CHAPTER - 6

SYNTHESIS AND CHARACTERIZATION OF TARTARIC ACID BASED POLYAMIDES

6.1 Preview

In past decades a great deal of research has been done on the synthesis of polymeric materials for long-term service. In particular, many kinds of thermally and chemically stable polymeric materials have been developed. Today however, a new field of research has developed, which is concerned with the synthesis of biodegradable polymers and studies of their properties[1]. This field of research is growing rapidly because of the strong demand for such polymers as packaging materials and biocompatible materials for specific medical applications due to the potential for their easy disposal and biodegradation under natural environment conditions. Hence, current attempts to develop biodegradable polymers have focused on the synthesis of new polymers and on the modification of natural polymers which show the excellent properties of strength and durability designed into plastics to meet end use requirements and to overcome / solve the problems associated with their disposal. For all these reasons, the interest for synthesis of polymers containing carbohydrate units in the main chain is steadily increasing because of their potential as biodegradable and biocompatible materials[2]. Moreover, the use of carbohydrate-based monomers open the access to polycondensates containing several asymmetric centers in the backbone of the repeating unit. The conformational properties of these stereoregular polyhydroxypolyamides are object of current investigation for their capacity to form helical structure containing large cavities of varying geometry depending on the configuration of parent carbohydrate (i.e. tartaric acid) moieties[3]. Several reviews dealing with these classes of polymers have appeared in the recent literature[4]. Moreover, tartaric acid-based polyamides had also been explored by Rodriguez-Galan et al.[5] to study their stereoregular optically active properties. Ducret et al.[6] reported enzymatic preparation of fatty esters of carbohydrates and studied their surface properties such as surface tension, interfacial tension and their ability to stabilize emulsion at 30^0C. Paredes and coworkers[7] reported synthesis of the alternating copolyamide consisting of l-alanine and 11-aminoundecanoic acid by the active ester method. This copolyamide showed its potential applications as a family of biodegradable and biocompatible polymers. Recently, Ennis and others[8] have suggested that polyamides, such as nylon-66 can be modified by N-acetyl substitution to produce materials that are both bio- and photodegradable. Beside, Arvanitoyannis and colleagues[9] have recently investigated the synthesis and properties of two novel series of biodegradable oligo(ester amide)s based on diacids (i.e. sebacic acid and octadecanedioic acid), 1,6-hexanediamine and ε-caprolactone, which could find applications either in reactive blending with polymers or as comonomer units (in prepolymers) for synthesizing '*tailor-made*' polymers. In this Chapter, we present the results of our study of the synthesis and properties of copolyamides from DL-tartaric acid with various diamines viz. ethylene diamine, hexamethylene diamine and benzidine.

6.2 Materials

DL-tartaric acid (DTA, Suvidhinath Lab., Baroda, India), hexamethylene diamine (HMDA, Fluka, Switzerland), benzidine (Suvidhinath Lab., Baroda, India) and sulphuric acid (98%, AR, SD's Fine-Chem Ltd., India) were used as received. Ethylene diamine (EDA, 97%, Fluka, Switzerland) was distilled prior to use and middle fraction was used. All solvent (Qualigens, India) used in this study were freshly distilled before use.

6.3 Experimental Methods

The FTIR spectra of the copolymers were recorded on a Bomem-MB140, Canada, FTIR spectrometer. For FTIR spectra, the solid polymers and KBr (spectroscopic grade) were thoroughly mixed and this mixture was pressed to form a pellet, whereas liquid sample was spread over the NaCl cells and the spectra were recorded. ^{1}H-NMR spectra of copolymers solutions were recorded under standard conditions at room temperature in deuterated D_2O on a JEOL JNM FX-100, FT-NMR spectrometer operating at 100 MHz. TMS was used as an internal reference and D_2O as an internal lock.

