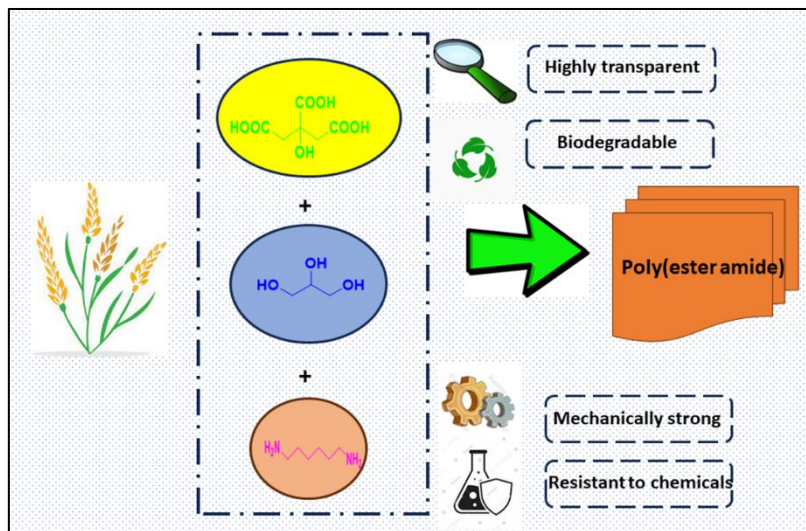


Chapter 2



Synthesis, characterization, and properties evaluation of bio-based poly(ester amide)s

Highlights

Poly(ester amide)s have cropped up as an interesting group of polymeric materials entailing excellent mechanical and thermal properties on ground of typical confinement of ester and amide connectivity in the polymeric chains. Howsoever, extensive usage of polymers derived from conventional petroleum sources is posing as a major threat to ecosystem sustainability. In this chapter, a bio-based water-soluble poly(ester amide) was synthesized using an environmentally benign route devoid of usage of any solvent. Citric acid, glycerol and hexamethylenediamine were used as the core raw materials for carrying out the synthetic reaction via a polycondensation route. The prepared polymeric resin was characterized using various spectroscopic techniques such as Fourier transform infrared (FTIR), ¹H NMR (Nuclear magnetic resonance), ¹³C NMR, etc. Poly(vinyl alcohol) was further incorporated to modify the resin. Different compositions of resin/poly(vinyl alcohol) were prepared and used to evaluate their performances. The polymeric films exemplified splendid mechanical attributes namely tensile strength (7-8 MPa), elongation at break (1380-2166%), and toughness (40.82-98.12 MJm⁻³) along with satisfactory thermal stability (above 210 °C) and good transparency. Moreover, they possess biodegradability aspects; thereby endowing the characteristics which can overcome the shortcomings of petroleum derived synthetic poly(ester amide)s and subsequently be utilized in various potential applications, especially as environmentally friendly coating materials.

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2.1. Introduction

Chapter 1 illustrated that over these years, synthetic polymers have garnered a cardinal position owing to the rampant advancement in today's materialistic world and they are considered as potent viable substitutes for their natural counterparts [1]. They render a crucial effect on our lives due to their extensive usage in diverse domains stretching from paints, packaging industries, textile industries, etc. to leading edge technological development [2-4]. Howsoever, these polymers are predominantly sourced from petroleum-derived feedstocks which are easily accessible and convenient to use. With the rapidity in depreciation of the conventional natural sources as discussed in **Chapter 1**, there's an urgent imperative to prioritize on the promotion of development and utility of biodegradable as well as bio-based polymers. Such environmentally sustainable polymers can abolish the growing concerns over release of toxic greenhouse gases, waste disposability issue and other ecological challenges [5-10].

In this context, poly(ester amide)s belong to an important category of polymers and possess significant relevance in diverse domains as expounded comprehensively in **Chapter 1**. They are of paramount interest to research fanatic persons on account of their versatile properties. Regular intertwinement of ester and amide linkages in the same polymeric backbone attributes to coupling of the favourable features of both polyamides as well as polyesters [11]. The presence of ester groups in the polymeric framework renders biodegradability trait to the poly(ester amide)s, wherein the high mechanical strength and thermal stability is assigned due to the double bond nature of the amide group combined with the strong influence of hydrogen bonding interactions [12-14]. In recent times, there has been a surge in the consumption of environmentally congenial materials on account of enhanced awareness amidst people regarding the use of "greener" alternative materials. Under this framework, it becomes pertinent to develop such materials which bear good durability in extreme environmental conditions such as different chemical media, ultraviolet radiation, etc. and also exhibit considerable mechanical strength [15]. Archives on synthesis of bio-based poly(ester amide)s state that mostly the reported ones are not completely biogenic in nature and thereby questing continue to synthesize a bio-based eco-friendly poly(ester amide). Furthermore, the extensive utilizations of organic solvents which are noxious in nature result in the discharge of VOCs which induce different respiratory disorders in living beings [16]. In this context, researchers are striving

to generate a paradigm shift from use of conventional petroleum-derived resources to bio-derived sources in order to carry out the synthesis of poly(ester amide)s. In this milieu, use of green solvents such as water, ethyl-lactate, etc. are highly encouraged along with low solvent driven systems such as dispersion systems, waterborne systems, etc. Water-soluble poly(ester amide)s provide manifold advantages including low toxicity, low flammability, ease of cleaning up, low odour content, etc. and are considered as apt eco-friendly materials [17, 18]. The presence of various hydrophilic or ionic groups on the end surface results in interaction of these functional moieties with water molecules and thereby conferring water solubility in these systems [19, 20]. Such systems can portray pivotal role in pharmaceutical, biomedical and bioengineering domains in view of their biocompatibility and biodegradability features.

Hence, in light of various considerations, it becomes imperative to synthesize a poly(ester amide) via an environmentally benign route to cater towards diverse applications. Therefore, in this present study, an attempt has been made to develop a water-soluble, biogenic poly(ester amide) involving the combination of citric acid, glycerol and hexamethylenediamine via a polycondensation reaction. Owing to the accord on environmental harmonization, there has been absolutely no usage of organic solvents; thereby negating the issue of VOC generation. The prepared polymeric system is found to be seasoned with various traits such as excellent solubility in water owing to its greater surface functionality, biodegradability, etc. To improve the performance of the synthesized poly(ester amide), it was modified by poly(vinyl alcohol). Assessments of the resultant systems were further carried out in order to analyse their specific properties and to understand their potential utility in different domains, especially as environmentally benign biodegradable coating materials.

2.2. Experimental

2.2.1. Materials

Citric acid (as indicated in **Chapter 1, Table 1.1.**) was obtained from Merck, India, with a purity level of 99.9%. It has a density of 1.66 g/cm³ along with a molecular weight of 192.12 g/mol. Prior to use, citric acid underwent a drying process under vacuum conditions for a time period of 5-6 hours at 50 °C.

Glycerol (as indicated in **Chapter 1, Table 1.4.**) was acquired from Merck, India. It is an odorless, clear, and viscous liquid that possesses trihydroxyl groups, boasting a minimum assay of 99%. It has a density of 1.26 g/cm³ along with a molecular weight of 90.09 g/mol. It exhibits hygroscopic properties, and consequently, underwent a drying process under vacuum conditions for a time duration of 5-6 hours at 50 °C before being employed for further usage.

Zinc chloride was procured from Sisco Research Laboratories Pvt. Ltd., India. It bears an empirical formula ZnCl₂, with a molecular weight of 136.29 g/mol. It is an odorless, white, deliquescent solid with density value of 2.90 g/cm³. It possesses purity level of 95%, with the remaining contents primarily consisting of oxychloride and water.

Hexamethylenediamine (as discussed in **Chapter 1, Table 1.2.**) was obtained from Sigma Aldrich, USA. It is a white crystalline solid having a boiling point and melting point of around 204-205 °C and 42-45 °C, respectively. It has a density value of 0.89 g/cm³ along with a minimum assay of 98%.

Potassium hydroxide (KOH) was obtained from Merck, India in the form of pellets. This compound demonstrates strong alkaline properties and possesses a molecular weight of 56.11 g/mol. KOH in the form of aqueous solution was used for evaluating the acid value.

