced from A.E. Felber et al)*[25].

4.1.4 pH RESPONSIVE WATERBORNE POLYURETHANES

Only few reports are available where such studies have been carried out, especially in the field of polyurethanes. However, each study involved a messy synthesis route, including synthesis of tripeptides [7,26]. The theme of their study was based on incorporation of hydrolyzable moiety in the PU structure, which can hydrolyze at acidic pH resulting in degradation of polymer, leading to drug release. In that case, the study of non-cytotoxicity of degraded materials becomes essential. Also, up to our knowledge, no reports are available which describe waterborne PUs with pH responsive characteristics.

As discussed in previous chapters, we have synthesized biocompatible carbohydrate crosslinked PUs [27–29]. Towards its more meaningful applications, we have modified it with acid chain extenders in order to make pH-sensitive waterborne PUs. To overcome difficulties associated with polymer degradation and harmful effects implied by its byproducts, we focused on preparation PUs with swelling controlled drug release, rather than degradation based drug release. We have also used biocompatible and non-toxic raw materials such as Polycaprolactone as polyol [30] and cellulose as crosslinker.

It has been known that polymers containing pendant acidic or basic groups either accept or donate a proton depending on the pH of the environmental media [31]. Considering this, we incorporated pendent acidic groups during synthesis of PUs by using α-hydroxy acids as chain extenders. The selection of α-hydroxy-acids like lactic acid, glycolic acid and dimethylol propionic acid for incorporation in the structure of PUs was done on the basis of the nature of hydroxyl groups. Moreover, the difference in their pKa values, chemical structure, polarity and affinity towards reaction with NCO of isocyante allows detecting the critical parameters towards the drug release efficiency of the resulting PU. The PUs synthesized herein possesses several advantages to be used in biomedical field as summarized above.

Owing to special characteristic of targeted drug delivery above pH 7 with respect to pH changes in GIT (gastro intestinal tract) passage, the use of pH sensitive polymers has found an important application in the field of colon specific drug delivery[32] based on pH sensitive nature of various polymers like N-(2-carboxybenzyl)chitosan [23], polyurethanes [33] and starch/methacrylic acid (MAAc) copolymer hydrogels [34]. The drug release specifically at pH 7.5 is useful for vaginal drug delivery [25] because of the pH changes occurring during sexual

intercourse[35,36]. Hence the polymer that can sense the difference in such pH range can be used as drug delivery systems such as microbicides and vaginal passaries for prevention of HIV-AIDS and other sexually transmitted diseases. PUs have been successfully used as intra-vaginal rings [37] and colon specific drug delivery systems [33,38]. Towards such applications, we successfully attempted synthesis of smart polyurethanes having stimuli responsive character with respect to pH, more specifically showing higher release at pH 7.4.

Felodipine (Fig. 4.2) was selected as a model drug for the drug release studies. Felodipine has a benefit of being selective as vasodilator and less cardiac effects as compare to other non-dihydropyridine calcium anatagonists. However, its low water solubility and poor bioavailability (as low as 15% after oral administration) are major concerns limiting its application. The controlled drug delivery systems are expected to improve its bioavailability at the site of action avoiding the first-pass metabolism [39].

