Surface Modification of CoCr alloy Using Vitamin-E for Cardiovascular Stents

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Statement of Purpose: Paclitaxel, sirolimus, and other "limus family" drugs are currently delivered from stents for inhibiting the growth of smooth muscle cells (SMCs) inside the arterial lumen and thereby preventing neointimal hyperplasia. However, these anti-proliferative drugs not only inhibit the growth of SMCs but also endothelial cells (ECs). The delayed or impaired re-endothelialization of stents causes a potentially fatal complication called late stent thrombosis in some patients.[1] Hence, there is a need to deliver drugs that can inhibit SMC growth and simultaneously encourage EC growth. Vitamin-E (α-tocopherol, α-TP) has been shown to inhibit SMC growth as well as promote EC growth when the drug was directly added to the cell cultures.[2] In this study, α-TP was directly coated on CoCr and the in vitro drug release was investigated for potential use in drug-eluting stents. The interaction of blood platelets with α-TP coated CoCr was also investigated in this study.

Methods: A solution of α-TP was prepared in chloroform at a concentration of 5 mg/mL. A 60 µL aliquot of the prepared α-TP solution was microdrop deposited on chemically cleaned CoCr alloy specimens (1 cm x 1 cm). The specimens were then dried under vacuum (-20 mmHg) for 48 hours at room temperature to obtain a thin α-TP film on CoCr. The specimens were surface characterized using contact angle, SEM, FTIR and AFM for studying the surface wettability, morphology, chemical composition and topography of the α-TP film on CoCr. For drug release studies, the drug coated specimens were immersed in PBS/Tween-20 for up to 28 days. The PBS/T-20 samples were collected at different time points to determine the amount of α-TP released using a HPLC. For platelet interaction studies, the blood was collected from healthy donors and the platelet rich plasma (PRP) was isolated using a standard procedure.[3] The platelets were then collected from the PRP and resuspended in Tyrode's solution. A 60 µL of platelet suspension was microdeposited on control CoCr and α-TP coated CoCr and incubated at 37 ºC for 1 hr. After that, the suspension was removed and used for the activation study while the platelets adhered on the specimens were used for the adhesion study. The platelet activation was quantified by flow cytometry using an antibody against P-selectin. The platelet adhesion was measured by quantifying the total amount of protein released by disrupting the adhered platelets by sodium dodecyl sulfate buffer. The protein released from the platelets was quantified by the bicinchoninic acid (BCA) protein assay.

Results: SEM showed a thin and transparent α-TP film was formed on CoCr (Fig 1a,b). AFM showed the 3D topography of α-TP on CoCr (Fig 1c,d). A sustained α-TP release was obtained from CoCr (Fig 2). The α-TP coating significantly reduced platelet activation on CoCr (Fig 3). The arrows in Fig 4A indicate multilayered aggregated platelets on controls while the spherical shaped unactivated platelets were seen on α-TP coatings.

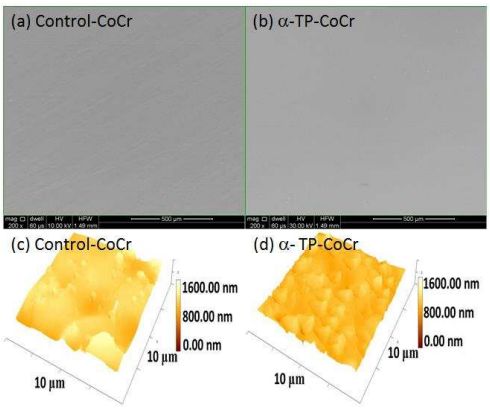

Fig 1. SEM (a,b) and AFM (c,d) images of control CoCr and α-TP coated CoCr.

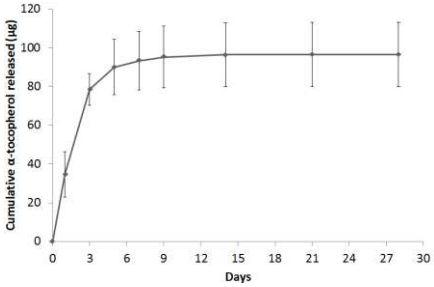

Fig 2. Cumulative α-TP release from CoCr alloy.

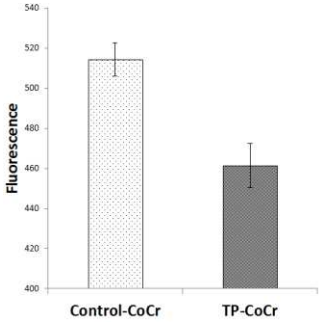

Fig 3. Platelet activation on control and TP coated CoCr.

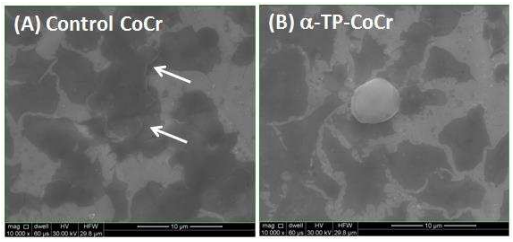

Fig 4. SEM images of platelets adhered on control and TP coated CoCr.

Conclusions: This study demonstrated that α-TP can be coated on CoCr and released for a period of time. Also, α-TP coating reduces platelet activation. Hence, α-TP is a promising drug for use in drug-eluting stents for reducing late stent thrombosis.

References: (1) Biomaterials 2007, 28, 1689-1710; (2) Eur J Nutr 2002, 41, 27-34; (3) JBMR 1998, 41, 304-311.