ELSEVIER

Thermochimica Acta 340–341 (1999) 195–203

thermochimica acta

www.elsevier.com/locate/tca

Degration of a starch based polymer studied using thermal analysis

Poonam Aggarwal

Department of Chemistry, University of Toledo, Toledo, OH 43606, USA

Accepted 6 August 1999

Abstract

Biodegradable polymers are being actively pursued as possible solutions to the solid waste disposal dilemma. Plastics require between 6% and 22% starch concentration in order to be classified as biodegradable; however the ability to monitor such levels are limited with poor quantitative correlation. The goal of this research was to introduce a quick and accurate method to measure the removal of the starch from the polymer matrix. A biodegradable copolymer was synthesized with polyacrylonitrile and the removal of starch by amylase treatment followed utilizing thermal analysis. © 1999 Elsevier Science B.V. All rights reserved.

Keywords: Biopolymer; Starch; Thermal analysis

1. Introduction

The persistence of petrochemical plastic materials in the environment beyond their functional life has resulted in a broad range of pollution, litter, and waste disposal problems for the society. Research to alleviate these problems include efforts to develop plastics that degrade more rapidly in the environment [1].

Recently plastic formulations containing 6–50% corn starch to increase the susceptibility of plastic products to biological degradation have been studied [2–5]. However, there is very little published information available on the rates, mechanism and extent to which starch containing plastics are degraded by living systems, making them biodegradable. Degradation can be followed by measuring changes in physical properties of the product, by studying chemical changes in the film or by assessing biological activity [6]. They are considered to be biodegradable because the removal of starch from the starch polymer composite can cause severe reduction in the mechanical strength of the remaining nonbiodegradable portions, so that it disintegrates readily into smaller pieces. Breakdown of the polymer weakens the materials by reducing the chain length (molecular weight) eventually to a level that can then be metabolized [7,8] by microorganisms.

Starch, especially from corn, is probably the most abundant and economical natural polymer available and its use in plastics production would greatly reduce the demand for petrochemicals and the negative impact on the environment caused by discarding plastics that do not biodegrade. Starch alone cannot be used in plastic production as it forms a brittle product once hydrated with water. It has to be combined with other materials in order to produce a satisfactory film. Methods to determine the degradation of such materials have proven inadequate either because of the length of time necessary to perform the tests or lack of their reproducibility and quantitative

0040-6031/99/$ – see front matter
PII: S0040-6031(99)00264-6

characterization caused by microbial growth and the production of metabolic products.

In this study a method using specific enzymes was developed for in vitro testing of the extent of degradation due to the removal of starch break down products by amylase bacteria.

The biodegradation of plastics and the additives which have been incorporated into these polymers can be thought of as taking place in two steps. The initial stage results in the formation of a deteriorated plastic, followed by a second phase degradation which results in the actual breakdown of the material. In the case of corn starch, hydrolyzing enzymes from the microorganisms convert the starch into breakdown products. This loss of starch from the polymer matrix weakens the polymer resulting in its partial breakdown. The objective of this study was to monitor and follow the primary breakdown events. The evaluation of the secondary degradation phase is inevitably a much slower event. It is hoped that the present study makes a contribution to a better understanding of the initial degradation step, specifically the role of microbial enzymes in the hydrolysis of biodegradable additives and fillers such as starch. In order to accomplish this objective, starch was copolymerized with acrylonitrile. Graft copolymers were prepared by first generating free radicals on starch and then allowing these free radicals to serve as macro initiators for vinyl or acrylic monomers [9–12].

The initial stage of degradation of this copolymer was then followed by treatment with glucoamylase. After the first stage of degradation the partially degraded material was characterized by FTIR, SEM, and TG by comparison with the undegraded copolymer and the individual polymers, namely starch and polyacrylonitrile.

2. Materials and methods

2.1. Starch preparation

Cationically modified corn starch was prepared as per Car and Bagby [13]. Sodium sulfate, nitric acid and ceric ammonium nitrate were from J.T. Baker, acrylonitrile was from Aldrich Chemical Company and glucoamylase at a concentration of 22 500 IU g^{-1} was from Sigma Chemical.

2.2. Synthesis of starch graft copolymer

The starch polyacrylonitrile graft copolymers were prepared by the method of Fanta and Bagley [14]. 22 g of cationic corn starch, was gelatinized in 500 ml distilled water under an atmosphere of nitrogen at 95°C for 10 min. The solution was then cooled to 25°C. At the same time the acrylonitrile was freed from the inhibitor by flash distillation under reduced pressure at 40°C and stored in the dark at 34°C. Ceric ammonium nitrate solution was prepared in 1N HNO_3 and aged 24 h before using. Ceric ammonium nitrate in nitric acid was added to gelatinized corn starch and stirred for 20 min, followed by addition of acrylonitrile. This was then stirred for 3 h under nitrogen. The copolymer was precipitated with ethanol and washed to remove excess reagents with distilled water. A portion of this copolymer formed was then mixed with dimethyl sulfoxide (DMSO). The extraction was carried out for 3 h under stirring. The solvent was then separated from the copolymer by vacuum filtration, washed free of solvent and dried at room temperature. Most of the copolymer remained insoluble in DMSO.

