

# Influence of electron irradiation on the crystallisation, molecular weight and mechanical properties of poly-(*R*)-3-hydroxybutyrate

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**Abstract** Biosynthetic poly[(*R*)-3-hydroxybutyrate] (PHB) undergoes a deleterious ageing process which has been attributed to progressive crystallisation. In this work an attempt has been made to arrest this crystallisation behaviour by an irradiation treatment which is known to induce cross-linking in many polymers. PHB films were irradiated with different doses of electrons and at different times after the initiation of crystallisation. The degree of crystallinity was subsequently determined as a function of time after irradiation using wide-angle X-ray scattering measurements. In addition, melting endotherms were obtained by differential scanning calorimetry (DSC) and molecular weights by gel permeation chromatography (GPC). Corresponding tensile stress–strain experiments were performed in order to obtain information on the mechanical properties of the sample films. The melting point and molecular weights were found to decrease linearly with radiation dose indicating that higher radiation doses cause degradation due to chain scission. However, lower doses applied to samples still in their amorphous state indicate a high degree of cross-linking, in which a network is formed

and the ageing process can be prevented to some extent. The tensile modulus and breaking strain were influenced strongly by the point in time at which the material was irradiated during crystallisation.

## Introduction

Biosynthetic poly[(*R*)-3-hydroxybutyrate] is a crystallisable, biodegradable polyester obtained via bacterial fermentation [1]. It was first commercially produced in the 1980s [2] and may in future prove to be an environmentally friendly alternative to many oil-based thermoplastics. Furthermore, its biocompatibility makes it potentially suitable for medical applications [3]. However, one disadvantage that lessens its attractiveness for industrial mass production is an ageing process that occurs in samples stored at room temperature, which leads to an increasing embrittlement of the material [4–7]. This has been attributed at least partially to a progressive crystallisation process [6, 8].

In this work an attempt has been made to prevent progressive crystallisation by subjecting PHB to an irradiation treatment with electrons, which may induce cross-linking in the amorphous regions of the polymer. Investigations on irradiated PHB samples have already been made by several groups, applying  $\gamma$ -rays [9–11] or swift heavy ions [12] that resulted in cross-linking effects as well as thermal degradation. In this work we were interested in the effects of electron irradiation of PHB films using different doses and applying the treatments at different stages during crystallisation.

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In order to obtain information on the mechanical properties of films irradiated at different times we carried out tensile testing of sample strips. In addition, the degree of crystallinity of all samples was determined from wide-angle X-ray scattering patterns. Differential scanning calorimetry (DSC) was used to obtain melting endotherms of the samples, and molecular weight distributions were measured by gel permeation chromatography (GPC).

## Experimental

### Material

PHB in powder form was provided by Biomer, Krailling, Germany. The material was a high purity grade polymer containing no additives or nucleating agents. Films of about 1 mm thickness were compression moulded at 190 °C for 3 min and then quenched from the melt to a temperature below the glass transition in order to prevent the material from crystallising, i.e., to keep it amorphous until crystallisation was desired. For this purpose the samples were kept in a bath containing an ice–salt mixture at approx. –7 °C.

### Irradiation

Irradiation with electrons was carried out at BGS (BETA-GAMMA-SERVICE) in Saal, Germany. The samples were exposed in air to different doses (33, 66 and 99 kGy) in a Rhodotron electron accelerator produced by IBA, Louvain-la-neuve, Belgium. The following procedure was adopted. Frozen samples were taken out of the ice–salt mixture and irradiated after being allowed to crystallise at room temperature for 5, 15, 45, 60 min and 1 day, respectively. This means that the primary crystallisation process, which takes approximately one and a half hours to complete (spherulite growth), was only complete in the last of the five samples.

### Sample analysis

Wide-angle X-ray diffraction patterns were obtained with a Siemens D5000 diffractometer using monochromatic Cu K $\alpha$  radiation and controlled by a Siemens DIFFRAC AT 3.2 measuring program. The samples were scanned in a 2 $\theta$ -mode between 6° and 55°. The degree of crystallinity  $\chi$  of each sample was evaluated using the area under the scattering curve, where

$\chi = (\text{total area} - \text{area under amorphous curve}) / (\text{total area})$ . The amorphous curve was obtained by scanning a fully amorphous sample that had been quenched from the melt below the glass transition temperature [13].

