# Enzymatic degradation of starch based blends thermoplastic compounds: Morphological changes in the material

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#### Introduction

Recently, different starch/synthetic polymer blends have been suggested to have potential for use in distinct biomedical applications [1]. The structure at the molecular level of those blends is very complex, involving possibly interpenetrating networking on other chemical reaction during the blend of the components [2]. A material to be used in bone repair should accumulate not only a set of mechanical properties (matching those of bone), with the adequate degradation kinetics, but also a bone-bonding capacity and biocompatibility.

#### Materials and Methods

The material studied was a thermoplastic blend of corn starch with a poly(ethylene-vinyl alcohol) copolymer (60/40 mol/mol), SEVA-C. The samples, were weighed and immersed for several pre-fixed ageing periods at pH 7.4 and 37°C±1°C in individual containers with a Hank's balanced salt solution(HBSS) with 50, 100 and 150 unit/l of  $\alpha$ -amylase. At timed intervals, twice a week, the increased sugar concentration was followed using the dinitrosalicylic acid method (DNS). The mass of glucose released to the solution was obtained for each enzyme concentration per specimen mass.

To observe the surface and roughness properties of SEVA-C specimens, as a function of immersion time, a NanoScope III atomic force microscope was used (Digital Instruments). Before analysis, the specimens were cleaned ultrasonically in acetone, rinsed with distilled water, and stored in a desiccators under constant relative humidity. The *in-situ* observation was performed in Tapping mode.

## **Results and Discussion**

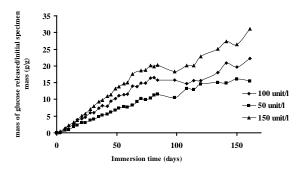


Figure 1 – Mass of glucose released to the solution for each enzyme concentrations, 50, 100 and 150 unit/l, per initial specimen mass (1.61 g) in 50 ml of solution, as a function of immersion time.

In order to study any SEVA-C structural material limitation to α-amylase degradation, 3 enzyme concentrations were applied to the same specimen (1.61 g). Figure 1 evidences the increased reducing sugars released per specimen mass, as a function of immersion time. After 100 days immersion, for 150 unit/l of enzyme the % of released saccharides was the highest. For 50 unit/l, there was a structural limitation, that tends to stabilize the % of released saccharides. Up to 50 days, the degradation rate tends to differ, increasing for the highest enzyme concentration. From the results we can conclude that the starch concentration present on SEVA-C material was limiting for enzyme binding. The α-amylase concentration normally present in human blood plasma (50 unit/l) was insufficient for the degradation of the material to be completed, until the formation of the newly grown bone tissue (3 months approximately).

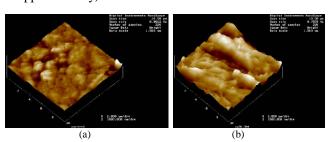


Figure 2. AFM tapping mode images: a) control and b) after 101 immersion days (data scale 1500 nm and scan size 10  $\mu$ m). The images present dried samples after immersion period.

The AFM tapping mode topographic images confirmed the increased porosity as a function of immersion time (Figures 2). Small protrusions were observed at the surface of the blends The specimen with longer immersion time (102 days) evidenced structural voids, an unquestionable sign of degradation. These results revealed also differences in the surface structure between the control and specimens with longer immersion time, evidencing the action of enzymatic solution on the material structure. The mean roughness changed from 47.2 nm for control to 384.2 nm for specimens with a longer immersion time (102 days).

### References

1. J.F. Mano, D. Koniarova, R.L. Reis, J. Mater Sci: Mater in Medicine 2003: 14: 127-135.

2. R.L. Reis and A.M. Cunha, in "Encyclopedia of Materials Science and Technology" (Elsevier Science Ltd, 2001).