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Comparison of Sequential Drug Release *in Vitro* and *in Vivo*

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Abstract

Development of drug delivery devices typically involves characterizing *in vitro* release performance with the inherent assumption that this will closely approximate *in vivo* performance. Yet, as delivery devices become more complex, for instance with a sequential drug release pattern, it is important to confirm that *in vivo* properties correlate with the expected “programming” achieved *in vitro*. In this work, a systematic comparison between *in vitro* and *in vivo* biomaterial erosion and sequential release was performed for a multilayered association polymer system comprising cellulose acetate phthalate and Pluronic F-127. After assessing the materials during incubation in phosphate-buffered saline, devices were implanted supracalvarially in rats. Devices with two different doses and with different erosion rates were harvested at increasing times post-implantation, and the *in vivo* thickness loss, mass loss, and the drug release profiles were compared with their *in vitro* counterparts. The sequential release of four different drugs observed *in vitro* was successfully translated to *in vivo* conditions. Results suggest, however, that the total erosion time of the devices was longer and release rates of the four drugs were different, with drugs initially released more quickly and then more slowly *in vivo*. Whereas many comparative studies of *in vitro* and *in vivo* drug release from biodegradable polymers involved a single drug, the present research demonstrated that sequential release of four drugs can be maintained following implantation.

Keywords

in vitro drug release; *in vivo* drug release; sequential drug delivery; cellulose acetate phthalate; Pluronic F-127

INTRODUCTION

A large number of publications describe the release of drug molecules from polymeric devices (*e.g.*, reviewed in [1–3]). Most of these studies have been logically aimed at determining effectiveness of the drug released for treating a particular condition or

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regeneration of a tissue. While the transition of a polymer-based drug delivery device from *in vitro* investigations to an *in vivo* setting is an important step in determining the designed device properties and performance, there are comparatively fewer reports on *in vivo* biomaterial degradation/erosion and drug release profiles from advanced devices [4–13].

Due to the dynamic physiological environment into which a device will be implanted, the material would be exposed to different cell types and numerous biomolecules [14], which can have considerable impact on the degradation and release kinetics. For instance, poly(glycolic acid) and poly(lactic acid) [15, 16] have been shown to possess different degradation rates *in vivo* when compared to *in vitro* conditions [10, 14, 17–20], ultimately resulting in altered drug release profiles. Understanding *in vivo* drug release will provide useful information by which delivery can be tailored more specifically for its intended purpose in disease treatment or tissue regeneration.

The surface-eroding association polymer system composed of cellulose acetate phthalate and Pluronic F-127 was previously designed for localized delivery of four drugs in a particular sequence for the directed resolution of chronic soft tissue disease (e.g., periodontitis) in a step-by-step manner [21]. The objective of the present studies was to quantitatively compare *in vivo* biomaterial erosion and sequential release from the cellulose acetate phthalate - Pluronic PF-127 (CAPP) association polymer system with the “programming” determined *in vitro*.

MATERIALS AND METHODS

Device fabrication

CAPP films were prepared by solvent evaporation after mixing cellulose acetate phthalate (Sigma-Aldrich, St. Louis, MO) and Pluronic F-127 (Sigma-Aldrich) in the weight ratio of 90:10, respectively. The mixture was dissolved in acetone to obtain an 8% polymer solution followed by the addition and dissolution of the drug of interest at either 1 wt.% (low dose) or 5 wt. % (high dose). The drug-polymer solution was poured into a Teflon dish and stored at 4°C for 24 hours for slow evaporation of the solvent. Blank CAPP films were prepared in the same way but without the addition of drugs. For the present study, CAPP films were loaded with metronidazole (Sigma-Aldrich), ketoprofen (Sigma-Aldrich), doxycycline (Sigma-Aldrich), or simvastatin (Haorui Pharma-Chem, Inc., Edison, NJ). Drug-loaded discs with a diameter of 6 mm and thickness of 0.5 mm were punched out of the films and arranged in the desired sequence with two blank discs between each drug layer. The stack of the CAPP films was bonded together by application of acetone between the layers. The multilayered device was then coated with poly(sebacic acid) (diacetoxo-terminated, PSA; Sigma-Aldrich), which acted as a barrier to enable unidirectional erosion and drug release. The completed device was wound with 4-0 poly(glycolic acid) suture (Oasis, Mettawa, IL) for circumferential reinforcement followed by compression at 1,500 N using a BOSE ELF 3300 mechanical testing system to consolidate the layers and reduce the overall thickness of the device. Blank (drug-free) multilayered devices were used for comparison. To increase the duration of erosion and release, a second type of device was fabricated with a thin PSA layer included between the two blank layers. Figure 1 shows a schematic representation of the process involved in fabrication of the two types of multilayered CAPP devices. For *in vivo*