2.3. Preparation of enzyme solutions

In the standard assay method the biodegradation of starch in plastic films was based upon the measurement of glucose produced as the starch was hydrolyzed by a mixture of amylases. The enzyme mixture was prepared as a ten-fold concentrate containing 200 IU ml^{-1} of glucoamylase (*Rhizopus sp*) in 0.1 M acetate buffer, at pH 4.8.

2.4. Degradation assays

For the determination of the rate and extent of starch degradation in the polymer samples containing starch, 500 mg of the copolymer was pelletized at 3000 psi. These pellets were then immersed in 5 ml of acetate buffer and treated with enzyme at a concentration of 200 IU ml^{-1}. As a control, pellets containing no enzyme were immersed in the same way with buffer. The samples were then incubated at 37°C. At the appropriate time intervals samples were removed and the glucose concentrations determined using glucose oxidase peroxidase method for glucose determi-

nation. In all the cases appropriate controls were run simultaneously and included samples containing the enzyme only, (no copolymer) the copolymer only, (no enzyme) and the enzyme with the starch based copolymer. This rate was followed for 24 h with samples collected every hour. When the experiments were conducted for a longer time (15 days), the enzyme solution was replenished every 2 days.

2.5. FTIR experiments

FTIR experiments were conducted using a Nicolet 5DX FTIR spectrophotometer at a resolution of $4\ cm^{-1}$ and were an average of 32 scans. For the analysis by FTIR, control and enzyme treated samples were used. The polymer pellets were washed and air dried.

The nitrogen content was calculated by elemental analysis using a Perkin–Elmer Elemental Analyzer, 2400 CHN. The percentage of nitrogen present in the copolymer, in polyacrylontrile, in the starch alone and in the enzyme treated copolymer determined.

2.6. Thermogravimetry data

The thermal analysis equipment used in this study was a simultaneous TG–DTA (thermogravimetry–differential thermal analysis) unit from TA instruments model number 2960, in which the TG and DTA signals are simultaneously obtained on the same sample being thermally treated. In all the experiments a steady flow rate of $100\ ml\ min^{-1}$ was maintained, in an atmosphere of air. Rising temperature experiments were conducted in which the heating rate was $10^{\circ}C\ min^{-1}$. A piece of the copolymer pellet, the enzyme hydrolyzed dried copolymer and polyacrylontrile were analyzed by TG–DTG.

3. Results and discussion

Incorporation of starch into a polyolefin matrix was proposed by Griffin as an effective means of accelerating the deterioration of plastics under biotic environmental exposure conditions [6,15]. The inclusion of starch, a readily biodegradable biopolymer, into the synthetic polymer is believed to result in rapid enzymatic hydrolysis of the starch under biotic exposure conditions, leading to a void containing matrix. The reduced mechanical integrity of the ensuing void containing matrix leads to its facile deterioration and perhaps even promotes subsequent biodegradation of the synthetic polymer, due to the increased surface area available for interaction with microorganisms [16].

Several studies have demonstrated that microorganisms are capable of complete or near complete removal of starch from such plastics. Regardless of the type of microorganisms used, it is the extra cellular microbial amylases which are responsible for the initial degradation of the starch. Therefore, such enzymes must be able to hydrolyze the starch within these polymers. In this study, a glucoamylase based assay was used as an enzyme system to demonstrate the effects that amylases have on the degradation of starch from starch based biopolymers. Glucoamylase is an exoacting enzyme that can act on both α-(1,4) and α-(1,6) glucosidic linkages present in starch and convert it to glucose.

The amount of starch removed from the films during the first 60 h of glucoamlyase treatment are plotted in Fig. 1. Control samples shaken in buffer solution in the absence of the enzyme also lost certain quantities of starch, which are reported as starch leaching. The contribution of glucoamylase activity to starch removal from the film in Fig. 1 can be computed from the difference of starch lost between the enzyme treated samples and their corresponding controls. As can be seen from the figure, the rate of hydrolysis were the highest during the first 24 h of incubation, and all the starch accessible to the enzyme were removed after 48 h of degradation, as there was very little weight loss after that. The standard used for this comparison was glucose. On the other hand the control sample showed barely any changes from the initial reading, indicative of the fact that the enzymes were responsible for the hydrolysis of the starch component of the copolymer.

FTIR spectroscopy was first used to confirm the formation of the copolymer and then to determine starch removal and the oxidative changes taking place in the polymer due to enzyme action. FTIR spectrum of the individual compounds used [17], starch and polyacrylonitrile in the copolymer production are also shown. The IR spectrum of starch is shown in Fig. 2. As can be seen from the plot, starch is characterized by

