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DISSOLUTION AND MECHANICAL PROPERTIES OF 3D PRINTED POLYLACTIC ACID FOR BIO-IMPLANTS[1]

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Abstract: In this paper preliminary results of dissolution of 3D printed meshes of PLA (polylactic acid) in a biologically active media are presented. The issue of biocompatible polymers dissolution is important as the knowledge of its mechanism and kinetics allows designing bio-implants with required and time controlled properties. Not only dissolution kinetics but also the change in mechanical properties of biocompatible polymers suitable to be used as implants is important. In this study, we investigated the influence of bioactive media on hardness and tensile behaviour of the PLA. Density, mass loss, hardness and tensile strength of the material after soaking in a bioactive media were determined. Mass loss was determined after different soaking times in order to establish a relationship between the rate of PLA dissolution and the geometry of meshes. The results draw some conditions for next experiments to study the kinetics of the PLA dissolution.

Keywords: Bio-implants, PLA, Polylactic acid, 3D printing, Polymer dissolution, Tensile test.

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INTRODUCTION

Traditionally, metals are the materials of choice when it comes to most engineering applications. However, with the advances made in chemical technology, the use of polymeric materials is rapidly gaining momentum, and their application for various engineering products is becoming much more frequent (Saba N. & M. Jawaid, 2018, Sun H. et all, 2018). Typical examples of the uses of polymers are custom goods, automotive parts, various parts for the oil and gas industry, or for marine applications. Due to their versatility polymers have found their way even into areas, where mechanical and thermal loadings have extremely high magnitudes such as the ordnance and munitions industry where such materials are used for various purposes including, but not limited to, components for small arms and large calibre artillery fuzes.

One of the quickly evolving branches of engineering technology, where polymer materials are widely used are the techniques for additive manufacturing, also known as Rapid Prototyping (RP). These methods are based on layer deposition of the material and its subsequent solidification, thus producing three-dimensional solid objects. Well known examples of such technologies are the Selective Laser Sintering (SLS), Stereolithography (SLA), and Fused Deposition Modelling (FDM) (Kalpakjian, S., & Schmid, S., 2013). These methods can use, among other materials, polymeric feedstock. Common areas of application of RP are production of ready to use parts and the casting of metals (Minev, R. et al., 2016). A very promising area of research is the use of 3D printing technology for medical purposes such as the production of implantable devices. All RP techniques can use synthetic polymers, which are a source of environmental pollution (Leja, K. & Lewandowicz, G., 2010). This is the cause of favouring biodegradable ones. In particular, biodegradability, and biocompatibility are of great importance to manufacturing of implants (Azevedo, H. et al., 2015, Ratner, B. et al., 2004, Gloria, A. et al., 2010). Examples of parts, made of biodegradable materials are: scaffolds for tissue repair, orthopaedic applications, stents, staples, sutures, tissue engineering devices, fracture fixation (pins, screws) in non-load-bearing bones, wound clip, adhesives, bloodcontacting devices, skeletal reconstruction, hernia repair meshes (Gregor, A. et al., 2017, Murphy, S. et al., 2014, Li, H. et al., 2016, Sanders, D. et al., 2012, Xiong, X. et al., 2010, Stephen, J. et al., 2017). Examples of natural polymers, used for such purposes, are collagen or alginate, which are natural fibres, and generally meet the requirement for compatibility by avoiding toxicity and reducing the probability for rejection of implanted devices. Due to certain advantages, synthetic materials have gained popularity for use in implantable device manufacturing. The most widely used synthetic biodegradable and bio-absorbable polymer materials for such purposes are: Polyesters such as Polylactic acid (PLA); Polyglycolic acid (PGA), or PLA/PGA co-polymers; Poly-α-hydroxy aliphatic esters; Polydioxanone (PDO); Polyhydroxbutyrate (PHB); Polygydroxyvalerate (PHV); Polycaprolactone; Polydioxanone etc (Li, H. et al., 2016, Sanders, D. et al., 2012, Nagarajan, J – S. et al., 2009).

