

EFFECT OF CHEMICAL TREATMENT OF POLYLACTIDE ON PROTEIN ADSORPTION

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Introduction

The biomaterials surface plays a crucial role in protein adsorption, cells adhesion and biological tissue response. One of the main surface parameters is its hydrophobic/hydrophilic character. Hydrophilic properties foster cells adhesion and proliferation [1]. This feature is important for bone regeneration. On the other hand, in cardio-surgery, hydrophobic surfaces counteract platelets aggregation [2]. Protein adsorption is the first stage of the biomaterial-tissue interaction and surface properties first decide on the type of the protein attached. The composition and structure of the adsorbed protein layer is considered one of the main factors determining the type of interaction between cells and biomaterials [3]. Albumin is an inhibitor of thrombotic processes, that is why it plays a key role in the case of biomaterials intended for contact with blood [2].

In this study, chemical treatment of polylactide (PLA) was approached. It was analysed how the changes in the surface wettability affect the albumin adhesion. The impact of a PLA manufacturing method (injection-moulding and casting from different solutions) on the surface wettability was also evaluated.

Materials and Methods

Poly(lactide) (PLA) Ingeo 3051D, NatureWorks@LLC was used. Injection-moulding was performed by using the screw injection moulding machine (MULTIPLAS) at 160°C. The second group of the samples were obtained by casting from the solution of PLA in CH₂Cl₂ (POCH) in proportion 1g/50ml and the third group from the solution of PLA in 1,4-dioxane (POCH), 1g/40ml. All the samples were shaped as round plates with a diameter of 1cm. One group of the PLA samples obtained from 1,4-dioxane solution were treated in 0.1M NaOH (POCH) and the second group in 0.1M HCl (POCH) for 20 sec. Next, the samples were rinsed with distilled water. In the next step, the samples were incubated in the albumin solution (albumin from chicken egg white, Sigma Aldrich) for 20min at 37°C, 1 sample per 2ml of the albumin solution and 8ml of the culture medium (Lonza, USA). The albumin solution was prepared by mixing 1g of albumin in 10ml of water. 4 samples were tested in every group. Contact angle was measured 10 times for every sample by using DSA10-Mk2, Krüss. Standard error was applied. The albumin was identified on the basis of infrared spectroscopy in ATR technique by using the BIO-RAD FTS 3000 Excalibur Series (PIKE) spectrometer. The MIRacle ATR diamond with ZnSe optics was used.

Results and Discussion

The influence of the manufacturing method on the surface wettability was observed (TABLE 1). PLA obtained from the 1,4-dioxane solution is characterised by more hydrophobic character (contact angle 90.7°) than PLA from the CH₂Cl₂ solution or the injected PLA. The contact angle measured for the injected PLA and for the CH₂Cl₂-based PLA is 72.2° and 76.5° respectively and these values are comparable to the literature characteristic of polylactide [2]. PLA obtained from

1,4-dioxane solution was treated in HCl and NaOH and next incubated in albumin because of the hydrophobic surface which is important for cardiovascular and cardiosurgical application. The treatment in HCl increased the contact angle to 107.3°. The NaOH treatment caused only slight changes of the surface wettability that are within the limits of statistical error. The obtained results prove the albumin adhesion on all the samples (untreated and treated in HCl and NaOH). The albumin adhesion is indicated by the strong hydrophilic character of the samples. The measured values of contact angles are approximately equal to 20°. The high dispersion of results after the albumin adhesion reveals no statistically significant differences between these samples. This can be related with the uneven albumin distribution on the PLA surface. The ATR spectra show that the amount of attached protein is different for the tested samples (FIG. 1). The bands connected with amid I (1655 cm⁻¹) and amid II (1540 cm⁻¹) are the most intensive for the spectrum of PLA treated in HCl after its incubation in albumin solution. In the spectra of the untreated PLA and the PLA treated in NaOH the intensity of bands connected with albumin is low, however the presence of these bands confirms the albumin adhesion. The mechanism of albumin adhesion on PLA was analysed by Kiss et al. [2]. The other bands observed on the spectra are typical for polylactide.

TABLE 1. Contact angle of samples.

SAMPLE	CONTACT ANGLE [°]
PLA injected	72.2 ± 2.2
PLA CH ₂ Cl ₂	76.5 ± 2.7
PLA dioxane	90.7 ± 2.5
PLA _{HCl}	107.3 ± 6.3
PLA _{NaOH}	94.8 ± 3.8
PLA with albumin	24.8 ± 10.1
PLA _{HCl} with albumin	22.5 ± 4.5
PLA _{NaOH} with albumin	16.4 ± 5.1

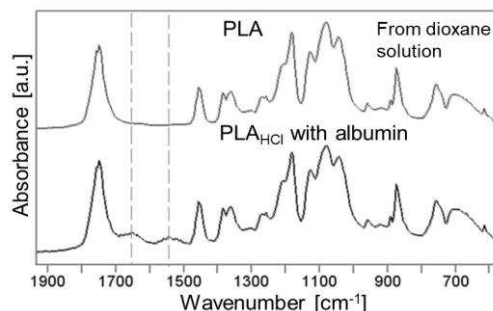


FIG. 1. ATR spectra of selected PLA samples.

Conclusions

The influence of the PLA solvent on the surface wettability was observed. The PLA dissolution in 1,4-dioxane resulted in the more hydrophobic surface. The PLA treatment in HCl increased the value of contact angle and affected the higher adhesion of albumin.

Acknowledgments

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