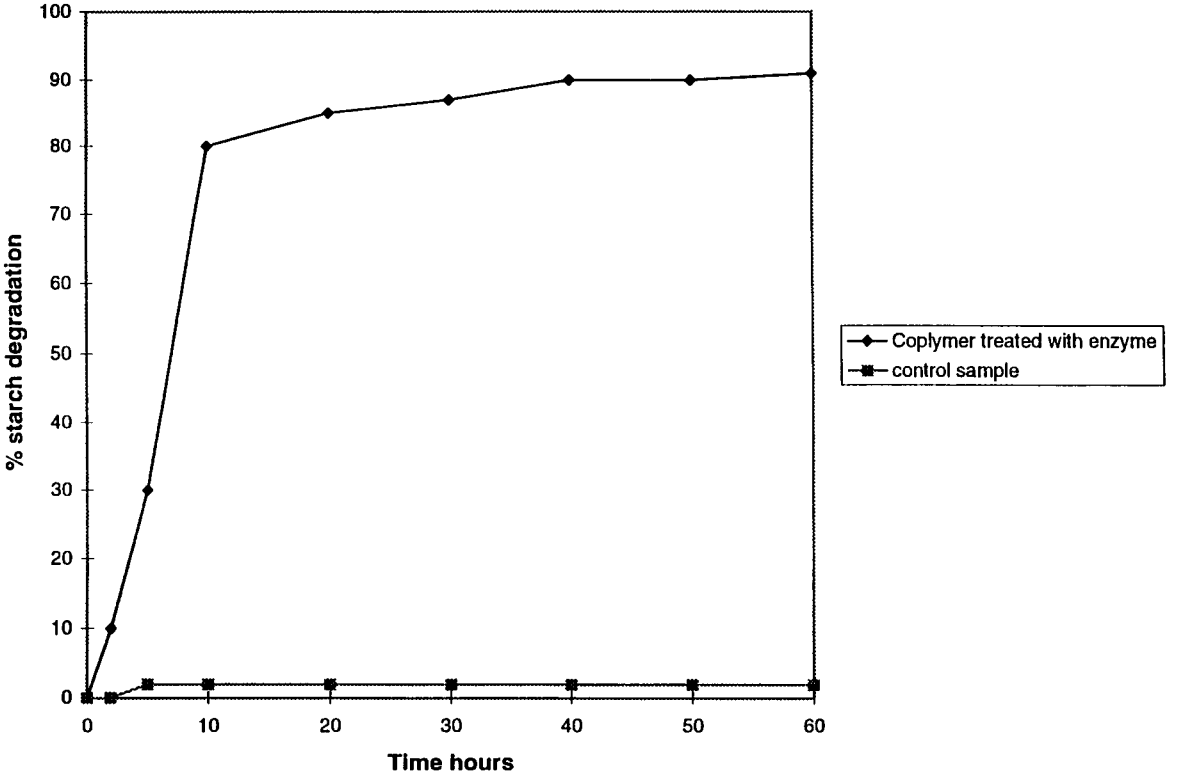

Fig. 1. Degradation of 20% starch/polyacrylonitrile copolymer using glucoamylase.

two strong and broad absorption bands occurring at 3900–3000 cm^{-1}, and at 1250–900 cm^{-1} which are due to O–H and C–O–C stretching. Starch was monitored by a broad OH stretching band centered around 3400 cm^{-1}, a minor C–H stretching band at 2921 cm^{-1} and a characteristic set of strong C–O stretching bands between 960 and 1190 cm^{-1} (C–O–C and C–O–H). These bands can both be used for qualitative starch analysis.

Polyacrylonitrile, on the other hand (Fig. 3) shows three strong bands. The first one at 2927 cm^{-1}, is due to CH_2 stretching, 1252 cm^{-1} due to along the chain bending (CH and CH2) and 1075 cm^{-1} (CH) across the chain bending. The band at 2240 cm^{-1} is due to CN strong stretching vibrations. The FTIR spectrum for the copolymer pellet after 24 h of incubation in sterile medium (Fig. 4) was essentially a composite of spectral features from each of the individual polymer components. Even after incubation for several weeks in sterile liquid medium, there was no apparent alteration in the FTIR spectrum observed. On the other hand, the samples which were treated with amylolytic enzymes, showed a dramatic loss of O–H and C–O absorption bands, indicating removal of starch from the polymer matrix (Fig. 5) due to enzyme hydrolysis. Since starch was the only component of the plastic that exhibited significant absorption bands in the 960–190 cm^{-1} region, it was possible to attribute this change in the spectral features to starch removal from the film. Absorption bands attributable to acrylonitrile were essentially unchanged. The thermal analysis study was conducted using TG. The partially degraded and undegraded polymer samples along with the individual copolymer materials were examined in this way. Thermal studies were designed to determine the effect of temperature on the degradation of plastics treated and untreated with enzyme. It was also used to investigate the possibility of detection of starch removal and presence in the copolymer matrix. The TG technique essentially involves continuous monitoring of the weight of the sample as a function of temperature, using a sensitive micro balance. A typi-