Tartaric acid (as indicated in **Chapter 1, Table 1.1.**) was procured from Loba Chemie, India. It is well characterized by its white crystalline structure. It has a density of 1.79 g/cm³ and a molar mass of 150.08 g/mol. Prior to utilization, it underwent a drying process for a time period of five hours at 50 °C due to its hygroscopic nature.

p-Toluenesulfonic acid was procured from Sisco Research Laboratories Pvt. Ltd., India. It is a white crystalline solid which is hygroscopic in nature. It is highly soluble in alcohols, water and other organic polar solvents. It has a molecular weight of 190.22 g/mol along with a density value of 1.24 g/cm³.

Poly(vinyl alcohol) was obtained from Central Drug House Pvt. Ltd., India. It is an odorless and colorless solid. It is highly soluble in water with a molecular weight of around 85,000-1,00,000. It was used for carrying out modification of the poly(ester amide) resin.

Oxalic acid was obtained from Rankem, India. It is a white crystalline solid bearing a molecular weight of 126.07 g/mol. It has a minimum assay of 99%. It is used for carrying out standardization of KOH.

Sodium hydroxide (NaOH) was procured from Merck, India in the form of pellets. It has a molecular weight of 39.99 g/mol along with 97% purity. NaOH in the form of aqueous solution was used for conducting the evaluation of chemical resistance of poly(ester amide).

Hydrochloric acid (HCl) was procured from Merck, India and diluted to 10% (v/v) for carrying out evaluation of chemical resistance test of poly(ester amide).

Sodium chloride (NaCl) was obtained from Merck, India. It was used in the form of aqueous solution to assess the chemical resistance parameter of poly(ester amide).

Ethanol (EtOH) was procured from Merck, India and was utilized in conducting the solubility as well as the chemical resistance test of poly(ester amide).

2.2.2. Methods

2.2.2.1. Synthesis of poly(ester amide) resin

The synthesis of bio-based poly(ester amide) was carried out by a melt polycondensation polymerization reaction of the core components i.e., citric acid, glycerol and hexamethylenediamine. *p*-Toluenesulfonic acid (*p*-TSA) was used as a catalyst to drive forth the reaction and was used in minuscule amount. Requisite calculated proportions of the reactants (**Table 2.1.**) were taken in a three neck round bottom flask furnished with an oil bath, a thermometer and a mechanical stirrer. The entire sequence of condensation reaction was brought into effect by continuous application of vacuum suction pressure in order to eliminate water generated during the course of the reaction. The concoction was heated at 140 °C for 3 h under vigorous mechanical stirring and the course of the reaction was closely monitored by drop in acid value. The reaction was eventually stopped before the attainment of gelation point, and a light brown viscous mass was thus obtained.

Since the texture of poly(ester amide) resin formed was found to be a bit brittle in nature, poly(vinyl alcohol) was introduced along with zinc chloride so as to provide strength and

agility to the films so formed. Water was added in minimal amount in order to facilitate the dissolution process effectively. The reaction mixture was heated at 75-80 °C for 1 h until homogeneity was achieved in the mixture. Four different compositions were prepared by varying the weight percentages of poly(ester amide) and poly(vinyl alcohol) contents and were labeled correspondingly as PEA 0%, PEA 50%, PEA 67% and PEA 77% (**Table 2.2.**).

Table 2.1. Compositions of the prepared poly(ester amide) resin.

Reactants	Amount (mM)	Possible reactive functionality (f)	Total functionality (Tf)
Citric acid	45	2	90
Glycerol	18	3	54
Hexamethylenediamine	18	2	36

2.2.2.2. Curing of resin

It involves a chemical reaction that employs the usage of a crosslinker in order to toughen the material so formed. In this context, tartaric acid, a dicarboxylic acid was used as the crosslinker. An amount of 5 wt% of tartaric acid was mixed uniformly with prepared poly(ester amide) resin along with poly(vinyl alcohol) and zinc chloride in a three neck round bottom flask to obtain a clear homogeneous solution at 80 °C. The mixture was stirred thoroughly for 1 h followed by transferring the contents into Teflon sheets. The sheets were dried at 50 °C for 24 h in a hot air oven succeeded by carrying out crosslinking reactions at 120 °C for 2 h.

Table 2.2. Compositions of poly(ester amide) and poly(vinyl alcohol).

Reactants (g)	Poly(ester amide) resin	Poly(vinyl alcohol)	Ratio
PEA 0%	-	1	-
PEA 50%	0.5	0.5	1:1
PEA 67%	0.5	0.25	1:0.5
PEA 77%	0.5	0.15	1:0.3

2.2.3. Characterization

2.2.3.1. Structural analysis

FTIR spectral analyses of the prepared poly(ester amide) resin and its different compositions with poly(vinyl alcohol) were carried out on a Nicolet FTIR spectrophotometer (Impact-410, USA) using KBr pellets lying within the range of 4000-500 cm^{-1} . ^1H and ^{13}C NMR spectra were recorded in a 500 MHz AV500 AVANCE III FT-NMR spectrometer using D_2O as the solvent and tetramethylsilane (TMS) as the internal standard. ^1H - ^1H COSY NMR spectrum was recorded in a 600 MHz AVANCE III HD FT-NMR spectrometer using D_2O as the solvent and tetramethylsilane (TMS) as the internal standard. Gel permeation chromatography (GPC) was conducted in order to determine the molecular weight of the polymeric resin using Waters Corporation, USA model equipment. Water was used as the eluent, and a constant flow rate of 0.75 mL/min was maintained. The analysis was carried out using a 515 HPLC pump and 2414 refractive index detector. Elemental analysis was driven forward using a CHN analyzer, PR 2400 series 11 Perkin Elmer.

2.2.3.2. Determination of physical properties

Acid value is determined to evaluate the number of milligrams of KOH required to neutralize the amount of free fatty acid present in 1 g of oil. The sample under consideration is weighed properly followed by carrying out titration against standardized KOH solution (using oxalic acid as primary standard) in an alcoholic medium. Herein 0.1-0.5 g of sample was taken and dissolved in 20 mL of water. Subsequently, 5 mL of aliquot was pipetted out from the reaction mixture and titrated against standardized KOH solution using phenolphthalein as the indicator. The appearance of faint pink color gauges the end point which reflects the amount of base required to neutralize the acidic components present in the sample. The acid value was calculated by using the following relation.

$$\text{Acid value} = \frac{\text{Volume of KOH} \times \text{Normality of KOH} \times \text{Equivalent weight of KOH}}{\text{Weight of the sample}}$$

In addition, the specific gravities of poly(ester amide) and its various compositions were measured as per the standard ASTM D792 liquid displacement method.

2.2.3.3. Determination of mechanical properties

Mechanical attributes namely tensile strength and elongation at break were recorded by a Universal Testing Machine (UTM); model number WDW10 (JINAN, CHINA) as per ASTM D 638 involving a load cell of 500 N along with crosshead speed of 10 mm/min. Scratch hardness tester (Sheen instrument Ltd., UK); model number 705 was used to measure scratch hardness feature of the prepared films by abiding standard ASTM G171-03 methodology. Impact hardness parameter was eventually calculated by using an impact tester (S.C. Dey & Co., Kolkata) by adhering to the standard ASTM D4272 falling weight methodology involving a maximum height of 100 cm. Thereby, impact strength is calculated by measuring energy per unit thickness against the maximum height traversed by the weight. The gloss properties of the synthesized polymeric films were measured using a mini gloss meter (Sheen instrument Ltd., UK) projected at an incident angle of 60°.

2.2.3.4. Determination of thermal properties

Thermogravimetric (TGA) as well as differential scanning calorimetry (DSC) analyses were executed in order to assess the thermal traits of the prepared poly(ester amide)s. TGA evaluation was carried forward by a TGA-system (Netzsch, Germany) conditioned under an inert environment of pure nitrogen gas falling in the temperature range of 30-600 °C and under the constant maintenance of gas flow rate of 30 mL/min against subsequent heating rate of 10 K/min. Moreover, differential scanning calorimetry study was brought off by a DSC 6000 instrument (Perkin Elmer, USA) equipped with a sophisticated cooling method. Constant heating rate of 10 °C/min against flow rate of 30 mL/min was sustained under an inert environment of pure nitrogen gas varying within the temperature ambit of -70 °C to 150 °C, thereby conforming to a heating-cooling-heating cycle.