The elemental analysis (viz., C and H) was carried out with a Coleman C, H analyzer and nitrogen was estimated by the Dumas method. The TGA was recorded on a Shimadzu thermal analyzer DT-30B. The TGA analysis was done in the presence of air at heating rate of 10 K min^{-1}. Viscosity studies of different solutions were carried out with the help of an Ubbelohde viscometer, placed vertically in a thermostat, at all required temperatures (± 0.05°C).

In the biodegradation studies, minimal salt medium (1L, pH 7.0) which contained [gL^{-1}] 4.3 K_2HPO_4, 3.4 KH_2PO_4, 0.3 $MgCl_2{\cdot}H_2O$, and 0.5 mL of trace element solution having [mgL^{-1}] 1.0 $MnCl_2.4H_2O$, 0.6 $FeSO_4.7H_2O$, 2.6 $CaCl_2.H_2O$, 6.0 $NaMoO_4$ was used[10] for growth studies. Ammonium sulphate [1.0 g L^{-1}] was supplied as a nitrogen source. Polyamide (NM-1A) containing DL-tartaric acid and ethylene diamine as monomers was added to the above medium as the sole source of carbon and energy and the medium was sterilised by autoclaving at 10 psi for 20 min. Minimal medium without the polymer was also inoculated and served as a control.

Following the conventional enrichment culture technique and using 1% (w/v) polymer (NM-1A) in minimal medium as a sole carbon and energy source a bacterial culture designated here as TED-1, capable of growth on the polyamide, was isolated after three enrichments in the minimal medium by streaking onto Luria agar.

For growth studies, cultures were cultivated in 5.0 mL of the minimal medium in 50 mL culture tubes at 30°C on a rotary shaker at 180 rpm. The growth was followed (i) turbidimetrically at 600 nm and (ii) by estimating the whole-cell protein. For whole-cell protein determination, cellular proteins were solubilised by treating the cells (approx. 100 μg dry weight in 0.5 mL) with 0.5 mL of 1.0 N NaOH at 100°C for 5 min and were subsequently estimated by the method of Lowry et al.[11].

6.4 Polymer Synthesis

The condensation polymerization process was used to synthesize polyamides using tartaric acid and various diamines viz. ethylene diamine, hexamethylene diamine and benzidine. These polyamides were synthesized by a two stage process.

(a) Synthesis of diethyl tartarate from tartaric acid and

(b) Synthesis of various polyamides using different diamines and diethyl tartarate.

(a) Synthesis of diethyl tartarate :

Diethyl tartarate was synthesized by refluxing a mixture of tartaric acid (0.1 mol), absolute ethanol (1.0 mol) and concentrated sulphuric acid (2.0 mL) for 24 hours. Excess alcohol was distilled off under vacuum and the reaction mass was poured in 200 mL diethyl ether. The crude ester layer was separated and traces of acid was neutralized by addition of sodium bicarbonate until effervescence ceases. The crude ester was passed through anhydrous sodium sulphate bed to remove traces of moisture. Transparent viscous diethyl tartarate was obtained on evaporation of ether in bulb oven at ~50^{0}C.

(b) Synthesis of polyamides:

Diethyl tartarate (0.01mol), diamine (0.011mol), N-methyl pyrrolidone (NMP, 25.0 mL) were taken in a 100mL round-bottom flask attached to a reflux condensor. The mixture was stirred magnetically for 6 hours in a water bath kept at 80°C. The mixture was then poured into excess of nonsolvent (acetone/water) to precipitate the polyamide.