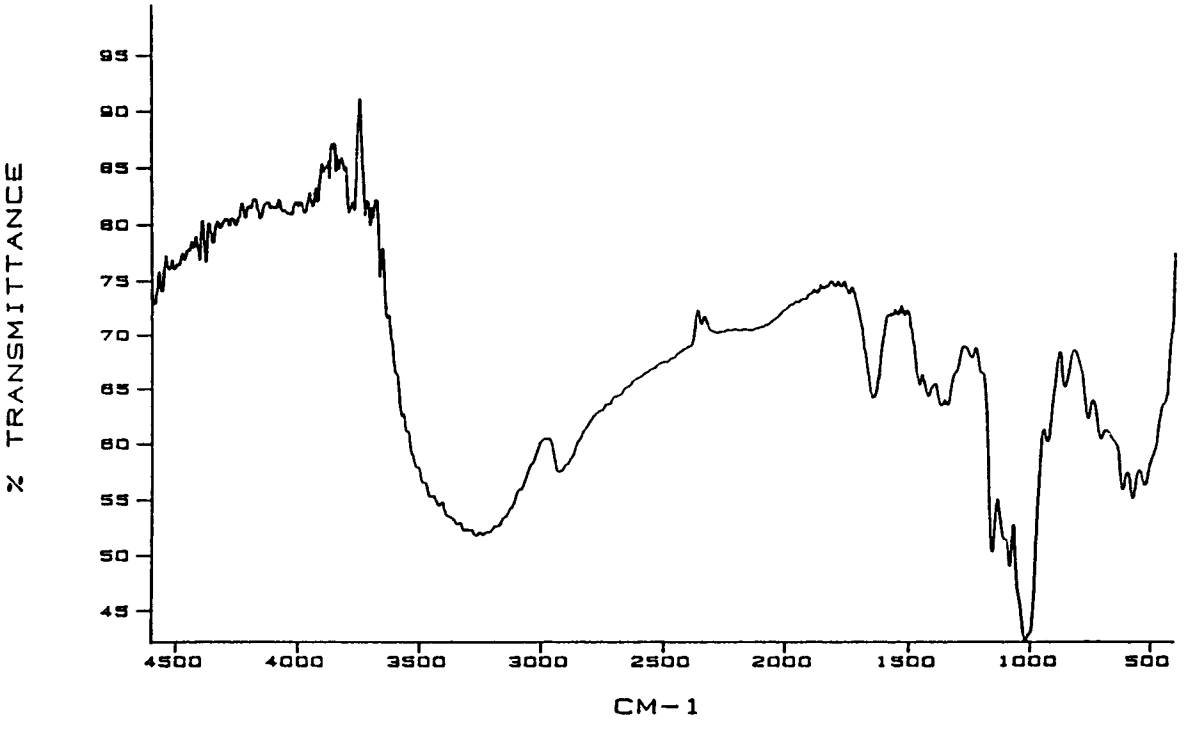

Fig. 2. IR spectrum of cationically modified corn starch (in a KBr pellet).

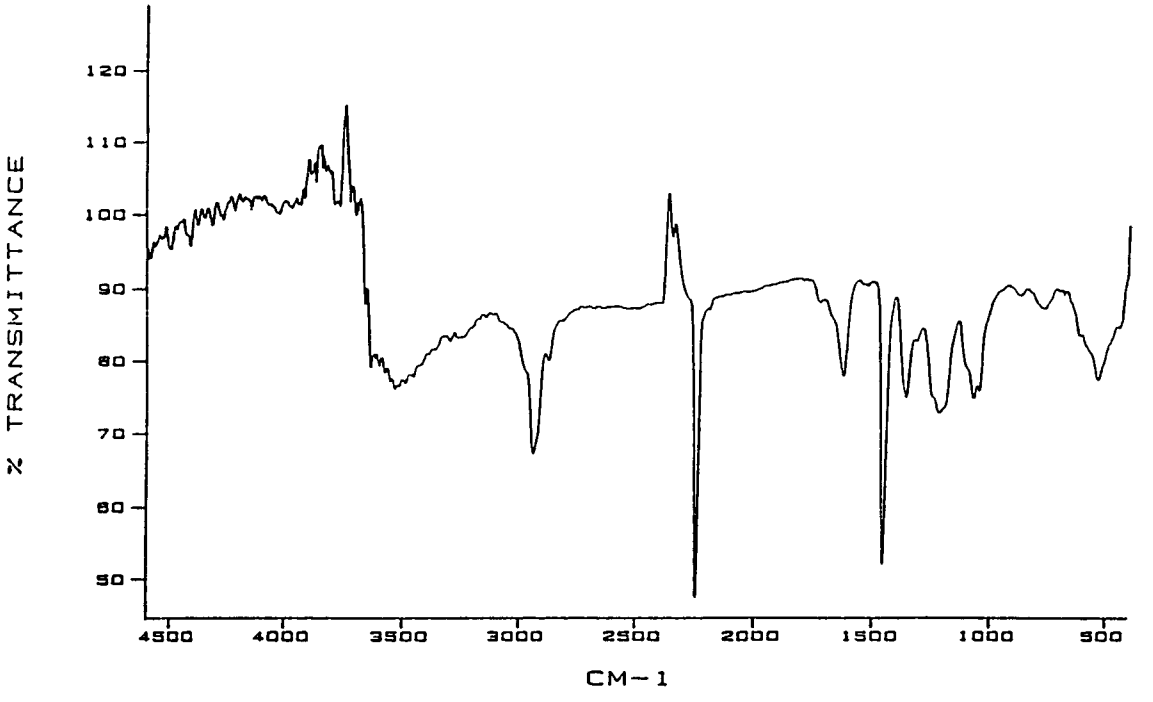

Fig. 3. IR spectrum of polyacrylonitrile (in a KBr pellet).