2.2.3.5. Chemical resistance test

In order to inspect chemical resistivity aspect, the polymeric films were exposed to diverse chemical conditions viz. water, HCl (5%), NaOH (5%), NaCl (10%) and EtOH (10%) at ambient room temperature for a period of 21 days and their weight loss profiles were specifically calculated after fulfillment of the prescribed regime. This study followed the standard ASTM D 543-95 protocol.

2.2.3.6. Transparency test

The transparency property of the polymeric films was determined by measuring the percentage transmittance using a UV-visible spectrophotometer (Evolution 300, Thermo Fisher, USA). The film samples of various polymeric compositions were cut uniformly into rectangular shaped pieces and eventually placed directly inside the cells of the spectrophotometer which are magnetic in nature. The test cell devoid of any polymeric film sample was considered as the reference standard. Percentage transmittance values were measured for wavelength ranging from 200 to 800 nm and the values were noted down for each composition respectively against the reference standard.

2.2.3.7. Biodegradation studies

The biodegradability attribute of the polymeric films was studied using *Bacillus subtilis* (a gram-positive bacterium) strain obeying the McFarland turbidity methodology. Furthermore, the growth or culture medium was prepared by combining a variety of salts viz. 2 g (NH₄)₂SO₄, 2 g of Na₂HPO₄, 4.75 g of KH₂PO₄, 1.2 g of MgSO₄ .7H₂O, 1 g of MnSO₄.5 H₂O, 0.1 g of CuSO₄.7H₂O, 10 mg of H₃BO₃.5H₂O, 70 mg of ZnSO₄.7H₂O, 10 mg of MoO₃, 1 mg of FeSO₄.7H₂O and 0.5 mg of CaCl₂.7H₂O into 1 L of demineralized water. The solution so obtained was consequently sterilized for a time period of 30 minutes subjected to a pressure of 15 lbs at 120 °C in an autoclave. This mixture was eventually utilized for culturing the bacterial strain at 37 °C for a period of 48 h in the incubator and a small amount of the growth media solution i.e., 100 µL aliquot (10⁸ microbes per mL) was added to 10 mL of the sterilized medium solution contained in a conical flask. The succeeding step involved sterilizing the polymeric films under UV radiation (254 nm) and adding them up into the conical flasks followed by carrying out incubation at 37 °C. Exact quantity of the above-mentioned bacterial culture media was employed as the control. Absorbances of the polymeric films containing the growth media were measured at 600 nm wavelength for a time period of 4 weeks in the form of optical density (OD) in order to track down the growth rate of the bacterial strain with time. Finally, the weight loss percentages were calculated for all the tested polymeric compositions after completion of the study and the surface morphology of the biodegraded films was assessed by a JOEL JSM-6390 LV Scanning electron microscope (SEM).

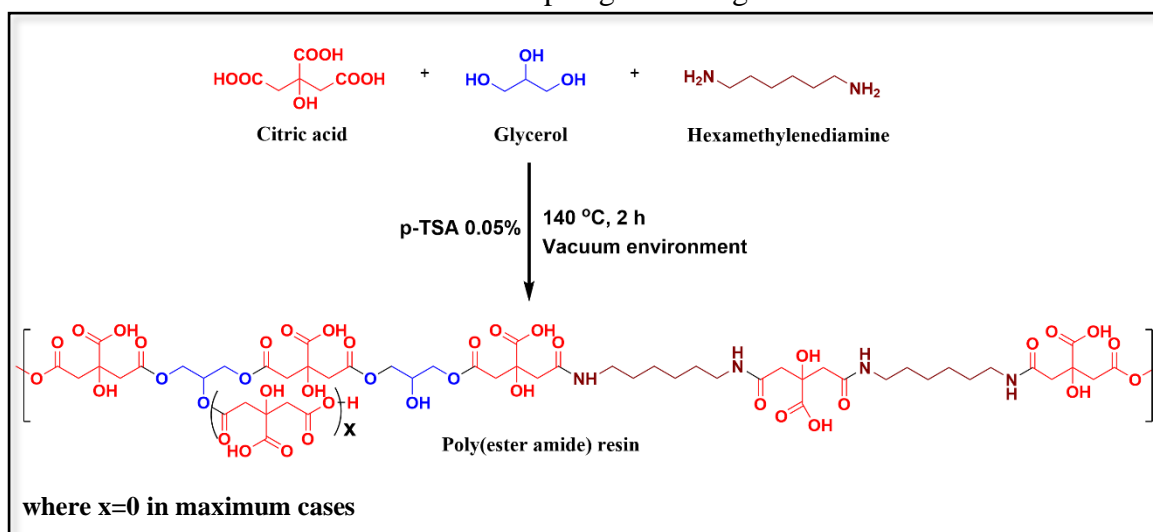
2.3. Results and discussion

2.3.1. Synthesis of poly(ester amide) resin

Synthesis of poly(ester amide) resin entails polycondensation reaction of the salient components i.e., citric acid, glycerol and hexamethylenediamine. The complete course of reaction was carried forward devoid of use of any solvent and implicated a one pot strategy. Citric acid is being a well-known tricarboxylic acid possesses two primary –COOH groups, one tertiary –COOH and one -OH groups. It is a customary well-known fact that primary –COOH group reacts faster than tertiary one [21]. Along the same line, glycerol also bears two primary –OH groups and one secondary –OH group and the reactivity of former is greater than latter [21]. These two primary functionalities i.e., –OH and –COOH react to form ester linkages coupled with discharge of water molecules. However, the reaction of secondary -OH of glycerol with primary -COOH group of citric acid cannot be completely ruled out, though formation of such products is not markedly noticeable, as indicated by the NMR spectral analyses (discussed later). On the other hand, hexamethylenediamine contains two primary amino-groups at its terminal position thereby facilitating the formation of amide linkages with primary –COOH functional groups of citric acid component. Ester and amide bond formations were confirmed by carrying out various spectroscopic studies as discussed later. One pot synthesis of poly(ester amide) resin is a propitious technique for industrial scale production as it hardly incorporates any polymer purification steps. Moreover, inspection of acid value was carried out time to time possibly after every half an hour to keep a check on the extent of reaction. It was clearly monitored that there was decline in acid value with time; howsoever, the fall off was quite steep in the beginning as compared to the end stage. This may be cited on grounds that with enhancement of chain length, effective collisions recede in number; thereby leading to increase in viscosity and reduction in acid value number unlike the beginning stage where there is rampant formation of ester linkage. The entire scheme representing the synthesis of poly(ester amide) resin is provided in **Scheme 2.1**.

In order to impart film forming ability to poly(ester amide) resin, poly(vinyl alcohol) was introduced along with zinc chloride. Poly(vinyl alcohol) is a well-known water-soluble polymer and is endowed with numerous useful traits such as good flexibility, non-toxic in nature, odorless, etc. [22]. The hydroxyl groups present in the framework of poly(vinyl alcohol) reacted with the carboxylic acid moieties of poly(ester amide) resin resulting in

esterification reaction. Zinc chloride herein acted as a complexing agent. Dipositive zinc ions exert affinity towards oxygen containing functionalities; thereby leading to increased molecular interactions. This amounts to upsurge in strength of the film so formed.