The polyamide of ethylene diamine was also synthesized using alcohol and benzene as solvent mediums. In this route, as condensation reaction proceeds, the polyamide gets precipitated. Various synthesized products are represented as follows:

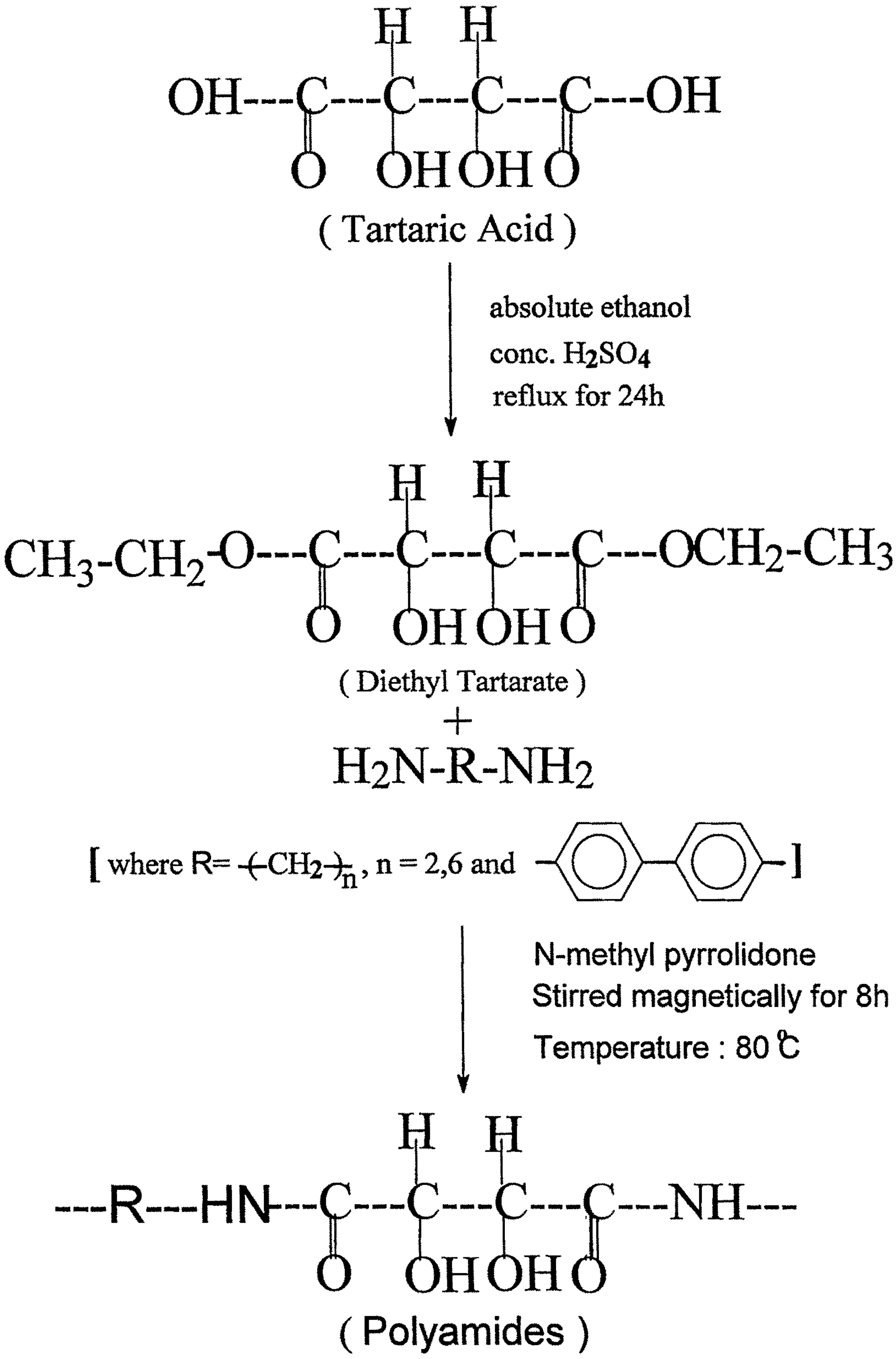
nylon-2,4 (A) / [NM-1A] : reaction product of ethylene diamine with tartaric acid in alcohol

nylon-2,4 (B) / [NM-1B]: reaction product of ethylene diamine with tartaric acid in benzene.

nylon - 6,4 / [NM-2] : reaction product of hexamethylene diamine with tartaric acid in NMP

nylon - 12,4 / [NM-3] : reaction product of benzidine with tartaric acid in NMP.