The goal of the current work is to investigate the suitability of PLA for manufacturing of implantable surgical meshes for tissue repair. Considering the intended in vivo application, the material in question has to be biocompatible, i.e. to be non-toxic to the host organism, with low corrosion and wear rates, it has to possess suitable mechanical properties, and it has to be easily and economically produced (Murphy, S. et al., 2014, Kang, Ch.-W et al., 2018, Wang, J. et al., 2012). Another requirement for the particular implanting devices in body interior is that of ageing, and resorption of the material. Over a given period of time the implanted device has to be dissolved and removed from the body by metabolic processes. Thus, the PLA must demonstrate ageing, and resorption rates within certain limits. Hydrolysis, oxidation, enzymatic interactions, and some mechanical effects (SuPing, L. et al., 2009, Deroinė, M. et al., 2014) govern these rates. Therefore, the present work aims to study properties such as density, tensile strength, and degradation of PLA samples, after exposing them to enzyme solution.

EXPOSITION

Materials and methods

All smaples in this study were produced using 3D printer Veleman K8200.

Degradation testing. The degradation of PLA was tested on meshes shown in Figure 1. Dimensions and design parameters of the specimens were as follows: Number of outline layers = 7; Mesh diameter = 37 mm; Outer diameter = 42 mm; Thickness = 420 μm. The most critical parameter of the meshes affecting the kinetics of the sample interaction with the enzyme solution is the surface to volume ratio (S/V). Two meshes were produced: with ratio S/V = 6.52 mm^{-1} and S/V = 5.28 mm^{-1}.

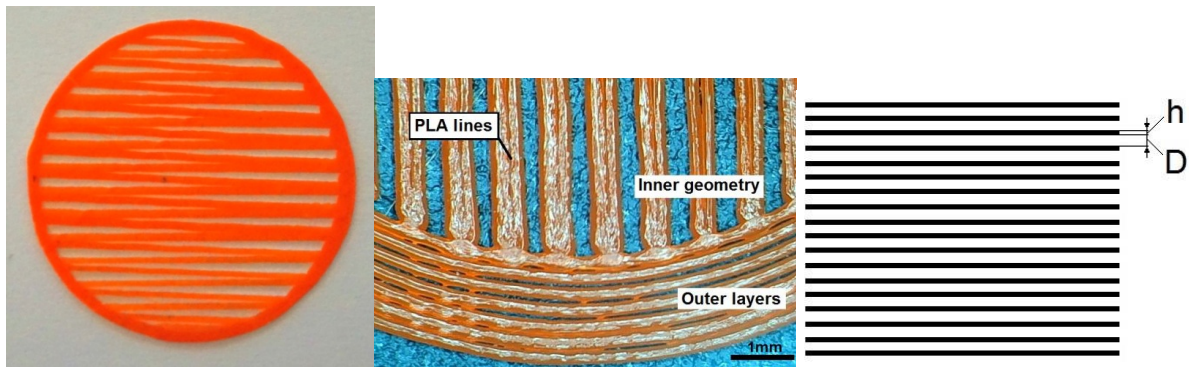Figure 1. Samples for degradation testing and their geometry: h=1150 μm, D=705 μm

The used degradation solution was 0.04 g/l lysozyme from chicken egg white from Alfa Aesar, buffered with 0.1M Tris-HCl at pH 7.4 and temperature of 37°C, open to air and stirred at 100 rpm. A magnetic stirrer LLG-uni STIRRER 3 with heating and setting accuracy ±1°C and ±10 rpm has been used to maintain the testing conditions. The specimens were hold for different testing periods then washed with deionized water and dried with filter paper. The wet mass m_w and the mass m_d after drying at 50°C in a vacuum dryer for 90 min were measured and the hydrophilicity was determined.

The degree of swelling (Castilla-Cortázar, J. et al., 2012) was calculated according to:

$$degree\ of\ swelling, \% = \frac{m_w - m_d}{m_d} * 100 \tag{1}$$

where m_w, g, is the wet mass at a specific time and m_d, g, is the dry mass.

Mass changes were calculated according to:

$$mass\ change, \% = \frac{m_0 - m_d}{m_0} * 100 \tag{2}$$

where m_0, g, is the initial mass.