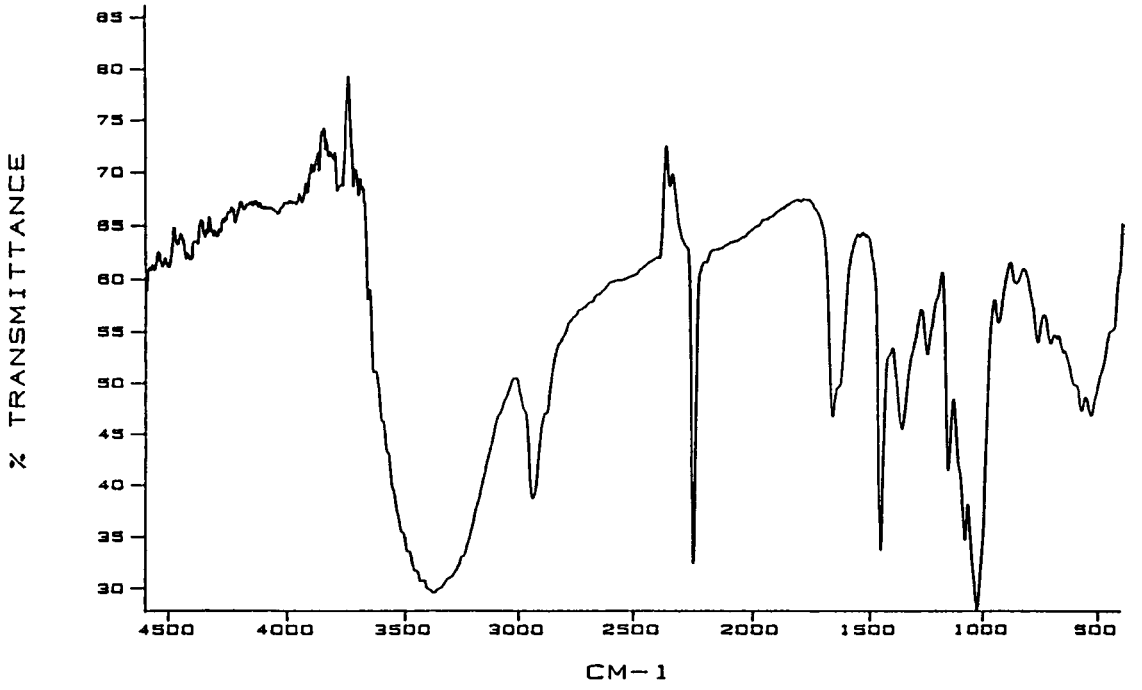

Fig. 4. IR spectrum of the starch–polyacrylonitrile copolymer after aging in sterile medium.

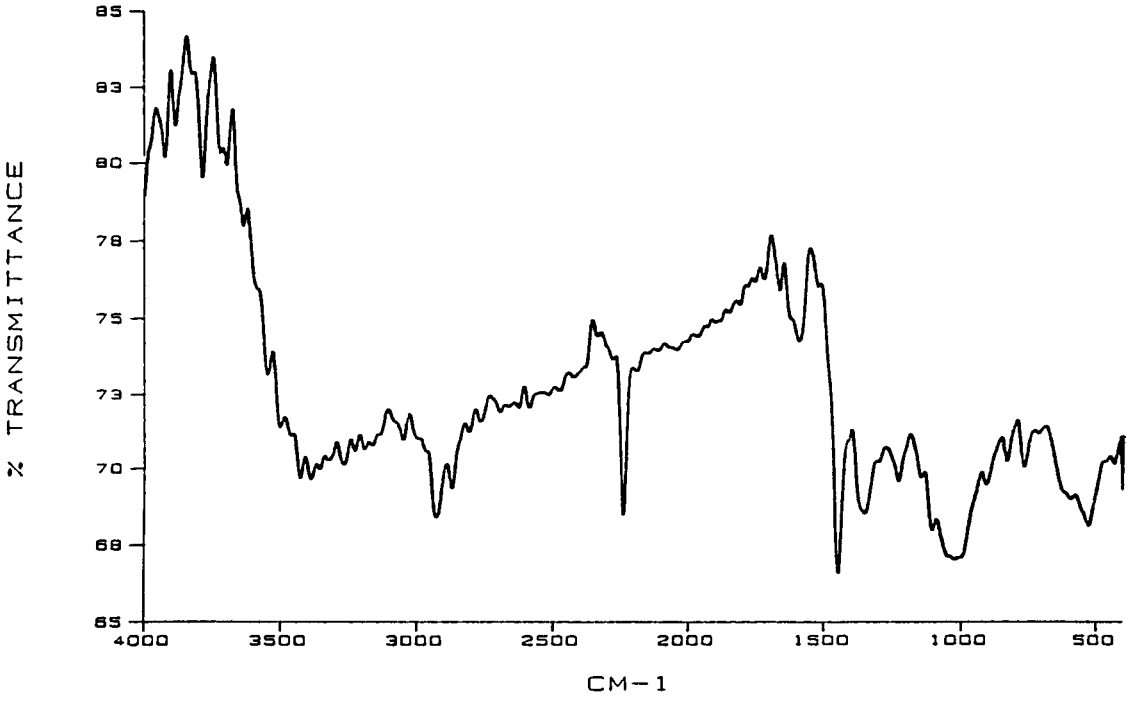

Fig. 5. IR spectrum of the starch–polyacrylonitrile copolymer after enzyme treatment.