The schematic representation of polyamides synthesis is given as follow.


(Tartaric Acid)
absolute ethanol
conc. H2SO4
reflux for 24h
(Diethyl Tartarate)
+
H2N-R-NH2
[where R= -(-CH2-)-n , n = 2,6 and
N-methyl pyrrolidone
Stirred magnetically for 8h
Temperature : 80 ℃
(Polyamides)

6.5 Results and Discussion

(a) Elemental analysis:

The percentage of nitrogen present in polyamides was estimated by Dumas method. The elemental analysis results (Table 6.1) indicate the polyamide formation.

(b) FTIR spectroscopic analysis of diethyl tartarate and polyamide:

Conformation and environmental changes of polymers at the molecular level were obtained by FTIR spectrum. Ester of tartaric acid (i.e. diethyl tartarate) shows very sharp ester stretching vibration at 1745 cm^{-1} (Fig. 6.1). Whereas hydroxyl (-OH) stretching was observed as a broad peak in the region 3250-3450 cm^{-1}. Besides this, other prominent peaks at their respective positions confirm diethyl ester formation. In case of polyamide (Fig. 6.2) the carbonyl stretching frequency was observed at 1640 cm^{-1}. Whereas secondary -NH- stretching and bending vibrations were observed at 3285 and 1548 cm^{-1}. These give clear evidences for amide formation. Other functional groups also showed characteristic IR absorption bands, which agreed well with those reported in the literature[12].

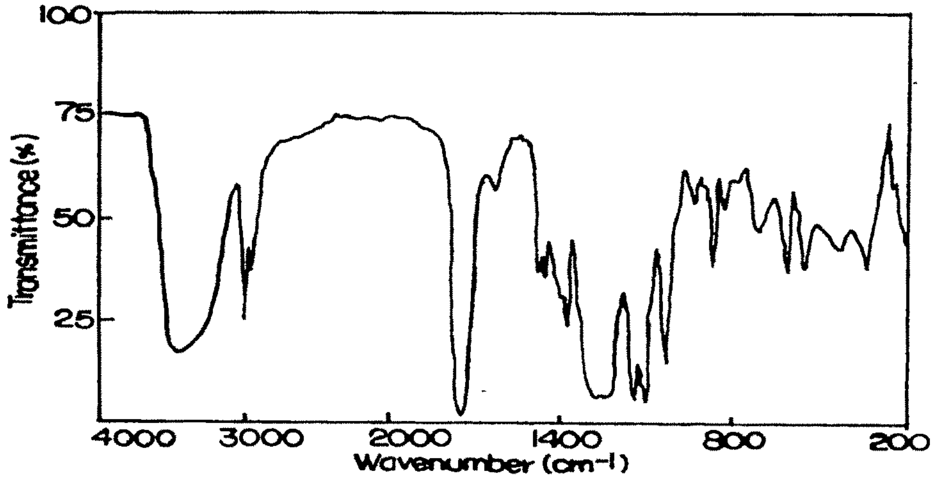


Figure 6.1 FTIR spectrum of diethyl tartarate.

Table - 6.1: Elemental Analysis Results for Various Polyamides.

Polymer Samples	N%	C%	H%
Nylon-2,4(A)	15.58	38.83	6.27
	(16.09)	(41.38)	(5.75)
Nylon-2,4(B)	15.55	40.98	6.49
	(16.09)	(41.38)	(5.75)
nylon-12,4	13.82	71.99	5.90
	(9.40)	(64.43)	(4.70)

Date in the parenthesis indicates theoretical values.