studies, the CAPP films and PSA were UV-sterilized for 1.5 hours, the solvents sterile-filtered, and assembly conducted in a laminar flow hood. Overall, four different types of devices were fabricated: 1) blank (B), 2) high drug dose (HD), 3) low drug dose (LD), and 4) low drug dose with PSA blank layers for slower erosion (LDS). The final dimensions were cylinders of 6 mm diameter and height ranging from 5 to 8 mm, depending on the particular type of device (*i.e.*, number of blank layers).

***In vitro* studies**

The multilayered CAPP devices were eroded in 4 mL of phosphate-buffered saline (PBS), pH 7.4, during incubation at 37°C with gentle shaking. At regular time intervals, the entire supernatant was collected, the mass of the samples measured, and 4 mL of fresh PBS added. The amount of drug in the supernatants was determined by high performance liquid chromatography (HPLC) using a Hitachi Primaide system equipped with a Kinetix C18 column (Phenomenex, Torrance, CA). For measuring the concentration of ketoprofen, an isocratic mobile phase of acetonitrile (ACN; 60%) and 0.1% trifluoroacetic acid (TFA; 40%) was used with UV detection at 260 nm, and for simvastatin, the isocratic mobile phase was acetonitrile (70%) and 0.1% TFA (30%) with detection at 240 nm. A gradient protocol was developed for measuring metronidazole and doxycycline. After initially maintaining the mobile phase at 100% 0.1% TFA for 3 min, ACN was introduced at 15%/min up to 70% 0.1% TFA : 30% ACN. After 3.5 min, TFA was increased at 20%/min back to 100% 0.1% TFA and maintained for 3 min. Metronidazole and doxycycline were measured at 318 and 350 nm, respectively.

***In vivo* studies**

All animal studies were conducted at the University of Kentucky in accordance with a protocol approved by the Institutional Animal Care and Use Committee (IACUC). Male Sprague-Dawley rats between the ages of 6 and 8 weeks were used. As described under Device fabrication, four different types of devices were implanted: 1) B, 2) HD, 3) LD, and 4) LDS. After anesthetization and skin preparation, a transverse incision was made between the ears of the animal, and the periosteum was slightly elevated to expose the calvaria. CAPP devices were placed with the drug-releasing side facing the bone, and the incision was sutured (Figure 2). Only one device was implanted in each rat. Animals were euthanized at specific time points (Table I) based on pilot studies evaluating the time course of polymer erosion, and the devices were retrieved for analysis. Any clots or tissue adherent to the exterior surfaces was gently removed with forceps and gauze, and tissue found within the well/cup-shaped was also partially removed while ensuring that the remaining drug loaded layers were not disturbed. The implants were then cross-sectioned using a razor blade and imaged for morphological analysis before measuring residual mass and thickness of the remaining CAPP layers. The retrieved devices were then completely dissolved in PBS, and the solution containing dissolved drugs and CAPP was filtered (0.45 μ m) before analysis using the same HPLC methods as described in the *in vitro* studies section. Because the subcutaneous nature of the implant site allowed exposure of a large area of soft and hard tissue to the drugs, the extractable amounts of drugs in the tissues were below the limits of detection. Consequently, *in vivo* release profiles were constructed by measuring the amounts of drugs remaining in the retrieved devices.