Mechanical testing was carried out with a Polymer Laboratories Miniature Materials Tester (Minimat). For the measurements sample strips with the dimensions 5 × 1 × 50 mm were stamped out of the films after the radiation treatment. Stress–strain curves were recorded with a crosshead speed of 3.5 mm/min and Young's Modulus determined from the initial slope, applying Hooke's Law.

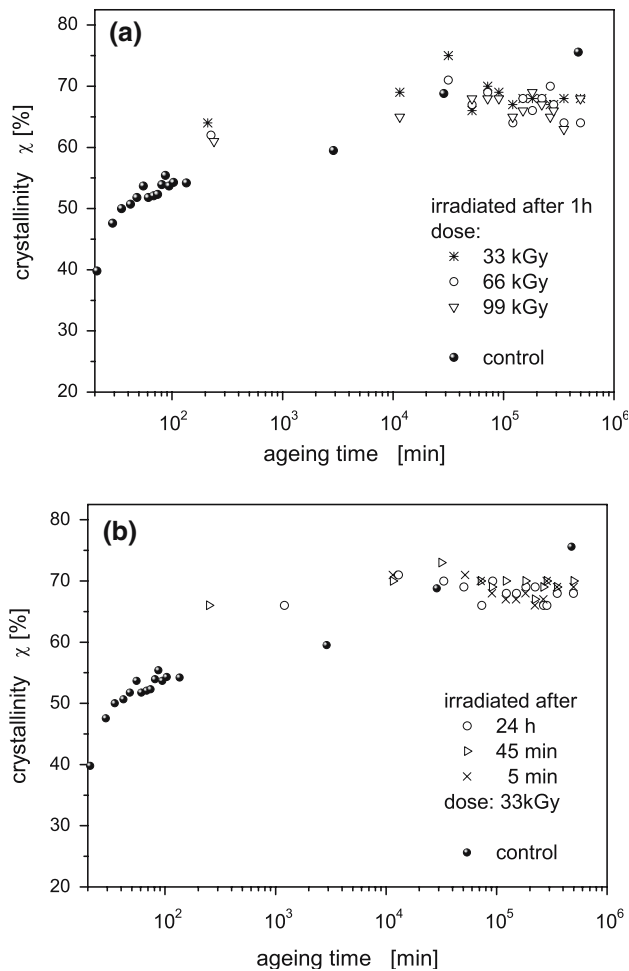
For differential scanning calorimetry a Du Pont DSC 912 was available. Indium was used for calibration. The sample was heated at a rate of 10 °C/min from room temperature to 220 °C, which is well above the melting point of PHB (approx. 178 °C).

Gel permeation chromatography (GPC) was carried out with a 10A VP HPLC system (Shimadzu) using a Phenogel 5  $\mu$ m linear column (Phenomenex) and chloroform (1 ml/min) as the eluent. Obtained molecular weights were calculated based on the used polystyrene standards.