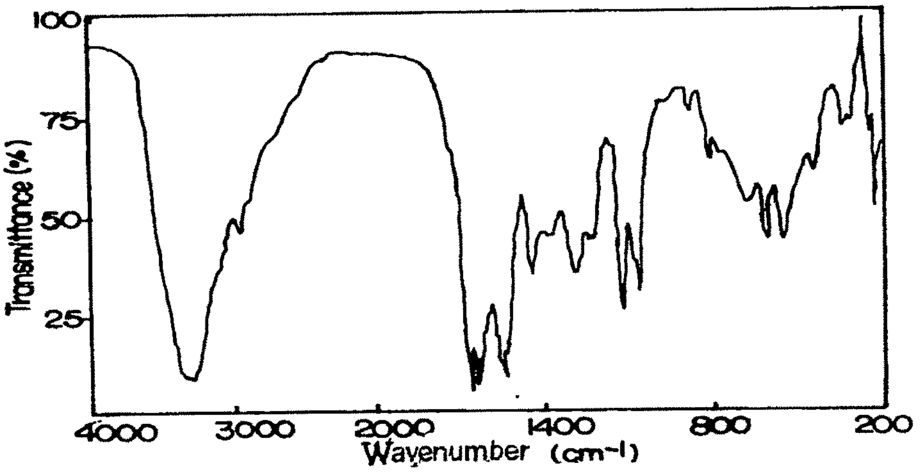
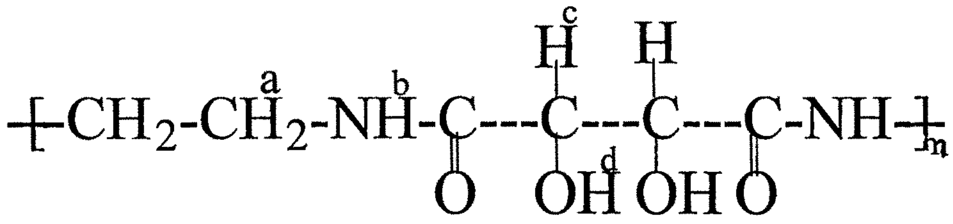


Figure 6.2 FTIR spectrum of polyamide (NM-1A).

(c) ^{1}H-NMR spectroscopic analysis:



$$\left[CH_2\text{-}C\overset{a}{H}_2\text{-}N\overset{b}{H}\text{-}\underset{\underset{O}{\|}}{C}\text{---}\underset{\underset{O\overset{d}{H}}{|}}{\overset{\overset{\overset{c}{H}}{|}}{C}}\text{---}\underset{\underset{OH}{|}}{\overset{\overset{H}{|}}{C}}\text{---}\underset{\underset{O}{\|}}{C}\text{-}NH \right]_n$$

Further evidence for the formation of polyamide was supported by nuclear magnetic resonance (NMR) spectroscopy. The peaks corresponding to structure given above were obtained. In PNMR spectrum (Fig.6.3) the signals that occur around δ=3.09 ppm are associated with the methylene protons (a), whereas methine protons (c) appear in a region of δ = 3.46-3.54 ppm .The signals at δ=4.38-4.52 ppm corresponding to hydroxyl protons (d) and δ=4.57-4.61 ppm represent the absorption of secondary amide protons (b). All these signals agreed well with those reported in literature[12].

