CHAPTER 2

POLYMER BIODEGRADATION

2.1 Testing for Biodegradation

The testing of materials for biomedical applications involves investigating the biocompatibility and/or the degradability of these materials. Test methods, test conditions and parameters relevant to the desired applications of the materials should be selected. Studies of biomedical polymer degradation are usually carried out in the sequence [18] :

(1) in vitro testing
(2) in vivo testing
(3) clinical trial

2.1.1 In Vitro Testing

In vitro investigations are of considerable importance in evaluating the functionality and durability of a material. In vitro testing of polymers for biomedical applications often involves degradation in blood serum and degradation with isolated tissues, e.g., with chick-embryo, liver tissue and the yolksac. Such testing is similar in certain ways to testing with microorganisms. The observed degradation might be a combination of enzyme-catalyzed degradations as well as nonenzyme-catalyzed degradations. In vitro degradation experiments are generally carried out before in vivo experiments to reduce the cost and time required for the latter [9].

2.1.2 In Vivo Testing

For biomedical applications, in vivo experiments must be carried out after in vitro testing. A polymer device is implanted for a certain period in an animal and is then explanted for analyses of chemical and physical changes. Degradation products released from the implanted device and excreted by the animal can also be analyzed.

When inserted in the aqueous environment of the body, polymers may be considered to undergo four stages of biodegradation (Table 2.1) [19].

Table 2.1 : Four stages of polymer biodegradation in vivo.

Stage	Effect	Molecular Changes
1	Hydration	Disruption of van der Waals forces and hydrogen bonds
2	Strength Loss	Initial cleavage of backbone covalent bonds
3	Loss of Mass Integrity	Further cleavage of covalent bonds to polymer molecular weight levels insufficient for mass coherence
4	Mass Loss (Solubilization)	Dissolution of low molecular weight species and phagocytosis of small fragments

2.1.2.1 Hydration

Hydration is variable in rate, degree and effect, and is dependent upon the nature of the polymer. Natural polymers such as collagen rapidly absorb appreciable quantities of water so that there is an almost immediate and significant reduction in strength compared to the unimplanted control. Synthetic materials, such as poly(glycolic acid), absorb little water and display negligible changes in physical properties during the hydration stage. This stage of absorption may be considered complete within minutes or hours after implantation unless, of course, the implant volume is so large that the diffusion of water into the mass simply takes longer. During this stage, few, if any, covalent bonds are broken. The primary effects result from disruption of secondary and tertiary structures stabilized by van der Waals forces and hydrogen bonds.

2.1.2.2 Strength Loss

The second stage of degradation is manifested by the irreversible loss of implant strength, usually as a result of covalent bond cleavage involving the polymer backbone. In the case of absorbable polyesters, the rate of strength loss is governed entirely by the rate of simple hydrolytic cleavage of the polymer backbone and is independent of any known enzyme systems. In this class of polymers, the strength loss rate is dependent upon temperature, pH and especially upon the degree of crystallinity of the polymer. More highly crystalline species may be expected to maintain their strength for longer periods of time compared to those which are more amorphous. At the end of this second stage, most if not all of the original mass is still present.

2.1.2.3 Loss of Mass Integrity

This stage involves the beginning of the absorption or mass loss process. In what may be considered to be a continuation of the stage 2 covalent bond breaking, the polymer is degraded to a molecular weight level below that required for mass coherence, and a friable or gelatinized mass which may fragment or partially solubilize results. It is during and after this stage 3 that the actual mass loss or absorption occurs.

2.1.2.4 Mass Loss (Solubilization)

The complete removal of polymer from the tissue may be considered as the fourth stage of absorption. The polymer may lose mass simply by the solubilization of low molecular weight species into the extracellular fluid. Alternatively, small fragments may be removed from the implant site by phagocytes and eventually carried to the lymphatic system for completion of the solubilization process.

It is also possible that polymeric masses may be removed from implant sites without actual reduction in the chain length through solubilization processes involving side chain modification rather than backbone scission. Poly(vinyl alcohol) derivatives are an example of this.

2.1.3 Clinical Trials

Clinical trials are the last step in the evaluation of a new material for use in a given biomedical application. However, as the long-term effects and the complications are seen only in patients, as many interim examinations of the implant as possible at re-operations should be made, not only when complications with the implant arise. In addition, as many autopsies as possible should be conducted, particularly when the patient died due to causes totally unrelated to the implant [18]. Permission to conduct clinical trials is

subject to legal procedures and necessitates close collaboration between the polymer scientists (the inventors) on the one hand and the medical practitioners (the end-users) on the other.

