

## Degradable Devices for Localized Delivery of Immunosuppressants to Prevent Transplant Rejection

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**Statement of Purpose:** Advances in surgical techniques and immunology have enabled transplantation of composite tissue allografts (CTAs, e.g., upper extremity, face, genitourinary tissues) to treat complex soft tissue injuries and extremity amputations. However, >85% of transplant recipients develop at least 1 episode of rejection within the first year, and >50% experience multiple such episodes. Systemic distribution of orally or intravenously delivered immunosuppressants increases the likelihood of widespread side effects. In addition, the effectiveness of oral administration is dependent on patient compliance. To address these limitations, we have developed a polymeric local drug delivery system using a biodegradable, bioresorbable tyrosine-derived polycarbonate (TyrPC) [1]. The devices are polymeric thin films loaded with the immunosuppressant tacrolimus (FK506). These devices are designed for implantation at the interface between host and donor tissue during transplant surgery to provide long-term, localized immunosuppression and reduce the incidence of rejection episodes.

**Methods:** A TyrPC known as E1218(1k) was chosen as the polymer based on prior release studies. E1218(1k), tacrolimus (LC Laboratories, Woburn, MA) and vitamin E (Sigma-Aldrich, Allentown, PA) were dissolved in 10 ml DCM, poured into a Teflon dish, and dried. Solvent-cast films were stacked, compression molded to the desired thickness and cut to desired dimensions. The final devices were sterilized by gamma irradiation at 22 kGy. *In vitro* drug release was monitored under sink conditions in PBS by HPLC. *In vivo* implants were placed subcutaneously in rats to measure systemic and localized release. Four non-human primates were given CTAs along with drug-loaded devices implanted at the site of surgery to investigate localized and systemic release as well as overall device effectiveness. Tacrolimus blood concentration was measured using an ARCHITECT tacrolimus assay. Tacrolimus tissue concentrations were measured by solvent extracting drug from tissue biopsies and analyzing the extract by LC-MS/MS. CTAs were observed for evidence of rejection.

**Results:** Adding vitamin E effectively stabilized the formulation by acting as a free-radical scavenger, thus preventing molecular weight loss and drug degradation during processing and gamma radiation sterilization. *In vitro* release showed a distinct three-stage elution profile with high release occurring over the first three days (>50  $\mu\text{g}/\text{cm}^2/\text{day}$ ), moderate release occurring over days 4-30 (20-40  $\mu\text{g}/\text{cm}^2/\text{day}$ ) and a consistent slow release beyond day 30 (5-10  $\mu\text{g}/\text{cm}^2/\text{day}$ ) (Fig 1). Laminating the devices with a layer of drug free polymer reduced the

initial burst release, which helps prevent drug-related toxicity while leaving extended release rates unchanged.

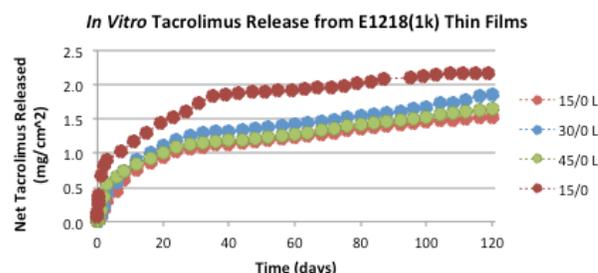


Figure 1. *In vitro* net tacrolimus release from 15%, 30% and 45% drug-loaded devices.

Tissue explants at two and three weeks from the regions surrounding subcutaneous implants in rats showed elevated local tacrolimus concentrations at least 1.5 cm from the device. This supports the hypothesis that these devices may provide localized immunosuppression. Systemic tacrolimus levels in both rats and non-human primates with implants placed subcutaneously showed systemic tacrolimus levels within the standard therapeutic range of 5-15 ng/ml for more than one week (Fig. 2). This resulted in non-human primate CTAs surviving more than one week without additional immunosuppressant injections.

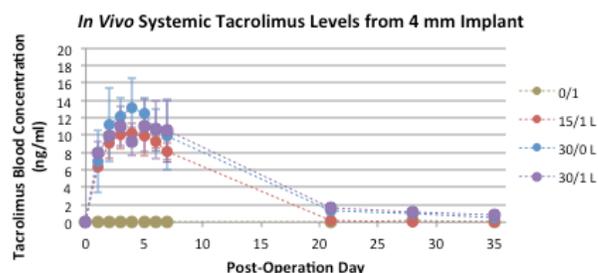


Figure 2. *In vivo* systemic tacrolimus concentrations following implantation of single 4 mm, 15% or 30% drug-loaded laminated devices in rats.

**Conclusions:** TyrPC devices loaded with tacrolimus were successfully fabricated, sterilized, characterized, and implanted into animal models. Target drug concentrations were reached systemically for one week *in vivo* while localized tissue tacrolimus levels remained measureable at three weeks post-implantation. Together, these results indicate that the devices provide systemic immunosuppression initially and localized long-term. Future work will adjust device formulation and implant number to extend the therapy duration and study the extent of localized immunosuppression achieved.

**References:** [1] Kohn, JK. *Adv Drug Delivery Reviews*. 2003 Apr; 55(4): 447-466