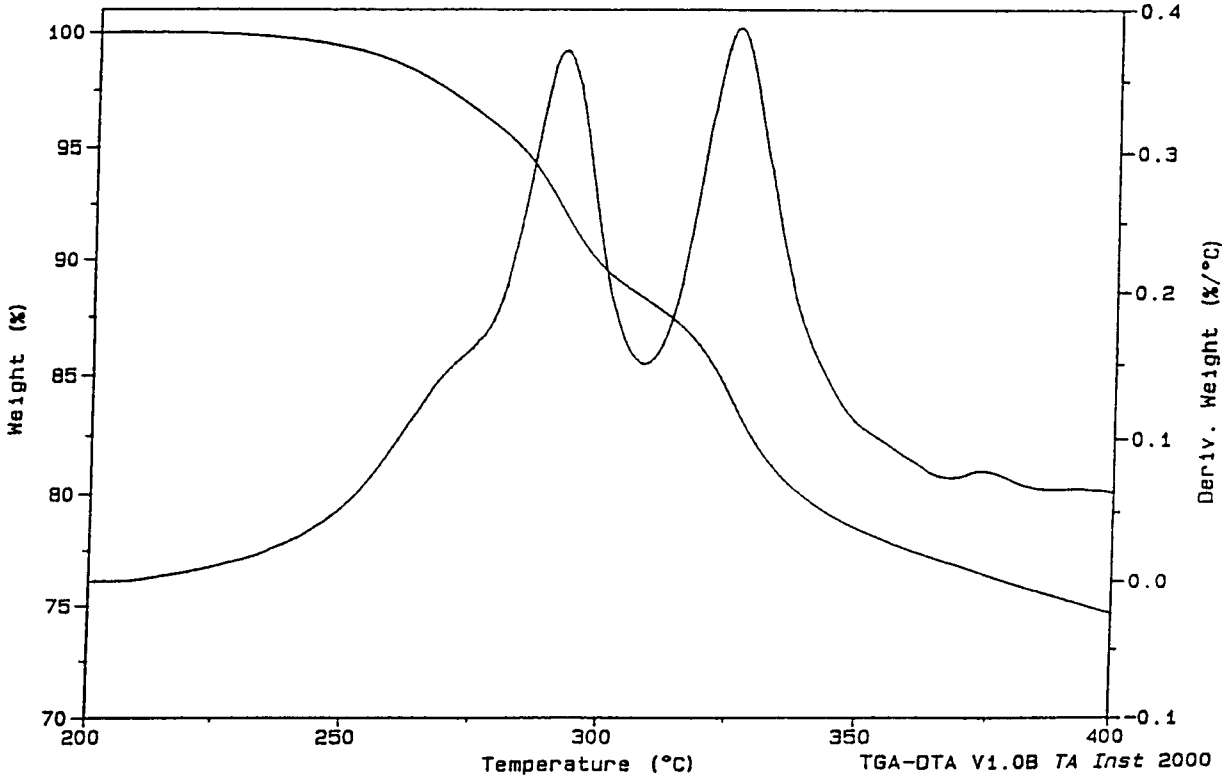

Fig. 6. TG–DTG plot of starch–polyacrylonitrile copolymer in an atmosphere of flowing dry air after aging in sterile medium.

cal TG–DTG trace obtained for the starch–polyacrylonitrile copolymer is shown in Fig. 6. A very distinct weight loss is seen in the temperature interval of 250–340°C. This is characterized as two distinct peaks on the DTG plot, with peak temperatures of 295°C and 330°C. The enzyme treated sample on the other hand (Fig. 7) showed only a single peak on the DTG plot at 330°C. Since starch shows a distinct weight loss around this region with a peak temperature of 311°C, the presence of only one peak in the enzyme treated sample was attributed to the removal of starch from the polymer matrix. Polyacrylonitrile on the other hand undergoes an exothermic reaction followed by a loss in weight (Fig. 8) at a higher temperature. There is so much heat given off that the system becomes hotter than the programmed heating rate. The reaction occurs from 295–400°C. The products include volatile gases such as HCN and less volatile materials such as monomers and dimers. The exothermic reaction is caused by the uncontrolled polymerization of nitrile groups, i.e. the polymer chain is destroyed. At 322°C the homo polymer melts [18]. The weight loss from 300°C to 340°C is 14.6%, the copolymer on the other hand shows a weight loss of 21.5%. The enzyme treated copolymer on the other hand shows a weight loss of 16.2% between this temperature range. This indicates a composition of 20% starch in the copolymer. Values obtained from elemental analysis for percent of nitrogen present (Table 1) indicate that there is 20% nitrogen present in the copolymer. Comparison with theoretical values and values obtained for polyacrylonitrile indicated a

