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Hydrolytic and enzymatic degradation of a Poly(ε-caprolactone) network

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Abstract

Long-term hydrolytic and enzymatic degradation profiles of poly(ε-caprolactone) (PCL) networks were obtained. The hydrolytic degradation studies were performed in water and phosphate buffer solution (PBS) for 65 weeks. In this case, the degradation rate of PCL networks was faster than previous results in the literature on linear PCL, reaching a weight loss of around 20% in 60 weeks after immersing the samples either in water or PBS conditions. The enzymatic degradation rate in Pseudomonas Lipase for 14 weeks was also studied, with the conclusion being that the degradation profile of PCL networks is lower than for linear PCL, also reaching a 20% weight loss. The weight lost, degree of swelling, and calorimetric and mechanical properties as a function of degradation time were obtained. Furthermore, the morphological changes in the samples were studied carefully through electron microscopy and crystal size through X-ray diffraction. The changes in some properties over the degradation period such as crystallinity, crystal size and Young's modulus were smaller in the case of enzymatic studies, highlighting differences in the degradation mechanism in the two studies, hydrolytic and enzymatic.

Keywords— PCL, hydrolytic, enzymatic, degradation, hydrolysis, network.

1. INTRODUCTION

The use of non-permanent scaffold materials that, over time, become completely replaced by natural extracellular matrix is central to the tissue engineering approach. The most important role of the use of scaffold *in vivo* is that it persists in a robust state for sufficient time to allow the formation of new tissue, but also ultimately degrading and becoming replaced by this tissue. For its successful implementation in applications such as surgical sutures, drug delivery systems, and tissue engineering scaffolds, hydrolytic degradation is of crucial importance [1,2].

The terminology used to define synthetic polymer breakdown is not consistent in the literature. In 1992, Vert *et al* [3] proposed the definitions of *biodegradable*, *bioresorbable*, *bioabsorbable* and *bioerodable*. According to these definitions, Poly(ε-caprolactone), PCL, is a semi-crystalline, bioresorbable polymer belonging to the aliphatic polyester family.

The hydrolytic degradation is an autocatalyzed process, where the carboxylic groups of the hydroxy acids produced catalyze further hydrolysis [4]. It is widely accepted that hydrolytic degradation of poly(α-hydroxy) esters can proceed via surface or bulk degradation pathways [2]. The diffusion–reaction phenomenon determines the means by which this pathway proceeds. The advantage of surface degradation is the predictability of the process, but surface and bulk degradation are ideal cases to which most polymers cannot be unequivocally assigned [2]. In tissue engineering, surface properties or porosity determine the performance of implantable scaffolds [5].

The enzymatic degradation of PCL polymers has also been studied, especially in the presence of lipase-type enzymes. [6-9] Three kinds of lipase were found to significantly accelerate the degradation

of PCL: Rhizopus delemer lipase [6], Rhizopus arrhizus lipase, and Pseudomonas lipase [7,8]. Highly crystalline PCL was reported to totally degrade in 4 days in the presence of Pseudomonas lipase [7,8], in contrast with hydrolytic degradation, which lasts several years.

Accelerated degradation could be achieved using an acidic or basic medium, which would enhance the hydrolysis of polyesters; this would also mimic physiological conditions better than other methods, such as temperature acceleration. The aim of accelerated degradation systems is to accomplish the degradation of polymeric devices within a shorter period of time, thus enabling the study of morphological and chemical changes during degradation in a more acceptable timeframe. [4,10,11]. In this way, researchers may anticipate analogous results by altering the rate of reaction whilst maintaining identical principal mechanisms of reaction. However, such data should always be verified against long-term data.

The rate of hydrolytic degradation of ester linkages is affected by a multitude of factors. In general, actions which increase the penetration of water accelerate the rate of hydrolysis (such as using, or blending with, a more hydrophilic polymer). Water absorption is thus a critical factor [12]. Two other important factors are the polymer's glass transition temperature (T_g) and crystallinity, both of which reflect the ability of water to access the polymer chains. A high T_g corresponds to relatively limited molecular motion and low free volume within the polymer network, meaning that less space is available for water molecules to penetrate. Similarly, a high degree of crystallinity limits hydration through the ordered packing of polymer chains. Any action reducing T_g and crystallinity will accelerate hydrolytic degradation.

PCL is one of the most widely studied synthetic polymers and has FDA approval in various devices for medical applications [12]. However, applications of PCL might be limited by the degradation and

resorption kinetics of PCL, which are considerably slower than other aliphatic polyesters due to its hydrophobic character and high crystallinity [13]. One way of avoiding crystallinity, and consequently increasing the degradation rate, is to prepare polymer networks. For example, PCL macromers have been synthesized through the reaction of PCL diol with acryloyl chloride and PCL networks were photopolymerized [14]. Thermal, mechanical, and morphological characteristics as well as the degradability of the PCL networks were studied. Bat *et al* [15] developed a method to obtain form-stable and elastic networks upon gamma irradiation based on high molecular weight (co)polymers of trimethylene carbonate and (ε-caprolactone). The in vitro enzymatic erosion behavior of these hydrophobic networks was studied using aqueous lipase solutions.