2.2 In Vitro Biodegradation Studies Performed in This Work

In this work, only in vitro testing was carried out. This was performed by immersing the samples in a 0.2 M phosphate buffer medium at the physiological pH and temperature of 7.40 and 37°C respectively. From the literature, various other in vitro media have also been used to simulate the extracellular fluid in the human body such as Krebs Solution, saline and blood plasma [9].

In order to follow the in vitro biodegradation of the synthetic homo- and copolyesters studied here, the following properties were monitored:

(1) weight loss (absorption)
(2) polymer morphology (% crystallinity)
(3) intrinsic viscosity (molecular weight)

2.2.1 Weight Loss

Weight loss resulting from the degradation of a polymer invariably requires the breakage of chemical bonds. Once chemical bonds start to break, reactive chain ends and free radicals are created. Degradation can then proceed either by depolymerization or by random chain scission. The dominant degradation mechanism may depend significantly on the details of structure and composition, including the types of end groups terminating the polymer chains, and the presence of structural defects, additives or impurities. In any case, loss of weight can only start after the first bond breakage event. The first bond to break will usually be the weakest bond in the polymer chain [20].

Dahlmann et al. [21] have studied the in vitro degradation of polylactides and poly(glycolide-co-lactide)s with different glycolide contents. The in vitro degradation, followed by weight loss as a function of time, is influenced decisively by the structure of the polyester (Fig. 2.1). While the weight losses of the polylactides were only small after 5 weeks, a drastic increase in the hydrolysis rate was observed by increasing the amount of glycolide units.

A careful study of the process of in vitro degradation revealed that hydrolysis not only sets in from the surface of the sample but that the polymer matrix also swells in an aqueous medium, relatively independently of the thickness of the layer, and is degraded equally over the entire cross-section. At the same time, degradation does not take place at the chain ends but is distributed statistically along the chain. Hence, it follows that a weight loss will only be observed when the degradation has proceeded relatively far and monomeric hydroxycarboxylic acids will occur only relatively late in noticeable quantities in the solution.

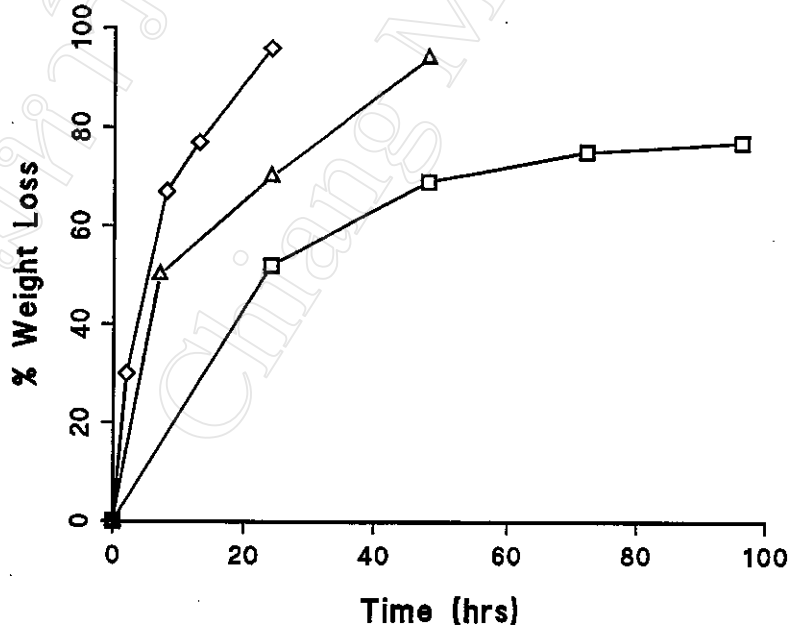


Fig. 2.1 : In vitro degradation of polylactides (PLA) and poly(glycolide-co-lactide)s (PGLA) with different glycolide contents [21].
□ D,L-PLA Δ PGLA (50/50) ◊PGLA (75/25).

2.2.2 Polymer Morphology [22]

Crystallization reduces the reactivity of polyesters. For example, the crystallinity of stereoregular poly(L-lactic acid) is associated with a several-fold reduction in the rate of chain scission relative to amorphous poly(DL-lactic acid). The crystalline phase, unlike the amorphous phase, is inaccessible to water and excipients and its degradation is restricted to the crystallite edges. Interestingly, chain scission of tie segments between crystalline segments of semicrystalline polymers can result in further crystallization of the amorphous phase. Increases in the crystallinity of poly(L-lactic acid), poly(ε-caprolactone) and poly(glycolic-co-L-lactic acid) 75:25, with increasing degrees of hydrolytic degradation have been described. Crystallization of L-lactic acid sequences in non-annealed poly(D-lactic acid-co-L-lactic acid) 4:96 has also been observed.