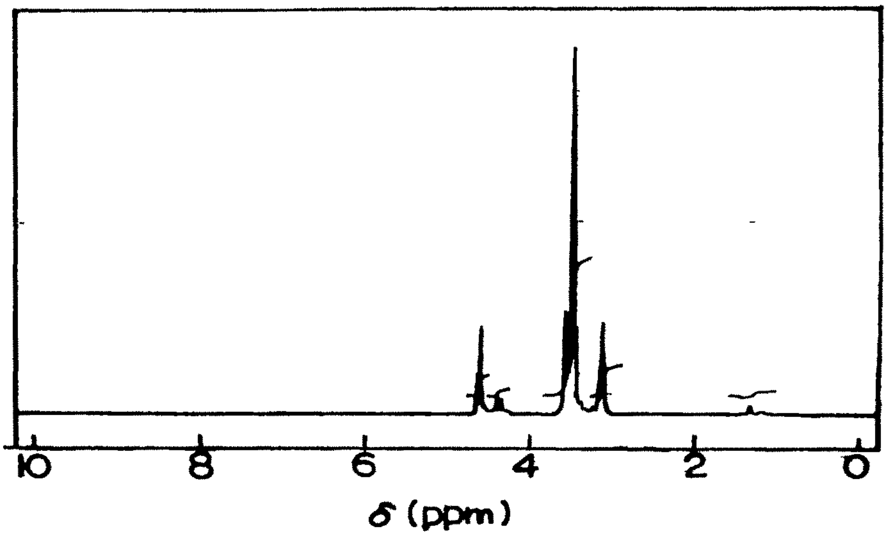


Figure 6.3: ^{1}H-NMR spectrum of polyamide (NM-1A).

(d) Solubility test :

All polyamides remain insoluble in methanol, acetone and benzene. However, they are soluble in polar solvents like N-methyl pyrrolidone (NMP), dimethyl sulfoxide (DMSO), dimethyl formamide (DMF), and 1,4-dioxane. Nylon-2.4(A) and (B) are soluble in cold water, whereas nylon-6,4 is soluble in hot water, however nylon-12,4 remains insoluble even in hot water.

(e) Thermal analysis:

The thermogravimetric data provide information regarding the thermal stability of the polymers. The thermogram of the polyamides nylon-2,4(A), nylon-2,4(B) , nylon-6,4 and nylon-12,4 are shown in Figure 6.4. All the polyamides showed two stage decomposition due to the rapid decomposition to water, carbon dioxide, ammonia, hexamethyleneimine and other volatile products[13]. The activation energy associated with each stage of decomposition was evaluated by the well-known Broido method[14]. The equation used for the calculation of the activation energy (E_a) was

$$\ln \left| \ln (1/Y'') \right| = (-E_a/R)(1/T) + \text{Constant}$$

where, $Y'' = (W_t - W_\infty) / (W_0 - W_\infty)$

that is, Y" is the fraction of the number of initial molecules not yet decomposed; W_t is the weight at any time t; W_∞ is the weight at infinite time (=zero) and W_0 is the initial weight.

On plotting ln $|\ln (1/Y'')|$ versus 1/T (Fig.6.5) a straight line is observed. The slope yielded activation energy. The calculated values for the activation energy of decomposition are listed in Table 6.2.

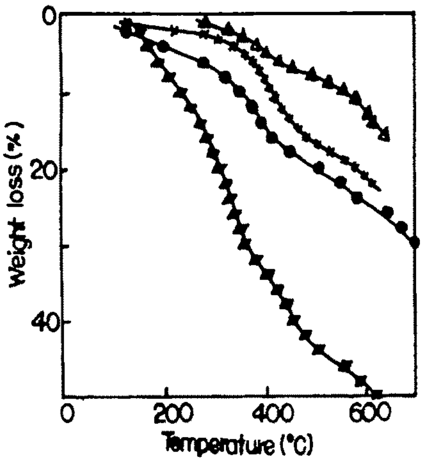


Figure 6.4 TGA plots for polyamides : (●) nylon-2,4(A), (■) nylon-2,4(B), (▲) nylon-6,4 and (x) nylon-12,4 at heating rate of 10^0 K min^{-1}.

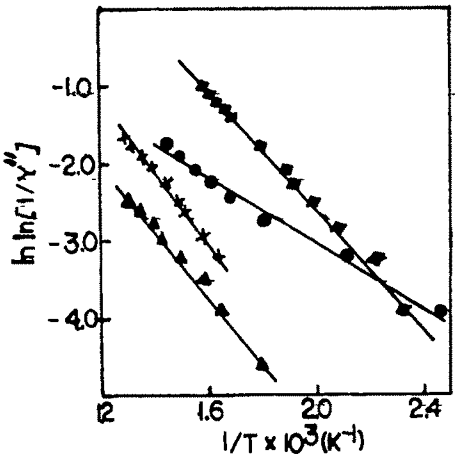


Figure 6.5 Activation energy plots for the polyamides: (●) nylon-2,4(A), (■) nylon-2,4(B), (▲) nylon-6,4 and (x) nylon-12,4 from the experimental data of Fig. 6.4.