A few years ago, our group reported a new strategy for crosslinking and obtaining a hydrophilic polycaprolactone[16]. A PCL macromer was synthesized through the reaction of PCL diol with methacrylic anhydride in order to obtain methacrylate end capped PCL. PCL networks were prepared by photopolymerization of the new macromer. Furthermore, the PCL macromer was copolymerized with 2-hydroxyethyl acrylate to improve the water sorption capacity of the system. The synthesis, characterization, and physical properties were described elsewhere[16,17]. Accelerated degradation studies in an alkaline medium have also been reported [18].

In this paper we present an enzymatic and long-term hydrolytic degradation study of these PCL networks, using phosphate buffered solution (PBS) and HPLC-grade water at 37ºC. From these parallel studies we were able to evaluate the effectiveness of the accelerated system [18] when compared with in vitro physiological conditions. The degradation process was monitored by determining the weight loss, X-ray diffraction analysis, thermal and mechanical properties, and surface morphology as a function of degradation time.

2. MATERIALS AND METHODS

2.1. Materials

α,ω-dihydroxyl terminated polycaprolactone with a molecular weight of 2000 Da and methacrylic anhydride (MA), were supplied by Aldrich. Benzoin (Scharlau, 98% pure) was employed as initiator. Dioxane (Aldrich, 99.8% pure), acetone (Aldrich, 99.5% pure), ethanol (Aldrich, 99.5% pure) and anhydrous ethyl acetate (Aldrich 99.8 %) were used as solvents without further purification. Lipase from Pseudomonas (PS) fluorescens (powder, EC 3.1.1.3, 40 units/mg) was purchased from Fluka, Sodium azide (NaN_3) 99% from Aldrich, the buffer solution (phosphate/di-Sodium hydrogen) pH =7,00 (20ºC) D=1,01 g/cm^3 and HPLC-grade water from Scharlau, and they were used as received.

The synthesis, characterization, and physical properties of the PCL network were published previously [16,17]. In brief, α,ω-dihydroxyl terminated PCL was endcapped with methacrylate groups to make a polymerizable macromer. The macromer was obtained by dropping the filtrate into an excess of ethanol, filtrated, recrystallized several times and also purified by column chromatography techniques, using silica gel 60 (70-230 mesh) as the stationary phase and ethyl acetate as the solvent. Finally, the precipitated PCL macromer was dried at 50 ºC for 24 hours under reduced pressure. PCL networks were prepared by UV polymerization. The PCL macromer was dissolved in dioxane, 35% (w/v) and mixed with benzoin (photoinitiator, 1 wt %). The reaction was carried out under ultraviolet light for 24 hours. Low molecular weight substances were extracted by boiling in ethanol for 24 hours and then vacuum dried to constant weight.

2.2. Hydrolytic and enzymatic degradation

Hydrolytic degradation at 37.0±0.5ºC and pH = 7.4 was carried out in two different media: phosphate buffered solution (PBS), and HPLC-grade water. The enzymatic degradation assay was carried out in a phosphate buffer saline solution (pH=7.4 and 37.0±0.5ºC) in the presence of PS lipase, 2.5 units/g of polymer (1mg/ml), and 0.02% of sodium azide to avoid bacterial proliferation. The buffer-enzymatic solution was changed twice a week to maintain the enzymatic activity. Specimens, discs of around 5 mm diameter made from films of 0.8 mm thick, were placed in tubes filled with degradation medium; the proportion of the sample to the degradation medium was 1/50 in mass. Three replicates were taken out of the solution at predetermined time intervals, then washed with distilled water and vacuum-dried at room temperature to constant weight.

The degradation process was followed by determining the water absorption and mass loss of the materials. Samples were washed with distilled water and gently wiped with paper. Wet weight was determined in order to evaluate the evolution of the samples' hydrophilicity. The degree of swelling was determined by comparing the wet weight (w_w) at a specific time with the dry weight (w_d) according to Eq.1

$$\text{degree of swelling}\,(\%) = \frac{w_w - w_d}{w_d} \times 100 \qquad (1)$$

The percentage of weight loss was determined after drying the samples in vacuo by comparing dry weight (w_d) at a specific time with the initial weight (w_0) according to Eq.2

$$\text{weight loss } (\%) = \frac{w_0 - w_d}{w_0} \times 100 \qquad (2)$$

A balance (Mettler Toledo) with a sensitivity of 0.01 mg was used to weigh the samples.

2.3. Scanning Electron Microscopy (SEM)

To investigate the surface and cross section morphology of dried network samples, SEM pictures of degraded and non-degraded samples were taken using a JEOL JSM-5410 scanning electron microscope.