An example of an increase in the rate of hydrolysis arising from a reduction in crystallinity is provided by the copolymer of ε-caprolactone and DL-lactic acid. This amorphous copolymer has been shown to undergo hydrolytic chain scission at a greater rate than either of the homopolymers. Similarly, copolymers of glycolic acid and lactic acid are degraded more rapidly than polyglycolic acid because of a reduction in the crystallinity; the 1:1 copolymer, which is totally amorphous, is hydrolyzed most rapidly.

It is possible that the increased reactivity of poly(DL-lactic acid-co-ε-caprolactone) is also a result of the reduction in the glass transition temperature (Tg) of the copolymer. A reduction in the Tg of the polyester is believed to increase its susceptibility to hydrolysis by increasing chain mobility and reducing the energy required to achieve the transition state. This possibility was first suggested by the temperature dependence of the rate of degradation of poly(glycolic-co-DL-lactic acid), as measured by the initial rate of tensile strength loss.

Gilding and Reed [23] have pointed out that the amorphous state of poly(glycolic acid) is ideal for applications where it is necessary to have mass loss simultaneously with molecular weight degradation, or for applications

such as drug delivery where it is important to have a homogeneous dispersion of the active species in a monophasic matrix. The partially crystalline morphology is relevant to use where high mechanical properties are required, i.e. in sutures and in orthopaedic or dental applications. It is expected that the degree of crystallinity will influence chemical and physical properties, such as swelling behaviour and hydrolytic sensitivity of the polymer and, consequently, its rate of biodegradation.

2.3 The Importance of Studying Polymer Microstructure [24]

The goal of most polymer research is to seek an understanding of polymer behaviour. To achieve this objective, it is necessary to gain some knowledge of structure-property relationships. The chemical microstructure of the polymer chain is related to the polymerization parameters used in the synthesis reaction. The synthesized polymer is then characterized, in the case of a copolymer, in terms of its composition and monomer sequence distribution and so on.

The process of characterizing a polymer is complex, even to determine the fundamental molecular structure of the polymer chains. These polymer chains often contain conformational variations, different crystalline phases, and a disordered or amorphous component, in addition to process-induced chain orientation. In the case of a copolymer microstructure characterization, NMR, IR, and UV spectroscopy, and pyrolysis-gas chromatography are among the techniques that exhibit sensitivity to differences in monomer sequence distribution.

2.4 Previous Work and its Relevance to This Study

There are various approaches to the design of biodegradable polymers for use in medical applications. The use of cyclic ester monomers in the formation of polyesters for the fabrication of synthetic surgical articles is well

known. Comonomers have often been employed to modify the characteristics of the various polyesters. The conventional polymerization method for forming polymers of cyclic esters is through ring-opening polymerization. The use of such monomers in the formation of polyester surgical articles has been discussed in a variety of patents and technical publications [25, 26].

Numerous copolymerizations of glycolide and other cyclic ester monomers such as β-propiolactone, β-butyrolactone, δ-valerolactone, ε-caprolactone and ethylene carbonate in the presence of various catalysts have been studied by many workers [27]. The method of synthesis usually involves sequential addition of the monomers in the polymerization whereby the glycolide monomer, the other cyclic ester monomer, or a combination of the two is substantially completely polymerized before the addition of the remaining monomers used to form the copolymer chain. By conducting the polymerization procedure in a stepwise or staged manner, the in vivo characteristics of the surgical articles produced can be modified prior to encountering the usual degree of interference in the ability of the copolymer to form dimensionally stable, highly crystalline, or highly oriented molecular structures. In a multi-stage process, a different catalyst may be employed at each stage if desired.

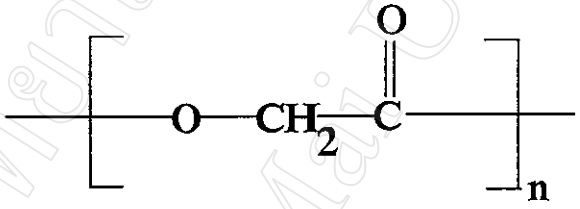
Since interest in biodegradable polymers of low toxicity seems likely to increase in the future, the demand for novel copolyesters with improved properties is increasing. Hydroxycaproic acid is another building block of relatively low toxicity and a suitable monomer is technically available in the form of ε-caprolactone. For example, the copolymerization of an equimolar mixture of glycolide and ε-caprolactone catalyzed by aluminium isopropoxide proceeds up to 25% for 2 h at 100°C without solvent to give a copolymer with a glycolide content of 85%. The average degrees of polymerization of the homoblock sequences for glycolide and ε-caprolactone are 14.0 and 2.5 respectively [17, 28].