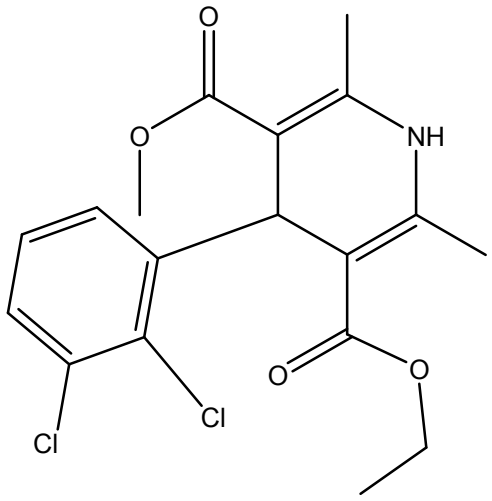


Figure 4.2 Structure of Felodipine

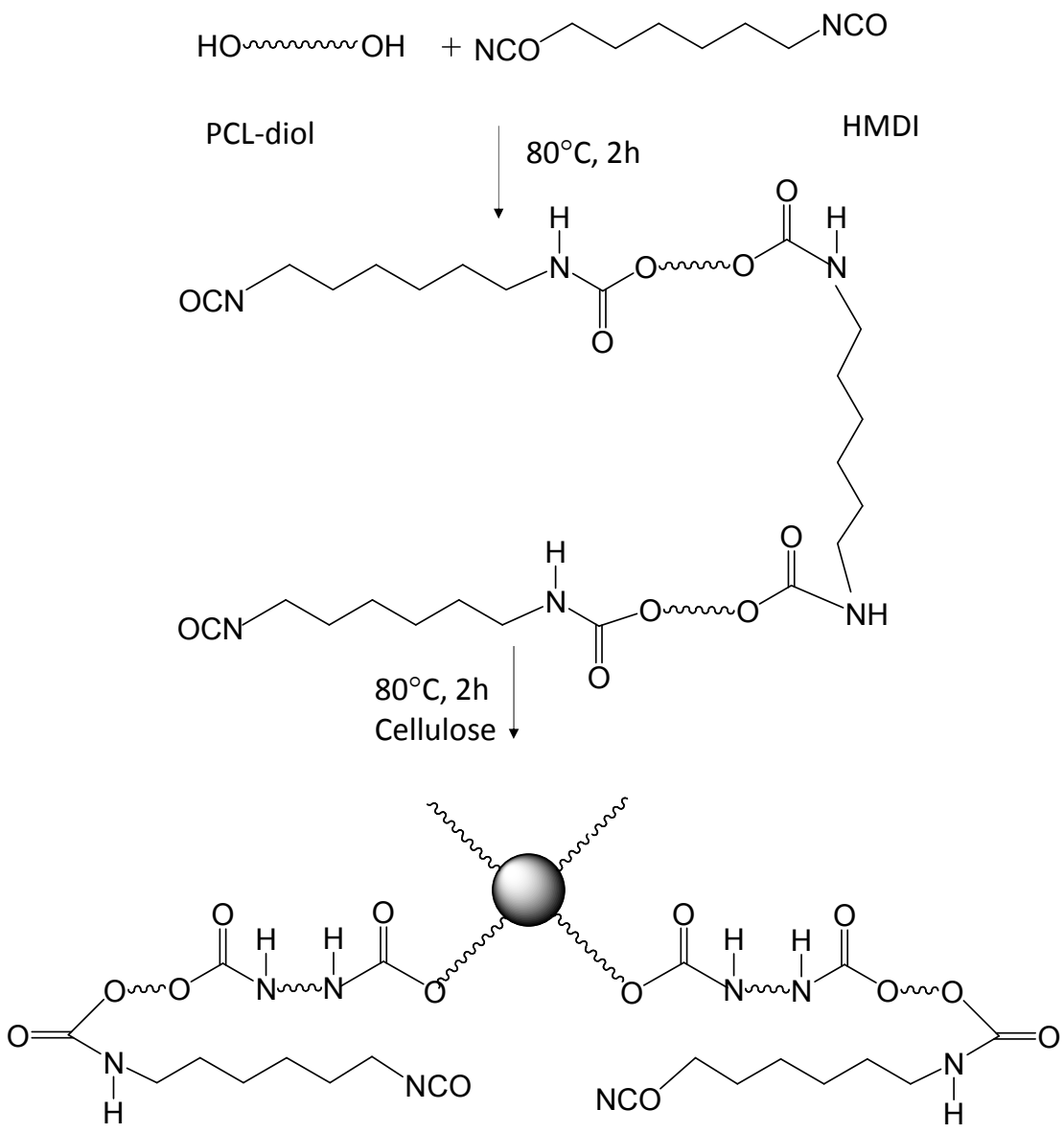
4.2. MATERIALS AND METHODS

4.2.1 MATERIALS

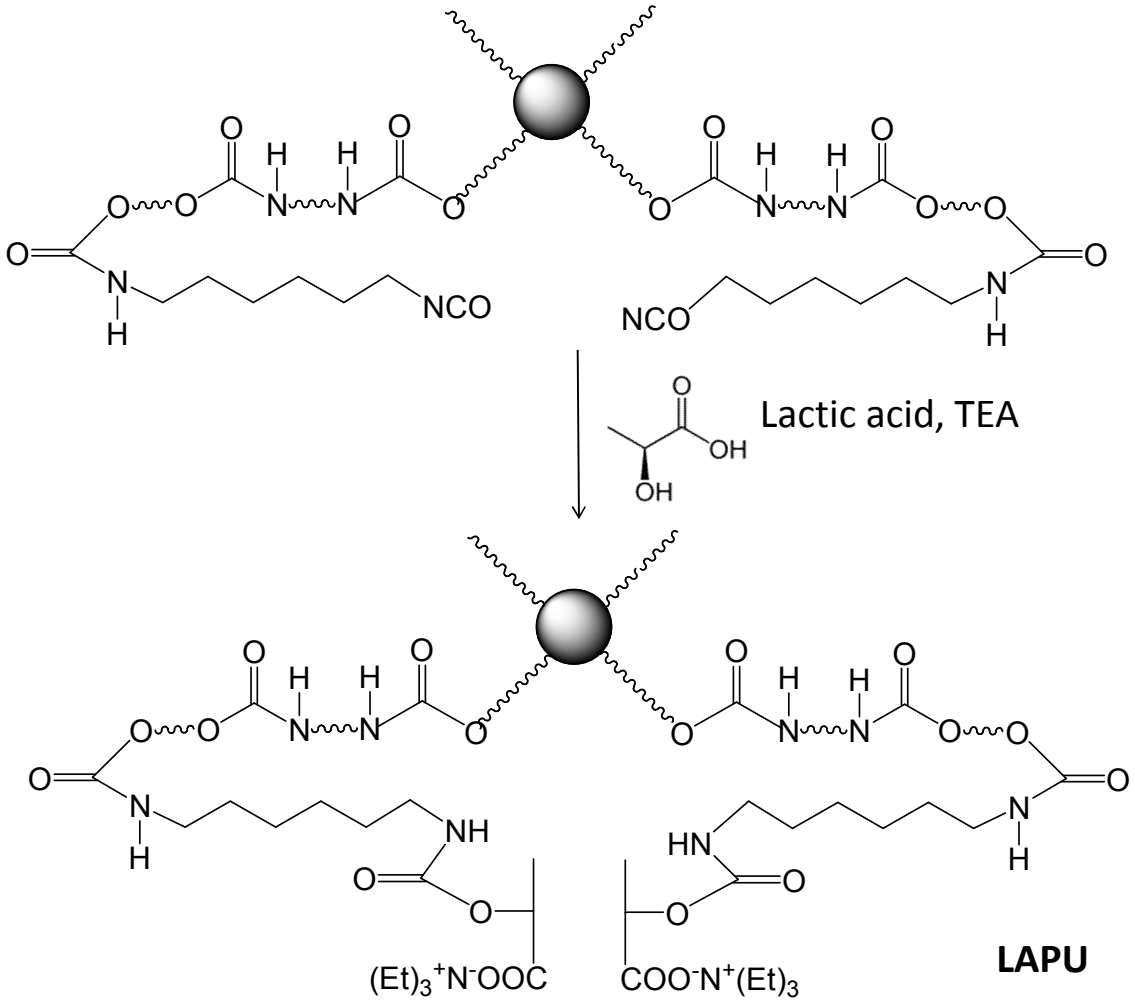
Polycaprolactone diol (PCL-2000), Hexamethylene diisocyanate (HMDI), dibutyl tin dilaurate (DBTDL), triethyl amine (TEA) and cellulose were purchased from Sigma Aldrich, India. PCL 2000 and cellulose were dried under vacuum at 80°C for 24h prior to use. Tetrahydrofuran (THF) was purchased from Qualigens, Bombay, India. THF was purified by distillation. Felodipine was kindly donated by Alembic Ltd, India and it was used directly as received. The buffer solutions of pH 1.2 and 4.5 were prepared as per method reported elsewhere [40]. Sodium chloride, Cetyl trimethyl ammonium bromide, potassium dihydrogen phosphate, and phosphate buffer saline tablets (for preparation of pH 7.4 buffer solution) were obtained from Sigma Aldrich, India. Hydrochloric acid was donated by GNFC Ltd, India.

4.2.2 SYNTHESIS OF WATERBORNE PU FILMS

In a 500mL five-neck reaction kettle fitted with a stirrer, a thermometer jacket, water condenser and nitrogen inlet, 25gm of PCL 2000 was charged. HMDI (5.65gm) was added drop wise over time interval of 15 min.



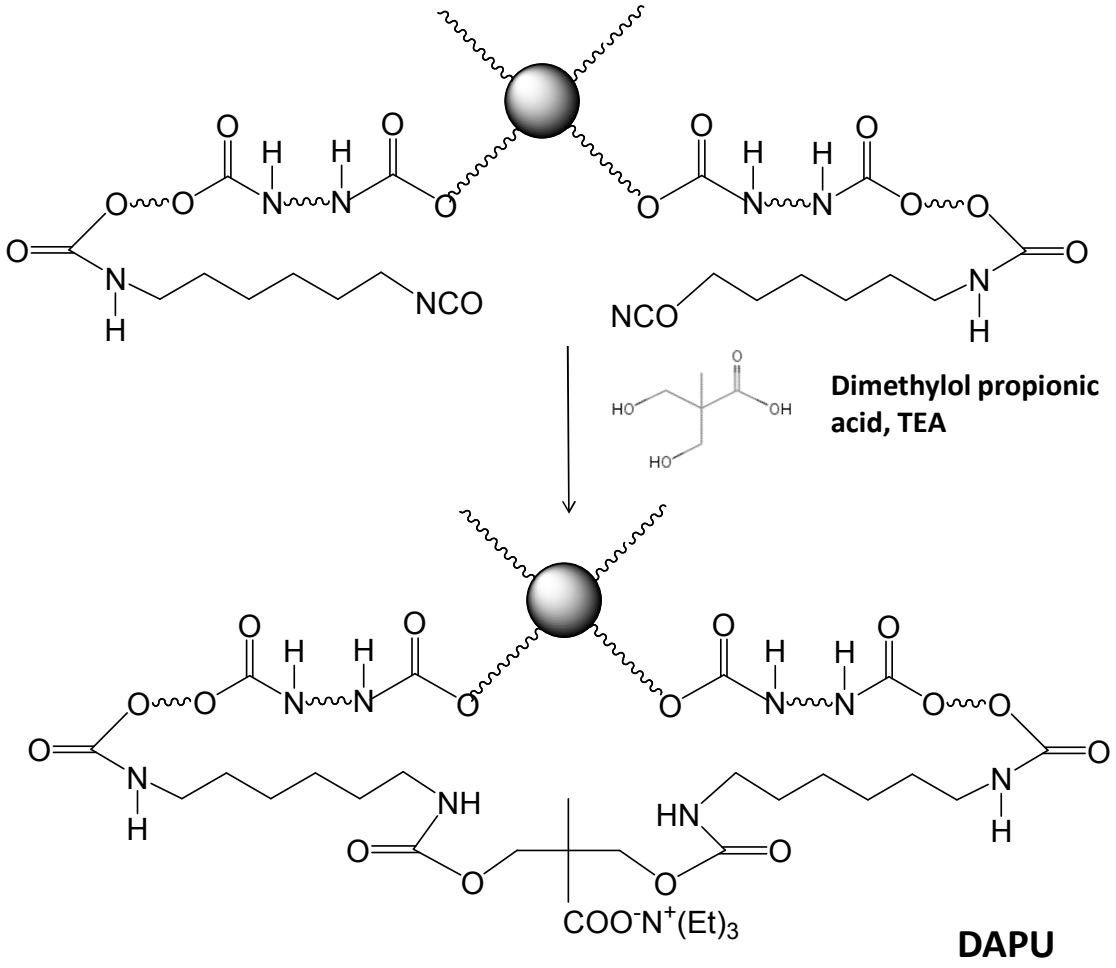
Scheme 4.1 Synthesis route for carbohydrate crosslinked PU, corresponds to carbohydrate with attached PU chains.



