Sequential drug delivery - *in vitro* and *in vivo*

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Statement of Purpose

A bioerodible drug delivery device has been developed with the capability of releasing four different drugs in a specific temporal sequence. Although adaptable to other conditions, the system is aimed at treating periodontitis and the related condition of peri-implantitis, which are oral diseases marked by bacterial infection, inflammation, and tissue loss. An ideal treatment to reverse this chronic condition would require multiple drugs delivered in the appropriate order. The present multi-drug delivery system is based on a cellulose acetate phthalate - Pluronic F-127 (CAPP) association polymer fabricated in the form of multilayered films loaded with antibiotic, anti-inflammatory, anti-resorptive drugs sequentially for treatment of infection, inflammation, and bone resorption stages of periodontitis and finally the release of an osteogenic agent to aid regeneration of bone. After extensive characterization of *in vitro* drug release from this device, *in vivo* drug release is being studied by supracalvarial implantation of the device in a rat model.

Methods

Metronidazole (antibiotic), ketoprofen (anti-inflammatory), doxycycline (anti-matrix destruction) and simvastatin (osteogenic) were loaded in CAPP films formed using the solvent evaporation technique. Films were made with a CAP:Pluronic ratio of either 70:30 and 90:10. Multilayered device were fabricated by alternating drug-loaded and blank films to achieve sequential release of different drugs in a particular order. Devices were also fabricated with thin layers of poly(sebacic acid) (PSA) between the blank layers to further slow erosion of the device. A PSA and poly(lactic acid) (PLA) cup-shaped coating was cast over the multilayered films to enable unidirectional degradation and drug release. The multilayered devices were eroded in phosphate-buffered saline (PBS) at 37°C. Aliquots of supernatant were collected and replaced with fresh PBS at regular time intervals. Drug release was monitored by UV spectroscopy and HPLC. The 70:30 CAPP devices without PSA blank layers were implanted under the periosteum of rat calvaria as a part of pilot animal studies. The animals were euthanized at particular time points to retrieve the devices and analyze the remaining amount of drug for reconstructing the release profile *in vivo*.

Results and Discussion

Both the multilayered 70:30 CAPP devices and 90:10 CAPP devices with PSA layers showed sequential release of the four drugs (Figure 1). The total erosion time of 90:10 PSA CAPP device extended up to 320 hours, thereby decreasing the erosion rate without compromising the sequential release pattern. The pilot *in vivo* study also showed a sequential release pattern (Figure 2). Device retrieved on day 1 showed that the metronidazole was completely released and 13% of ketoprofen was still present. Till day 5 more than 90% of doxycycline and simvastatin were measured, indicating that only significant release of metronidazole and ketoprofen had occurred till that point of time. On day 7 only 4% of doxycycline and 89% of simvastatin was present in the retrieved device, and by day 9 all the CAPP layers had been eroded and there was no presence of drugs in the retrieved device. More *in vivo* studies are being conducted to further characterize the *in vivo* drug release.

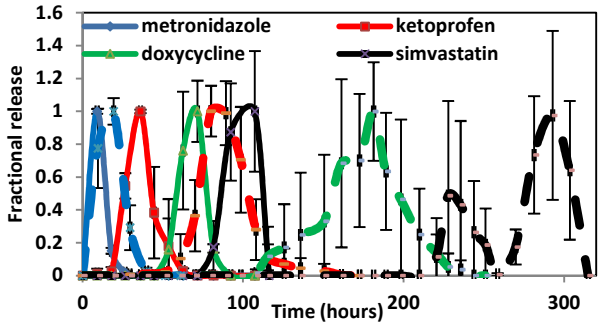

Figure 1. Sequential release of four drugs from multilayered 70:30 CAPP devices (——) and 90:10 CAPP device with PSA blank layers (- - -). Data are mean ± standard deviation (n=3).

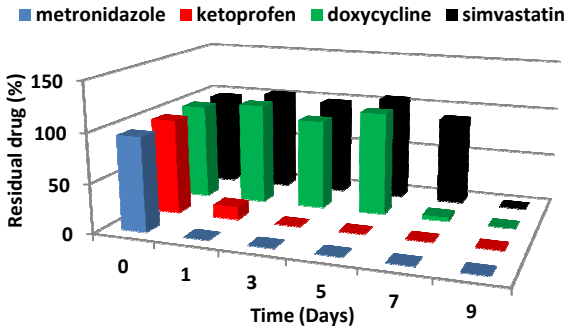

Figure 2. Residual drug content in multilayered 70:30 CAPP devices retrieved from the animals.

Conclusion

The bioerodible CAPP devices released four different drugs in a temporally separated sequence. Furthermore, release kinetics can be adjusted, such as by changing the ratio of CAP to Pluronic and/or inserting blank PSA layers. Results from the pilot animal studies confirmed the sequential release pattern *in vivo*.

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