Kricheldorf et al. [29] have studied the copolymerizations of glycolide and β-propiolactone carried out in nitrobenzene solution catalyzed by various

initiators. Low glycolide/β-propiolactone ratios were found when acidic initiators were used. In contrast, a high glycolide/β-propiolactone ratio was obtained with aluminium isopropoxide. Many similar examples are to be found in the polymer literature.

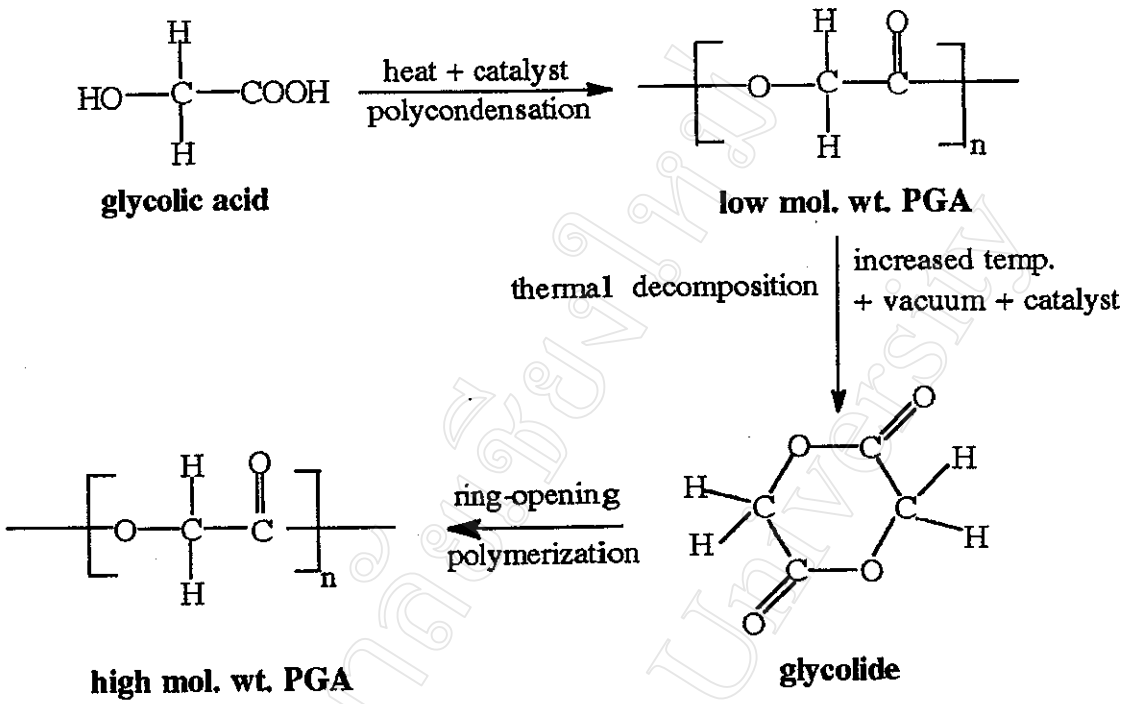
2.5 Poly(glycolic acid) [30]

The most widely studied biodegradable polymer, since interest in these materials began, has been poly(glycolic acid) (PGA). The molecular formula of PGA is as shown below:



poly(glycolic acid)

PGA is the simplest member of the poly(α-ester) series. In its synthesis, glycolic acid (hydroxyacetic acid) is converted to PGA by initially reacting glycolic acid with itself to form the cyclic diester glycolide which, in the presence of heat and a catalyst, is then converted to a high molecular weight, linear-chain polymer from which filaments can be extruded (Scheme 2.1). The commercial PGA multifilament suture (trade name DEXON : Davis & Geck, American Cyanamid Co.) is manufactured by orienting these filaments by means of stretching and braiding.

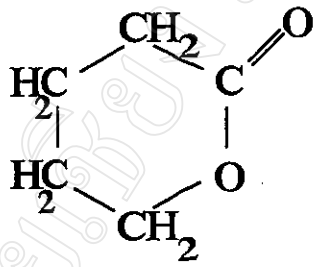


Scheme 2.1 : Conversions involved in the synthesis of high molecular weight poly(glycolic acid) (PGA).