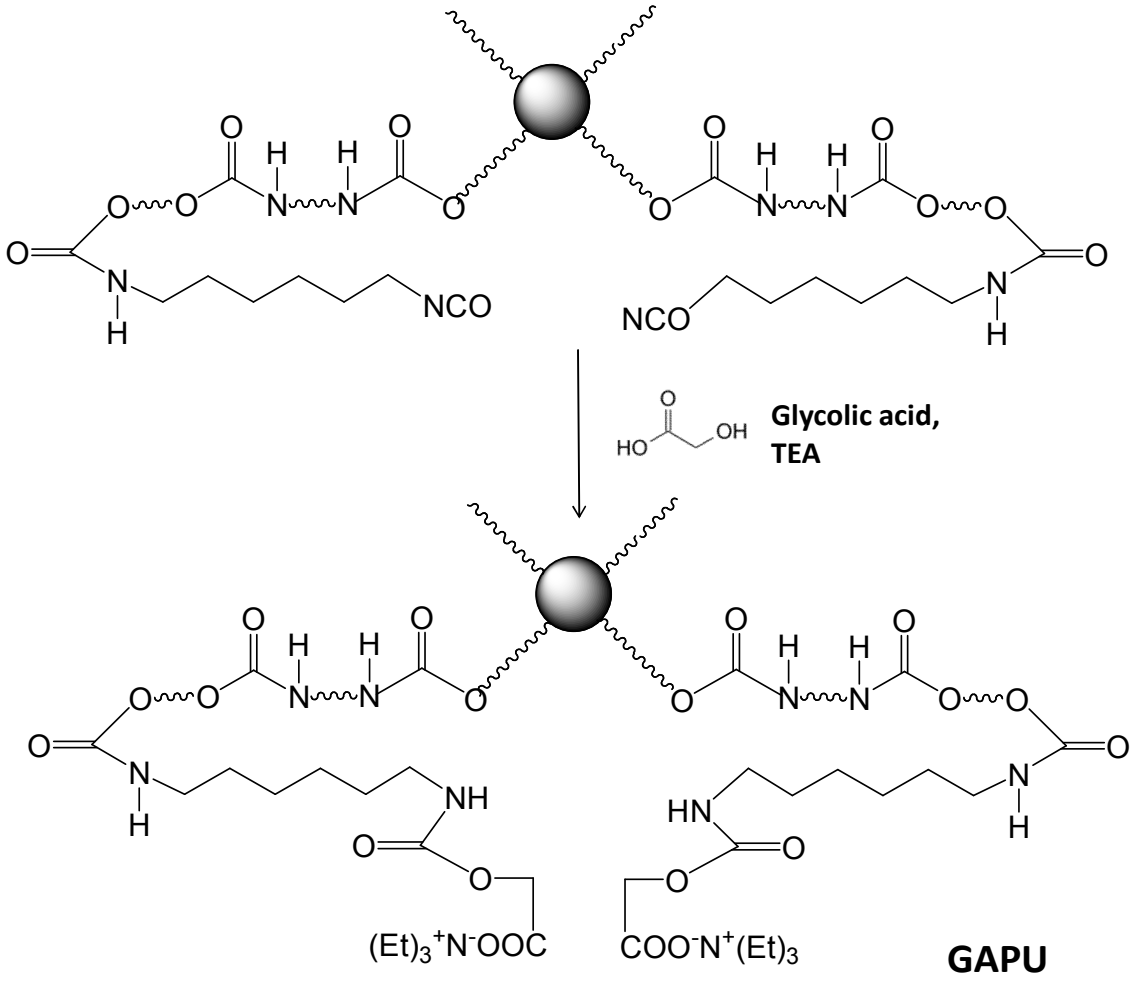
Scheme 4.2(A) Synthesis route for LAPU

The reaction was allowed to proceed at 80°C for 2h under nitrogen atmosphere. Cellulose (0.575gm) was dispersed in THF, and the resulting dispersion was added drop wise to the reaction mixture. After mixing the contents for 2h at 80°C, the calculated amount of acid chain extender and 40mg of DBTDL was added. The reaction was continued for another 1h at 80°C. The contents were cooled to room temperature and a calculated amount of TEA was added to neutralize carboxyl groups of acid, followed by 30min stirring. The dispersion thus obtained was added drop wise in 500mL distilled water, resulting in 30% W/V PU emulsion. The PU emulsion

was casted on the glass molds at room temperature and allowed to dry for 8days. Afterwards the films were kept in a vacuum oven at 60°C for 24h and stored in desiccators till further use. The synthetic route of PUs is outlined in Scheme 4.1 and Scheme 4.2. The mole ratio PCL/HMDI/cellulose/acid chain extender was kept as 1/2.7/0.5/1.



Scheme 4.2(B) Synthesis route for DAPU



Scheme 4.2(C) Synthesis route for GAPU

4.2.3 CHARACTERIZATION

The polymers were characterized for thermal, morphological and structural properties as follows. The FTIR spectra of the PUs were recorded on a PerkinElmer IR spectrophotometer at room temperature. For this purpose, the 2% (W/V) solution of samples in dimethyl acetamide was prepared. It was cast onto KBr pellets followed by drying at 40°C for 24h. Afterwards, the pellets were dried under vacuum at 60°C for a further 24h in order to remove the residual solvent. The pellets thus prepared were used further for FTIR analysis. TGA was carried out on

TG–DTA 6300 INCARP EXSTAR 6000 in temperature range of 30–450°C in nitrogen atmosphere; at a heating rate of 10°C/min. Differential scanning calorimetry (DSC) thermograms were recorded on a NETZSCH DSC at a rate of 10°C /min under 30–40mL/min gas flow rate of nitrogen. The temperature range of -100 to 100°C was selected for DSC analysis. The analysis was carried out under both cooling and heating cycles. For SEM analysis, JSM 6380LV, JEOL was employed. The samples were cryogenically fractured and the morphology was studied at different magnifications after coating samples with a thin layer of gold.

4.2.4 SWELLING STUDIES

The swelling studies were carried out as per reported method [41] in buffer solutions of pH 1.2, 4.5 and 7.4 at 37°C. Pre-weighed polymer films were kept in 50mL swelling medium for 24h (considered to be time required for complete equilibration swelling) in individual vials. The films were then blotted with tissue paper to remove excess droplets from surface and reweighed. The Equilibrium swelling (Q) was calculated as per following equation.

$$Q = \left(\frac{W_2 - W_1}{W_1}\right) \times 100 \text{---------- (1)}$$

Where W_1 denotes initial weight of the polymer before swelling and W_2 is the weight of polymer after equilibrium swelling. The swelling experiments were performed in triplicate and the mean value of Q was considered for the present study.

4.2.5 DRUG LOADING AND RELEASE FROM PUS

Felodipine was incorporated into the PU films by following solvent evaporation technique [42]. A known weight of the drug was dissolved in THF. The homogeneous drug solution thus obtained was added to 5wt% solution of PU in THF. The drug-polymer mixture was sonicated

for 2h. The resulting blend was cast on pre-dried glass plate and allowed to dry at room temperature for 48h. It was again dried under vacuum at 40°C until constant weight to confirm complete removal of residual solvent. Thus we obtained PU loaded with 5wt% of Felodipine. The films were trimmed in order to obtain disc shaped films with 1cm diameter and 100 μm thicknesses. The loading efficiency (%LE) of Felodipine in the PU film was determined spectrophotometrically from the calibration plot constructed at 362 nm after extensive extraction of drug loaded film in ethanol. Loading percentage was calculated by using following equation [29].

$$\% \text{ Loading } = \frac{\text{Wdrug}}{\text{Wpolymer+ Wdrug}} \times 100 \text{ ---------- (2)}$$

Where W_{drug} is weight of drug incorporated into PU and $W_{polymer}$ is weight of PU film before drug loading. The loading efficiency was found to be similar for all PUs, i.e. 5.0562%, 5.0532% and 5.0548% for LAPU, GAPU and DAPU respectively. The drug release study of individual film was carried out in PBS with varying pH. A known weight of PU film was kept in individual vials containing 50mL PBS of pH 1.2, 4.5 and 7.4. The buffers were thermostated at 37°C in a shaking water bath. Since Felodipine is reported to be a light sensitive drug, the system was well protected so as to avoid exposure to light. At a certain time interval, 3mL of aliquots were taken out to measure the amount of drug released. A corresponding quantity of thermostated buffer was added to respective vials. The absorbance of each aliquot was measured at 362 nm and the concentration was measured using the calibration plot constructed for Felodipine at 362 nm on UV spectrophotometer. A similar quantity of corresponding PU without drug, for each polymer, was also kept in respective buffer solution under similar conditions. The absorbance from this polymer was determined for each measurement and it was considered as a blank. The obtained

value of absorbance for blank was subtracted from the absorbance value of corresponding drug loaded polymer for all the observations. This was done to ensure that the observations are not affected by the absorbance of any unwanted material leaching out from the polymer during release experiments

4.2.6 DEGRADATION STUDIES

The degradation studies of PU films were carried out in alkaline solution in order to carry out the hydrolysis of films [42]. The PU films were trimmed in a disc with 0.3mm diameter. Each film was weighed and kept in a vial containing 10mL 1M NaOH solution. The vials were thermostated at 37°C in a bacteriological incubator. The films were taken out at a specific time intervals, dried until constant weight and evaluated for its weight. The films were then kept in respective vials. The study was carried out for 90days. A plot was constructed for %weight loss of PU films against time in order to observe degradation. The measurements were carried out three times and mean values were considered for study.

4.3. RESULTS AND DISCUSSION

4.3.1 SYNTHESIS OF WATERBORNE PU FILMS

The synthesis of PU films was carried out by prepolymer method. The reaction between PCL-diol and excess HMDI led to NCO terminated prepolymer which was allowed to react with cellulose to form a crosslinked PU network as outlined in scheme 4.1. The polymer thus obtained still contained excess NCO groups which can react further with hydroxyl group/s of acid to yield an acid terminated PU. TEA is added at this stage to neutralize the acid groups. The emulsion is poured in the distilled water in order to obtain waterborne polyurethane. Here it is essential to note that the acids contain both hydroxyl and carboxyl groups. Both the groups have a tendency

to react with NCO groups. Since the reaction rate of primary hydroxyl groups is 2.5 times higher than that of carboxylic groups [43], we hypothesized that the extent of reaction between carboxyl groups and NCO groups is negligible. The synthesis procedure explained above, thus led to PU having the structure as elaborated in scheme 4.2. Three different PUs using α-hydroxy acids, namely lactic acid, glycolic acid and dimethylol propionic acid as chain extenders were synthesized and labeled as LAPU, GAPU and DAPU respectively.

4.3.2 CHARACTERIZATION

4.3.2.1 FTIR STUDIES

As shown in Fig. 4.3, the structure of synthesized PUs was confirmed by FTIR spectra. Absorption peak corresponding to the NCO group at 2270 cm−1 was absent which assured that there is no untreated isocyante in the synthesized PUs. Since all the PUs prepared herein are carbohydrate crosslinked PUs with acid chain extenders, we found similar spectra for all PUs. The two splitting absorption bands at 1700-1710 cm−1 and 1745-1755 cm−1 could be assigned to a carbonyl group for hydrogen-bonded and free carbonyl groups, respectively as cited by Reddy et al [44]. The formation of urethane linkage (NH-CO-O) can be assured by C-O band and N-H stretch. In the present study, the appearance of the C–O band at peak value 1644 cm−1 and strong absorption peak centered at around 3312-3340 cm−1 for hydrogen bonded N-H stretch confirmed the formation of urethane linkage. However, it can be concluded that no free N-H was present in our PUs since no peak was observed for free N-H as reported by Liu that could appear around 3420 cm−1 [45]. Since the acid chain extenders were used, hypothesizing that –COOH group will remain unreacted, we should get an absorption peak corresponding to it. The absorption peak obtained at 1680 cm−1 corresponding to –C=O group of acid and shoulder

peak at around 3500 cm−1 which could arise due to the OH group of acid confirms presence of acid group supporting our hypothesis. The absorption bands at 2904 and 2863 cm−1 were attributed to -CH2- stretching vibrations. The appearance of shoulder peak at around 3500 cm−1 could arise due to the OH group of acid.