## Results and discussion

### Wide-angle X-ray measurements (WAXD)

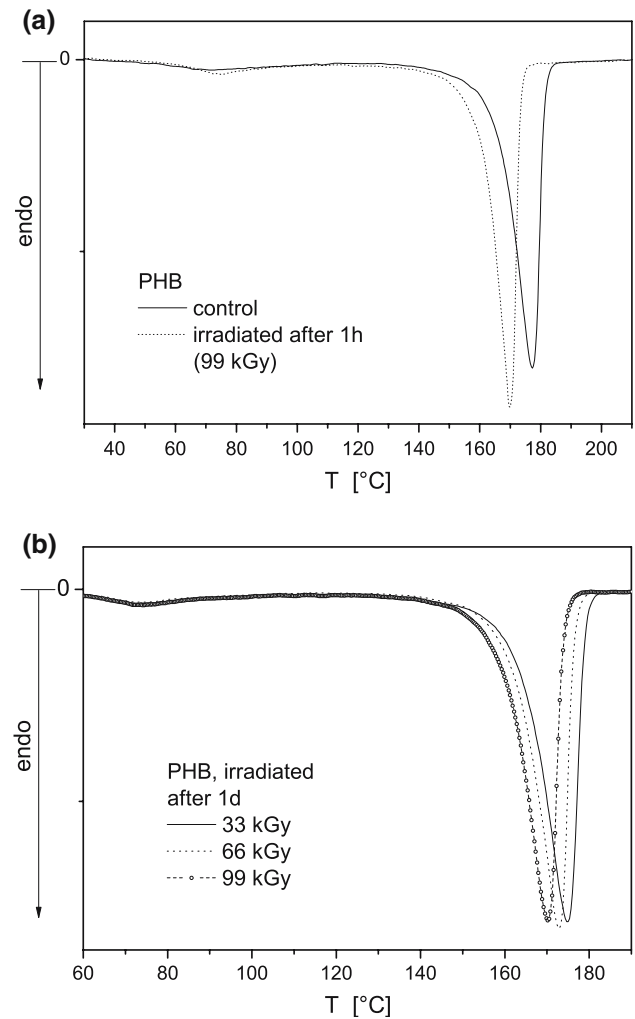
A characteristic of pure PHB crystallised and stored at room temperature is the progressive development of crystallinity with ageing or storage time. Firstly, in Fig. 1 this behaviour is shown for a control sample that remained non-irradiated. During primary crystallisation (up to 10<sup>2</sup> min)  $\chi$  reaches approximately 55%, but then it continues to increase indefinitely with time during the progressive crystallisation stage until it reaches almost 76% after 1 year of storage. Fig. 1a also shows  $\chi$  for samples irradiated with 33, 66 and 99 kGy 1 h after the initiation of crystallisation (after removal from ice-water bath). It can be seen that there is no significant difference between the results for the three doses. At short storage times  $\chi$  exceeds that of the control sample (approx. 4 h after irradiation it is just greater than 60%). However, no remarkable increase with ageing time is subsequently evident. After 1 year the average degree of crystallinity is still below 70%. As can be seen in Fig. 1b, similar results were obtained for the series of samples that were irradiated with 33 kGy for crystallisation times of 1 day, 45 min and 5 min. The results reach a plateau below the value for the non-irradiated sample, indicating that the radiation seems to have caused progressive crystallisation to



**Fig. 1** Degree of crystallinity for samples (a) irradiated with three different doses and for an untreated control sample and (b) irradiated with 33 kGy after different crystallisation times

stop. No significant difference is observable between the irradiated samples, no matter what dose was used or whether they had been irradiated during the primary or the secondary crystallisation process.

Initially, however, the degree of crystallinity in irradiated samples is higher than that for an equivalently aged control sample. This could be a result of a heating effect, which was observed as a short-term temperature rise of approx. 20–30 °C during irradiation. The crystallisation rate of PHB is actually increased by temperature in this temperature range, so that the sample merely has a higher crystallinity because it effectively crystallised at an elevated temperature. However, it should be noted that the radiation itself could also be a cause for this effect, since shorter polymer chains make crystallisation easier. This would be the case if chain scission occurred (see below).



**Fig. 2** DSC curves of PHB (a) comparison between irradiated and non-irradiated material (b) irradiated after 1 d with three different doses

### Thermal analysis

DSC measurements were carried out on aged samples (i.e., more than 1 year after the irradiation treatment), since we were interested in ageing effects (see also mechanical measurements).

The thermal behaviour of irradiated and non-irradiated PHB material is illustrated in Fig. 2. Fig. 2a shows the DSC curve of an irradiated (after 1 h) and a non-irradiated sample for comparison. First of all, a remarkable decrease of the melting point from 178 °C in the untreated sample to 170 °C by irradiation with a dose of 99 kGy is observable. Furthermore, a slight shift of the endotherm in the lower melting region (melting of small, unstable crystallites) to higher temperatures in the irradiated material can be noticed. This can be attributed to the above-mentioned tem-

perature rise of about 20–30 °C during the irradiation procedure, creating an additional annealing process.

In Fig. 2b, the DSC curves for samples that were irradiated 1 day after the start of crystallisation with three different doses are shown. The lower melting endotherm remains unchanged for each dose, whereas the melting point decreases with higher doses. However, the melting temperatures for the 99 kGy samples in Fig. 2a and b do not exhibit different values, although one was irradiated after and one during primary crystallisation. This indicates that the melting behaviour is only affected by the dose, and not by the time when the material was exposed to the radiation. Table 1 gives a summary of all samples and their melting temperatures, taken from the DSC curves.