Scheme 2.1. Synthesis of poly(ester amide) resin with most probable structure

2.3.2. Curing of poly(ester amide)

This study primarily involves usage of a crosslinker to carry forth the chemical crosslinking process in order to obtain thermoset. Citric acid and glycerol are multifunctional monomers, and their reaction along with hexamethylenediamine resulted in synthesis of the main polymeric matrix under the given conditions, where only the primary functional groups were reacted. Although the main condensation reaction ensued formation of a tough polymeric network; the texture was found to be somewhat brittle in nature. This stemmed the need to form a polymeric blend with poly(vinyl alcohol). The resulting system is presumed to contain unreacted functionalities; thereby posing as great explicit sites for crosslinkers such as tartaric acid to react via formation of some ester linkages, etc. This enabled further toughening of the material via formation of a strong three-dimensional polymeric network leading to good mechanical features. The carboxylic acid functionalities of tartaric acid can react with the hydroxyl groups present on the periphery of polymeric system to form a crosslinked entity [23]. Furthermore, the cross-linked films were meant to undergo curing steps at moderately high temperature in order to facilitate the formation of ester connectivity. The curing time of all the poly(ester amide) compositions are tabulated in **Table 2.5**.

2.3.3. Structural analysis

The confirmation of the poly(ester amide) structure was carried out through different spectral analyses which are inclusive of FTIR, ^1H NMR, ^{13}C NMR, etc.

2.3.3.1. FTIR spectral study

The presence of disparate chemical functional groups within the polymeric framework is confirmed by carrying out FTIR spectral analysis. The FTIR spectra shown in **Figure 2.1**. enumerated the functionalities existing in the prepared poly(ester amide) resin. The broad band around 3400 cm^{-1} represented the stretching O-H vibrations. This reaffirmed the presence of large hydroxyl groups on the polymeric surface which are hydrophilic in nature. Moreover, it is a well-known fact that $-\text{OH}$ stretching vibration is much more intense than $-\text{NH}$ stretching vibration on account of $-\text{OH}$ bond being much more polar than the latter one. In addition, there is a strong possibility of $-\text{NH}$ and $-\text{OH}$ band resonances to get overlapped. So, distinct prominent peaks of $-\text{NH}$ stretching vibrations are not observed. In turn, distinct IR absorption peaks at 2835 and 2928 cm^{-1} corresponded

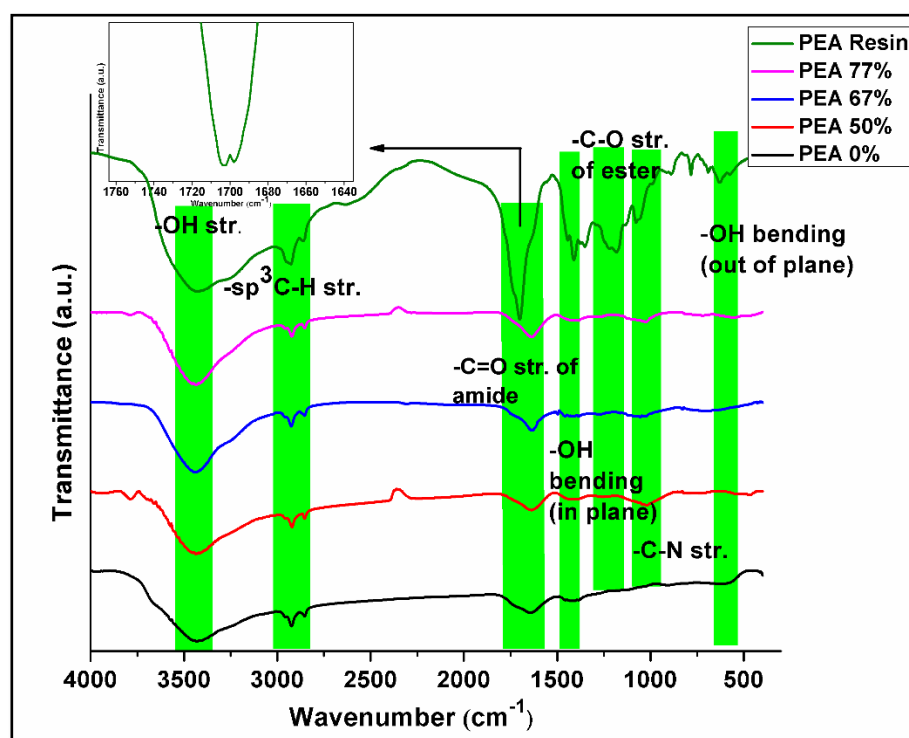


Figure 2.1. FTIR spectra of poly(ester amide) at different compositions with poly(vinyl alcohol).

to symmetric and asymmetric C-H stretching vibrations, respectively in an aliphatic system. The characteristic IR absorption bands at 1701cm^{-1} and 1640cm^{-1} ascribed to C=O stretching of esters and amide, respectively. This briefed up the formation of ester linkages between carboxylic acid and hydroxyl moieties as well as amide bond (also known as Amide (I)) between amine and carboxylic acid functionalities. The inset diagram further shown in the IR plot expounded explicitly the carbonyl stretching vibrations of ester at around 1700cm^{-1} . In addition, absorption frequencies at 1189cm^{-1} and 1068cm^{-1} illustrated C-O and C-N stretching, respectively. The characteristic absorption bands at 1418cm^{-1} and 627cm^{-1} are attributed to -OH bending in plane and out of plane, respectively. Thus, the aforementioned FTIR spectral studies supported the formation of poly(ester amide).

2.3.3.2. CHN analysis

Carbon, hydrogen and nitrogen being crucial elements in an organic sample were analyzed by using a CHN analyzer. This technique is considered to be of the oldest characterization methods in order to determine the elemental composition in an organic sample [24]. The elemental composition of carbon, hydrogen and nitrogen in weight percentage were documented in **Table 2.3**. The experimentally found values agreed well with the calculated ones devoid of any branching unit.

Table 2.3. CHN weight percentages of poly(ester amide) resin.

Elemental analysis	Carbon	Hydrogen	Nitrogen
Theoretical value			
(With branching unit)	46.09	5.59	3.98
(Without branching unit)	46.75	5.89	4.54
Experimental value	46.87	5.55	4.31

2.3.3.3 ^1H NMR spectral study

Protons present in diverse chemical environments were studied and analyzed using ^1H NMR spectroscopy. The ^1H NMR spectrum of prepared poly(ester amide) resin is shown in **Figure 2.2**.

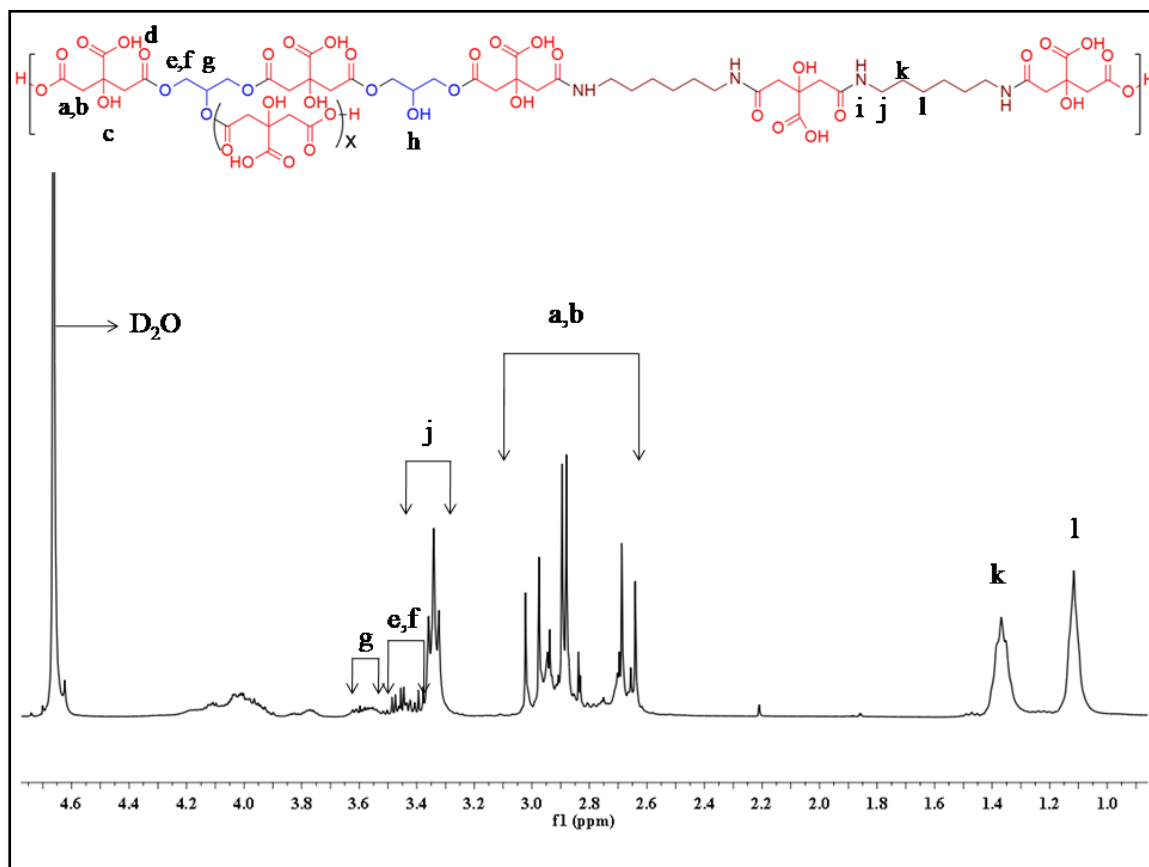


Figure 2.2. ^1H NMR spectrum of poly(ester amide) resin.