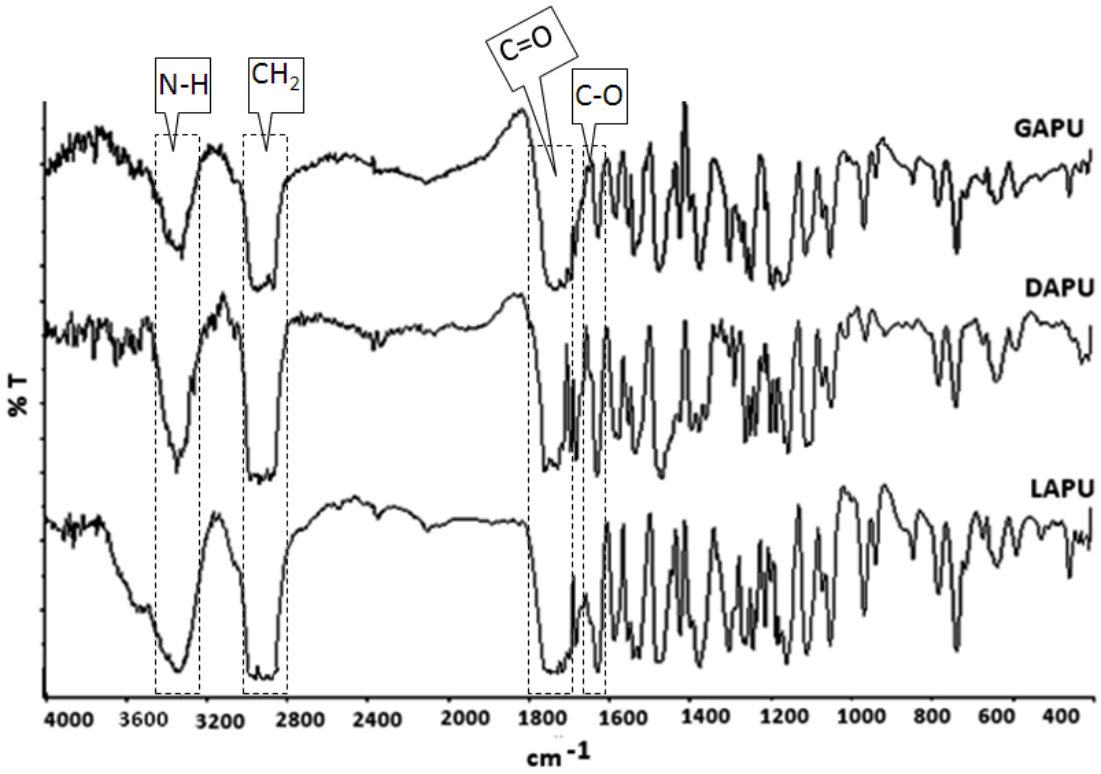


Figure 4.3 FTIR spectra of synthesized PUs

4.3.2.2 THERMAL ANALYSIS

The PUs were analyzed for thermal degradation behavior using TGA (Fig. 4.4(a)). As reported in our earlier study, despite the use of carbohydrate with low thermal stability, in the present case also, the PUs showed reasonable thermal stability. The use of acid chain extender did not show significant difference in thermal stability as all the three PUs behaved similarly with respect to degradation in function of temperature. The initial weight loss observed could be attributed to

loss of volatile contents such as water. The later degradation was attributed to the degradation of urethane bonds that may result in dissociation in isocyanate and alcohol, the formation of secondary amine and carbon dioxide or formation of primary amine and Olefin, as cited by Rueda-Larraz et al [44].

Table 4.2 Glass transition temperatures for PUs

Polymer	T_{gSS}	T_{gHS}
LAPU	-33.3	56.8
DAPU	-5.8	50.9
GAPU	-1.4	48.2

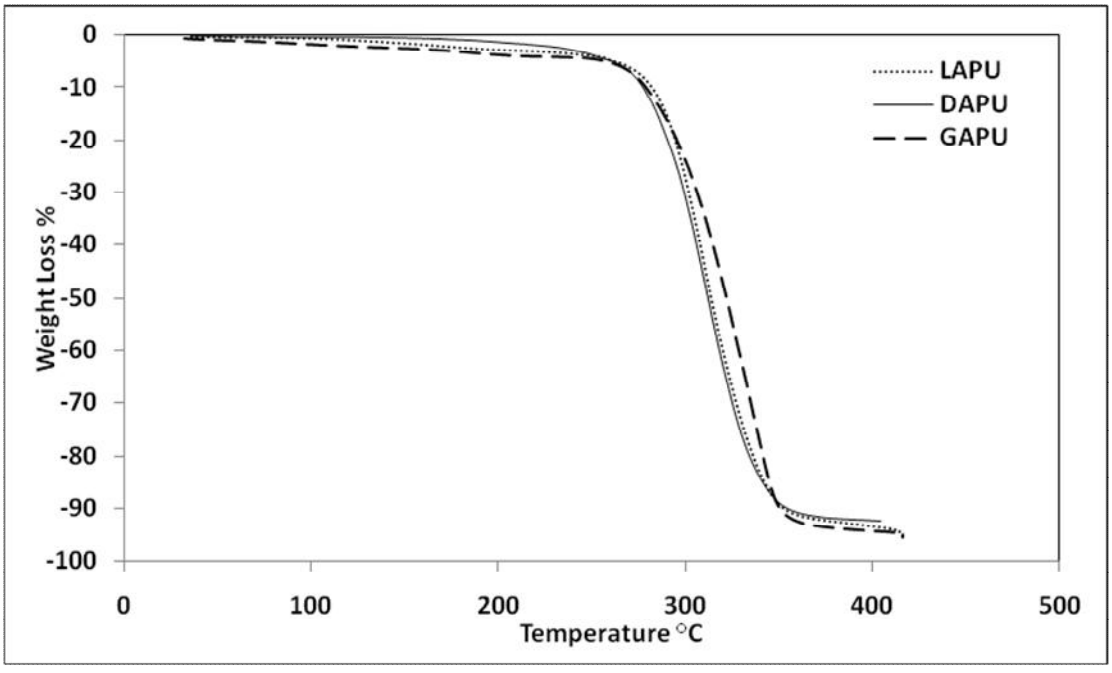


Figure 4.4(a) Thermal degradation plots of PUs

The DSC analysis revealed the glass transition temperature of all PUs. The DSC analysis was performed both under heating and cooling cycle. The glass transition temperature was considered as a midpoint of the glass-transition process (Table 4.2). The peaks at lower temperatures as shown in Fig. 4.4b-(A) were assigned to glass transition temperature of soft segments (T_{gSS}).