The solution was changed with fresh one after every soaking period. Mass measurements were carried out with analytical balances Kern ADB 600-C3 with readability of 0.1 mg and reproducibility of 0.2 mg. For statistical representability of the resuls 10 measurments have been done for each of the testing periods.

Tensile testing was performed before and after soaking for 2640 hours in the enzymatic solution, according to ASTM D638, with an Instron universal testing machine at ambient temperature of 22°C and 72% relative humidity. The crosshead speed was set at 5 mm/min. Samples for tensile testing were conditioned at 22°C for 48 h before testing. 48 h before tensile testing, the soaked samples were washed with deionized water and dried in a vacuum dryer at 50°C for 90 min. Tested samples are shown in Figure 2. Two building strategies were used: 1) cross-hatched layers and 2) layers, parallel to tension direction.

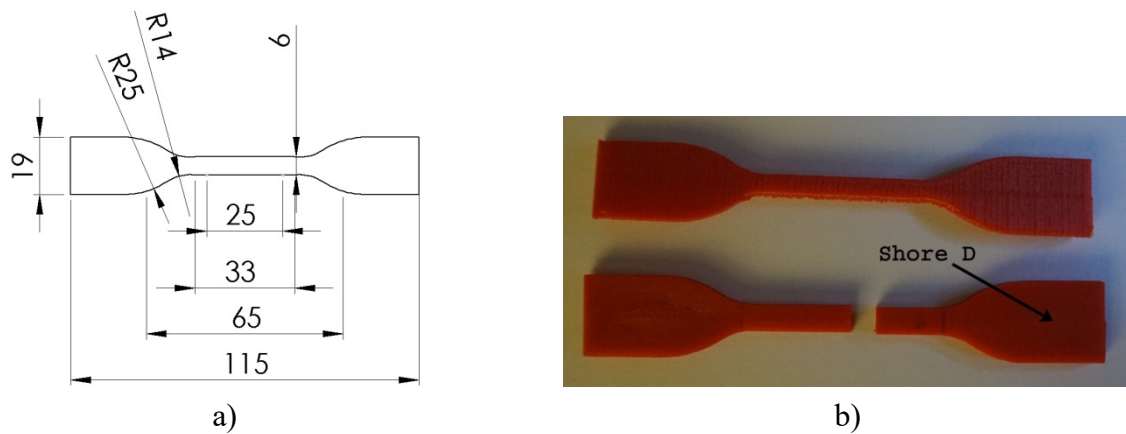a)   b)

Figure 2. Samples for tensile testing: a) dimensions and geometry; b) two samples before and after tensile testing. The Shore hardness testing position is also specified.

Shore D hardness testing was carried out according to ASTM D2240 at temperature of 22°C and 72% relative humidity with Shore D durometer and load of 5 kg. The clamping sections of the tensile test samples (before and after 2640 hours exposure to the lysozyme solution) were used to perform the Shore tests as shown in Figure 2, b.

The **PLA density** was determined for PLA in delivery sate, and before and after soaking for 1056 hours in the working media according to ASTM D792-98, using Gay-Lussac pycnometer and spherical specimens of PLA with diameter of 5 mm – Figure 3.

Results and discussion

In contrast of our expectations the density of the PLA samples did not change significantly after the extrusion process performed by the 3D printer (Figure 3) which is a positive circumstance giving grounds for using the material for load bearing applications in ortopeadic implantology. However, after soaking the samples the density of the material decreased slightly.
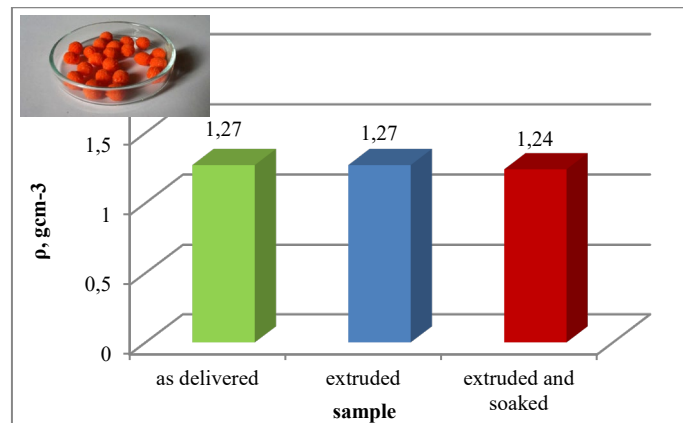