Statistical analysis

Data are plotted as mean $\pm$ standard deviation for $n = 3$. Slopes of the *in vitro* and *in vivo* mass loss and thickness loss profiles were statistically compared by analysis of covariance using Graphpad Prism software. Differences were considered significant when $p < 0.05$.

RESULTS

In vitro mass loss

When incubated in PBS, LD CAPP devices with either two blank layers or with a PSA layer between the two blank layers followed linear erosion profiles (Figure 3). The slopes of these mass loss curves, which represent erosion rate, were significantly different ($p < 0.0001$). Mass loss occurred at a faster rate for devices with just two blank layers when compared with devices also having the PSA layer. The 90:10 CAPP layers eroded at a rate of $0.04 \text{ mg/mm}^2/\text{hour}$ when sink conditions were maintained by replacing PBS at regular time intervals.

In vitro drug release

Figure 4A shows the instantaneous release profiles for four drugs released from the multilayered delivery system with two blank layers between each drug layer (LD devices). The sequence of drug release followed the order of loading in the surface eroding system. Metronidazole (in the outermost layer) was released during the first 30–40 hours of incubation in PBS, followed by ketoprofen (40–90 hours), doxycycline (70–150 hours) and ending with simvastatin (125–240 hours).

The use of the PSA between the blank CAPP layers further increased the erosion time of LDS devices to 300 hours (Figure 4B). Release of metronidazole occurred within the first 40 hours, as in the case of LD devices, but the presence of PSA layers before the other drug layers slowed erosion and extended the release periods of other three drugs. Release of ketoprofen occurred through 125–135 hours, which was close to half of the total erosion time of the device. Release of the third drug, doxycycline, started at around 115 hours and lasted until 235 hours, and this was followed by simvastatin from 220 hours through complete erosion of the devices at around 300 hours. The times at which the peaks were observed *in vitro* for all four drugs in the two different device types are summarized in Table II.

In vivo thickness and mass loss

Qualitative, visual observations showed the presence of fibrous tissue associated with the bone side of implants retrieved at later time points (not shown). Cross-sections of retrieved devices from all three groups (HD, LD, and LDS) showed a gradual reduction in thickness of the CAPP layers (Figure 5). The HD and LD groups with two blank layers followed a similar decrease in thickness, with almost all the CAPP eroded by day 10. The LDS group with PSA eroded at a slower rate and had residual CAPP layers even at day 18. Figure 6A shows the thickness loss profiles over the course of study. The HD and LD devices followed a biphasic pattern with thickness loss occurring at a faster rate until day three or four (approximately spanning loss of the first four layers, which include metronidazole and ketoprofen) followed by a slower rate until complete erosion by day 10. There was no

statistically significant difference between the thickness loss profiles for the HD and LD groups. The LDS group followed a similar biphasic pattern, but at a decreased rate that resulted in only 80% of thickness loss occurring by day 18. The slope of the thickness loss profile for the LDS devices was significantly different from those for devices with just two blank layers ($p=0.04$).

The mass of devices measured after retrieval was used to construct the mass loss profiles shown in Figure 6B. The HD and LD groups followed similar mass loss profiles, with no significant difference between the slopes of the curves. The LDS devices exhibited a similar mass loss profile but with a slower rate that was significantly different from that for devices with two blank layers ($p=0.002$).