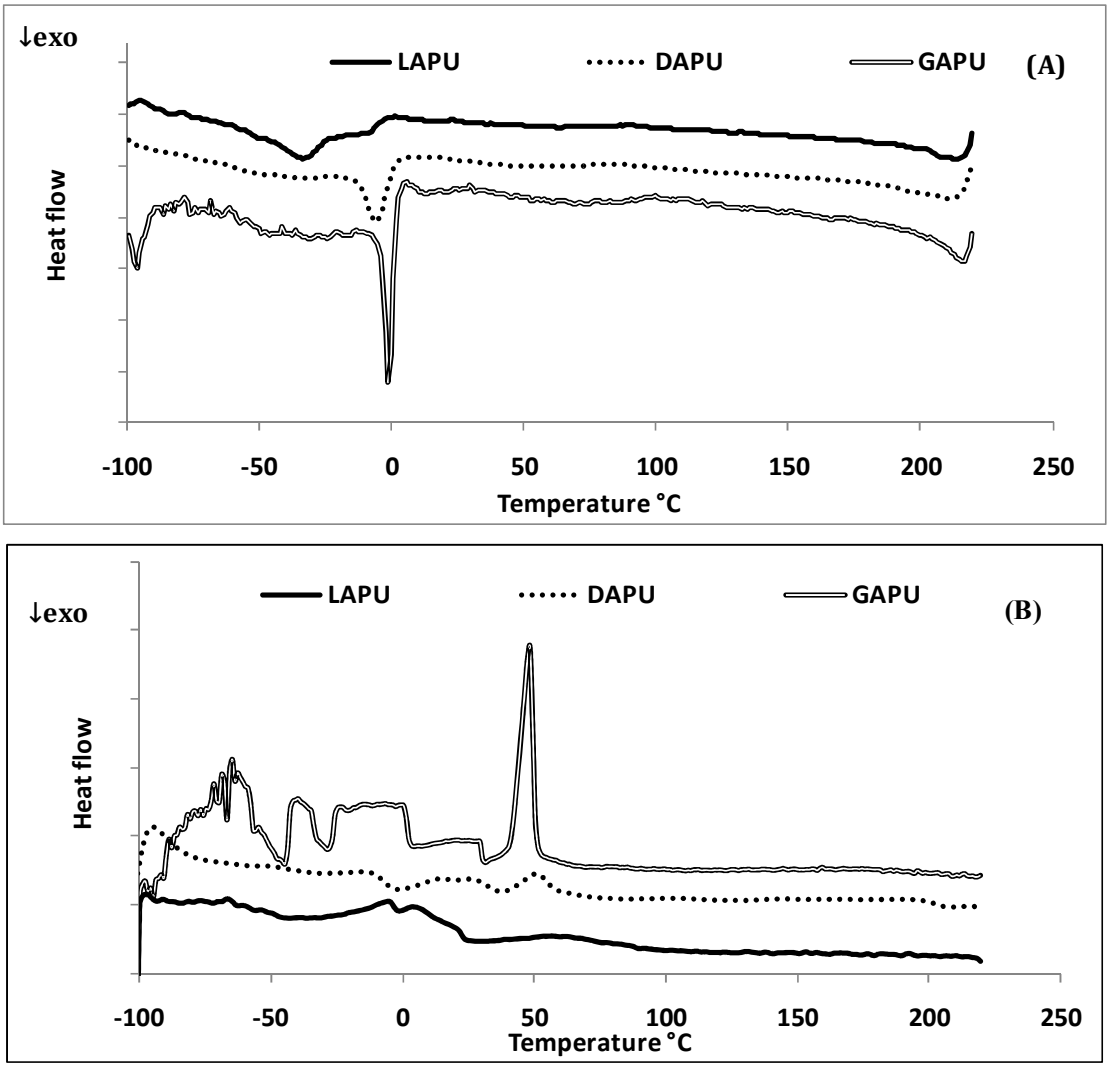


Figure 4.4 (b) DSC thermograms of PUs

On the other hand, the peaks obtained at higher temperatures were assigned to glass transition temperature of hard segments (T_{gHS}) as shown in Fig. 4.4b-(B). It was observed that T_{gSS} for PUs

was highest for GAPU, followed by DAPU and LAPU. This can be explained on the basis of the fact that the glass transition temperature of PUs depends strongly on the molecular mobility [44].

The polyurethane with high crosslinked structure results into the lower mobility of chains resulting into high Tg. Also, reactivity of NCO towards the primary hydroxyl group is about 3.3times higher than that of secondary hydroxyl groups [43]. In case of GAPU, the primary OH group available on the glycolic acid molecule could have reacted faster as compared to a secondary OH group of lactic acid resulting in a more crosslinked structure of GAPU as compare to LAPU. While in case of DAPU, though there are two hydroxyl groups available in dimethylol propionic acid, one primary and other secondary, the crosslinking could not be as efficient as GAPU, may be due to steric hindrance offered by both hydroxyl groups to each other. Amongst LAPU and DAPU, even though under the effect of steric hindrance, a primary OH of dimethylol propionic acid is more reactive as compared to a secondary OH group of lactic acid, providing a more crosslinked structure of DAPU as compared to LAPU. The crosslinking density in synthesized PUs thus is hypothesized to be in order of GAPU > DAPU > LAPU. Hence the above findings of glass transition temperature could be explained on the bases of structure-property relationship as explained above.

4.3.2.3 MORPHOLOGY

Fig.4.5(a) shows SEM images of pure and drug loaded films for LAPU and DAPU at different magnifications. The difference that can be observed at first sight between pure polymer for LAPU and DAPU is that LAPU showed uniform surface with some exceptional appearance of white dots while DAPU showed a string like structure along with bigger particles. The particles can be seen due to some part of unreacted cellulose particles that could be leached out during

cryogenic fracture. Also, this can be compared with "sea-island structure" which can result in higher phase separation between soft and hard segments of PU as proposed for the PUs with cellulose whiskers [46] and PUs with cellulose nanocrystals [47]. Hence LAPU, showing uniform morphology assures our assumption of well crosslinked structure as compared to DAPU. The drug loaded films of both LAPU and DAPU showed the presence of drug in the form of particles distributed non-uniformly on the surface of the film [48].

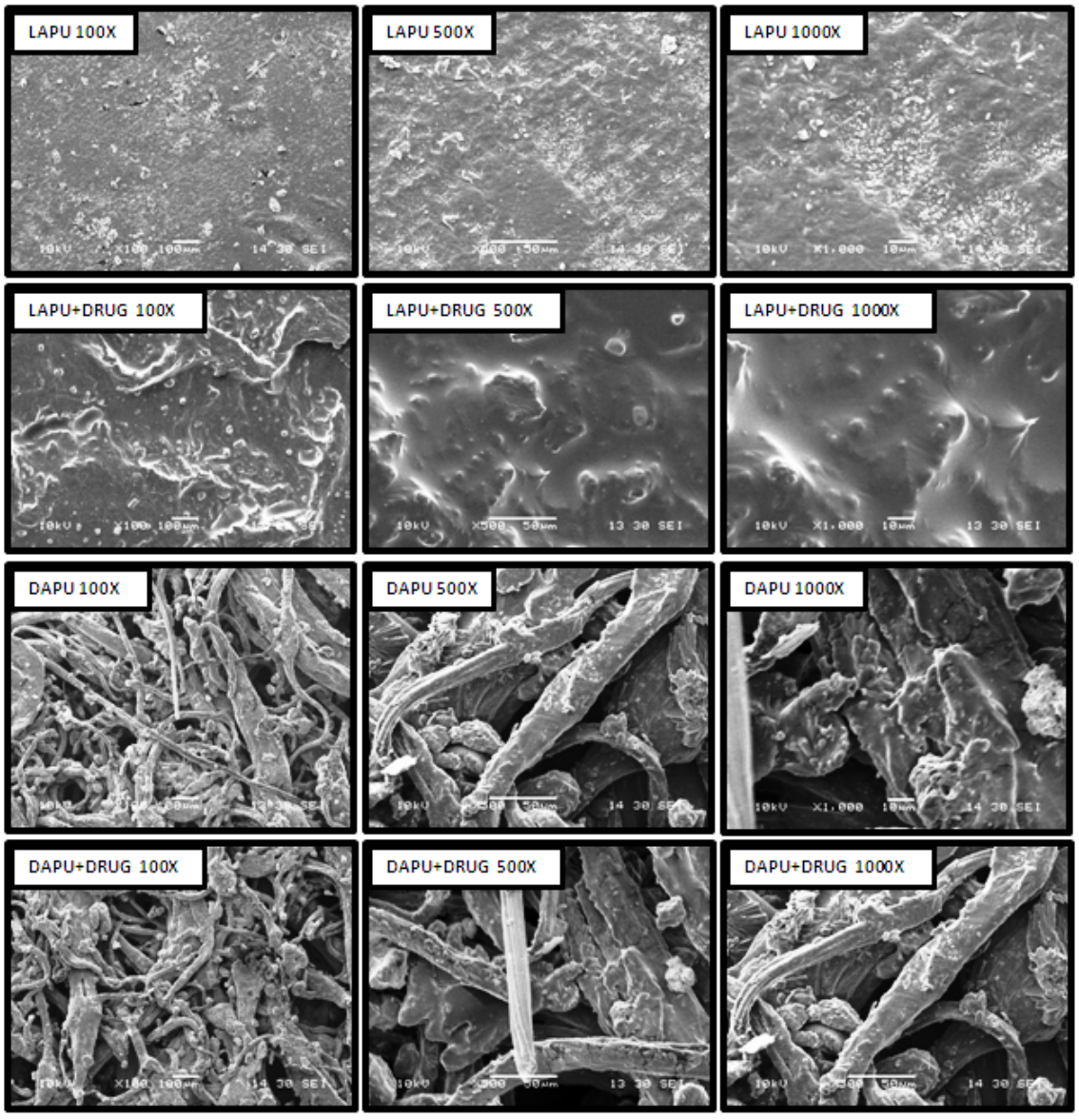


Figure 4.5(a) SEM micrographs of pure and drug loaded PUs

Fig. 4.5(b) shows the morphology of polymers after release of drug. In order to observe the effect of drug release on the polymer with respect to pH of the surrounding medium, LAPU was selected for SEM studies. After drug release experiment, morphology of LAPU in buffer solution with all three different pH values was observed under SEM. The images revealed that the polymer surface was severely damaged after drug release, may be due to the vacated space after swelling of polymer and drug release.

