Inhibition of Smooth Muscle Cell Growth by Paclitaxel Releasing Polyethylene Oxide films for Drug-Coated Balloons

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Statement of Purpose: Drug-coated balloons (DCB) are commonly used for treating peripheral vascular disease (PVD).[1] However, a significant amount of drug is lost from the balloons before it reaches the target site.[2] This results in the delivery of inadequate amount of drug at the diseased site. Hence, there is a great need to control the delivery of drugs from balloons in such a way that no or minimal drug should be lost during its transit but then all the drug should be delivered at the diseased site within a few minutes. Also, since the time required to advance the balloon to different arteries in the leg (iliac, femoral, popliteal, and tibial) is expected to be different, there is a need to tailor the drug release depending on the arterial location. In this study, we have developed polyethylene oxide (PEO) films at different wt/vol% to tailor the release of paclitaxel (PAT). The PAT loaded PEO films developed in this study were investigated for their drug release as well as for their inhibitory effect on smooth muscle cell growth.

Methods: Four different groups of PAT loaded PEO (PAT-PEO) films were prepared using a solvent casting method. PAT-PEO films were prepared with 10, 15, 20, and 25 w/v% of PEO that contained 1.5, 1, 0.75, and 0.6 wt% of PAT, respectively. Initially, the PEO was stirred in deionized water for 6 hrs. After that, a PAT solution in ethanol was added to the PEO solution and allowed to stir for 18 hrs. The PAT-PEO solution was then poured into a petridish and dried in an oven for 48 hrs. Control PEO films without PAT were also prepared. All PAT-PEO and control-PEO films prepared were characterized using SEM, FTIR, DSC, and tensile tester for studying the morphology, chemical composition, thermal properties, and mechanical properties, respectively. For the drug release studies, the PAT-PEO films were immersed in PBS/T-20 (pH 7.4) at 37 °C. The PBS/T-20 samples collected at 30 sec, 1 min, 2 min, 3 min, 5 min, and 7 min were analyzed for the quantity of PAT released using a HPLC. A density of 15 x 10^3 human aortic smooth muscle cells (SMCs) was seeded in a well plate. After 24 hrs, the films were added to the cells and the growth of cells was measured using a resazurin fluorometric assay at days 1, 3, and 5. The morphology of the cells was investigated by staining the cells with fluorescein diacetate and imaged using a fluorescence microscope. A one way ANOVA was used to determine statistical significance at $p < 0.05$.

Results: SEM images showed that PAT-PEO films were free of any drug physically present on the surfaces (Fig 1). FTIR spectra also showed that there was no drug present on the film surfaces. These results suggested that PAT was incorporated well inside the PEO films. Also, the cross-sectional SEM images did not show the presence of drug crystals inside the films. DSC spectra suggested that the PAT was molecularly dispersed in the PEO films. Drug release studies showed that only <5% of drug was released from PAT-PEO-10% by 1 min (Fig 2). However, >90% of drug was released between 1-2 min. Similarly, >90% of drug was released only between 2-3 min, 3-5 min, and 5-7 min from PAT-PEO-15%, PAT-PEO-20%, and PAT-PEO-25%, respectively (Fig 2). Thus, these results demonstrated that PAT-PEO films can be tailored to prevent initial loss of the drug and then deliver it at required time points for artery-location specific DCB applications. Both PAT-PEO and control PEO films significantly inhibited the growth of SMCs (Fig 3). The inhibitory effect of PAT-PEO was significantly greater than that of control PEO films. FM images showed characteristic spindle shaped SMCs with well spread morphology in the control well while the cells treated with control PEO and PAT-PEO films showed a poorly spread discoid shaped morphology.

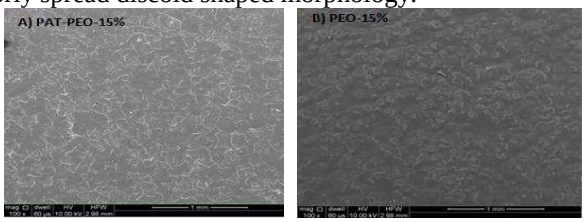

Fig 1. SEM images of PAT-PEO-15% (A) and control-PEO-15% film surfaces.

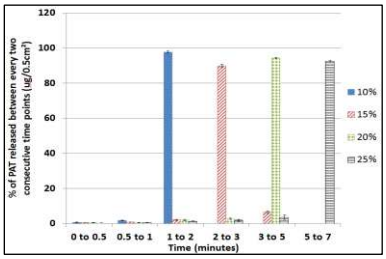

Fig 2. Percentage of PAT released from four different groups of PAT-PEO films.

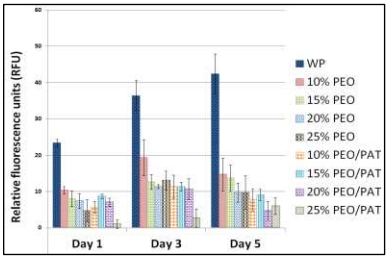

Fig 3. SMC viability and proliferation for different groups of PAT-PEO and control PEO films.

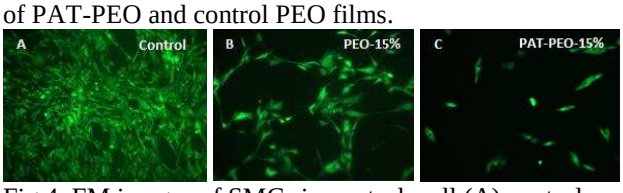

Fig 4. FM images of SMCs in control well (A) control-PEO-15% (B) and PAT-PEO-15% (C) films after 5 days.

Conclusions: This study demonstrated that PAT-PEO films can be tailored to deliver drugs at different time points and to successfully inhibit SMC growth for drug-coated balloon applications.

References: (1) Circulation 2009; 2: 352-358. (2) Mat. Sci. and Eng. 2013; 33: 4244-4250.