***In vivo* drug release**

HPLC measurement of the amount of drug present in the retrieved devices was used to construct *in vivo* drug release profiles. For HD devices (Figure 7A), metronidazole was essentially gone at 12 hours, but 100% of the other three drugs was present. After one day of implantation, about 50% of ketoprofen remained and nearly 100% of doxycycline and simvastatin. There was no significant amount of ketoprofen observed after the second time point. Doxycycline was observed until day 4. Approximately 100% of the simvastatin was detected until the final stages of the study (day 9–10). In the case of LD devices (Figure 7B), a similar release pattern was observed, with metronidazole completely released by first time point (24 hours) and nearly 100% of the other three drugs still present. Implants retrieved at day 2 showed retention of 100% doxycycline and simvastatin, with ketoprofen almost completely released. There were traces of doxycycline observed in the remaining time points, with simvastatin measured until the last time point.

LDS devices followed a similar sequential release pattern but at an overall slower rate (Figure 7C). While metronidazole was completely released within 24 hours, ketoprofen was observed at trace amounts through day 5. At day 12, nearly 100% of doxycycline was measured in the retrieved devices, and at day 18 (the last time point), approximately 50% of the doxycycline was observed. With the devices not being completely eroded by day 18, almost 100% of simvastatin was observed in all the retrieved devices. Release of the last drug (simvastatin) was observed to be more delayed in all the three types of devices when compared with the other three drugs released. Table III gives an overview of the time points through which the specific drugs were observed *in vivo* for the different types of devices.

Comparison of *in vitro* and *in vivo* erosion and drug release

Figure 8A compares *in vitro* and *in vivo* release profiles for LD devices with two blank CAPP layers. For these faster-eroding devices, metronidazole release (from the outermost drug layer) was completed within the first hours 30 *in vitro* and by 12–24 hours *in vivo*. Ketoprofen release *in vivo* occurred faster than was observed *in vitro*, but in both the cases, release was complete within 90–100 hours. For the third drug, doxycycline, *in vivo* release occurred at a slightly slower rate when compared to that measured *in vitro*. Doxycycline was completely released by 150 hours *in vitro*, compared to release of 80–90% at this time *in vivo*. Simvastatin release (from the last layer) *in vivo* was much slower than under *in vitro*

conditions. By 240 hours, simvastatin was completely released *in vitro*, but drug-loaded layers remained in the devices retrieved at 10 days of implantation.

Figure 8B compares *in vitro* and *in vivo* cumulative release profiles for LDS devices with PSA layers that slowed erosion. Similar to the faster-eroding devices, metronidazole release occurred within 24 hours. Release of ketoprofen again occurred faster *in vivo* when compared to *in vitro*. Whereas ketoprofen release was complete in 150 hours *in vitro*, approximately 90% of the drug was released by 72 hours *in vivo*. Similar to the faster-eroding devices, doxycycline release *in vivo* was slower than its *in vitro* release. Only 53% of doxycycline was released *in vivo* by 288 hours compared to complete release *in vitro* by 240 hours. Simvastatin release from slow-eroding devices *in vitro* was completed by 300 hours, but all devices retrieved at day 18 (last time point) had complete simvastatin layers still present, indicating that simvastatin release had not yet started before 432 hours. Table III summarizes the temporal differences between the faster- and slower-eroding devices *in vitro* and *in vivo* based on the time points through which release of a particular type was measured.

Figure 9 directly compares *in vitro* mass loss and *in vivo* thickness loss for the fast- and slow-eroding devices. *In vivo* thickness loss was used instead of mass loss because blood and tissue adherent to the retrieved devices made *in vivo* mass measurements inaccurate. If polymer erosion was the same under both conditions, the data points would follow a line at 45°, but this was not observed. These plots show an increased erosion rate during the initial stages and relatively slower erosion rate that was observed during the final stages for both types of devices *in vivo*.