It is observed from the Table 6.2 that the activation energy associated with second stage of decomposition is lower than that of the first stage. In case of nylon-6,4, having a long flexible chain showed lower weight loss and higher activation energy of decomposition indicating a higher thermal stability.

Table 6.2: Activation Energy of Decomposition for Various Polyamides by Thermogravimetric Analysis.

Polymer Samples	Decomposition Temperature Range (⁰C)	Weight Loss (%)	Activation Energy[a], E_a ($kJmol^{-1}$)
nylon-2,4(A)	132-416	16%	17.0
	454 -701	30%	13.9
nylon-2,4(B)	155-360	30%	28.3
	380-620	50%	11.7
nylon-6,4	286-495	08%	37.2
	530-615	14%	33.5
nylon-12,4	342-501	17%	40.7
	529-614	22%	15.8

[a] Calculated using Broido method at a heating rate of 10 K min^{-1} in air

(f) Biodegradation studies:

Biodegradation studies of the polyamide [NM-1A] of DL-tartaric acid (DTA) and ethylene diamine was carried out in inorganic minimal salt medium. When the individual polymer (NM-1A) was supplied as a sole carbon and energy source in a synthetic medium at a concentration of 0.5% (w/v), the isolate TED-1 was found to grow on the polymer as shown in Figure 6.6. The above isolate was found to be a gram negative, rod shaped bacterium. The isolate did not grow in the absence of the polymer in the synthetic medium. Thus, for the growth of the isolate TED-1, the presence of the polymer in the medium was found to be essential and thereby indicating the biodegradable nature of this polymer. Using a similar strategy, degradation of copolyesters of citric acid-1,2,6-hexane triol[15], succinic acid-glycerol-polyethylene glycol-200 copolymer[16] and tercopolymers of acrylamide, acrylic acid and acrylonitrile[17] has been reported. The isolate grew without a lag period and the maximum growth reached at ~ 30 h is shown in Figure 6.7. This observation was further confirmed by estimating the biomass of growth in terms of whole-cell protein, where the increase in biomass followed a similar pattern as above.