2.4. Differential Scanning Calorimetry (DSC)

The thermal properties of the samples were measured by using a Mettler Toledo differential scanning calorimeter (DSC) calibrated with indium. The measurements were carried out at a scan rate of 10 ºC/min between –10 ºC and 100 ºC. To keep the same thermal history, each sample was first heated to 100ºC and cooled down to -10ºC at a rate of 10 ºC/min. Then a subsequent heating run was performed from -10 ºC to 100 ºC at a rate of 10 ºC/min. The data from all three scans were collected for subsequent analysis. Crystallinity was calculated assuming proportionality to the experimental heat of fusion, using the reported heat of fusion of 139.5 J/g for the 100% crystalline PCL [19]. The first scan measures the effect of the degradation process on the sample; the second heating scan analyses the melting of the crystals once the first heating scan has erased the previous history of the material [19]. As all samples have crystallized under the same conditions, the melting peak corresponding to the second heating scan depends on the material properties, not on their history.

2.5. X-Ray Diffraction (XRD)

X-ray diffraction spectra of degraded samples were obtained on an X-ray diffractometer, Rigaku Ultima IV, in the Bragg-Bentano configuration using the Kα radiation of a Cu anode. The samples were scanned from 2θ=15 to 35º at a speed of 2º/min. Crystallite size determination was carried out using the Debye–Scherrer equation [20].

$$D = 0.9\frac{\lambda}{\beta \cos\theta} \qquad (3)$$

where D is the apparent particle size, β is the full-width at half-maximum (FWHM) of the X-ray diffraction line (additional peak broadening) in radians, λ is the wavelength used, and θ is the angle between the incident ray and the scattering planes. The constant 0.9 in Eq. (3) depends to an extent on the symmetry of the crystal and has been discussed by several authors [21,22].

2.6. Mechanical testing

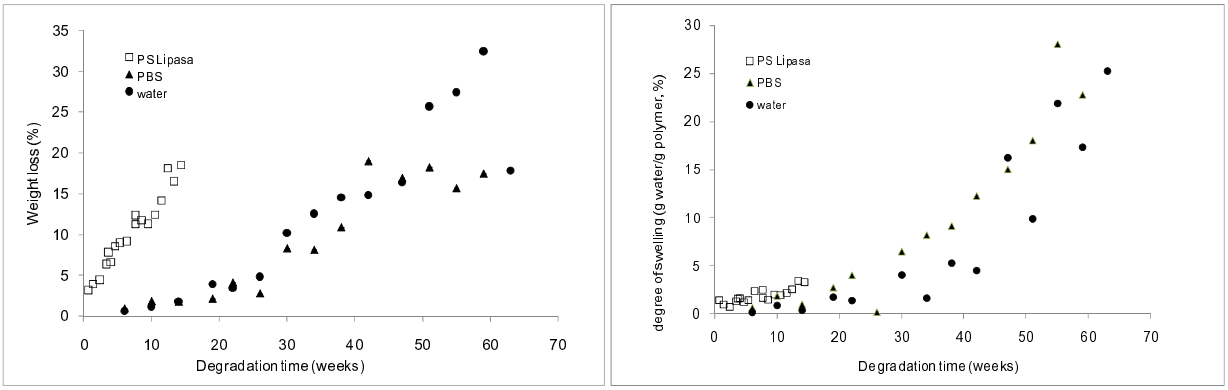
Mechanical properties were evaluated using a Seiko TMA/SS6000 by applying three consecutive compression ramps from 1 to 400 g at 50 g/min. Sample deformations were collected every 5 s. Between each compression ramp, the sample was allowed to recover for one minute. The section of the apparatus probe was 0.785 mm^2 (probe diameter 1 mm), with 5.1 MPa being the maximum value of stress applied to samples. Three samples of each degradation time, including non-degraded samples, were measured, with all samples being 0.75±0.1 mm thick at the beginning of tests.

3. RESULTS AND DISCUSSION

3.1. Weight loss and swollen degree

Figure 1a shows the mass loss as a function of degradation time. The mass losses for samples immersed in water and PBS were rather similar until 40 weeks after which the degradation rate slowed down for samples immersed in PBS and increased for samples degraded in water. Relatively little mass (<5%) was lost during the first 25 weeks for both hydrolytic degradations. After week 25, a slight increase in mass loss was observed for samples degraded in water and PBS, reaching 32% mass loss after 60 weeks for samples in water and 18% after 60 weeks for samples in PBS. For samples degraded with PS lipase, a small mass loss of <5% was observed in the first weeks. After week 3, a rather constant increase in mass loss was observed, reaching 18% after 14 weeks.

Figure 1b shows a high correlation between the degree of swelling and the mass loss for hydrolytic degradation (both in water and PBS). The water uptake increases during the degradation period reaching about 18-28% after 60 weeks. The enzymatically degraded samples absorbed a lower amount of water, less than the 4%, and showed no significant change in water uptake during the 14 weeks of degradation. These results are consistent with some bulk degradation for the hydrolytic degradation (water and PBS).



a) b)

Fig. 1. Weight loss a) and degree of swelling b) of hydrolytic and enzymatic degradation of PCL networks.