The methylene protons (k, l) present in hydrocarbon framework of hexamethylenediamine exhibited chemical shift values at δ 1.11 and 1.36 ppm, respectively. In addition, methylene protons (j) attached to basic nitrogen atom of amine functionality showed a typical downfield shift and produced a signal at δ 3.3 ppm. Again, the protons of methylene groups (a, b) present in citric acid reflected chemical shift values in the range 2.64-3.02 ppm. Glycerol unit also bears methine (g) and methylene protons (e, f) and their corresponding signals produced were observed in the range of 3.4-3.6 ppm. The absorption peaks present in the range 3.9-4.2 ppm ascribed to the trace impurities present in the polymeric sample after the subsequent extraction process and as the intensity is very low,

they are neglected. Further, D₂O was used as the solvent to carry forward the NMR experiment and it displayed its characteristic NMR peak at 4.6 ppm. Since D₂O was used as the solvent, the carboxylic acid (-COOH) proton of citric acid (d), hydroxyl (-OH) protons of glycerol (h) and citric acid (c) as well as amino (-NH) proton (i) of hexamethylenediamine underwent deuterium exchange on account of being acidic in nature; henceforth their signals could not be recorded in the ¹H NMR spectrum.

2.3.3.4. ¹³C NMR spectral study

Likewise, the presence of carbon in different chemical environments was substantiated using ¹³C NMR spectral analysis. A representative ¹³C NMR spectrum of prepared poly(ester amide) is shown in **Figure 2.3**. The carbonyl carbon (i, a) of carboxylic acid moiety produced NMR signals at δ 179.91 ppm. In turn, carbonyl carbon of ester (d) and amide (g, l) generated chemical shift values at δ 177.7 and 172.85 ppm, respectively. The tertiary carbons (j, b) of carboxylic acid component exhibited signals at δ 72 ppm and 71.09 ppm. In addition, the methylene and methine carbons of glycerol unit (f, e) displayed NMR signals at δ 65.49 and 62.18 ppm, respectively wherein the methine carbon (e*)

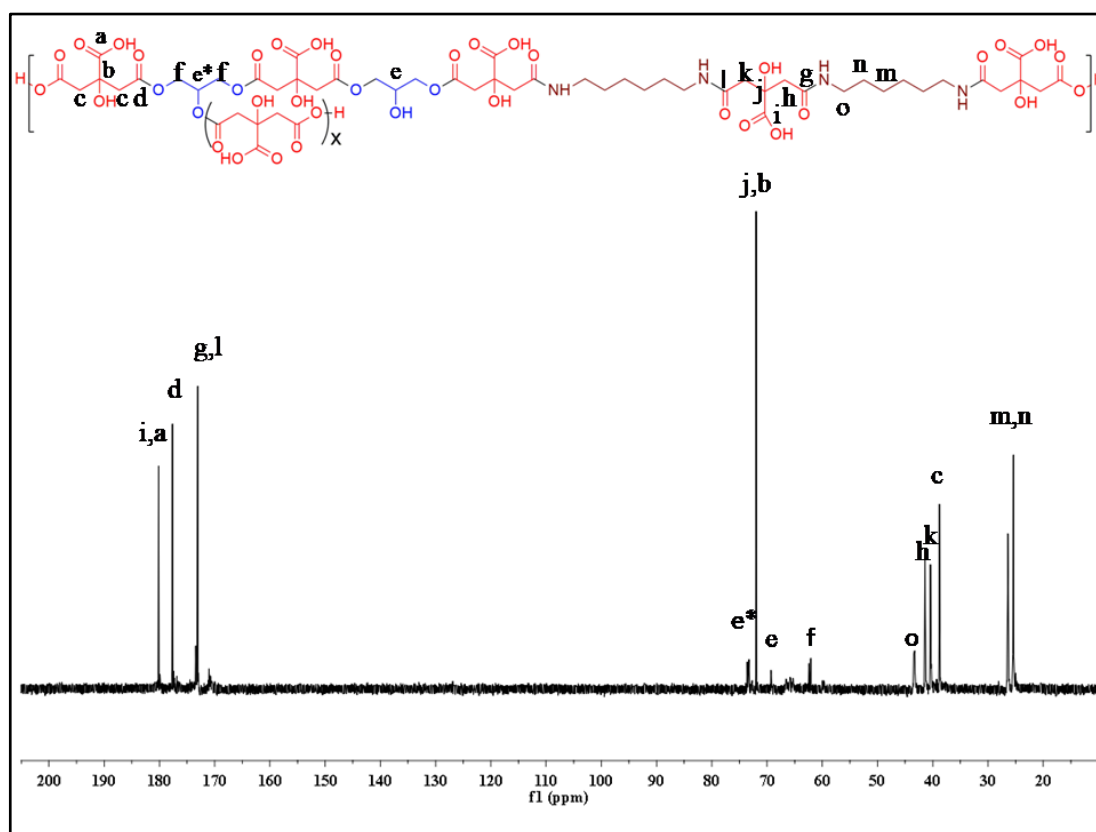


Figure 2.3. ¹³C NMR spectrum of poly(ester amide) resin.

attached to the citric acid branching unit exhibited signal at δ 72.6 ppm. Moreover, the methylene carbons belonging to citric acid unit (h, k) incorporating both ester and amide linkages showed up signals at δ 40.34 and 41.44 ppm. Under the same pretext, citric acid component bearing ester linkages at both ends displayed chemical shift value at δ 38.54 ppm. The long hydrocarbon chain framework of hexamethylenediamine (m, n) and (o) exhibited prominent peaks at δ 26.43, 25.28 and 43.10 ppm, respectively.

2.3.3.5. 2D NMR spectral study

2D NMR spectral analysis has also been carried out (as shown in **Figure 2.4.**) in order to wave off the issue of limited spectral resolution in 1D NMR. Moreover, peak assignments at times become cumbersome in 1D NMR on account of overlapping peaks due to the presence of similar monomeric units. Accordingly, homonuclear ^1H - ^1H COSY (Correlated Spectroscopy) was performed to address the above-mentioned issue.