Figure 3. Samples for PLA density determination and the masured density of the studied samples

Figure 4,a shows load-elongation curves from the tensile test, and Figure 4,b – the calculated from force-elongation data yield strength $R_{0,2}$ and ultimate strength R_m. From the figures, it is evident that the calculated ultimate strength was related to the building strategy. When the layers were no cross-hatched and parallel to the tension, the ultimate strength was nearly one and a half as much as that of cross-hatched layers. It showed that the connection between the lines in the layers that were transverse to the tension was marginal and did not resist tension.

The yield strengths of samples were virtually equal except for the soaked cross-hatched sample. The data from tensile test did not allow calculating yield strength for the soaked specimen with cross-hatched layers as the fracture occurred below yield strength and was brittle. The fracture

for the rest of the specimens occurred at low degree of plastic deformation and was practically brittle.

A small increase in ultimate strength was observed only after soaking in the enzime solution-Figure 4,b. A possible explanation of this phenomenon could be associated with the previously discussed decrease of the material density (see Figure 3) after soaking. The change of the density was very small and no significant voids and imperfections in the material structure were observed after extrusion. It can be assumed that this rather small increase of the material's density is associated with some "swelling" effect that took place in the material during the soaking. The swelling led to denser alignment of the material structure not only on the molecular level but also on the macro level affecting the contact between the lines and layers of the 3D printed samples, and leading to the observed increase in the ultimate strength.

The soaking of the samles under the observed conditions (see the parameters: time, solution temperature and composition described in the beginning of this section) did not significantly influence the Shore hardness of the PLA material. The investigated building strategy and the corresponding layers' alignment (parallel layers and crosshatched layers of material deposition) also did not affect the hardness. The Shote hardness of all investigated samples remained within the range of 79-80 units no matter of the soakng procedure and build strategy. This result is important for the medical users of this type of 3D printing technology as it means engineering friendly build preparation that does not neceserrely take into accont any anisotropy of the parts. More particularly the X-Y-Z sample orientation (as well as "sorted field" options) would be of less importance when the mechanical properties are in concern although this conclusion needs to be taken into account more cautiously when thiner samples are prepared (e.g. meshes build of only 2-3 layers).

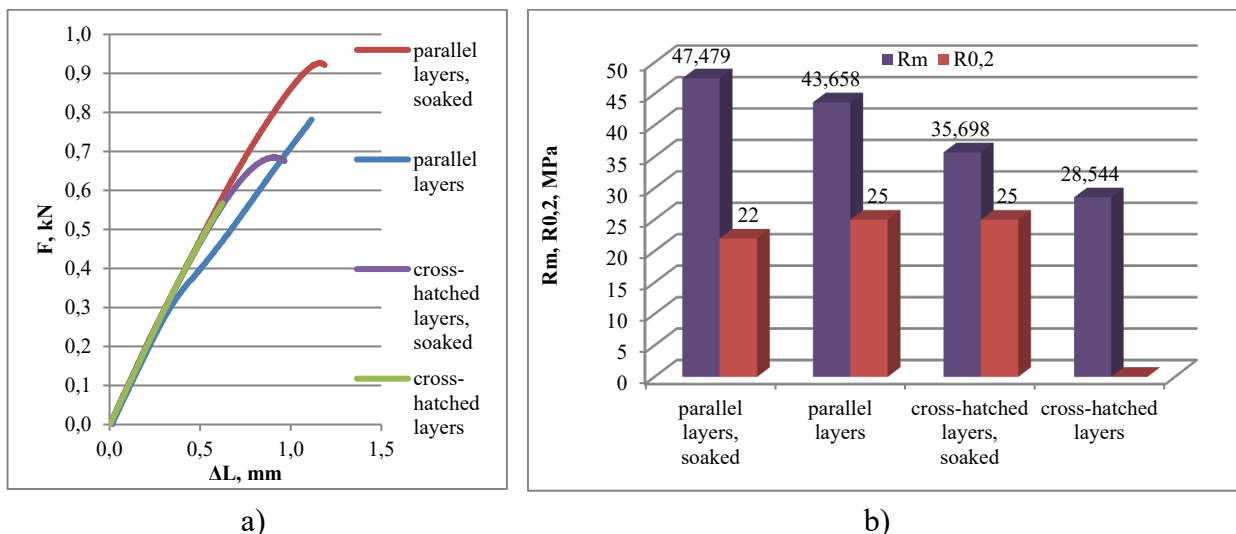a)   b)