Furthermore, hydrolytic degradation, whether with water or PBS, of PCL networks is faster than that of linear PCL [4]. Previous accelerated degradation studies also have demonstrated that PCL networks degrade faster than linear PCL when immersed in an alkaline medium [18]. Theiler *et al* [23] related an increase of the degradation rate with the higher hydrophilicity and lower crystallinity of PCL networks as compared with linear PCL. Harrison *et al* [24] studied how crystallinity and water absorption affect the degradation rate. They demonstrated that water penetrates the lamellar regions of the crystallites.

However, the enzymatic degradation of PCL networks is lower than that of linear PCL [25]. Enzymatic degradation studies [26,27] show that erosion occurs at the material surface without depending on hydrophilicity. This is consistent with the fact that hydrophilicity does not change with degradation (Fig 1b). By contrast, it is widely accepted that enzymatic degradation mainly affects the amorphous phase.

3.2. Scanning Electron Microscopy (SEM)

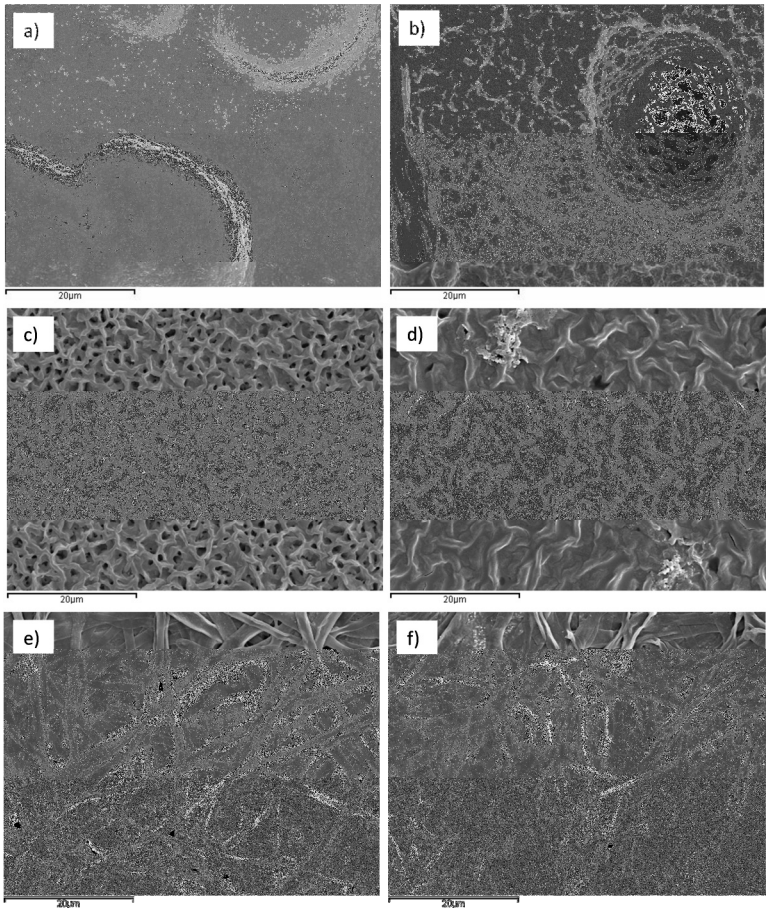


Fig. 2. Superficial SEM microphotograph of: a) a non-degraded sample; b) a sample degraded in enzymatic medium for 11.5 weeks; c) a sample after 26 weeks of degradation in water; d) a sample degraded for 26 weeks in PBS; e) a sample after 59 weeks of degradation in water; f) a sample degraded for 59 weeks in PBS. Bar=20µm

Figure 2 shows SEM microphotographs of non-degraded and degraded samples in different media at different degradation times. During the degradation period the sample surface became rougher. Superficial erosion seems to happen in all cases, although the morphology is different. The sample shown in Fig 2b was degraded for 11.5 weeks in an enzymatic medium, presenting a weight loss of 14 %. In this case, SEM examination revealed surface erosion over the whole surface. In addition, rounded holes on the surface were observed. However, a transversal cut did not present any change in

morphology (figure not shown). These results would be in agreement with the commonly accepted fact of superficial erosion in the case of enzymatic degradation.

The samples subjected to hydrolytic degradation, after 26 weeks of degradation, when the weight loss in water and PBS was 3% and 1.5% respectively, also presented a rough surface (see Fig 2c and 2d). With further degradation, after 59 weeks, a fibrous structure appeared (Fig 2e and 2f). The interior also presented a fibrous structure (figure not shown), which could indicate a certain amount of degradation in bulk as a consequence of the penetration of the degradation media inside the sample. After more than 60 weeks, the samples became very fragile, breaking easily, and occasionally fragmenting into smaller pieces.