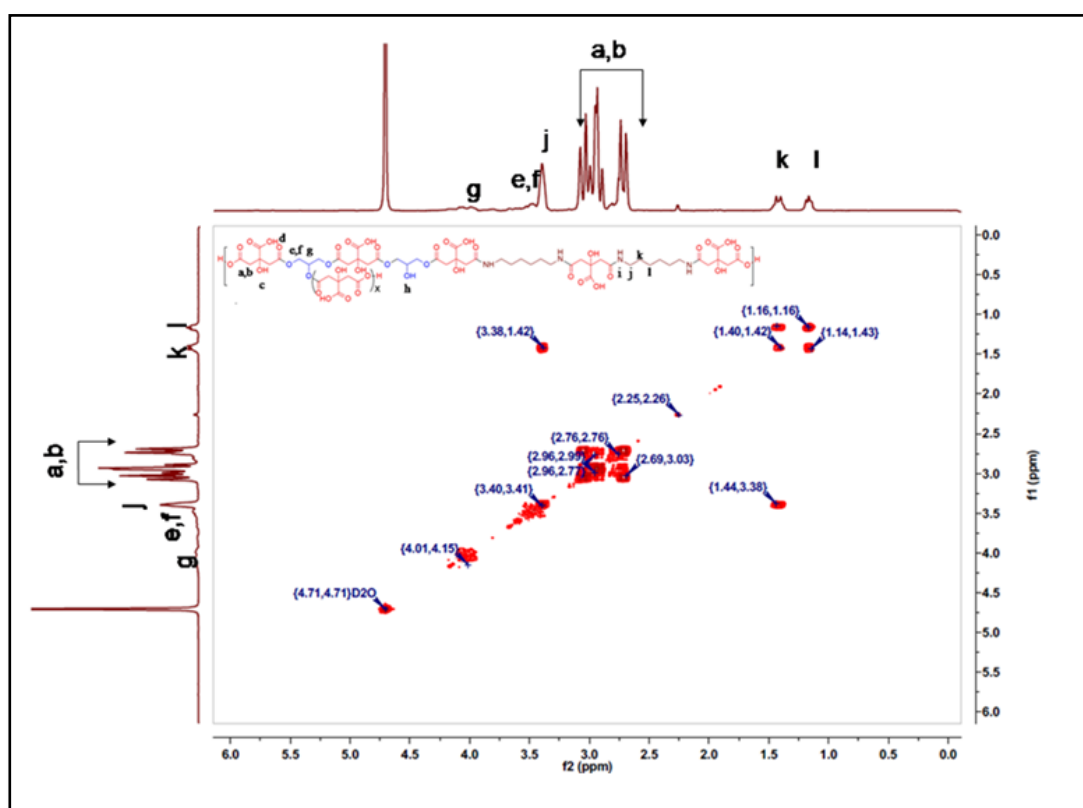


Figure 2.4. ^1H - ^1H COSY spectrum of poly(ester amide) resin.

^1H - ^1H COSY is considered as one of the vital tools to elucidate structure of polymers. It rests on the principle of J -coupling via one, two, three or four chemical bonds in general. Correlations are established between those proton nuclei which are bonded via J -coupling.

In addition, cross peaks are obtained in the spectrum only for those proton nuclei which are mutually bonded to other proton nuclei present in close proximity. As depicted in the simulated COSY spectrum (**Figure 2.4.**) above, three prominent cross peaks were obtained which highlighted the fact that these proton nuclei were mutually coupled via chemical bonds. The first cross peak at (1.44, 3.38) ppm signified the fact that the proton at 1.44 ppm (k) is coupled to proton at 3.38 ppm (j). Similarly, data points or cross peaks at 1.14, and 1.43 ppm in the plot revealed coupling between the protons (l) and (k). Another set of data points at (2.69, 3.03) ppm reflected homogeneous coupling between (a) and (b) protons. Therefore, the aforementioned details proposed the formation of polymeric material.

2.3.4. Physical properties

The prepared poly(ester amide) resin was found to be brown in color, whereas the films prepared from different compositions were yellowish brown in color. Physical properties such as specific gravity and acid value were determined using the standard methodologies

and the data are specifically tabulated in **Table 2.4.** It was found that with the increasing amount of citric acid portion, there was a slight hike in the value of density. The probable reason can be traced back to the fact that rise in polar functionalities resulted in greater molecular interactions leading to a more compact and dense system. Solubility tests carried out dictated that poly(ester amide) resin was solvable in highly polar solvents such as dimethylformamide (DMF), aniline, dimethyl sulfoxide (DMSO), acetonitrile (ACN), water, ethanol etc. and insoluble in non-polar solvents for instance hexane, toluene, xylene, etc. The solubility in polar solvents can be attributed to the presence of polar functionalities on the periphery of the resin. In turn, GPC results (number average molecular weight (g/mol): 3340, weight average molecular weight (g/mol): 3405 and polydispersity index (PDI): 1.01 exhibited low molecular weight of poly(ester amide) which ascribed to low viscosity and in turn resulted in facile handling and processing technique.

2.3.5. Mechanical properties

Assessments of the mechanical properties namely tensile strength, elongation at break, toughness, impact resistance and scratch hardness were carried through, and the results accomplished were tabulated in **Table 2.5.** These properties are known to rely on diverse

parameters; for instance-molecular weight of the polymeric framework, type and constitution of the components involved, presence of inter and intra molecular forces of interaction, existence of hydrogen bonds, mesh network formation by polymeric chains, omnipresence of polar functional groups on the surface of polymeric resin, etc. [25, 26]

Table 2.4. Physical properties of poly(ester amide) at different compositions with poly(vinyl alcohol).

Property	PEA resin	PEA 0%	PEA 50%	PEA 67%	PEA 77%
Acid value (mg/g)	359	-	-	-	-
Density at 25 °C (g/cm ³)	0.90	0.91	0.93	0.94	0.96

It can be contemplated further from mechanical study (as shown in **Figure 2.5.**) that with the change in weight percentage of poly(ester amide) resin so formed, there is a subsequent alteration in the parameters. Better crosslinking promotes extensive forces of attraction to come into play, thereby making the system strained as well as providing adequate rigidity to the structure. It was closely observed that with increment in poly(ester amide) content, there is a plunge in tensile strength value. This can be elucidated in the context, that subsequent increases in poly(ester amide) amount with reduction in poly(vinyl alcohol)

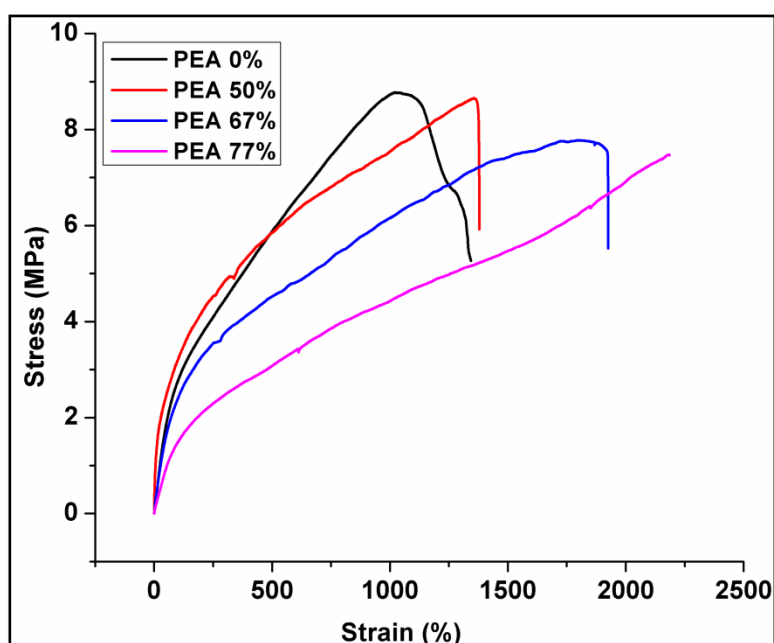


Figure 2.5. Stress-strain profiles of poly(ester amide)s.

content resulted in insufficient amount of crosslinking, henceforth dip in tensile strength value. On the other hand, it was detected that surge in poly(ester amide) proportion ensued uprise in elongation at break value with decrease in poly(vinyl alcohol) content. This can be unfolded on the ground that with the increase in poly(ester amide) composition, there is extension of the hydrocarbon chain thereby contributing towards enhanced elongation and flexibility. Likewise, toughness values were also studied and analyzed from stress-strain profiles and exhibited a similar trend as stated above. Moreover, slight decrease in scratch hardness and impact resistance parameters were noted down with further rise in poly(ester amide) content. This implies inadequate synergism between tensile strength and elongation attributes. Besides, the films also displayed fine gloss properties; thereby indicating a smooth surface domain and great dimensional constancy.

Table 2.5. Mechanical properties of poly(ester amide) at different compositions with poly(vinyl alcohol).