Figure 4. Tensile testing results: a) Load – elongation curves of PLA; b) ultimate strength and yield strength of PLA

Figure 5,a shows dry mass changes of specimens with surface to volume ratio $S/V = 6.52$ mm^{-1} and 5.28 mm^{-1} as a function of soaking time. Initially mass loss was observed for both samples and the initial mass losses for sample with $S/V = 6.52$ mm^{-1} were more prominent. The analyses and explanation of this behaviour should follow the degradation mechanism of the material, which was recently extensively studied. According to the literature (Azevedo, H.S. & Reis, R.L., 2004, Li, S., 1999, Karamanlioglu, M., 2013, Elsawy, M.A. et al., 2017, Prema, S.& Palempalli, U.M.D., 2015), the PLA biodegradation starts with water absorption and hydrolysis that leads to scission of ester bonds in PLA macromolecules (Karamanlioglu, M., 2013):

$$-COO- + H_2O \longrightarrow -COOH + HO- \tag{3}$$
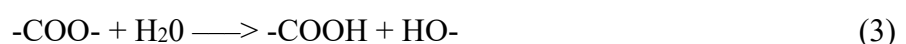
This process is later followed by enzymatic degradation. During the initial stages of degradation, the formed products are with "significant" mass and size that do not allow them to be quckly dissolved and they remain in the material structure. Therefore, the measured mass losses are small (Azevedo, H.S. & Reis, R.L., 2004) in the beginning of the process. That beahviour of PLA was observed and confirmed in the present study. The degradation products were observed even by naked eye preferably on the surface of the polymer filaments. As Figure 5,a shows, during the earlier stages of our experiment mass losses even increased slightly, and maximum mass loss for both specimens was measured only after 144 hours of soaking. More particularly the figures show that in the observed realatively short time scale the further soaking led to increase in dry mass - mass changes started to decrease and become negative for the specimen with higher S/V, the last fact indicating mass gain.
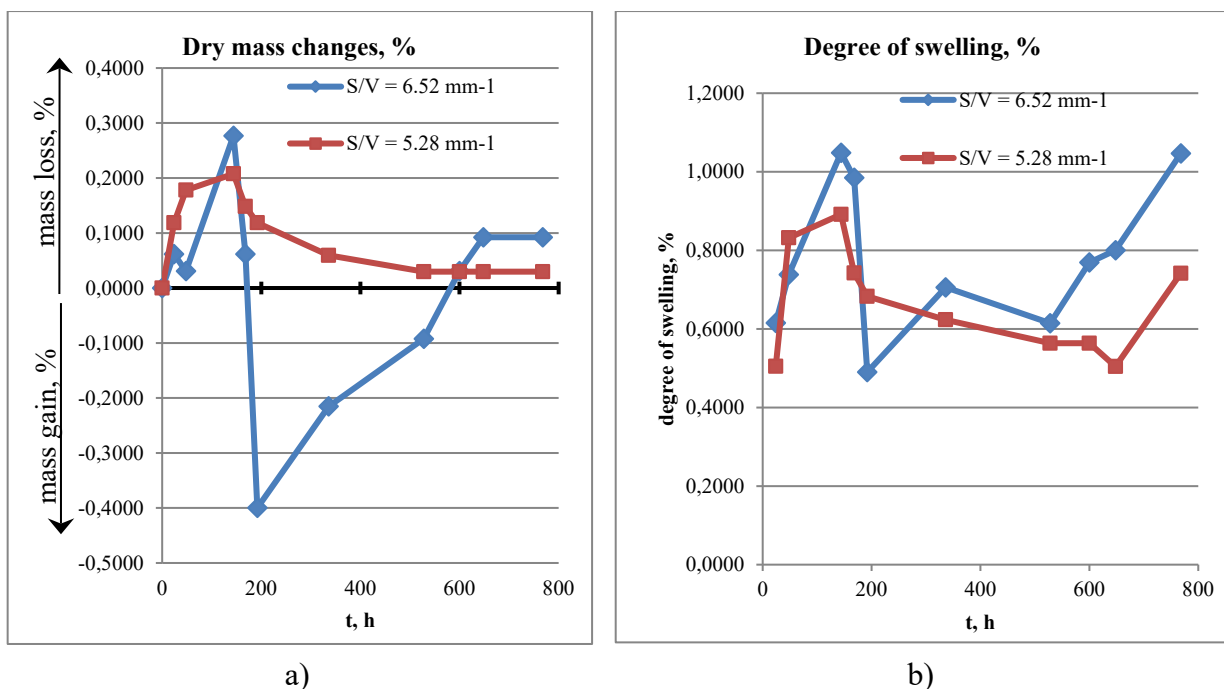