In vitro tests on PGA sutures (DEXON) have shown that degradation occurs in two stages. The first stage consists of diffusion of water into the amorphous regions of the polymer leading to random hydrolytic chain scission of the ester groups. The second stage starts when most of the amorphous regions have been degraded, hydrolytic attack then focussing on the crystalline domains. An apparent increase in crystallinity with (degradation) time is explained by the chain fragments of the hydrolysed amorphous regions having a lesser degree of entanglement and by their ability to re-align themselves into a more ordered crystalline state [2]. PGA sutures are synthetic, non-collageneous, absorbable sutures with surgical handling properties similar to those of silk. The material has been shown to be completely non-toxic upon implantation into animals [31] and to function satisfactorily when used in a wide variety of surgical operations.

2.6 Poly(δ-valerolactone)

The six-membered ring cyclic monoester analogue of glycolide is δ-valerolactone. The chemical structure of δ-valerolactone is as shown below:

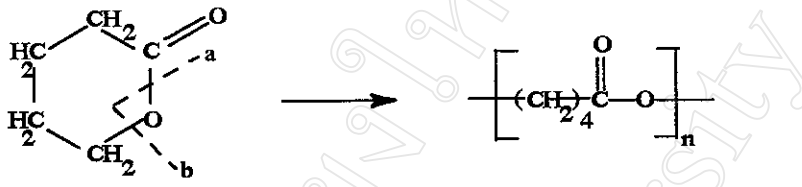


δ-valerolactone

Carothers [31] was able to polymerize δ-valerolactone by heating at 80°-85°C for a period of about 13 days, or by contacting δ-valerolactone with potassium carbonate catalyst at a temperature of 80°-85°C for a period of about 5 days. The resulting polymers were soft waxes possessing average molecular weights of approximately 2000 which had relatively low thermal stability.Poly(δ-valerolactone) is a flexible material which has a low melting point (≈ 53°C), a low crystal density, and good solubility in most organic solvents.

Carothers et al. [31] originally postulated that δ-valerolactone might polymerize through rapid ester interchange. Stannet and Szwarc [32] suggested that the spatial configurations of monomer units and polymer chains were very similar and, thus, interchange processes could occur with little movement of the carbon center. They further suggested that, with δ-valerolactone, cleavage

could occur at the acyl-oxygen bond (a) or the alkyl-oxygen bond (b), as shown below:



δ-valerolactone poly(δ-valerolactone)

Based on the studies of model compounds, Stannet and Szwarc went on to conclude that the alkyl-oxygen bond cleavage might be more feasible energetically. However, Cherdron and co-workers [32] reported that acyl-oxygen cleavage was the preferred polymerization mechanism. A great number of anionic and cationic compounds have been examined as catalysts for δ-valerolactone polymerization with varying results. For example, the polymerization of δ-valerolactone initiated by ethylene glycol at 160°C was found to form only low molecular weight polymer with substantial amounts of monomer remaining in equilibrium with the polymer.

2.7 Poly(glycolic acid-co-δ-valerolactone)

Copolymerizations of glycolide with other cyclic monomers such as ethylene carbonate, trioxane and δ-valerolactone have been studied [33]. It was found that, compared to ethylene carbonate and trioxane, δ-valerolactone appeared to be more reactive and therefore more competitive in its copolymerization with glycolide. Whereas poly(glycolic acid) is characterized by a high melting point (Tm ≈ 220°C) and poor solubility in organic solvents, poly(δ-valerolactone) has a low melting point (Tm ≈ 55°C) and good solubility. The potential therefore exists for the copolymerization of glycolide and δ-valerolactone to lead to a new copolyester with new improved properties.

In other previous work, Fukuzaki et al. [34] synthesized a copolyester of glycolide and δ-valerolactone in the presence of water without catalysts at 200°C under N_2. A relatively low molecular weight poly(glycolic acid-co-valerolactone) was obtained.

Kricheldorf et al. [27] also synthesized poly(glycolic acid-co-valerolactone) by using three different classes of catalysts: acidic catalysts, complexing catalysts, and anionic catalysts at 100°C in bulk. The results showed that acidic initiators favored the incorporation of δ-valerolactone, whereas the complexing initiators favored the incorporation of glycolide.

It has been on the synthesis, microstructural characterization, and in vitro biodegradability of poly(glycolic acid-co-valerolactone) that this research project has focussed its attention.