3.3. Differential Scanning Calorimetry (DSC)

DSC was used to monitor degree of crystallinity and thermal properties of the samples before and after degradation. The melting point (T_m) and the heat of fusion (ΔH_f) were screened during the first and second heating scans. As an example, Figure 3 shows the first (left) and second (right) heating scans of samples degraded in PBS for different degradation times. The same general tendency observed in Figure 3 for PBS is present in water and Lipase degradation. Changes of area, position and half-width of the peak were measured as a function of degradation time.

Figure 4 shows the crystallinity of PCL samples as a function of degradation time in water, PBS and the enzymatic medium, from the first heating scan. An appreciable increase can be seen in crystallinity, around 10% after immersing the samples in water and PBS degradation media for 59 weeks.

The same correlation has been found in samples degraded in PS lipase (Fig. 4), showing that the degree of crystallinity increases during hydrolysis, in this case around 6% after 11.5 weeks. The fact

that the enzymatically degraded samples presented a lower crystallinity at time zero could be due to anisotropies in the crosslinking density produced during sample preparation.

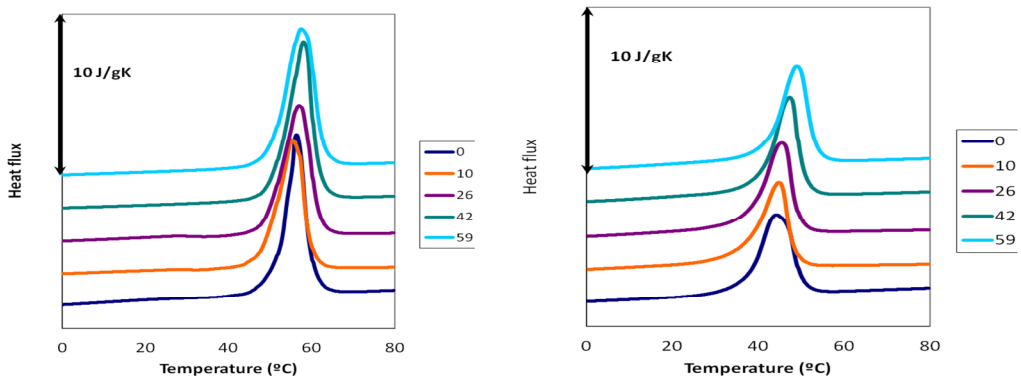


Fig. 3. First and second heating scan of samples degraded in PBS. Labels indicate the degradation time in

As shown in Figure 4, the degree of crystallinity increases during hydrolysis. It is a well-known phenomenon that the amorphous regions are more susceptible to hydrolysis, leading to increasing crystallinity during degradation of semicrystalline polyesters. The shorter chains formed during degradation also have higher mobility, allowing for reorientation of the crystalline phase, which

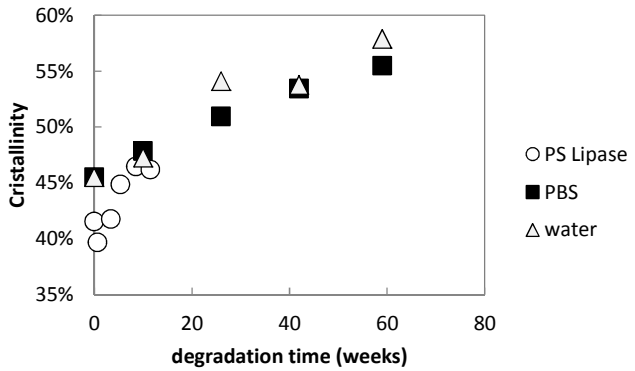


Fig. 4. Crystallinity of degraded samples in water (triangles), in PBS (black squares), and in Lipase (circles) against degradation time from the first heating.

increases crystallinity.

The crystallinity in the first scans was higher than in the second heating scans, the difference being 8% on average. This could be explained by recrystallization during storage at room temperature (25ºC) and during degradation (at 37 ºC) and it has been demonstrated that this depends on the fabrication process and storage conditions [28,29]. Both temperatures are close enough to the melting temperature of the PCL networks, around 50ºC, and the PCL chains could easily be converted into new crystals. There is no statistically significant change in crystallinity measured from the second heating scan during degradation (data not shown).

The melting temperature (T_m) of all samples (degraded in water, PBS and with lipase) increased with degradation time. T_m may increase as a result of the formation of thicker and more perfect lamellae [30]. The increase in T_m is explained by continued crystallization, where the crystalline thickness increases during hydrolysis. The same trend was observed in the second heating. The increasing T_m from the second heating scan could be due to chain scissions of the materials that may loosen their network structures to some extent, facilitating the crystallization process.

The peak half-width changes from 4.8ºC for the non-degraded sample to 11.5 ºC after 59 weeks of degradation in water and to 7.7 ºC after 59 weeks of degradation in PBS. The peak half-width did not significantly change from the non-degraded to degraded samples after 11.5 weeks in Lipase. The position and width of the peak can be associated with the thickness of the crystals; higher width corresponds to greater dispersion in crystal thickness distribution [31].