Property	PEA 0%	PEA 50%	PEA 67%	PEA 77%
Curing time (min)	120 ± 5	130 ± 5	140 ± 6	140 ± 5
Tensile strength (MPa)	8.73±0.013	8.62 ±0.013	7.72±0.008	7.46 ±0.013
Elongation at break (%)	1340±0.086	1380 ± 0.15	1925 ± 0.101	2166 ± 0.12
Toughness (MJm⁻³)	45.83±0.2	48.02 ±0.2	79.85 ±0.4	98.12 ±0.1
Impact resistance (kJ/m²)	14.92 ± 0.6	14.2 ± 0.5	10.89 ± 0.5	10.50 ± 0.3
Scratch hardness (kg)	7.5 ± 0.5	7 ± 0.3	7 ± 0.3	6.5 ± 0.2
Gloss at 60°	95 ± 5	85 ± 5	85 ± 6	80 ± 5

2.3.6. Thermal properties

TGA and DSC studies were conducted in order to investigate the thermal behavior of prepared poly(ester amide) at different compositions with poly(vinyl alcohol). By scrutinizing the thermal degradation curves, it was found that all the four compositions resorted to a similar degradation pattern. All of them exclusively followed a two-step degradation profile, as shown in **Figure 2.6**. The thermal degradation parameters i.e.,

degradation onset temperature (T_{on}), degradation peak temperature at two stages and glass transition temperature (T_g) were illustrated and clearly documented in **Table 2.6**. The initial degradation started off with loss of small moieties such as miniscule amount of water adsorbed on the polymeric surface. The actual thermal degradation which initiated in the temperature range 217-233 °C can be credited due to the rupture of aliphatic segments along with loss of ester units which are known to be thermo-labile. In addition, the second degradation peak which falls in the temperature range 417-445 °C can be associated with the scission of poly(vinyl alcohol) framework as well as degeneration of thermally stable amide linkages. The thermal region after 445 °C is due to formation of mainly carbonaceous or charred residue products of black carbon. The weight loss remnants after degradation at 600 °C were found to be high i.e., 43.92%, 35.13%, 38.24% and 35.31% for PEA 0%, PEA 50%, PEA 67% and PEA 77%, respectively. These signify that the degradation got completed 50-60% only even at such high temperature (600 °C).

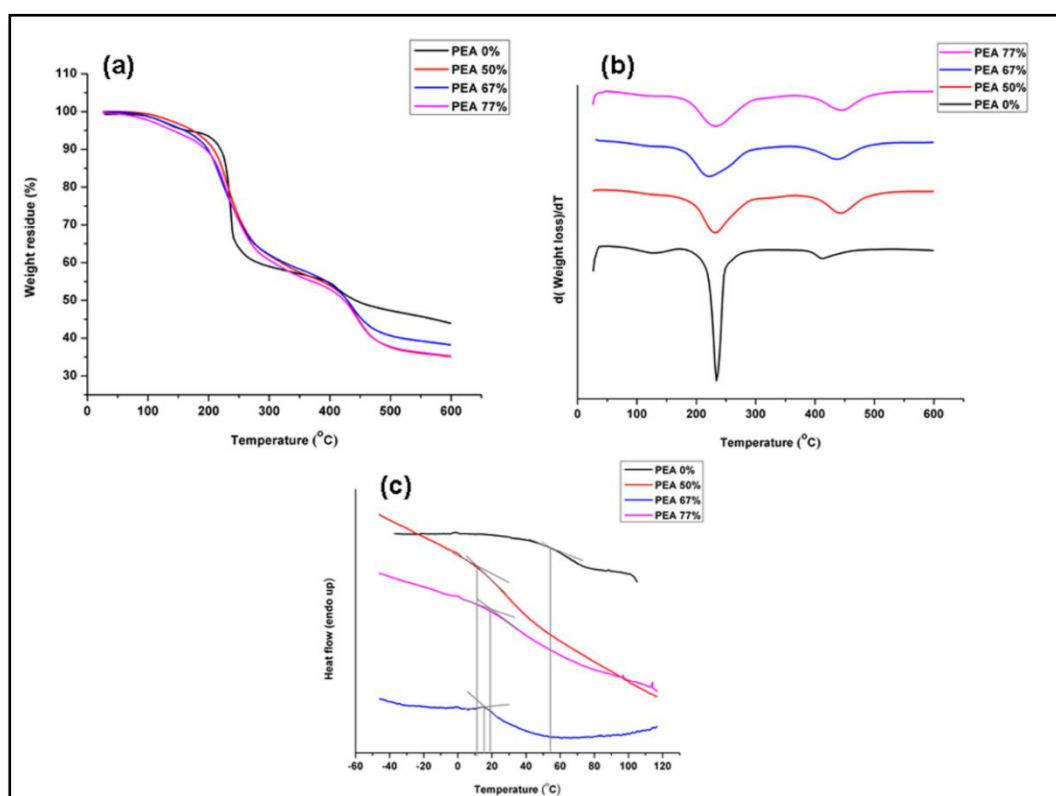


Figure 2.6. (a) TGA thermograms, (b) DTG curves and (c) DSC curves with T_g of polymeric films of poly(ester amide)s.

Moreover, assessment of the DSC curves as shown in figure (**Figure 2.6.(c)**) resulted in computing T_g which was found to lie within the temperature ambit of 55-11.17 °C. It was

noted that with the introduction of poly(vinyl alcohol) in the polymeric network, there was a dip in the T_g value. This could be adjudged to the fact that addition of poly(vinyl alcohol) resulted in enhancement of free volume in the polymeric network since it is known to slide in between the polymeric chains and rip them off apart. This in turn makes the polymer to glide past each other freely without any constraints; thereby resulting in lowering the T_g value.

Table 2.6. Thermal degradation parameters for poly(ester amide) at different compositions with poly(vinyl alcohol).

Parameters	PEA 0%	PEA 50%	PEA 67%	PEA 77%
T_{on} (°C)	233	232	226	224
1 st stage degradation peak temperature (°C)	330	328	334	299
2 nd stage degradation peak temperature (°C)	410	438	443	445
Weight residue (%) at 600 °C	43.92	35.13	38.24	35.31
Glass transition temperature, T_g (°C)	55	11.17	15.05	18.93

2.3.7. Chemical resistance study

Chemical resistance is considered to be a vital trait in order to test the resistance of poly(ester amide) in diverse chemical backdrops. The chemical resistance study was executed by exposing the poly(ester amide) thermosets in various coarse chemical environments for 21 days at room temperature and the weight loss outcomes were demonstrated in **Table 2.7**. It was noted that poly(ester amide) and all its compositions displayed fairly good resistance towards aqueous, acidic (5% HCl), saline (10% NaCl) and alcoholic (10% EtOH) media but relatively poor resistance towards basic (5% NaOH) medium. This could be substantiated on the pretext that poly(ester amide) resin contained ester linkages which are prone to hydrolysis under alkaline conditions [12]. Besides, presence of polar functionalities on the polymeric surface resulted in greater molecular interactions leading to enhanced defiance against aqueous, acidic, saline as well as alcoholic environments.

Table 2.7. Chemical resistance data (weight loss %) of poly(ester amide) at different compositions with poly(vinyl alcohol).

Sample	5% NaOH	5% HCl	10% NaCl	10 % EtOH	Water
PEA 0%	1.34	0.41	0.21	0.11	0.15
PEA 50%	1.67	0.98	0.45	0.35	0.26
PEA 67%	2.64	1.13	0.61	0.65	0.58
PEA 77%	2.89	1.36	1.12	0.89	0.87

2.3.8. UV-transparency

Transparency is considered to be an essential feature in polymeric films. It basically administers the see-through trait of a material and is considered to be a crucial attribute in packaging applications. It is a well-known fact that the greater the transmittance value, higher is the transparency as more visible light can get access through the films [27]. In this present study, transmittance values for all the polymeric compositions were tested and recorded with wavelength ranging from 200-800 nm as elucidated in **Table 2.8**. PEA 0% composition (devoid of any poly(ester amide) content) rendered transmittance value of almost 70% at 800 nm, wherein PEA 77% composition exhibited transmittance of 74% at 800 nm. Howsoever, increment of poly(vinyl alcohol) amount did not affect transmittance values enormously. Overall, all the polymeric films laid out good transmittance values and displayed transparency traits as shown in **Figure 2.7**.

