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Multifunctional-Dual Drug Delivery Poly-Lactic Acid Biocomposite Coating with Hydroxyapatite for Bone Implants

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Abstract. The new novel implant designs include the functionality of advanced drug delivery systems in order to improve osteointegration and to inhibit the implant-related post-operative infections. In this research, Ti6Al4V metallic implants were coated with the multifunctional-dual slow drug delivery coating which includes a polymeric matrix system based on a poly-lactic acid thin film and bioactive ceramic hydroxyapatite. This system consists of two different kinds of pharmaceuticals, which are gentamicin and simvastatin. It is reported in this paper that the simvastatin and gentamicin containing poly-lactic acid coating which was applied uniformly and successfully with and without hydroxyapatite microspheres. Most importantly, the release rate of drugs was controlled with poly-lactic acid matrix and hydroxyapatite microspheres which have different dissolution rates in order to prevent the high dosages causing adverse side-effects of drugs.

1. Introduction

The last few decades have seen the development of the multidisciplinary fields of tissue engineering including nanotechnology and regenerative medicine. The number of studies bringing together tissue engineering and nanotechnology has increased in recent years in order to develop a new generation of multifunctional implantable devices. In order to improve surface properties of dental and orthopedic implants, biocomposite-coating systems are commonly used. Therefore, next generation multifunctional implantable systems which include the functionality of drug delivery systems have advanced to increase osteointegration of bone implants, to improve bioactivity and to prevent toxic metallic ions [1-3].

Controlled drug delivery systems have very diverse applications for biomedical development. These systems can be modified with the selection of biomaterials which are used as the matrix to incorporate different types of pharmaceuticals according to the various types of treatment. Based on the type of application, the implantable drug delivery systems are designed to enhance the effectiveness of the drug by controlling long-term drug release rate while reducing or limiting negative side effects [3, 4].

Recent research in combinational drug delivery system and metallic medical implants including dental and orthopedic implants show those natural or synthetic biodegradable polymers such as collagen, poly-lactic acid (PLA) which can be used with other additives to deliver controlled dissolution rates.

The most important challenges during the surgery of bone implants are implant-related post-operative infections, lack of osteointegration and difficulty of bone regeneration after surgery [5, 6].

The design of our novel coating material in this study to address the challenges in bone implant designs includes incorporating antibiotics (gentamicin (Gm)) and simvastatin (SIM), and HAp

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microspheres within a biodegradable PLA thin film composites for metallic dental and orthopedic implants. PLA is an attractive biodegradable polymer for drug delivery system as it is non-toxic and biocompatible. Simvastatin which is a cholesterol treatment drug stimulates bone regeneration [7, 8]. Low concentration of simvastatin support bone growth however high dosage of simvastatin causes negative side-effects including bone related problems such as reducing bone growth [9].

Gentamicin is used as a systemic antibiotic therapy in orthopedics, for example in osteomyelitis treatment, but gentamicin has been reported to be ototoxic and nephrotoxic at the high concentration. Therefore, the development of a local or targeted slow drug delivery system for simvastatin and gentamicin can prove to be highly beneficial to prevent adverse side-effects or post-operative complications [5, 8, 9].

In this research, PLA-HAp containing biocomposite coating material which contain two types of drugs were applied to the metallic implants in order to increase biological activity and surface properties. Additionally, drug release rates and surface characterisation after coating were analysed, respectively.

2. Experimental

2.1 Materials

All chemicals were obtained from Sigma Aldrich, Australia. High purity Ti-Al-V discs were obtained from the Good Fellows UK.

2.2 The coating of the Ti6Al4V disc with PLA biocomposite thin films

Ti6AL4V metallic implant discs were used as a substrate in this study. The Ti6AL4V discs were cleaned with 100% acetone by ultra-sonication before the application of the coating method. The Ti6AL4V discs were coated with PLA biocomposites film which includes 30% Gm and 0.01% SIM drug particles within and without HAp microspheres. PLA biocomposites were prepared by chloroform solution in order to follow solvent casting method. The spin coating method was applied to this study. Ti6AL4V discs were rotated under vacuum at 5000rpm during the PLA biocomposite coating process.