3.4. X-Ray Diffraction

X-Ray diffraction profiles for different degradation times in water and PBS are shown in Fig. 5. The diffraction pattern showed characteristic PCL peaks at 2θ=21.3º and 23.7º, which correspond to (1 1 0) and (2 0 0) crystallographic planes, respectively [32]. It can be observed that there were only slight changes in the diffraction profiles as degradation proceeded. The corresponding XRD diffraction profiles for enzymatically degraded samples are shown in Fig. 6; the obtained spectra for different

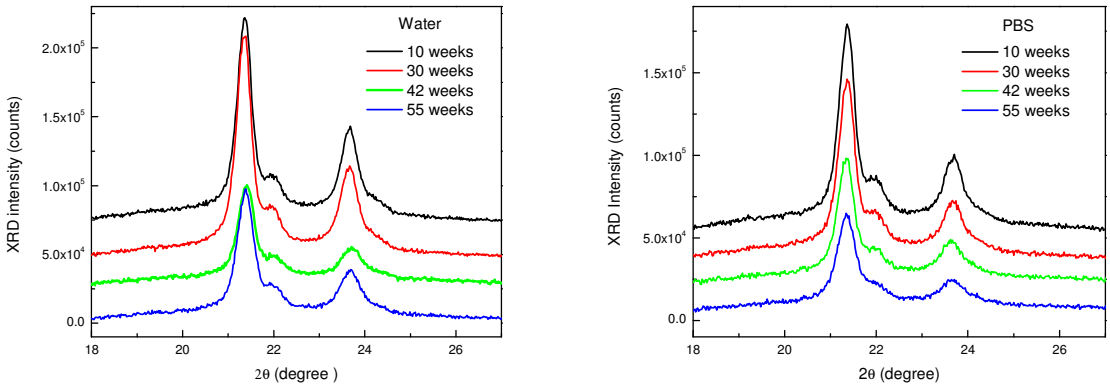


Fig. 5 a-b. XRD patterns for samples degraded in a) water and b) PBS at different degradation times. For the sake of clarity, they have been shifted.

degradation times are almost identical.

The crystallite size obtained by using the Debye–Scherrer formula (3) for hydrolytically and enzymatically degraded samples is shown in Fig. 7 and 8. A slight decrease can be observed in the crystal size when degraded hydrolytically while the crystal size remains constant in the case of enzymatic degradation.

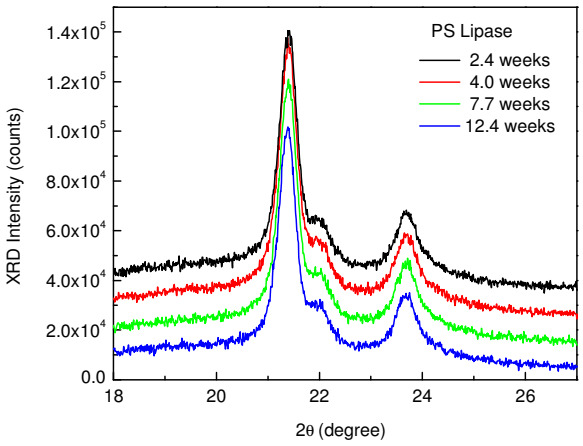


Fig. 6. XRD patterns for samples degraded in PS Lipase at different degradation times.

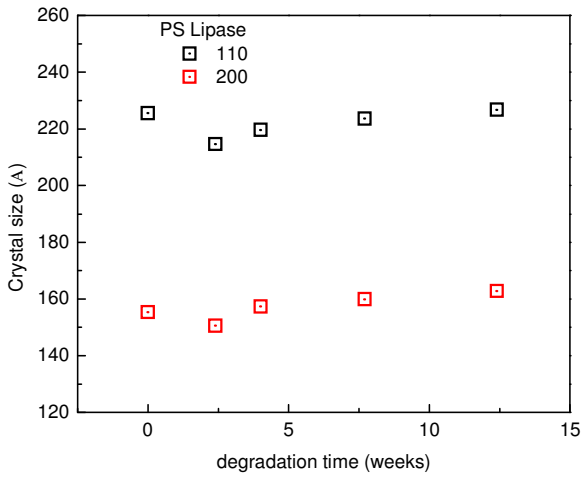


Fig. 7. Crystallite size calculated from the Scherrer equation for samples degraded in PS Lipase at different degradation times.

By comparing the results obtained by DSC and XRD, we found that, as far as enzymatic degradation is concerned, the DSC results indicated the increase in melting temperatures and no significant changes in the peak half-width, whereas the XRD results showed no changes in crystal size.