Table 1
Amount of nitrogen present in individual components and the grafted copolymer, before and after treatment with glucoamylase

Material	N from elemental analysis (%)
Polyacrylonitrile	26.40 (theory 26.39)
Cationic starch	0.03
Copolymer synthesized	21.11
Copolymer treated with enzyme	24.42

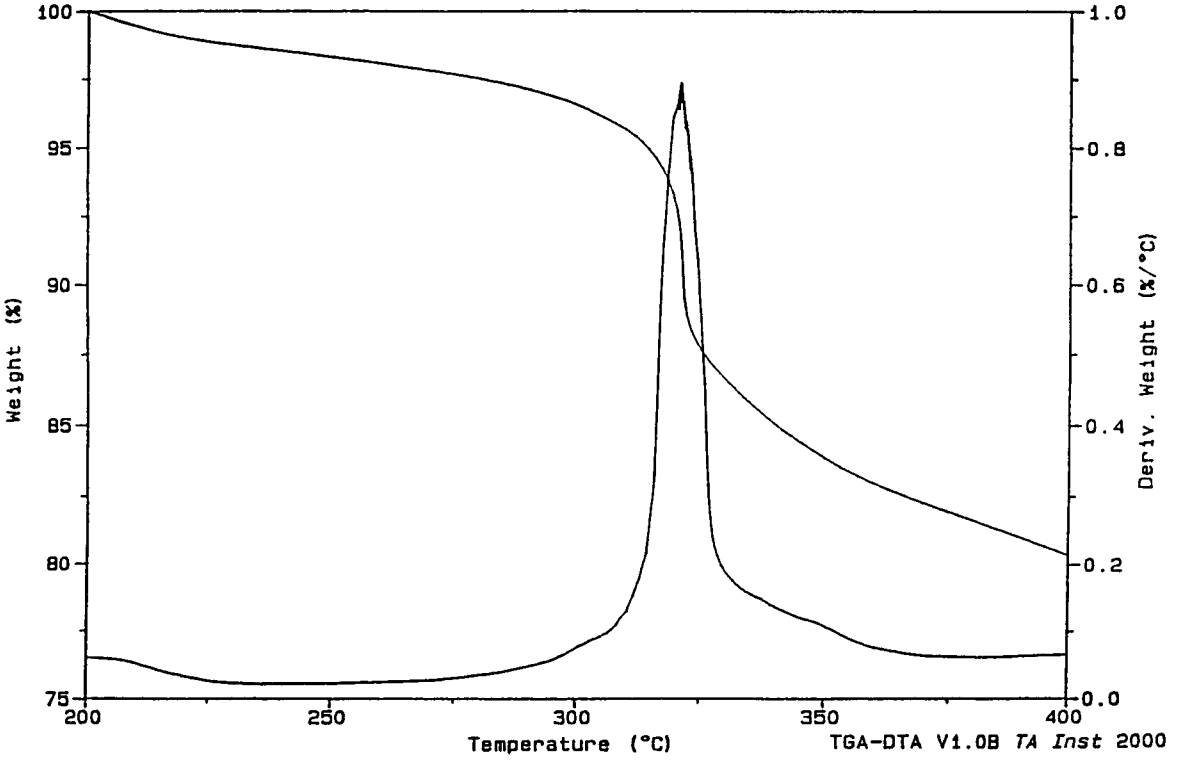

Fig. 7. TG–DTG plot of enzyme treated starch–polyacrylonitrile copolymer in an atmosphere of flowering dry air.