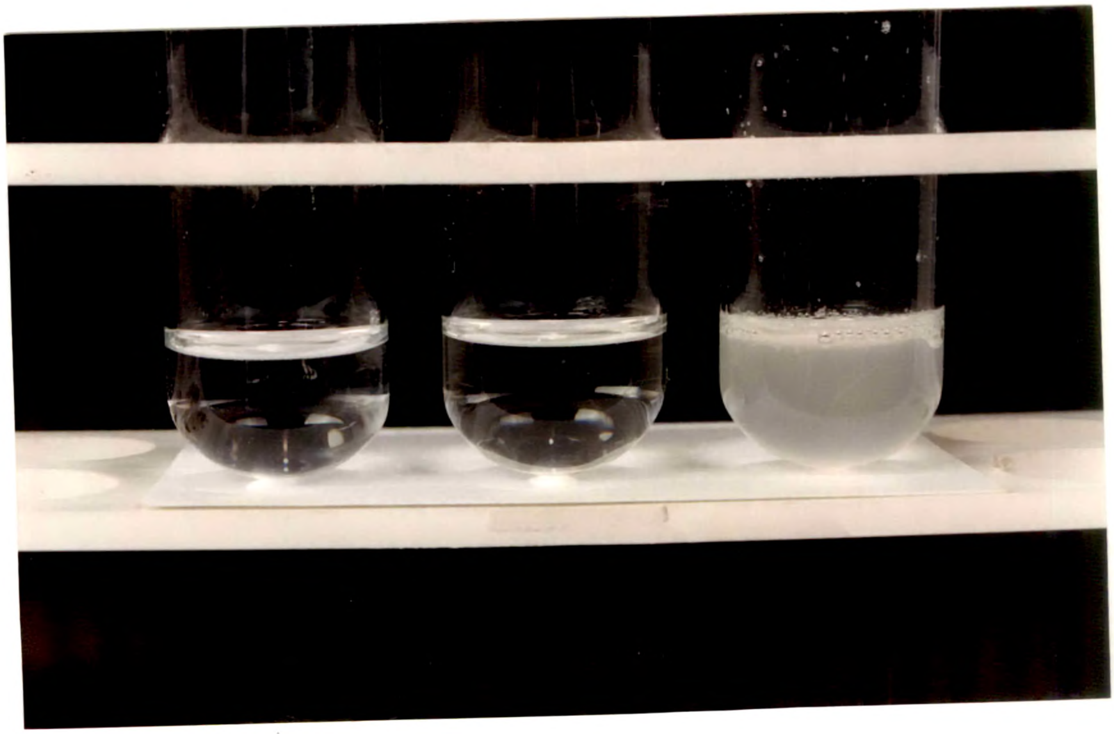


Figure 6.6: Growth* of the organism TED-1 on polyamide (NM-1A) after 72h.

***From left to right :**

a) minimal medium without polymer inoculated,
b) minimal medium with polymer uninoculated and
c) minimal medium with polymer inoculated.

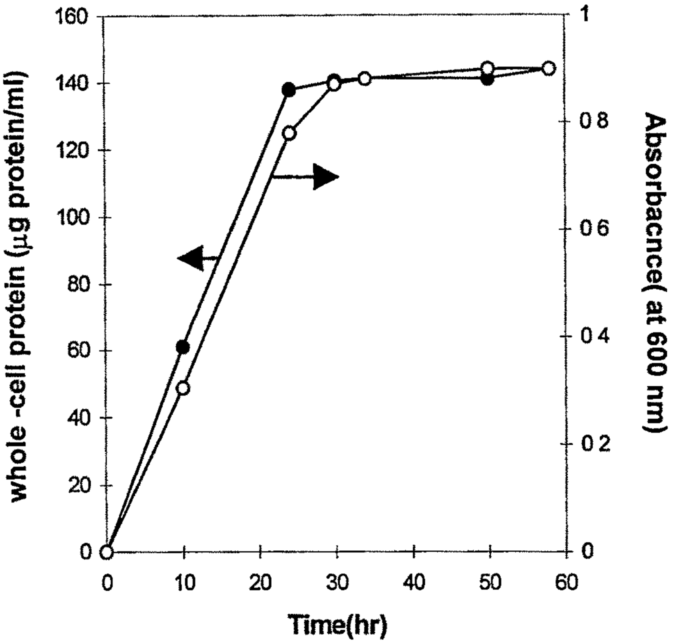


Figure 6.7: Time course of the growth* of isolate TED-1 on polyamide (NM-1A)#.

* Growth was determined in terms of absorbance at 600 nm (O) and whole-call protein (●).

\# Polyamide (NM-1A) was supplied as a sole carbon and energy source in minimal medium at a concentration of 0.5% (w/v). [Control had no polymer and was inoculated.]

6.6 Conclusion

A series of polyamides were synthesized using condensation technique. The properties of polyamides vary with respect to their constitution. All the polymers were characterized by elemental analysis, FTIR, PNMR, solubility test, thermal analysis and biodegradation studies. The spectroscopic techniques provide information regarding the structure and environmental change of the polymers. Polyamides of ethylene diamine are soluble in cold water while that of hexamethylene diamine was soluble in hot water. However, the polyamide of benzidine remain insoluble even in hot water. The activation energy of decomposition (E_a) associated with second stage of decomposition is lower than that of the first stage. Higher thermal stability was observed for nylon-6,4 in comparison to other polyamides. Preliminary studies carried out in terms of growth supporting ability of the polymer indicated the biodegradable nature of the polymer.

6.7 References

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