DISCUSSION

The main aim of this research was to investigate the performance of the CAPP association polymer system under *in vivo* conditions and relate the findings to what occurs during standard *in vitro* release experiments. Multilayered devices based on the 70:30 CAPP composition were previously shown to deliver molecules with different properties (*e.g.*, solubility) in a zero-order fashion [21, 22]. Furthermore, a predetermined sequence of release was shown *in vitro*, and there was no diffusion of drugs between the layers of CAPP even when incubated at 37°C [22]. In this particular study, the system was modified to have a ratio of 90:10 to achieve longer erosion times, which was verified *in vitro* as well as in pilot *in vivo* studies (data not shown). Based on those pilot *in vivo* results, multilayered devices were fabricated with 90:10 CAPP films for a larger scale study to compare *in vitro* and *in vivo* material responses and drug release.

In addition to the primary component of CAPP, the surface-eroding polyanhydride PSA was a part of the delivery system [23]. PSA was used as an external barrier in all devices and as an internal blank layer in the LDS devices. The rate of erosion of PSA is slower than that of CAPP, making it an appropriate material for a barrier layer to achieve unidirectional erosion. Furthermore, the thin (approximately 300 µm) intermediate layers between the blank CAPP layers further decreased the overall rate of mass loss, thereby increasing the total erosion time of the device with only a slight increase in total thickness of the device (< 1 mm) when

compared to the devices with just two blank CAPP layers. The adjustable nature of the delivery system with respect to the erosion time creates the possibility of designing devices based on specific disease conditions.

***In vitro* mass loss and drug release**

In vitro, LD and HD devices showed sequential release of the four drugs metronidazole, ketoprofen, doxycycline, and simvastatin. As intended, LDS devices also showed sequential release of the drugs, with better distinction between the drug release peaks compared to devices without PSA layers. The final drug released from the device, simvastatin occurred at an even slower rate when compared to the other drugs. Mixing of the slower-degrading PSA with CAPP during attachment of the final simvastatin-loaded CAPP layer to the PSA barrier likely affected the erosion rate of this particular layer, which resulted in the less uniform and slower release of simvastatin compared to the other drugs. The hydrophobic nature of simvastatin contributed to variability in measuring this drug. This can be resolved in future device fabrication by incorporating a thin blank CAPP layer between the simvastatin layer and PSA barrier. The simvastatin and doxycycline release peaks were also broader, which could be because of the cylindrical, well-shaped design of the PSA barrier leading to retention of PBS within the shell as the initial CAPP layers eroded. The resulting reduced circulation of fluid near the final drug layers would lead to slower erosion of polymer and release of drugs.

***In vivo* thickness loss, mass loss, and drug release**

The bottom CAPP layers were seen at day 18 in LDS devices compared to near complete erosion in LD and HD devices by day 10. This observation shows that the PSA-containing devices retained their slower-eroding properties *in vivo*, as it was previously demonstrated *in vitro*. Quantitatively, the loss of thickness followed a pattern comparable to that seen qualitatively in the cross-sectional images. The biphasic thickness loss patterns observed for both the LD and HD devices were not significantly different, indicating that the amount of drug loaded in the device did not affect the erosion rate. Thus, dosing can be adjusted for different future applications. LDS devices also followed a biphasic thickness loss pattern but at a slower rate, demonstrating the ability of the PSA layers to slow erosion without drastically altering the pattern. Even though the presence of blood and tissue attached to the remaining CAPP layers and PSA barrier of retrieved devices made it difficult to accurately measure mass, the pattern of mass loss observed was comparable to that of the thickness loss. The biphasic thickness and mass loss patterns could be mostly due to reduced circulation of fluid because of cylindrical, well-shaped PSA barrier as described in the previous *in vitro* mass loss and drug release section. Along with the reduced circulation of fluid after erosion of the initial CAPP layers, growth of fibrous tissues into the PSA shell could have further slowed erosion of CAPP layers in the deeper end of the devices *in vivo*. Although a central supracalvarial pocket just large enough to fit the implant was created, slight movement inevitably occurred as the animals returned to normal activity in their cages.