2.3 Surface Characterisation of PLA Biocomposite Thin Films Coated Ti6Al4V Discs

Scanning electron microscope (SEM) (ZEISS Supra55VP, Zeiss, Germany) was used to observe the Ti6Al4V surface after coating at 5kV acceleration voltages. Fourier transform infrared spectroscopy (FT-IR) (Thermo Fisher Scientific, Madison USA) analysis was applied for PLA, PLA-Gm, PLA-Gm-(HAp-Gm) and PLA-Gm-SIM coated Ti6Al4V discs in order to determine the thin film characteristic between the range 3000-500 cm^{-1}.

2.4 Determining Simvastatin and Gentamicin Drug Release Rates

UV-Spectrophotometer was used to determine the concentration of gentamicin and simvastatin, separately. In order to measure the gentamicin and simvastatin release rate from the PLA biocomposite matrix on the Ti6Al4V disc, phosphate-buffered saline (PBS; 0.1M and pH 7.4) was used as releasing medium. PLA coated discs were placed into the PBS solution and then kept in a temperature controlled water bath shaker at 37C°, 100rpm. The release rate of gentamicin particles which were loaded into HAp or without HAp into the PBS solution was measured with an O-phthaldialdehyde reagent for predetermined time intervals by UV-Vis Spectrophotometer at 332nm. The preparation of O-phthaldialdehyde reagent is described within the research of Chou *et al.* in details [10]. A calibration curve for gentamicin in PBS was plotted before each measurement. Simvastatin release into PBS solution was measured with 100% ethanol. Although simvastatin is insoluble in water, it is soluble in ethanol and chloroform. Therefore, SIM-PBS solution was mixed with ethanol in a ratio of 40:60. The absorbance value of the mixture was measured at 238nm. Elghnimi *et al.*, reported the detailed standard curve method [11].

3. Results and Discussion

3.1 Surface Characterisation of PLA Biocomposite Thin Films

The surface of PLA, PLA-Gm PLA-Gm-(HAp-Gm) and PLA-Gm-SIM coated Ti6Al4V discs with and without HAp microspheres were analysed with high magnification Scanning Electron Microscopy (SEM). In Fig. 1, the images of PLA (Fig. 1-A), PLA-Gm (Fig. 1-B) PLA-Gm (Fig. 1-C) with HAp microspheres and PLA-Gm-SIM (Fig. 1-D) without HAp microspheres coated samples are illustrated. According to these results, all of the coatings were applied successfully and uniformly on the Ti6Al4V surface. While the individual Gm particles can be observed on the PLA-Gm coated sample, the more agglomeration is observed for the particles on the PLA-Gm-SIM coated samples. On the other hand, after drug loading process, the shape of Gm particles changed, and they combined with HAp particles which were used as a drug carrier in this system. Although gentamicin didn`t dissolve in the chloroform solution during the coating process, the simvastatin powder dissolved.