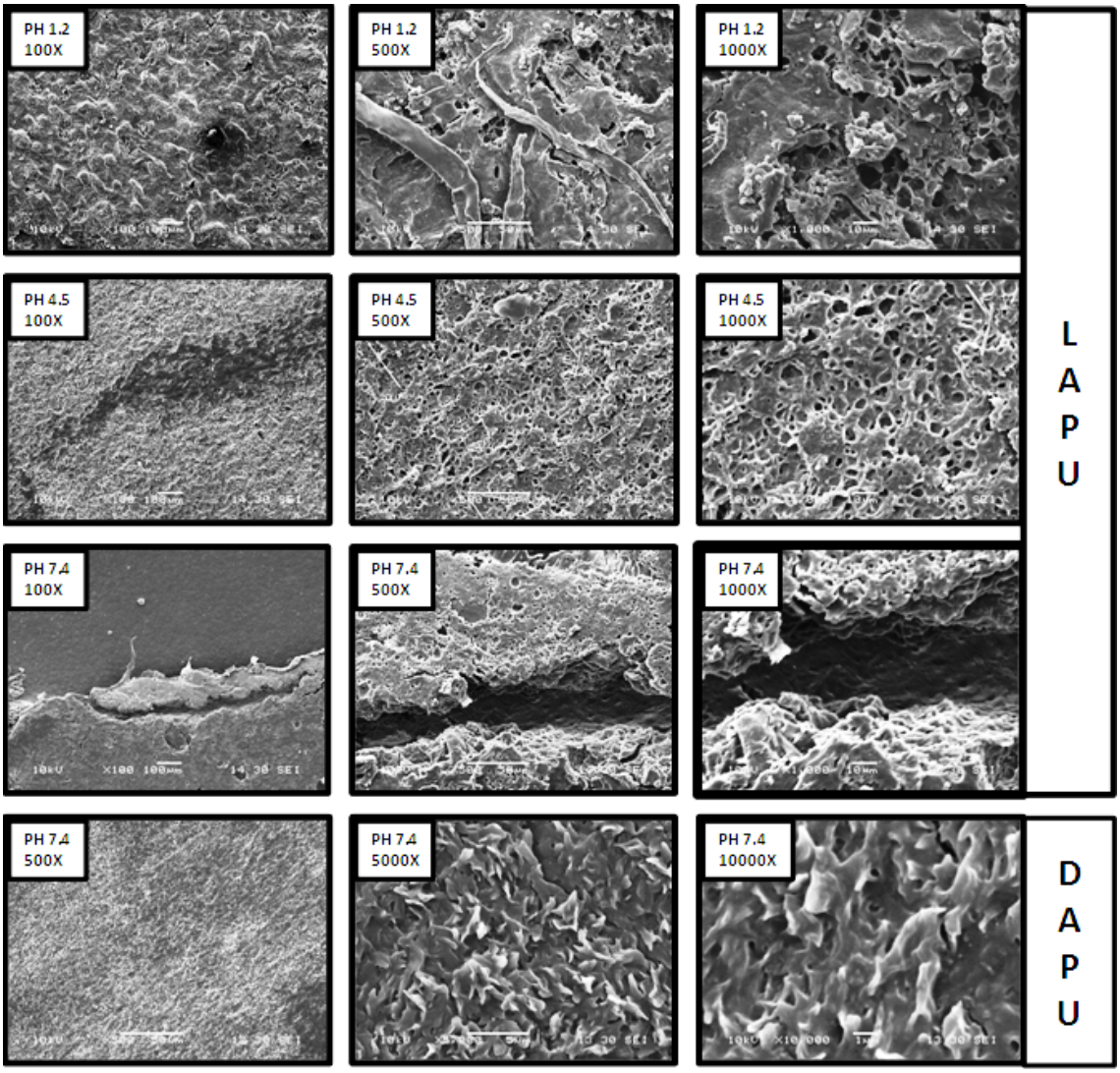


Figure 4.5(b) SEM micrographs PUs after drug release

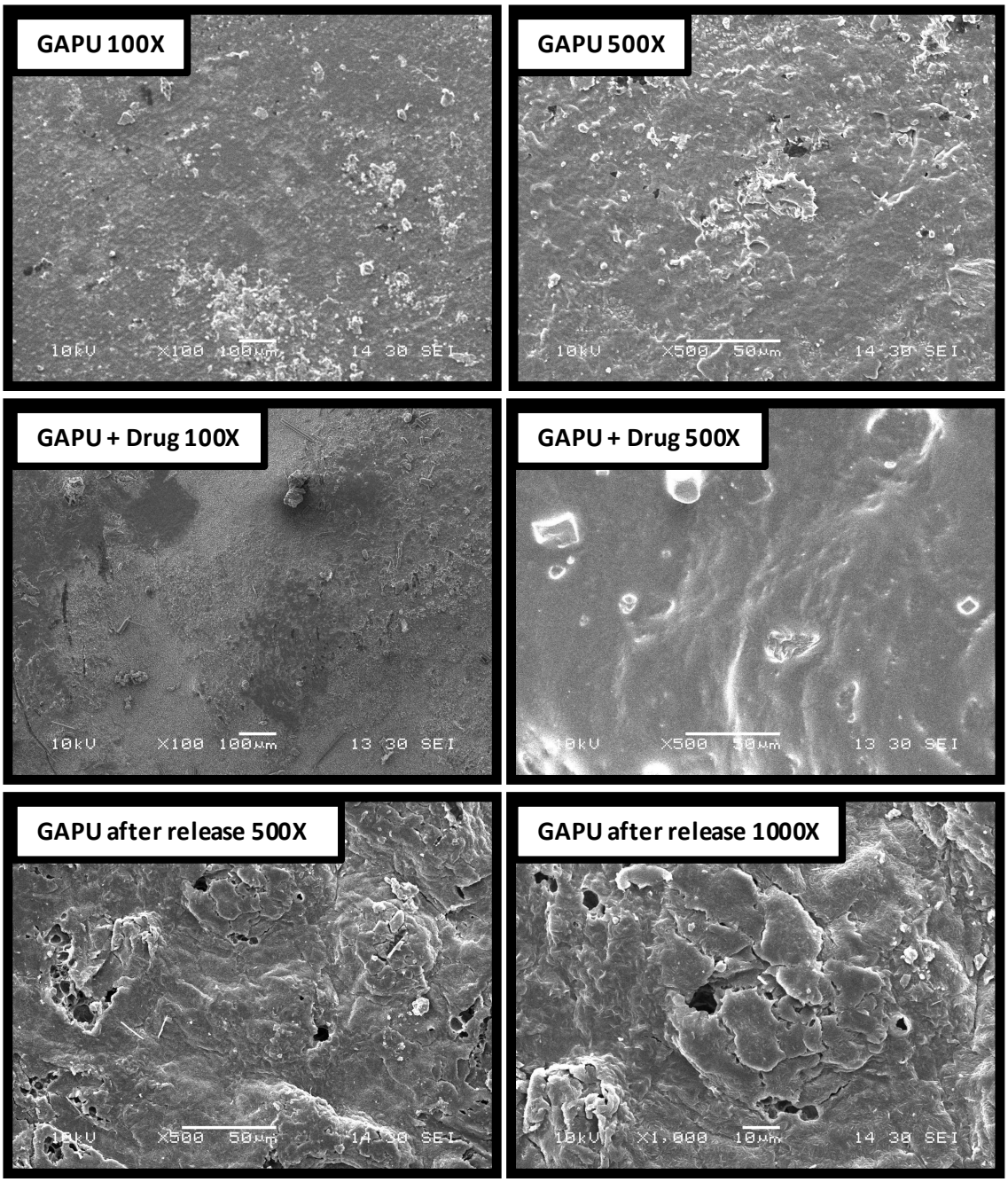


Figure 4.5(c) SEM micrographs of GAPU (Pure, drug loaded and after drug release

The important observation was that as the pH value of external media increased from 1.2 to 4.5 to 7.4, the severity of damage increased. As can be seen in Fig. 4.5(b), for LAPU with external medium pH 1.2, small pores were observed on polymer surface. With the increase in pH, the pores grow bigger and deeper reaching ultimately from the surface to the core of polymer (at pH 7.4). This might be due to higher rates of diffusion of drug at pH 7.4 from the PU. The similar surface damage was observed in case of DAPU and GAPU as shown in Fig. 4.5(b) and Fig. 4.5(c) respectively.

4.3.2.4 SWELLING STUDIES

The polyurethanes prepared herein having α-hydroxy acids represent anionic polymer with obvious pH-sensitive nature. According to swan et al, the degree of swelling of ionic polymers depends on several factors like charge on the polymer, its concentration, crosslink density and hydrophilicity of polymer [49]. The swelling behavior of synthesized PUs hence depends mainly on crosslink density and concentration of acidic groups on the PU, which varies depending on the type of acid chain extender. As shown in Table 4.3, the equilibrium swelling of PUs increases with increasing pH of a buffer solution. This is an obvious phenomenon for most of the carboxyl groups on the polymer surface remain unionized at acidic conditions. Under such stimuli, there is very little chance of electrostatic repulsion between acidic groups. Hence very little swelling was observed at pH 1.2. However, as pH changes to basic condition, the carboxyl groups get ionized leading to increased charge density [50]. The change in pH value leads to increased swelling of anionic polymers due to two important phenomena that occurs simultaneously. First, the increase in the charge density due to ionized acidic groups leads to

high cationic concentration differences between inner and outer surface of polymer which results in increased osmotic pressure between polymer and external solution. This ultimately causes easier penetration of external solution in the polymer network resulting in higher swelling. A second phenomenon is electrostatic repulsion amongst anions on polymer surface which also fasten the expansion of polymer chains leading to higher swelling.

Table 4.3 Swelling coefficient and drug release kinetics' constants for PUs

Polymer	pH value	Q∞	n value	k value
LAPU	1.2	2.8366	0.19	0.87
LAPU	4.5	3.2133	0.50	0.19
LAPU	7.4	6.4627	0.32	0.39
DAPU	1.2	2.5362	0.46	0.20
DAPU	4.5	2.9862	0.44	0.26
DAPU	7.4	5.6932	0.37	0.34
GAPU	1.2	1.3694	0.44	0.22
GAPU	4.5	2.1563	0.49	0.23
GAPU	7.4	5.1236	0.21	0.53

4.3.2.5 DEGRADATION STUDIES

The degradation pattern of PUs in 1M NaOH is plotted as %degradation of PUs against time and represented in Fig. 4.6. All the PUs were observed to be degradable with degradation of 41.41%, 22.84%, and 12.58%for LAPU, DAPU and GAPU during 90 days of time respectively. The degradation rate depends on crosslinking density and hydrophilicity of the polymer [51]. The fashion of degradation rate in the present case is very obvious since the crosslinking density decreases as we move from GAPU to DAPU to LAPU as discussed above. The degradation is also attributed to PCL soft segments which get hydrolysed in alkaline conditions. As per a previous study [52], the alkaline hydrolysis of PCL soft segment lead to introduction of OH and COOH groups on the periphery of polymer giving hydrophilic substrates that can result in increased degradation. The alkaline hydrolysis of polymers is advantageous in the context that it can form pores on polymer surface which is an important feature for biomedical applications [52].