As far as hydrolytic degradation is concerned, we found, on the one hand, an increasing melting temperature measured by DSC, commonly related with an increase of the crystal size, and an increasing

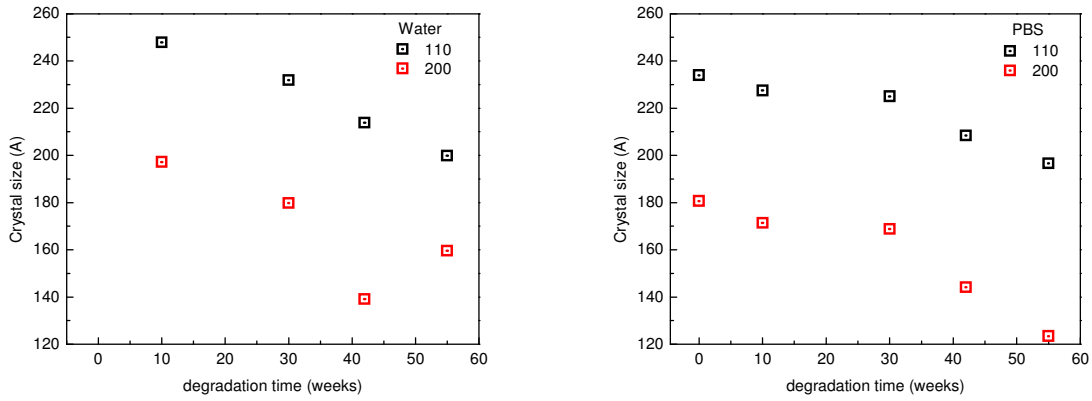


Fig. 8 a-b. Crystallite size calculated from the Scherrer equation for samples degraded in a) water and b) PBS at different degradation times.

half-width, related with a wider distribution of crystal size. On the other hand, XRD results indicate a decreasing crystal size, which could indicate a greater proportion of small crystals.

3.5. Mechanical testing

To evaluate the mechanical properties, stress-strain tests were conducted. For each degradation time, three consecutive compression scans were measured for three different samples. Although all the measurements show a similar behavior, in some cases, a different behavior can be observed, due to the brittle fracture of samples; in general, after a “toe” region, the stress-strain curve shows a linear region, until the maximum stress is reached. When the sample was unloaded, a plastic deformation appeared that remained unaltered at the beginning of the third scan. The curves corresponding to the second and third scans were very similar. As an example, the three compression scans corresponding to the samples

of 19 weeks' degradation in water and 38 weeks' degradation in PBS are shown in Fig. 9 (the last one corresponding to a brittle fracture at the first scan).

In order to evaluate Young's modulus, the slope of the stress-strain curves in the 4-5 MPa range was calculated. The Young's modulus values corresponding to the first and third compression scans are shown in Table 1, for water, PBS and enzymatic degradation. The values shown represent average values and standard deviations for the samples measured.

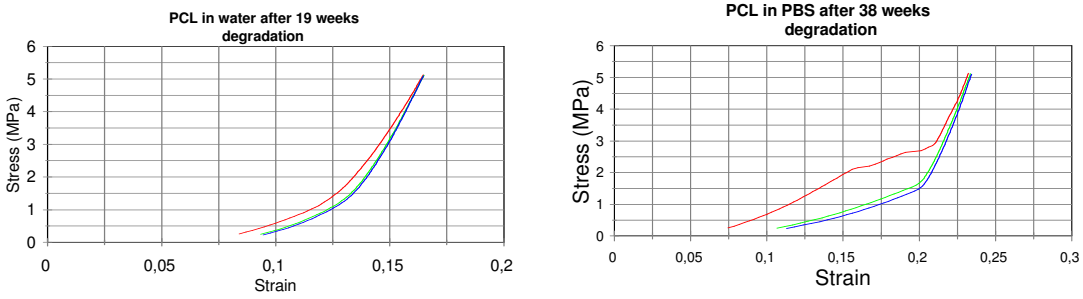


Fig. 9. Stress-strain curves for the PCL samples degraded for 19 weeks in water and degraded in PBS for 38 weeks.

Degradation media	Degradation time (weeks)	E_1(MPa)	Plastic deformation (μm)	E_3(MPa)
Water	0	83±9	25±2	98±11
	19	120±3	6.9±0.9	140.3±0.5
	38	106±10	19±11	130±6
	63	134*	21*	160*
PBS	0	83±9	25±2	83±9
	19	95±11	10±3	117±15
	38	110±3	34±20	130±3
	63	**	**	**
enzymatic	0	57.0±1.2	27±3	78±2
	1.4	74±3	57±11	93±3
	4.7	74±3	36±10	91±3
	9.5	70.8±1.1	31±1	88±1
	13.4	70±2	32±18	89±4

Table 1 PCL network degraded in different media. Young's modulus and plastic deformation for different degradation times. E_1 corresponds to the Young's modulus values calculated from the first scan while E_3 corresponds to the Young's modulus values calculated from the third scan.
* Only one sample could be measured. ** No sample could be measured.

Samples subjected to enzymatic degradation present slightly lower Young's modulus values at zero time compared to samples subjected to hydrolytic degradation, probably due to anisotropies produced during sample preparation.