2.3.9. Biodegradation study

Biodegradation study of poly(ester amide) at different compositions with poly(vinyl alcohol) was accomplished against bacterial strain, *Bacillus subtilis*, an aerobic and gram-positive soil bacterium for a period of 25 days. It was revealed from thorough investigation that the polymeric surface got eroded in an extensive manner as displayed in **Figure 2.8**. (a) & (b). This is indicative of the bottom-line fact that the polymeric sample acted as a catabolic source [28]. Since the poly(ester amide) resin is composed of ester linkages which are prone to hydrolysis, biodegradation implied rupturing of the polymeric framework into smaller chunks. Herein, poly(ester amide) and its varied compositions with

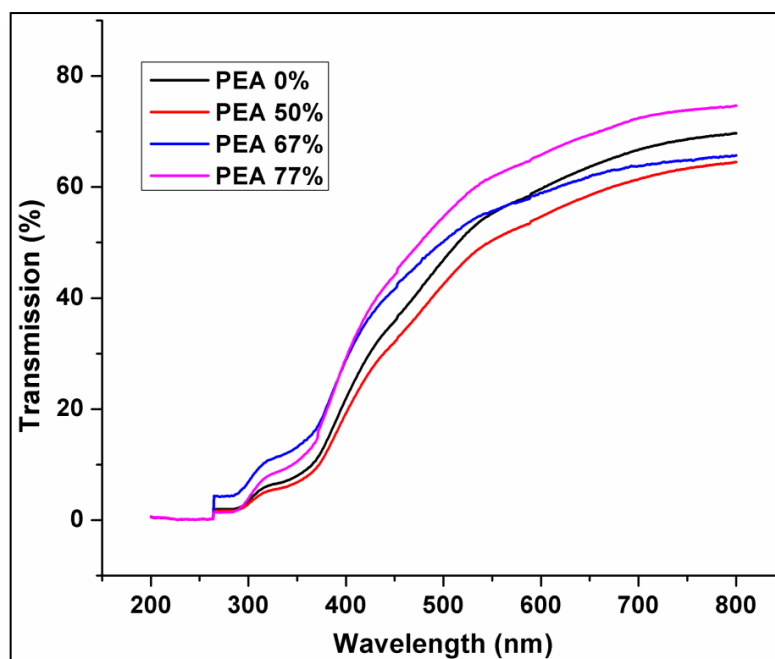


Figure 2.7. UV-transparency curves of poly(ester amide)s.

Table 2.8. Transmittance values of poly(ester amide) films at different compositions with poly(vinyl alcohol).

Poly(ester amide) compositions	Transmittance (%) (at 300 nm)	Transmittance (%) (at 800 nm)
PEA 0%	3.42	69.68
PEA 50%	2.86	64.50
PEA 67%	6.86	65.69
PEA 77%	3.88	74.64

poly(vinyl alcohol) were introduced into a basal media which resulted in subsequent growth of bacteria, followed by disruption of the polymeric surface by carrying out scission of ester linkages into tiny fragments or moieties which were further diffused and solubilized to produce by-products such as water, CO₂, etc. [29, 30]. Furthermore, it could be interpreted that increment of poly(ester amide) content resulted in enhancement of biodegradation rate as predicted by the weight loss profile diagram showcased in **Figure 2.8. (b)**. This inferred that incorporation of citric acid content resulted in greater number of ester linkages ensuing considerable biodegradation rate.

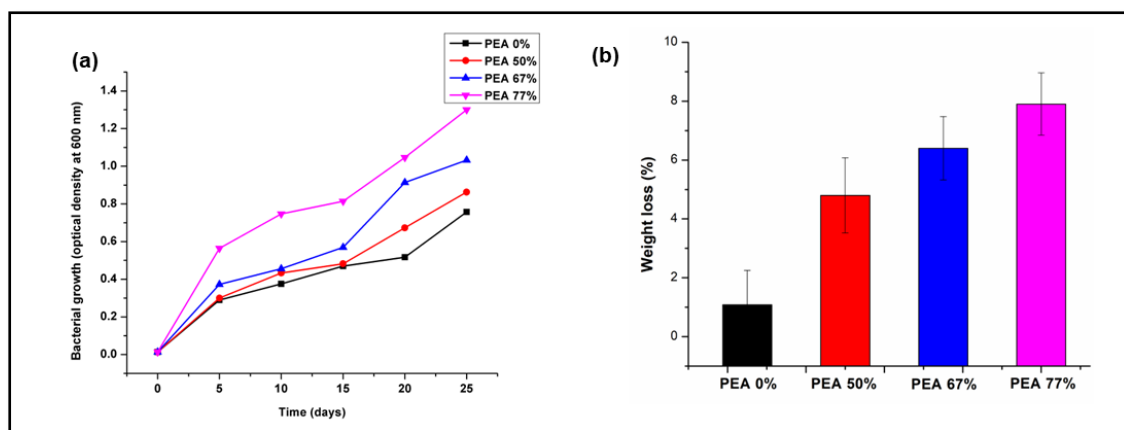


Figure 2.8. (a) Variations of *Bacillus subtilis* growth, (b) weight loss (%) profiles of polymeric films of various compositions against exposure time.

SEM images of original and degraded poly(ester amide) are shown in **Figure 2.9. (a) & (b)** which further elucidated the expanse of degradation after carrying out the biodegradation study for a time period of 25 days. The profound rate of degradation can be further confirmed by comparing the SEM image of degraded one with that of the controlled one (devoid of any bacterial strain).

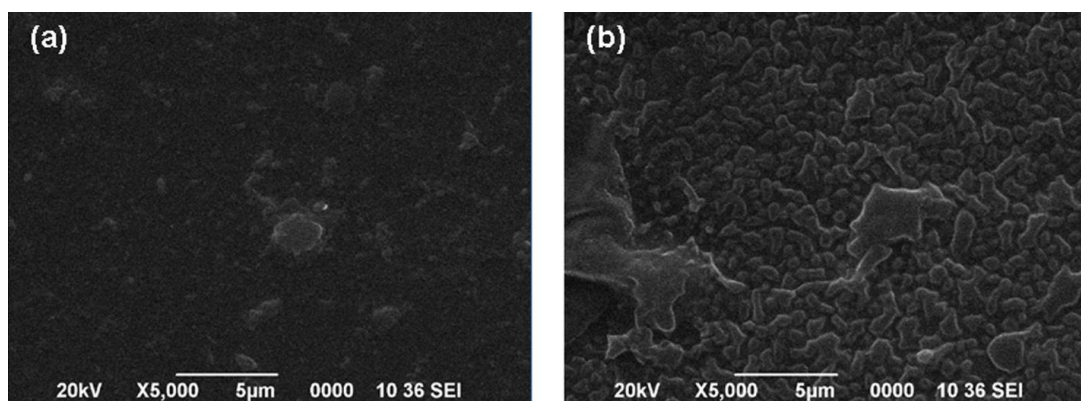


Figure 2.9. SEM images of PEA 77% composition (a) control and (b) biodegraded by *Bacillus subtilis*.

2.4. Conclusion

From an overall inclusive study, it can be contemplated that a bio-based, tough, water-soluble poly(ester amide) can be fabricated utilizing an environmentally congenial route bereft of any organic solvent. Citric acid, glycerol and hexamethylenediamine are

considered as the potent key components in synthesizing the neutral functionalized poly(ester amide) resin. Most of the reactants are biodegradable in nature; thereby adhering to the norms of clean synthesis coupled with conferring desirable characteristics to the poly(ester amide) and its varied compositions with poly(vinyl alcohol). The prepared polymeric compositions demonstrated decent mechanical properties, aqueous solubility, thermal properties, chemical resistance as well as notable biodegradation features, which revealed their potential applications as coating materials. The presence of the aforementioned attributes can credit their usage as a potent viable sustainable material and can be further employed in diverse commodity utilities, especially as protective coating materials.

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