Figure 5. PLA degradation as a function of time: a) mass changes – loss and gain;
b) degree of swelling

The observed degradation products were white in colour and they deposited onto the specimens' surfaces leading to the mass increase. This type of deposits are typical for the aerobic biodegradation of polymers which leads to formation of water, carbon dioxide, biomass and residues (Leja, K. & Lewandowicz, G., 2010):

$$Polymer + O_2 -> CO_2 + H_2O + biomass + residue(s) \qquad (4)$$
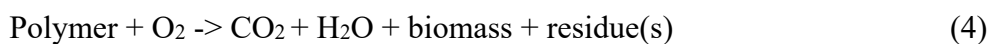
The degradation products are later converted to gases, a process known as mineralization (Leja, K. & Lewandowicz, G., 2010). The mineralisation started after 600 hours of soaking when the deposits disappeared from specimens' surfaces and positive mass losses were measured again. The hydrolytic degradation followed by enzymatic degradation after 600 hours of soaking lead to positive mass losses due to the catalytic effect of the lysozyme.

Figure 5,a and 5,b demonstrate a correlation between the mass loss and the degree of materials swelling. The water uptake depended on the change of the mass of the samples: it increased with mass loss and decreased when mass gain was in place. That correlation indicates that degradation process altered surface conditions: presence or absence of degradation deposits. The surface deposits acted as a barrier for the water molecules; they reduced the water absorbtion and obstructed the process of hydrolysis.

In summary, dry mass changes of specimens during soaking were governed by two competing processes: (i) degradation by hydrolysis and enzymatic degradation, and (ii) deposition of degradation products. These two competing processes lead to complex behavior of the material subjected to the enzymatic solution illustrated by the kinetic curves in Figure 5. The greater mass loss of the specimen with higher S/V (6.52 mm^{-1}) compared with the sample with lower S/V (5.28 mm^{-1}) confirms that the morphology of the PLA mechanical structures plays an important role for the speed of the dissolution process. As observed in other studies (Karamanlioglu, M., 2013) the increase of the surface to volume ratio of the dissolved material lead to a higher rate of hydrolyses.

CONCLUSION

The experimental results allow the following conclusions to be made:

1. No significant changes in the mechanical behaviour of the PLA material after soaking in the lysozyme solution for 2640 hours were registered. The tensile strength, yield strength and hardness do not show significant change. The PLA fracture remained brittle.

2. The PLA density decreases slightly after a 1056 hours of exposure to the lysozyme solution, which could be attributed to some swelling processes in the material that take place on different material structure scale.

3. Within the studied time span the PLA mass decreases by a negligibly small amount. During this initial treatment period the PLA degradation competes with the deposition of degradation products which leads to complicated dissolution kinetic curves.

4. The capability for water intake after 816 hours of exposure of the PLA to the solution is found to be rather small (app. 1%).

5. The calculated mass loss and degree of swelling for the samples with about 20% greater surface to volume ratio (S/V = 6.52 mm^{-1} vs. 5.28 mm^{-1}) are larger by 30-40 %. This proves the importance of the mechanical morphology of the material concerning the dissolution rate.

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