As degradation proceeded, Young's modulus measured for the first scan (E_1) seemed to increase in samples degraded in water and PBS, although it did not present a consistent trend. For samples under enzymatic degradation, Young's modulus presented a similar trend; it increased at first and decreased slightly after week 5 of degradation, as reported in a previous paper [18]. The samples with the longest degradation times in water and PBS were very difficult to measure, because they were too brittle and broke easily. After 63 weeks of degradation in PBS, it was impossible to measure any samples due to their fragility. Results obtained for the Young's modulus corresponding to the third scan (E_3) followed a similar trend to that of the first one, although with higher values.

As has been said in previous papers [33], moduli for the second and third scans are higher than that for the first scan, due to pore collapse and the densification effect. This effect is related to the increase in the deformation measured between the first and third scans.

This change in modulus is due to two opposite effects [18]. On the one hand, calorimetric measurements reveal faster degradation of the amorphous phase of material than the crystalline phase, increasing the crystallinity and therefore the mechanical modulus. But, on the other hand, degradation produces mass loss and porosity in the sample, resulting in a decrease of Young's modulus [34]. The porosity can be produced inside the sample, if bulk degradation occurs, or near the surface of the sample, if superficial erosion is the degradation mechanism [25]. In both cases, porosity produces deformation between the first and third compression scans, as can be seen in Table 1.

The results of calorimetric measurements showed an increase in crystallinity of around 10% in samples degraded hydrolytically, in water and in PBS. Then, the increase of the Young's modulus suggests that the effect of increasing crystallinity is more important than the effect of porosity. It must be noted that the degradation in PBS and water produces more brittle samples than enzymatic degradation. In fact, no sample after 63 weeks of degradation PBS could be measured, and samples degraded during 38 weeks showed curves (Figure 9) with a plateau zone (at the first scan) revealing a non-recoverable crashing. For this reason, deformation values must be examined carefully.

Enzymatic degradation is faster than hydrolytic degradation, but the weight lost at the end of the test is roughly the same in all experiments. Calorimetric measurements indicate an increase in crystallinity that is lower (around 5%) than in samples under hydrolytic degradation. This could be the reason why the modulus of these samples does not increase as much as hydrolytically degraded samples.

The hydrolytic degradation of PCL network is faster than that of linear PCL, probably due to the higher hydrophilicity and the lower crystallinity of the PCL network. The same result was found in accelerated alkaline degradation [18]. On the other hand, enzymatic degradation of network PCL is slower than that of linear PCL. A more in-depth study would be necessary to understand the reasons for this.

4. CONCLUSIONS

Hydrolytic and enzymatic degradation of PCL networks were studied in terms of weight loss and swelling ratio, also using DSC, XRD, SEM and mechanical properties. Two hydrolytic media were used, HPLC water and PBS showing similar results. Enzymatic degradation was faster than hydrolytic degradation, and its mechanism was different. According to the degree of swelling and morphology results, the enzymatic degradation seemed to follow a superficial erosion mechanism, while the hydrolytic degradation affected the whole sample via a bulk erosion mechanism. In all cases an increase in crystallinity was found, although the change was greater in hydrolytic degradations. This increase in crystallinity produced an increase in the Young's modulus values, leading to those hydrolytically degraded samples becoming brittle and fragile at the end of the degradation period.

By comparing the hydrolytic and enzymatic degradation of PCL networks, the results show that the rate of enzymatic degradation is higher than that for hydrolytic, but the enzymatically degraded samples presented fewer changes in properties than the hydrolytically degraded samples when the same weight loss is considered.

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FIGURE CAPTIONS

Fig. 1. Weight loss a) and degree of swelling b) of hydrolytic and enzymatic degradation of PCL networks.

Fig. 2. Superficial SEM microphotograph of: a) a non-degraded sample; b) a sample degraded in enzymatic medium for 11.5 weeks; c) a sample after 26 weeks of degradation in water; d) a sample degraded for 26 weeks in PBS; e) a sample after 59 weeks of degradation in water; f) a sample degraded for 59 weeks in PBS. Bar=20μm

Fig. 3. First a) and second b) heating scan of samples degraded in PBS.

Fig. 4. Crystallinity of degraded samples in water (triangles), in PBS (black squares), and in Lipase (circles) against degradation time.

Fig. 5 a-b. XRD patterns for samples degraded in a) water and b) PBS at different degradation times. For the sake of clarity, they have been shifted.

Fig. 6. XRD patterns for samples degraded in PS Lipase at different degradation times.

Fig. 7. Crystallite size calculated from the Scherrer equation for samples degraded in PS Lipase at different degradation times.

Fig. 8 a-b. Crystallite's size calculated from the Scherrer equation for samples degraded in a) water and b) PBS at different degradation times.

Fig. 9. Stress-strain curves for the PCL samples a) degraded for 19 weeks in water and b) degraded in PBS for 38 weeks (○ first scan, _____ second scan, ------- third scan).

TABLES

Table 1. PCL network degraded in different media. Young's modulus and plastic deformation for different degradation times. E_1 corresponds to the Young's modulus values calculated from the first scan while E_3 corresponds to the Young's modulus values calculated from the third scan.
* Only one sample could be measured. ** No sample could be measured.