Analysis of the drugs present in the retrieved devices indicated that the drugs were released in a step-by-step manner *in vivo*. For example, when the first drug, metronidazole, was

completely released, the other three drugs were still present in the device. Importantly, the other drugs were not released by diffusion. This type of behavior was observed for the rest of the four drugs. For all three types of devices, *i.e.*, LD, HD and LDS, drug release occurred in the intended sequential manner *in vivo*. This indicates that the dose of the drug or the rate of device erosion did not have a substantive effect on the sequential drug release pattern. Even *in vivo*, the CAPP layer loaded with simvastatin attached to the PSA barrier resulted in non-uniform erosion and drug release as observed *in vitro*. The extended release of doxycycline and simvastatin relative to the release of first two drugs, metronidazole and ketoprofen, was comparable to the *in vitro* release of these drug in the particular sequence. This also corresponds to the biphasic mass loss and thickness loss that was observed *in vitro*. It must also be noted that the particular drugs released could modulate the erosion profile, *e.g.*, by altering the inflammatory response and resulting volume of exudate and cellular populations.

Comparison of *in vitro* and *in vivo* performance

On the whole, CAPP initially eroded and released drugs more quickly, but during the later stages, the device design and physiological conditions played major roles in reducing the rates of polymer erosion and drug release when compared to their *in vitro* behaviors. The faster-eroding devices were predicted to last 270–300 hours and the slower eroding devices for 500–600 hours *in vivo*. Formation of fibrous tissue, observed in the devices retrieved after one week of implantation, combined with the previously described retention of fluid within the cylindrical, cup-shaped PSA barrier appeared to have slowed the clearance of erosion byproducts and the consequent erosion-dependent release of drugs. These effects were more distinct in the case of slower-eroding devices.

Along with differences in overall erosion rate *in vitro* and *in vivo*, the release rate of specific drugs was affected by their position in the multilayered device. Prior studies showed that the type and amount of drug did not affect polymer erosion [22]. The first two drugs were released faster and the last two drugs were released more slowly *in vivo* when compared to the corresponding *in vitro* release profiles. This comparative information could be valuable for precisely designing devices capable of releasing multiple drugs in a predetermined sequential order. For example, the particular drugs selected make the system relevant for potential treatment of periodontitis [21]. Knowing the effect of *in vivo* implantation on the erosion and release profiles enables “tuning” the system to deliver the active agents at a particular time. The *in vivo* results clearly suggest that the last drug released from the device would be delayed when compared to the corresponding *in vitro* release from the same device. Based on this, the PSA layer before the simvastatin layer can be removed to counter the delay that occurs *in vivo*, or the blank CAPP layers can be replaced by thicker PSA layers to further delay release if required.

Limitations of the present study include the relatively small number of samples per group and the sometimes dissimilar time points at which samples were collected. At least three samples were analyzed for each type of device and each time point, however, to provide data that would enable comparison of polymer erosion and drug release between the two conditions (*in vitro* and *in vivo*). Ideally, all samples would be evaluated at identical times,

but the different erosion rates for the devices (*e.g.*, LD vs. LDS) and in the incubation and animal environments necessitated use of dissimilar time points to encompass the duration of erosion and release.

Comparisons of *in vivo* and *in vitro* drug release have generally been conducted for delivery systems containing a single drug, with the *in vivo* drug release calculated by the measuring the amount of drug in the plasma or by measuring the amount of drug remaining in the device/delivery system [4][8–10]. Based on the type of polymer used, type of delivery system, and physiological effects on the device, *in vitro* and *in vivo* release profiles are similar in some cases [4]. In others, however, *in vitro* release was different from what was measured *in vivo* [10]. Other layer-by-layer drug delivery systems [*e.g.*, 24–27] generally incorporate a single drug, and the *in vivo* release profiles have not been compared with those observed *in vitro*. The present study involved an *in vivo-in vitro* comparison of drug release for a complex delivery system involving sequential release of multiple (four) drugs. Such information can be useful for designing complex multiple drug delivery systems and understanding how they are affected by pathophysiological conditions, such as periodontitis [21].