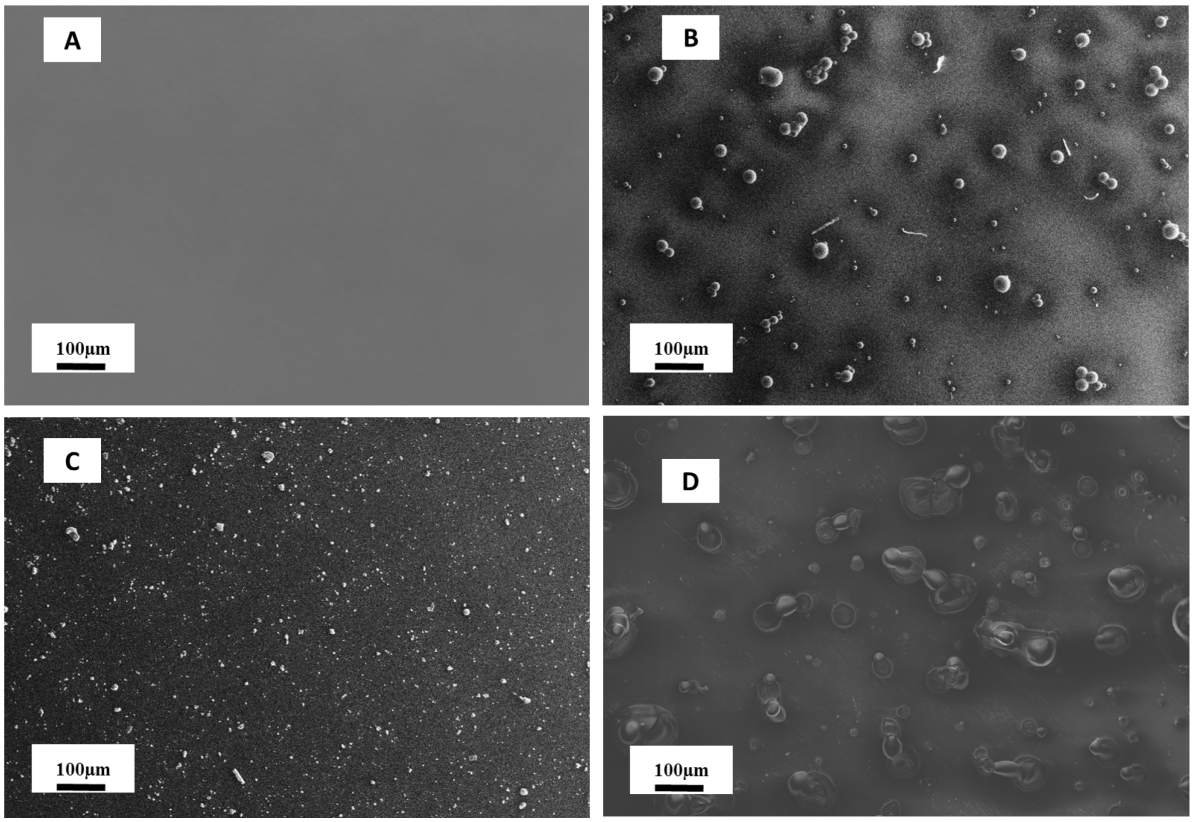


Fig.1 The surface SEM images of PLA (A), PLA-Gm (B), PLA-Gm-(HAp-Gm) (C) and PLA-Gm-SIM (D) coated Ti6Al4V samples are illustrated by high magnification.

3.2 Fourier transforms infrared Spectroscopy (FT-IR)

FT-IR spectroscopy was used to observe the chemical properties of the PLA biocomposite thin film coating materials on the Ti6Al4V samples. The FT-IR results in Fig. 2 showed that the spectra of PLA (Fig. 2-A), PLA-Gm (Fig. 2-B), PLA-Gm-SIM (Fig. 2-C) and PLA-Gm-(HAp-Gm) (Fig. 2-D) coated Ti6Al4V discs. The specific PLA peaks which are –C=O (1746 cm^{-1}), -C-H deformation (1451-1358 cm^{-1}), -C-O ester (1180-1043 cm^{-1}) and, -C-O stretch (867-751 cm^{-1}) peaks can be clearly observed for the entire samples. The characteristic peaks of gentamicin in Fig 2B-C-D are obtained Amide groups (N-H stretch) which are 1619 $cm^{-1}$and 1523 cm^{-1}. The characteristic Gm peaks were seen clearly in Fig. 2-C. At the same time, these peaks were also observed as a minor peak which refers to amide I and II groups in Fig. 2-B and D. The results are also supported by the literature [12].

On the other hand, the characteristics peaks of simvastatin are the free OH stretching vibrations (3540-3550 cm^{-1}), the C-H asymmetric stretching (2995 cm^{-1}), the - C=O (1757 cm^{-1}) stretching and 1269-1287 cm^{-1}which is due to lactone –C-O-C stretches according to the literature [13-14].

The -C=O which was also observed PLA coated sample, and lactone –C-O-C bands are observed in Fig. 2-C. However, hydroxyl and carbonyl groups exist in both the structures of the drug and the polymer. When simvastatin dissolves into the polymer solution, the drug combined with the polymer OH stretching vibration band illustrated in the spectrum which shows a broad band at 2942 cm^{-1} in Fig. 2-C. Additionally, In Fig. 2-D, PO_4 minor peaks show between 602 cm^{-1} – 478 cm^{-1} which are the characteristic peaks of HAp.