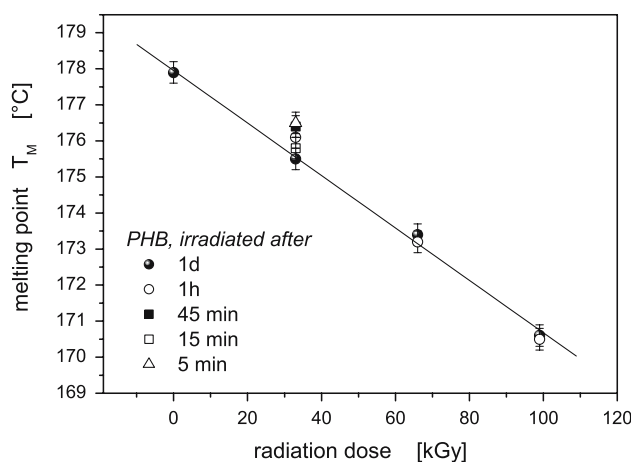
Regarding the table as a matrix it can be easily seen that the values of the melting points in the vertical direction representing an increasing radiation dose decrease continuously for all samples. Horizontally, each row depicting a certain dose, the values are similar, i.e., no significant difference between samples irradiated after different crystallisation times is noticeable. The results of Table 1 are plotted graphically in Fig. 3, which shows the relationship between melting point and radiation dose for all the samples.

A linear relationship between melting temperature and dose is evident. This effect has already been observed by Mitomo et al. using  $\gamma$ -radiation [9]. The observed decrease in melting point indicates a thermal degradation process of the polymer due to chain scission involving a decrease of molecular weight.

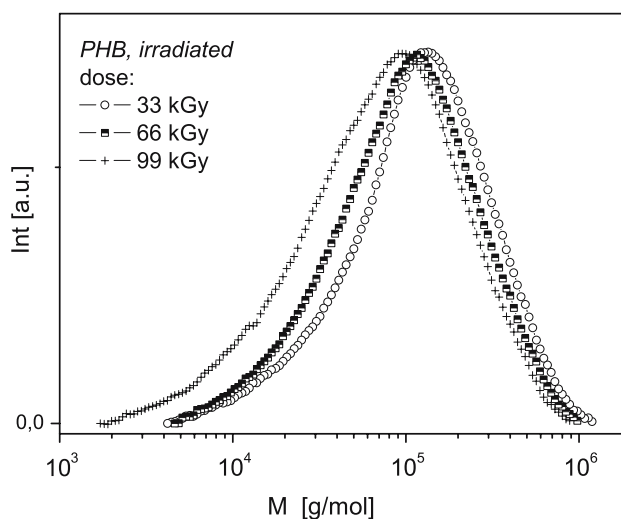
### Molecular weight analysis

GPC measurements were carried out in order to obtain information on the molecular weight distributions of the irradiated samples. The molar mass of the as-received powder was  $M_w = 327$  kg/mol (data provided by supplier).

Figure 4 shows the distributions for samples irradiated 1 h after the onset of crystallisation with 33, 66 and 99 kGy, respectively. Note the logarithmic scale. The detector signal, which displays changes in the



**Fig. 3** Melting point as a function of radiation dose



**Fig. 4** Molecular weight distribution of samples irradiated with different doses directly after primary crystallisation

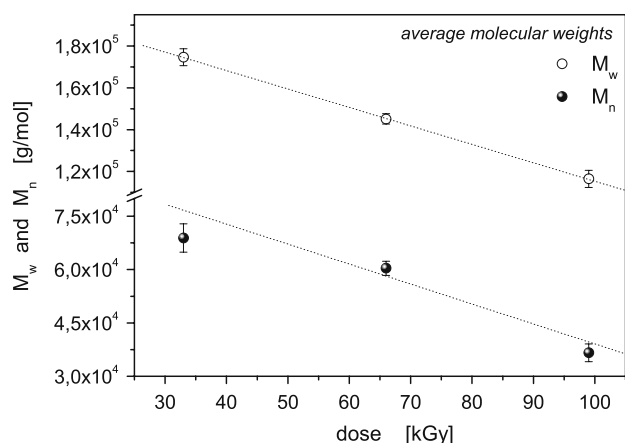
refractive index of the solvent, was normalised and given in arbitrary units. No conclusions should be drawn from the absolute signal voltage, since it was influenced by various factors such as the fluctuating detector temperature or the sample quantity. From Fig. 4 it is clear that the molecular weight tends to become lower and its distribution broader with increasing radiation dose.

In Fig. 5, the number and weight average molecular weights of these three samples are plotted against the dose to illustrate this. Just like the melting points, the molecular weights exhibit a linear correlation with the radiation dose.