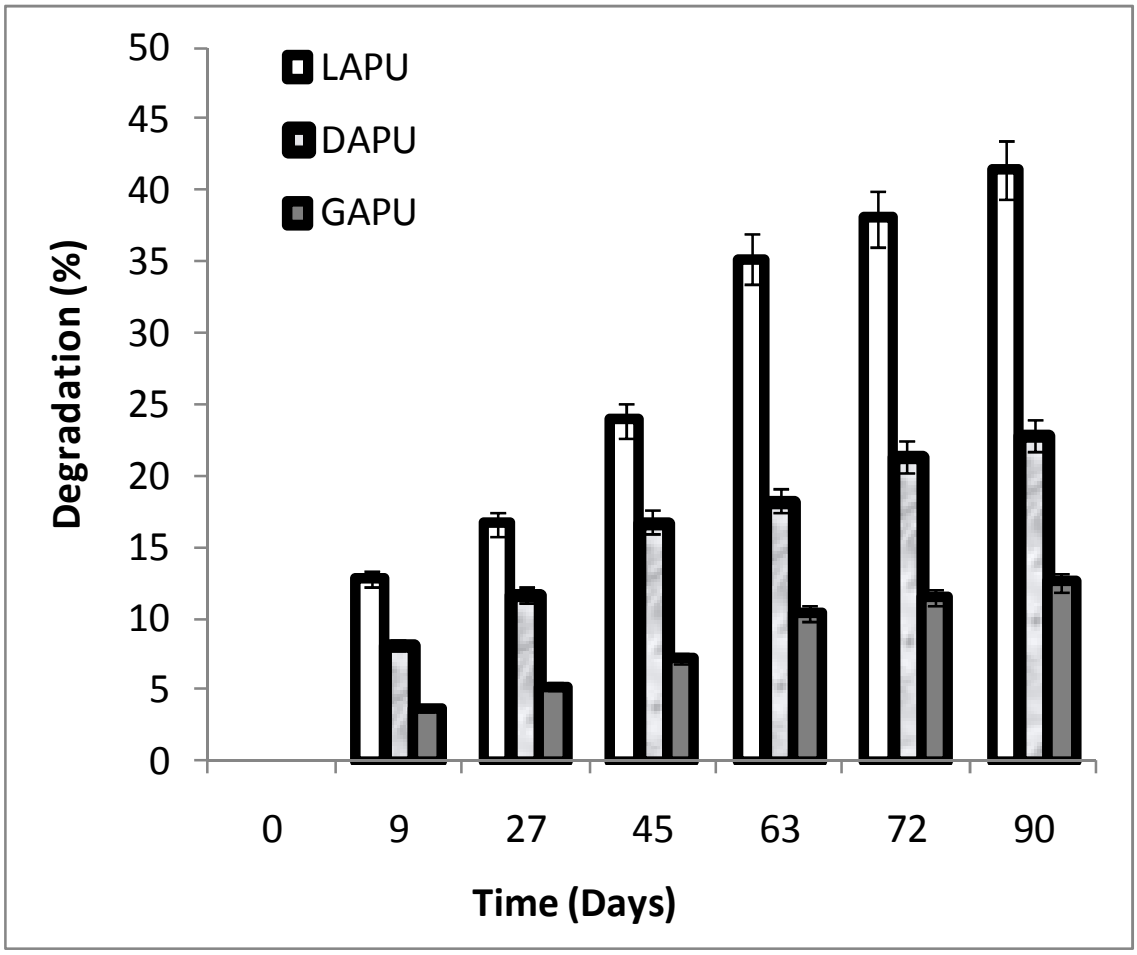


Figure 4.6 Degradation plot of PUs in 1M NaOH solution

4.3.2.6 DRUG RELEASE STUDIES

The drug release study was carried out at three different pH values i.e. 1.2, 4.5 and 7.4 for all the polymers. The experiment was performed thrice and data were collected. Mean values were considered to plot a graph of cumulative drug release vs. time. The rate of release of the drug was increased when the pH of buffer increased from 1.2 to 4.5 and again increased when pH was 7.4. All the polymers behaved in a similar manner in terms of change in the rate of release of drug with respect to pH. The severe damage to polymer morphology that was studied after

release experiment, for PH 7.4 is in agreement with this phenomenon. The pH responsiveness of the PUs was attributed to acid chain extenders that forms anionic polymer when subjected to PBS. At pH 1.2, the release of the drug was observed due to swelling in PBS. The response with respect to pH is triggered due to presence of insoluble -COOH groups that get ionized at pH 4.5. The presence of a large number of anionic groups on the polymer makes the polymer chains repel one-another leading to expanded dimension or higher swelling of the polymer. The phenomenon becomes more effective when the pH increases to 7.4. As reported by Bajpai et al, the swelling of anionic polymer increases with increase in external pH due to the increased ionic strength of external media [53]. The polymer, in such stimuli, can exchange ions with the solution; hence maintaining the charge neutrality. The concentration of free counter ions inside polymer increases. This results in increased osmotic pressure difference between the polymer and the solution. This finally causes swelling of the polymer.

It can be concluded that there is no effect of variation of acid chain extender in terms of drug release rate with respect to pH, i.e. For all PUs, the rate of release of drug with respect to pH follows same order 7.4>4.5>1.2 irrespective of type of acid chain extender. However, it was observed that with change of chain extender from lactic acid to dimethylol propionic acid to glycolic acid, the rate of drug release decreased for constant pH value. More specifically, during the period of 25h, the amount of cumulative drug release was 98.52%, 80.29% and 54.69% for LAPU, DAPU and GAPU at pH 7.4 respectively (Fig. 4.7). A similar trend was observed for pH 4.5 and 1.2 also. This behavior can be explained on the basis of swelling of PUs as well as T_g of corresponding PUs. The drug release was observed to be a direct function of swelling and inversely proportional to the T_g of PUs. This is attributed to the more crosslinked structure of GAPU showing highest T_g, as discussed in section of thermal properties. Higher crosslinking

leads to slower diffusion of drug molecules that can migrate from the polymer matrix, leading to a slower release of the drug. The pKa values of lactic acid, dimethylol propionic acid and glycolic acid are 3.86, 4.41 and 3.83 respectively. No correlation was found between pKa values of acids and drug release characteristic of PU containing corresponding acids, as also observed by Claeys et al in the study of the effect of acid modifiers in release properties of PU tablets [54].