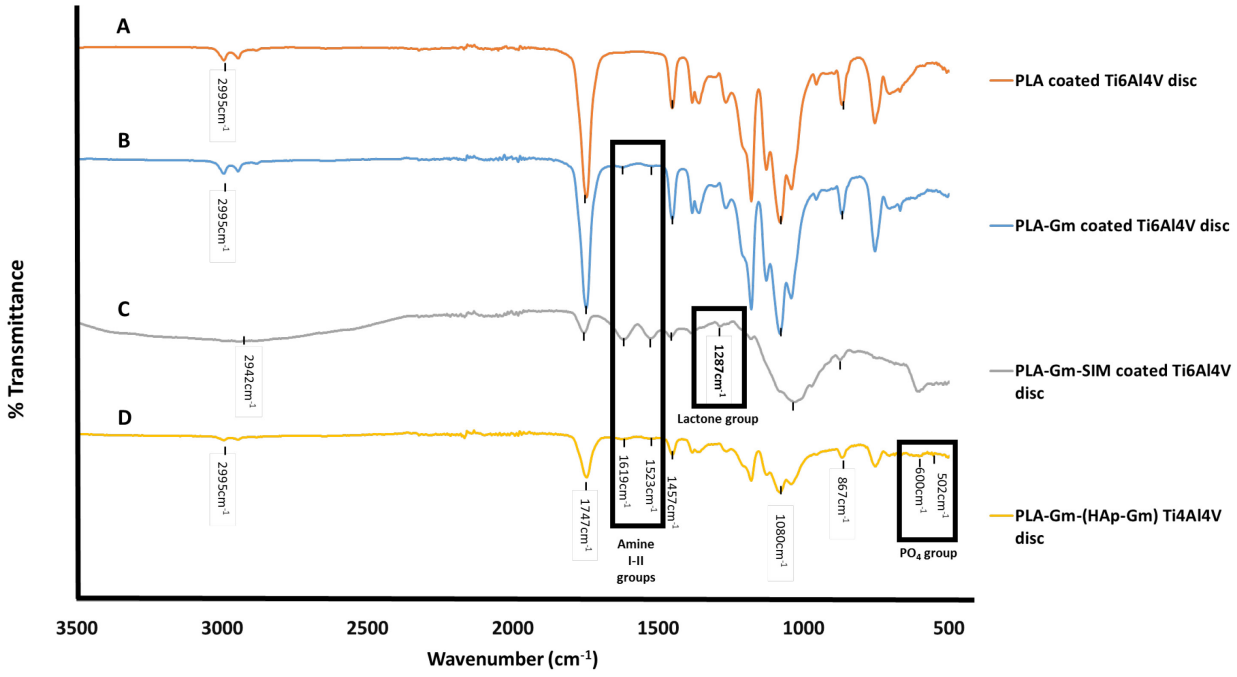


Fig.2 The FT-IR results show that the spectrums of PLA (A), PLA-Gm (B), PLA-Gm-SIM (C), PLA-Gm-(HAp-Gm) (D) coated Ti6Al4V samples with characteristic Gm (1619-1523 cm^{-1} which are amide group I and II) peaks, characteristic SIM (1287cm^{-1} which is lactone group) peaks, and characteristic HAp (600-502 cm^{-1} which are PO_4 group) peaks, respectively. Specific peaks for Gm, SIM, and HAp are labeled with boxes on the spectrum.

3.3 Determining Drug Release Rate

In this slow drug delivery system, PLA matrix controls the release of both simvastatin and gentamicin by its degradation rate through its molecular weight which can be adjusted to give a specific dissolution rate. Fig. 3-A shows the simvastatin release rate and 10% of the total simvastatin released in the first week, which refers to the initial burst release from this system. In Fig. 3-B the graph illustrates the release rate of gentamicin with and without HAp microspheres in the first week. In Figure3-B shows that the initial burst release of Gm particles was faster than the initial burst release of Gm particles with HAp microspheres which means HAp microspheres affect the drug release rate as a drug carrier and they are appropriate for the slow drug delivery system. The initial burst release suggests that the surface-bound Gm antibiotics or SIM drug particles released as part of the dissolution of the PLA coating. After the initial burst release, the second drug release profile will depend on the degradation of the PLA thin film matrix which will contribute to slow drug delivery.

