FULLY IMPLANTABLE AND RESORBABLE WIRELESS MEDICAL DEVICES FOR POSTSURGICAL INFECTION ABATEMENT

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ABSTRACT

We present a therapeutic application of a microfabricated implantable and resorbable medical device made out of fully degradable materials by demonstrating in vivo elimination of bacterial infection by wireless activation of the device after implantation. The device disappears upon its completion, requiring no retrieval.

INTRODUCTION

For many years a principal objective of developing implantable devices was to guarantee that such devices would resist degradation within the human body and do not induce immune responses[1]. However, recently the demand for materials that degrade with precisely controlled characteristics (i.e. degradation rate and products) has increased dramatically along with a rapid surge of interest in degradable devices[2]. Especially, implanted devices and materials for thermal therapy to manipulate the body or tissue temperature for the treatment of disease can be traced back to the earliest medical practices and flourishes more than ever in modern research thanks to the advances in both materials (especially organic and inorganic nanoparticles)[3-5] and energy sources (ultrasound, radiofrequency waves, microwaves, and lasers)[6-9]. One fundamental challenge/requirement for non-invasive "on demand" heat delivery is to achieve spatial precision, i.e. conforming a barely working dose of heat in small tissue volumes, vertically and horizontally, leaving minimum degradation (sub)products and little damage to surrounding tissues[3].

We present here an electronically addressable, biologically degradable medical device that combines resorbable electronics and thermo-therapy through wireless control. A potential application of such devices for infection abatement (for example, surgical site infection treatment) is demonstrated.

METHOD AND RESULTS

The device is manufactured by using degradable and bioresorbable components. Silk - known and used as a high quality textile material for thousands of years - has extended its splendour to the field of biomedical engineering, as a "versatile" biomaterial, for its remarkable biocompatibility and unique mechanical and optical properties[10]. Here we report a silk-based integrated medical device for wirelessly controllable localized thermo-therapeutic treatments with all components degradable and resorbable, including a serpentine magnesium (Mg) resistor serving as the heating element that is connected with a inductively coupling Mg coil for remote heating, fabricated on a flexible silk substrate and further encapsulated in a silk "pocket" (not shown) to control the lifetime of the device (Fig. 1).

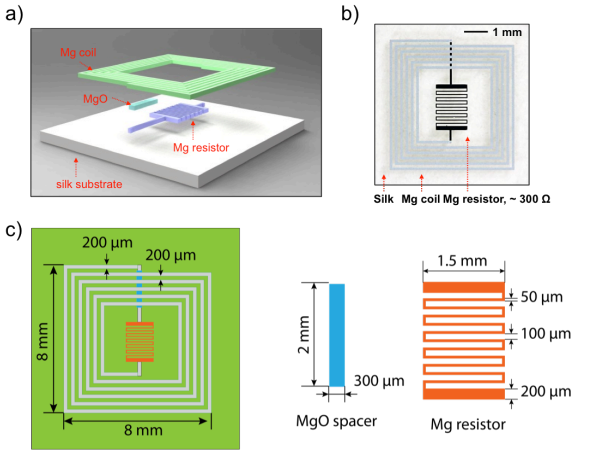

Figure 1: Geometries and dimensions of the device used in the experiment.

Silk fibroin aqueous solutions were prepared as previously described. Briefly, *Bombyx mori* cocoons were boiled for certain period of time ranging from 15 minutes to 60 minutes (varying with different applications and lifetime of devices), in an aqueous solution of 0.02 M sodium carbonate, followed by a throughout rinse using deionized water. After 2 days drying in a chemical hood, the silk fibroin was dissolved in an aqueous solution containing 9.3 M lithium bromide at 60 °C for 4 hours. The solution was then injected in Slide-a Lyzer dialysis cassettes (MWCO 3500, Pierce, Rockford, IL) and was dialyzed against deionized water for 48 hours (8 water changing in an interval of 6 hours).