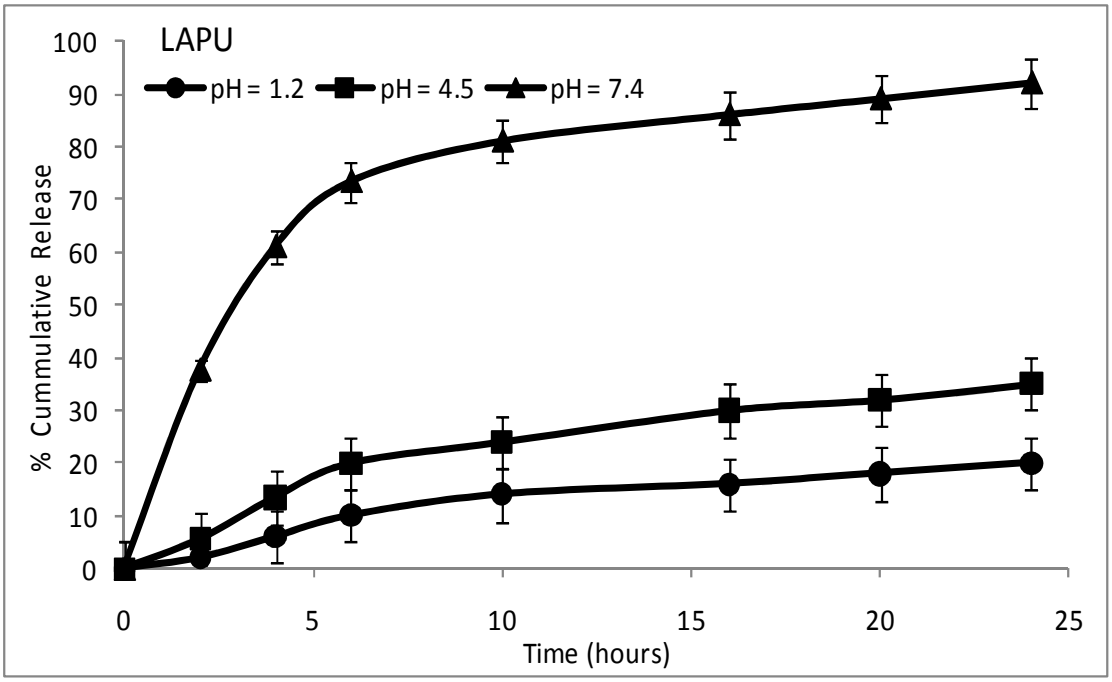


Figure 4.7(a) Drug release profiles for LAPU

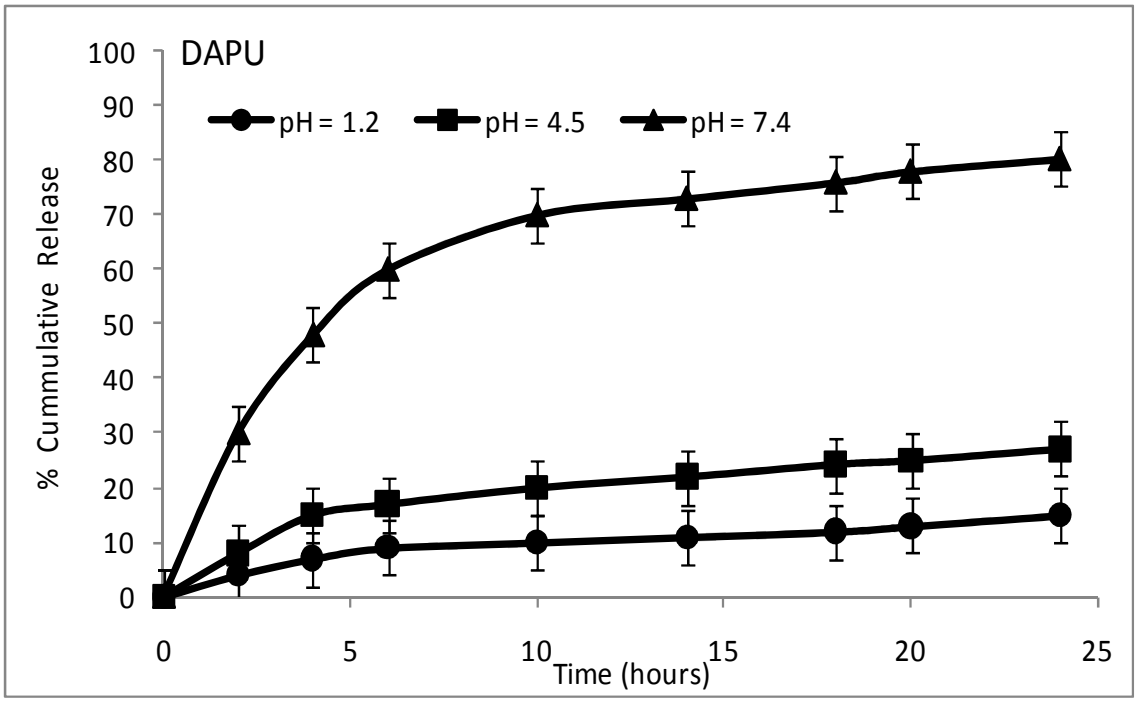


Figure 4.7(b) Drug release profiles for DAPU

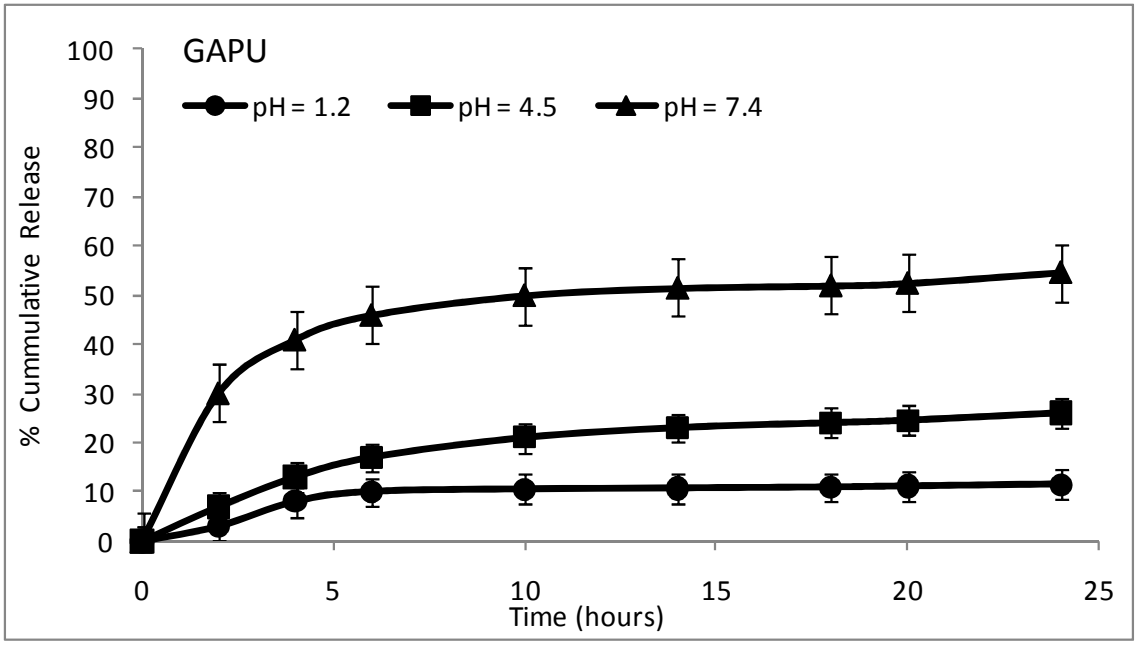


Figure 4.7(c) Drug release profiles for GAPU

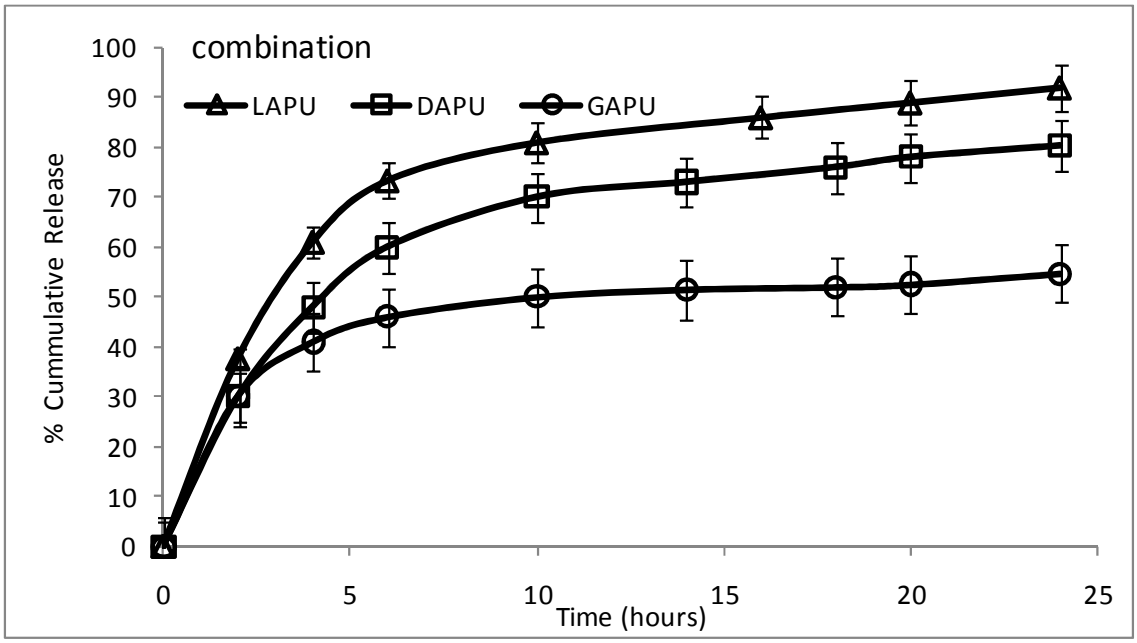


Figure 4.7(d) Drug release profiles for PUs at pH 7.4

The drug release kinetics was studied by using the following equation [55].

$$\frac{M_t}{M_\infty} = kt^n \qquad \text{---------- (3)}$$

Where M_t/M_∞ denotes a portion of drug released at time t (M_∞ is considered same as the amount total drug loaded in each polymer), k is constant of release rate and n is an important exponent value which can be used to define release mechanism. Normal Fickian diffusion for a thin polymer membrane is defined by $n<0.5$, while Case II transport is characterized by $n = 1.0$.

The results obtained from the plot shown in Fig. 4.7, were used to plot a graph of log (Mt / M_∞) versus log (t). Linear regression was used to find the intercept and slope which gave values of k and n respectively. All release profiles presented n values that are less than 0.5 as listed in Table

4.3. These values of n suggest that the release of Felodipine from all PUs in the present study follows the Fickian diffusion.

4.4. CONCLUSIONS

Cellulose crosslinked waterborne polyurethanes (PUs) based on poly ε-caprolactone were prepared with different α-hydroxy acids. The thermal degradation, structural and morphological properties of all PUs were identical, irrespective of type of chain extender. The α-hydroxy acids acted as drug release modifiers and imparted pH responsive characteristic to the PUs. PU with glycolic acid as a chain extender, showed highest crosslinked structure, highest T_g and slowest drug release at a given pH. PUs were loaded with Felodipine and drug release was monitored at different pH values. The encapsulation of dug inside PU matrix and the morphology of polymer after drug release were studied by using SEM. All PUs showed the highest release rate at pH 7.4, considered to be pH of human organs such as the colon as well as vagina after sexual intercourse. An added advantage is control of release rate depending on the type of acid chain extender used. The PUs were observed to degrade under highly basic conditions. The reported PUs, being "smart" polymers, can find attractive application in the field of drug delivery involving pH responsive characteristics such as microbicides and vaginal passaries for prevention of HIV-AIDS and other sexually transmitted diseases.

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