This behavior is consistent with our previous studies using HAp microspheres and Gm embedded within the PLA matrix and the HAp microspheres (15, 16)

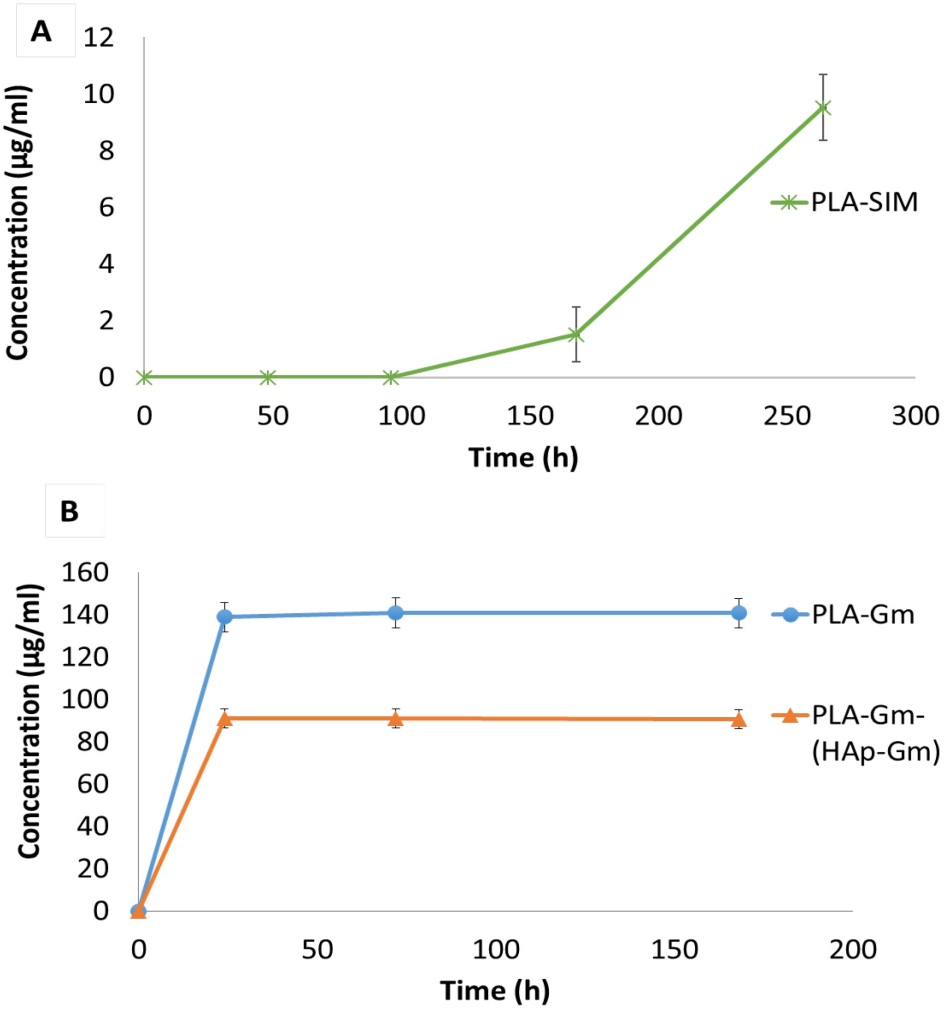


Fig.3 The drug release rate results of Simvastatin (A) and Gentamicin (B) with HAp and without HAp microspheres are given as a concentration versus time graphs for a week.

4. Conclusions

These results demonstrated that Ti6Al4V implants could be coated with biodegradable PLA-HAp and PLA only composites containing Gm and SIM which has the potential to act like an as a dual drug delivery system for the enhancement of bone implant osteointegration and against the post -operative implant infection.

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References

[1] J. Chou, T. Ito, D. Bishop, M. Otsuka, B. Ben-Nissan, B. Milthorpe, Controlled Release of Simvastatin from Biomimetic β-TCP Drug Delivery System, *Plos one*, 8 (2013).