The above results indicate the occurrence of chain scission in the irradiated polymer samples, especially in those samples irradiated after an hour or more of

**Table 1** melting point of samples irradiated with different doses

	Melting point $T_M$ [°C]				
	1 d	60 min	45 min	15 min	5 min
0 kGy	177.9				
33 kGy	175.5	176.1	176.4	175.8	176.5
66 kGy	173.4	173.2			
99 kGy	170.6	170.5			



**Fig. 5** molecular weight as a function of radiation dose

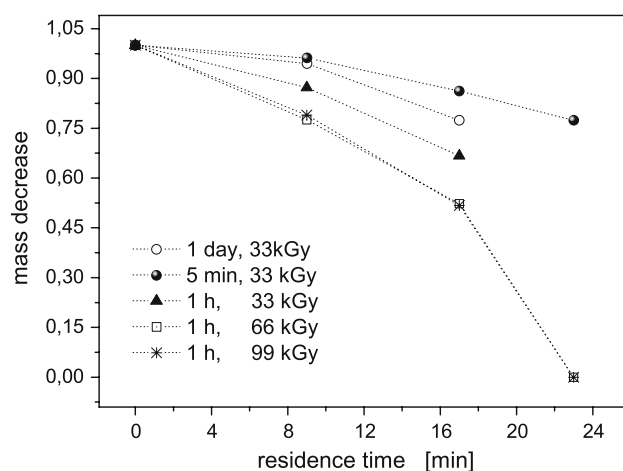
crystallisation. The average molecular weights for all measured samples are listed in Table 2.

Particularly eye-catching, however, are the relatively high values for the sample that was irradiated while still in a highly amorphous state (after 5 min of primary crystallisation). These high values suggest that cross-linking rather than degradation (chain scission) occurs in this sample. In order to assure ourselves of this, we carried out a simple swelling experiment. About 100 mg of each sample were added to chloroform and the mass decrease measured as a function of time. The results are shown in Fig. 6.

The samples exposed to higher doses (66 and 99 kGy) were quickly dissolved. After 23 min they had completely disintegrated. This was expected since all these samples showed signs of degradation (from GPC), i.e., chain scission produces shorter chains that are less entangled and can more easily be dissolved. (Two values at 23 min are missing because the samples could not be measured, since they were partially dissolved into lumps of material and soft.) The sample suspected of forming a network structure (5 min, 33 kGy) could not be dissolved. After 23 min, 80% of the sample was still solid, and a few days later we could observe that the sample had formed a swollen gel.

#### Mechanical measurements

Mechanical measurements for samples irradiated during primary crystallisation showed relatively large



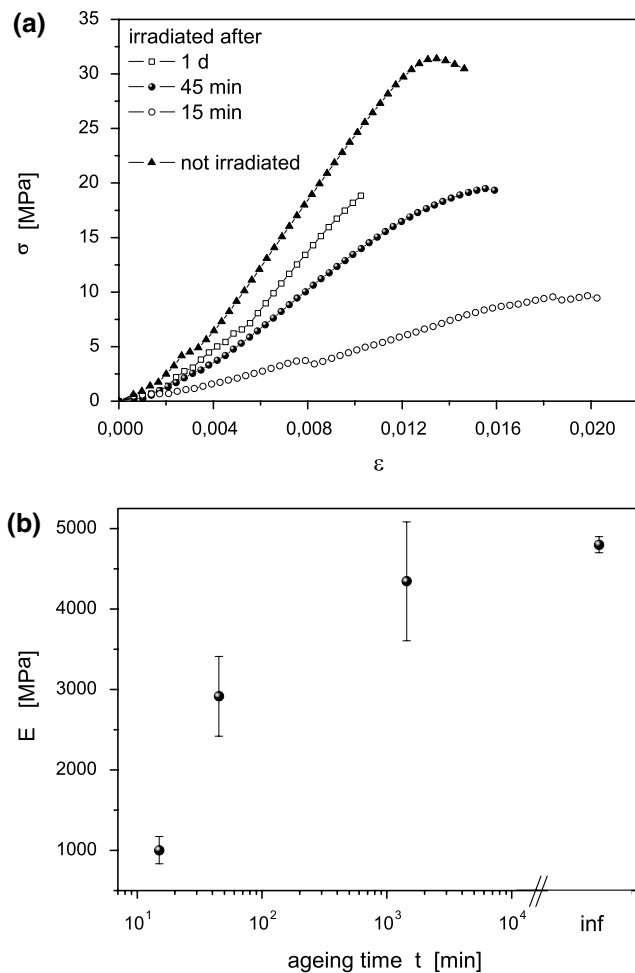
**Fig. 6** mass decrease of all samples as a function of residence time in chloroform

errors, since the sample films were still soft during the irradiation treatment and had become partially distorted during the procedure. As a consequence it was not always possible to obtain completely planar sample strips suitable for mechanical measurements. Nevertheless, satisfactory measurements could be made for several samples. At the time the stress–strain experiments were carried out, the samples had all been stored for more than 1 year after irradiation, in order to find out whether the ageing process could be halted by radiation. Figure 7a shows stress–strain diagrams for samples irradiated 1 day, 45 min and 15 min after initiation of crystallisation, and for an aged non-irradiated control sample.