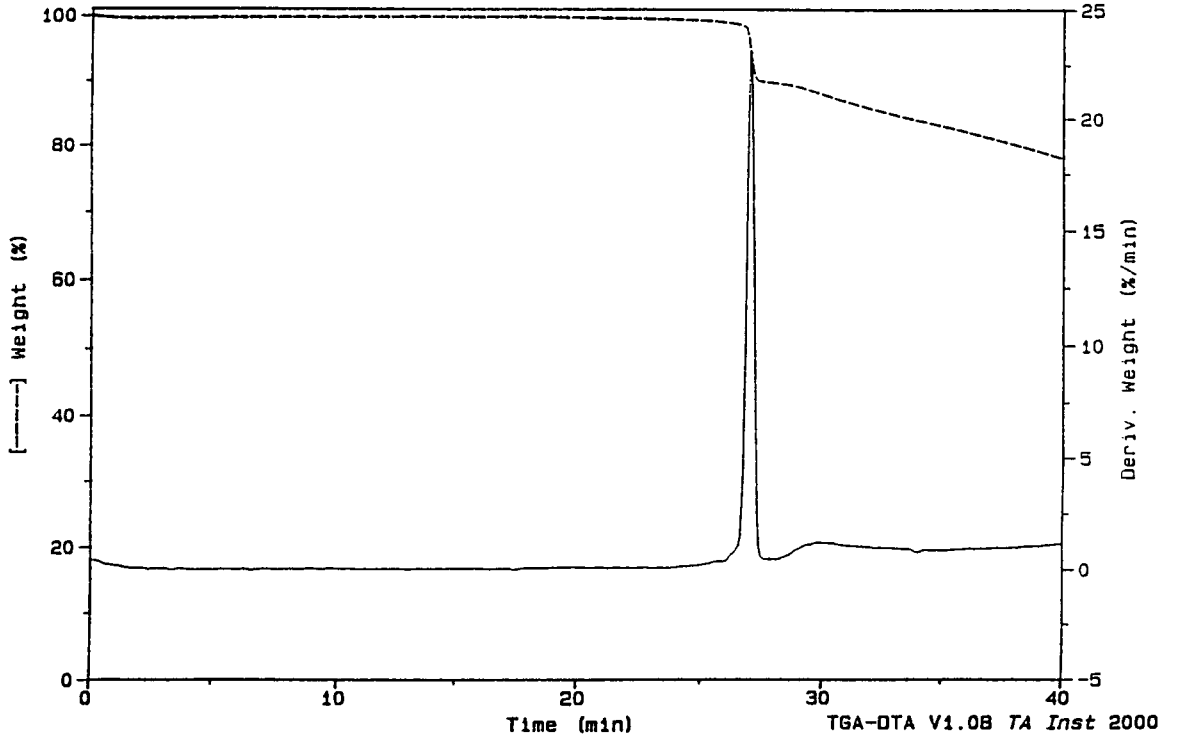

Fig. 8. TG–DTA plot of polyacrylonitrile.

loss of approximately 92% starch from the enzyme treated samples. This indicated that starch removal from the enzyme treated copolymer could be readily observed by TG. The values obtained from elemental analysis have a close resemblance to the values obtained from the TG plots. This indicates that TG can be used to determine the existence or removal of starch from grafted copolymers where the levels of starch need to be determined.

4. Conclusions

The results show that enzyme based assays can be useful for the determination of degradability of starch based polymers. Enzyme based assay systems can be standardized easily, are quantitative and rapid. These assays can be useful for the determination of relative degradability or susceptibility to biological mechanisms of deterioration for a variety of biodegradable polymer products. Although these methods do not truly mimic conditions commonly found in natural environments or landfills, extra cellular enzymes are the agents of starch hydrolysis in each of these environments. Besides, it is extremely difficult for any one single standardized assay system to accurately represent all the natural environments. The use of TG to measure accurately the starch content in polymer films or materials has been shown as an effective method for rapid quantitative determination. Starch content is essential to the issue of biodegradability and accurate methods of analysis are the key to product performance.

References

[1] A.C. Albertsson, S. Karlsson, in: J.E. Glass, G. Swift (Eds.), Agricultural and Synthetic Polymers: ACS Symposium Series, vol. 433, American Chemical Society, Washington, DC, 1990, p. 64.
[2] F.H. Otey, R.P. Westoff, W.M. Doane, Ind. Eng. Chem. Prod. Res. Dev. 19 (1980) 592.
[3] F.H. Otey, R.P. Westoof, W.M. Doane, Ind. Eng. Chem. Prod. Res. Dev. 26 (1987) 1659.
[4] F.H. Otey, W.M. Doane, in: Proceedings Symposium on Degradable Plastics, Soc. Plastics Industry, 1987, p. 39.
[5] F.H. Otey, R.P. Westoff, C.P. Russell, Ind. Eng. Chem. Prod. Res. Dev. 16 (1977) 305.
[6] W.J. Maddever, P.D. Campbell, in: S.A. Barenberg, J.A. Brash, R. Narayan, A.E. Redpath (Eds.), Degradable Materials: Perspective, Issues and Opportunities, CRC Press, Boca Raton, FL, 1990, p. 237.
[7] J.E. Potts, Encyclopedia of Polymer Science and Engineering, vol. 2, Willey, New York, 1985, p. 632.
[8] P.P. Klemchuk, Modern Plastics 66 (1989) 48.
[9] L.A. Gugliemelli, C.L. Swanson, F.L. Baker, W.M. Doane, C.R. Russel, J. Polym. Sci. 12 (1974) 2683.
[10] L.A. Gugliemelli, M.O. Weaver, C.R. Russell, C.E. Rist, J. Appl. Polym. Sci. 13 (1969) 2007.
[11] L.A. Gugliemelli, M.O. Weaver, C.R. Russell, C.E. Rist, J. Macromol. Sci. Chem. 4 (1970) 331.
[12] L.A. Gugliemelli, M.O. Weaver, C.R. Russell, C.E. Rist, J. Macromol. Sci. Chem. 1 (1967) 1381.
[13] G. Mino, S. Kaizerman, J. Polym. Sci. 31 (1958) 242.
[14] G.F. Fanta, E.B. Bagley, Encycl. Polym. Sci. Technol 2 (suppl.) (1977) 242.
[15] C.G.L. Griffin, A.C. Albertsson, S. Karlsson, in: J.E. Glass, G Swift (Eds.), Biodegradable Fillers in Thermoplastics, ACS Advances in Chemistry Series, vol. 134, American Chemical Society, Washington, DC, 1975, p. 159.
[16] S. Nakatsuka, A.L. Andrady, J. Polym. Sci. 45 (1992) 1881.
[17] H. Spedding, Advan. Carbohydr. Chem. 19 (1964) 23.
[18] D. Dollimore, G.R. Heal, Carbon 5 (1967) 65.