[2] A.H. Choi, S. Cazalbou, B. Ben-Nissan, Nanobiomaterial Coatings in Dentistry, *Frontiers of oral biology*, 17 (2015) 49–61.

[3] J. Chou, J. Hao, B. Ben-Nissan, B. Milthorpe, M. Otsuka, Coral exoskeletons as a precursor material for the development of a calcium phosphate drug delivery system for bone tissue engineering, *Biological & pharmaceutical bulletin*, 36 (2013) 1662–1665.

[4] N. Rajgor, M. Patel, V.H. Bhaskar, Implantable Drug Delivery Systems: An Overview, Systematic Reviews in Pharmacy, 2 (2011) 91-95.

[5] B. Ben-Nissan, I.J. Macha, S. Cazalbou, Calcium phosphate nanocoatings and nanocomposites, part 2: thin films for slow drug delivery and osteomyelitis, Nanomedicine, 11 (2016) 531-544.

[6] I.J. Macha, B. Ben-Nissan, J. Santos, S. Cazalbou, B. Milthorpe, Hydroxyapatite/PLA Biocomposite Thin Films for Slow Drug Delivery of Antibiotics for the Treatment of Bone and Implant-Related Infections, *Key Engineering Materials*, 696 (2016) 271–276.

[7] S. Silva, M. Kaduri, M. Poleya, O. Adira, N. Kriskya, J. Shainsky-Roitmana, A. Schroedera, Biocompatibility, biodegradation and excretion of polylactic acid (PLA) in medical implants and theranostic systems, Chemical Engineering Journal 340 (2018) 9–14

[8] J. Chou, T. Ito, M. Otsuka, B. Ben-Nissan, B. Milthorpe, Simvastatin Loaded β -TCP Drug Delivery System Induces Bone Formation and Prevents Rhabdomyolysis in OVX Mice, Advanced Healthcare Materials, 2 (2013) 678-681

[9] J. Chou, T. Ito, D. Bishop, M. Otsuka, B. Ben-Nissan, B. Milthorpe, The effectiveness of the controlled release of simvastatin from b-TCP macrosphere in the treatment of OVX mice, Journal of Tissue Engineering and Regenerative Medicine, 10(2016) E195-E203

[10] J. Chou, S. Valenzuela, D.W. Green, L. Kohan, B. Milthorpe, M. Otsuka, B. Ben-Nissan, Antibiotic delivery potential of nano- and micro-porous marine structure-derived β-tricalcium phosphate spheres for medical applications, Nanomedicine, 9 (2014) 1131–1139.

[11] T.Y. Elghnimi, M.A. El-Majri, R. Bennouh, A. Enkessa, M.M. Siaan, Comparatıve *ın-vıtro* Evaluatıon of Commercıal Simvastatin Tablets (20 MG), *IJPSR,* 7(2016) 2402-2409.

[12] A. Sionkowska, B. Kaczmarek, R. Gadzala-Kopciuch, Gentamicin release from chitosan and collagen composites, Journal of Drug Delivery Science and Technology, 35 (2016) 353-359.

[13] G.M El-Maghraby, M.M. Abdelzaher, Formulation and evaluation of simvastatin buccal film, Journal of Applied Pharmaceutical Science, 5 (2015) 070-077.

[14] B. Brahmaiah, P.K. Desu, S. Nama, Sd. Khalilullah, S.S. Babu, Formulation and evaluation of extended release mucoadhesive microspheres of simvastatin, *Int J Pharm Biomed Res*, 4(2013), 57-64.

[15] I.J. Macha, S. Cazalbou, B. Ben-Nissan, B. Milthorpe, Marine Structure Derived Calcium Phosphate–Polymer Biocomposites for Local Antibiotic Delivery ,Marine Drugs, 2015, 13 (2015) 666-680.

[16] I. Karacan, I.J. Macha, G. Choi, S. Cazalbou, B. Ben-Nissan, Antibiotic Containing Poly Lactic Acid/Hydroxyapatite Biocomposite Coatings for Dental Implant Applications, Key Eng. Mater. 758 (2017) 120–125.