The corresponding values of tensile modulus can be seen in Fig. 7b. In this diagram the non-irradiated sample is defined as “inf” (infinity) on the time scale. Obviously, there is a correlation between the time during crystallisation when irradiation was carried out and the tensile behaviour. The sample irradiated 1 day after it was allowed to crystallise, which is long after completion of primary crystallisation, exhibits a modulus comparable to the untreated sample (taking experimental errors into account). Samples that were exposed to the radiation during the primary crystallisation process (45 and 15 min) show a much lower tensile modulus and maximum stress, and a larger maximum strain. This means that irradiation during

**Table 2** Molecular weight of various samples

	Age: 1 d (33 kGy)	Age: 5 min (33 kGy)	Age: 1 h (33 kGy)	Age: 1 h (66 kGy)	Age: 1 h (99 kGy)
$M_w$ [g/mol]	178384	212929	174660	145139	116431
$M_n$ [g/mol]	78931	98024	68885	60370	36609



**Fig. 7** (a) Stress–strain-diagrams and (b) Young’s modulus for samples irradiated at different crystallisation times with 33 kGy, and a control sample

primary crystallisation can arrest the tendency towards brittleness. It is also evident that this behaviour is more pronounced the sooner irradiation takes place.

This result seems exceptional, since our previous investigation has shown that a linear relationship exists between Young’s modulus and crystallinity in PHB [13]—and we have already shown that all irradiated samples have approximately the same degree of crystallinity (see Fig. 1). The moduli are different here, however, due to the different degrees of cross-linking, while the crystallinity itself appears to play a subordinate role. We therefore suggest that irradiation causes cross-linking in the melt, which may significantly alter the resulting polymer morphology after crystallisation. The final structure may thus contain, in addition to spherulites of crystalline–amorphous layers, regions of networked chains with elastic properties. The decreases in melting point indeed suggest that the resulting crystallites are smaller or more defect-rich than in non-

irradiated material. The modulus may, therefore, no longer be determined solely by the spherulitic architecture but also by an elastic network of cross-linked chains. A further striking result is that samples irradiated in the amorphous state seem not to be prone to ageing. However, just as intriguing is the possibility demonstrated here of obtaining a highly crystalline material (in the region of 70%) with elastic properties.

## Conclusions

Irradiation treatment during primary and secondary crystallisation causes the degree of crystallinity to stabilise to an average value of approximately 70% in all irradiated samples, whereas without irradiation the progressive crystallinity continues indefinitely. The resulting tensile properties depend on the time during crystallisation at which the material was irradiated; a decrease in Young’s modulus and an increase in maximum strain were observed, which are more pronounced the larger the amorphous fraction of the material was at the time of irradiation. Cross-linking was found to occur preferentially in samples irradiated near the onset of primary crystallisation, i.e., when they were still virtually in the molten phase. We suggest that in those samples an elastic network is formed which dominates the final mechanical behaviour.

DSC measurements show a linear decrease in melting point with increasing radiation dose, and GPC measurements a decrease in molecular weight. This indicates a reduction in crystal size and/or increased crystal defects as well as the occurrence of chain scission at higher doses. The GPC results relate only to the fraction of material that was not cross-linked by the irradiation. Solubility experiments indicate that a large gel fraction was present in samples irradiated (virtually) in the amorphous state. Therefore, electron irradiation seems to be causing both chain scission and cross-linking, where the cross-linking occurs preferentially in molten or amorphous regions of the material.

The aim of this work was to ascertain whether the detrimental ageing process in PHB can be prevented by irradiation treatment. This seems to be a feasible proposition provided the treatment is applied to the polymer while its chains are still in a molten state or when the amorphous fraction is as high as possible so that cross-linking rather than chain scission occurs. We have demonstrated here that ‘melt irradiation’ is potentially a useful technique for stabilising the properties of PHB and preventing its progressive embrittlement.

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