CONCLUSION

Multilayered, erodible CAPP devices were successful in delivering four drugs in a sequential manner both *in vitro* and *in vivo*. The polymer erosion rate can be adjusted to shift release peaks, and drug dose can be changed while maintaining the temporal nature of the profile. Although the overall time course was longer *in vivo*, erosion and release were initially faster but then slowed in comparison to standard *in vitro* conditions. This type of comparative information will be useful for modifying the delivery system to obtain programmed sequential release profiles.

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Figure 1.
Fabrication of the device.

Figure 2. Implantation of a CAPP device over the rat calvaria. (A) Site of implantation during the surgery and (B) after closure.

Figure 3.

In vitro mass loss of devices with two blank layers (LD; low dose, 1 wt%) and devices with a PSA layer between the blanks (LDS; low dose with PSA for slower erosion). Data are mean $\pm$ standard deviation (n=3–4).

Figure 4.

Sequential drug release from *in vitro* studies: (A) LD (low dose) and (B) LDS (low dose with PSA) devices. Data are mean $\pm$ standard deviation (n=3-4).

Figure 5. Cross-sectional images of devices retrieved following implantation for increasing durations. (A) HD (high dose, 5 wt%), (B) LD (low dose, 1 wt%), and (C) LDS (low dose with PSA) devices.

Figure 6.

In vivo (A) thickness and (B) mass loss for HD (high dose, 5 wt%), LD (low dose, 1 wt%), and LDS (low dose with PSA) devices. Data are mean $\pm$ standard deviation (n=3).

(A)

(B)

Figure 7.

Sequential drug release from *in vivo* studies as reflected by the amount of drug remaining the devices: (A) HD (high dose, 5 wt%), (B) LD (low dose, 1 wt%), and (C) LDS (low dose with PSA) devices. Data are mean $\pm$ standard deviation (n=3).

Figure 8.

Comparison of *in vitro* and *in vivo* cumulative release of metronidazole (M), ketoprofen (K), doxycycline (D) and simvastatin (S) from (A) LD (low dose, 1 wt%) and (B) LDS (low dose with PSA) devices. Fractional release was calculated as M/M_{∞} . Data are mean $\pm$ standard deviation (n=3).

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(A)

(B)

Figure 9.

Comparison of *in vitro* mass loss and *in vivo* thickness loss of (A) LD (low dose, 1 wt%) and (B) LDS (low dose with PSA) devices. The dotted line at 45° represents equal erosion under the two conditions.

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Time points at which samples were retrieved during the *vivo* study (number of animals at each time point n=3).

Table I

Device type	Time points of device retrieval (days)											
	-	-	-	3	-	-	-	6	-	-	9	-
Blank (B)	-	-	-	-	-	-	-	-	-	-	-	-
Low dose (LD)	-	1	2	-	4	-	6	-	8	-	10	-
High dose (HD)	0.5	1	2	3	4	5	-	7	-	9	10	-
Low dose with PSA (LDS)	-	1	-	3	-	5	-	8	-	-	12	18

Table II

Time of metronidazole, ketoprofen, doxycycline, and simvastatin peaks for the two types of devices tested *in vitro*. Note: times given in hours.

Device type	Metronidazole	Ketoprofen	Doxycycline	Simvastatin	Total erosion time
Low dose (LD)	19	61	99	173	235
Low dose with PSA (LDS)	19	90	181	293	300

Comparison of *in vitro* and *in vivo* release, indicating the duration of release for each drug in the different devices. Note: times given in hours.

Table III

Drug type	Faster eroding devices		Slower eroding devices	
	<i>in vitro</i>	<i>in vivo</i> (high dose)	<i>in vivo</i> (low dose)	<i>in vitro</i>
Metronidazole	30	12	24	24
Ketoprofen	90	72–96	72–96	120
Doxycycline	150	144–180 (only traces observed at later times)	144–180 (only traces observed at later times)	432
Simvastatin	235	>240	>240	300