The silk solution was cast on a flat surface (i.e. the bottom of polystyrene petri dishes) and was left drying at ambient conditions for 12 hours, resulting in silk fibroin films. The thickness of silk film can be precisely controlled by adjusting the volume and concentration of the silk solution and the casting area. For example, a dose of 0.2 mL/cm^2 of 6 wt% silk solution produces films of ~ 100 µm thick.

The fabrication involves a series of chemical-free processes including shadow mask deposition for the resistor and coils, thin film casting for the silk substrates, and low temperature embossing (T~85°C, for the formation of the encapsulating silk pocket). The Mg serpentine resistor was fabricated on a silk substrate of ~ 50 µm thickness and had a resistance of ~ 300 ohms, determined by the thickness (i.e. ~ 200 nm) of the 1st Mg deposition. After the deposition of a passivation layer of MgO, a 6 turn receiving coil (~ 2 µm) was deposited through a 2nd deposition step in order to connect and power the serpentine resistor. This was accomplished by inductive coupling to an external coil of ~ 5mm in diameter.

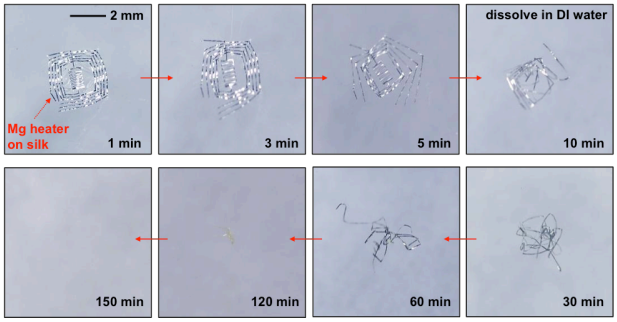

Figure 2: Optical images of full dissolution of the device (without any encapsulation) in DI water at room temperature within 2.5 h.

The device (prior to encapsulation) rapidly disintegrates (in ~ 5 mins) and fully dissolves (in ~ 150 mins) when immersed in DI water (Fig.2). The lifetime of the same device could be potentially prolonged by several orders of magnitude with suitable encapsulation strategies. Thermal embossing/lamination technique was used in this work for thermal control of silk fibroin film crystallinity. Moreover, with reflow upon heating, silk can act as a glue by controlling its thermal state. Briefly, the device to be encapsulated was placed in between two pieces of pre-treated (i.e. annealed to be water-insoluble) silk films. A few tiny drop of silk solution were applied around the edges to help sealing the silk "pocket". A detailed description of this technique (including embossing parameter optimization and the relationship of life time and embossing conditions) will be given elsewhere.

A series of *in vitro* and *in vivo* experiments were conducted to evaluate the performance of as-fabricated devices with a commercial IR camera (FLIR SC645). This device can act as an option for embedded infection management by thermal treatment when systemic antibiotic treatment alone is insufficient due to the rapid emergence of antibiotic-resistant infectious strains. The performances of the device were first evaluated *in vitro*, followed by *in vivo* studies in mice. An *in vitro* setup was used to explore the parameter space related to the therapeutic function of the device, specifically the effect of temperature and duration of heat treatment on bactericidal performance.

This was carried out by placing the devices underneath bacterial cultures of *Staphylococcus aureus* grown on agar plates and wirelessly powering them for heating through inductive coupling via a primary coil (Fig.3a).

Lyophilized *S. aureus* cultures were reconstituted and expanded according to instructions provided by ATCC. To test susceptibility, bacteria cultures were grown in liquid Tryptic Soy Broth for 18–24 h to an optical density (OD_{600}) between 0.8 and 1 (corresponding to a viable count of approx. 10^7 –10^8 CFU/mL).

Antibacterial effect *in vitro* was estimated based on the principle of the Kirby-Bauer Susceptibility Test where antibacterial effect is assessed by comparing zones of clearance in bacterial lawns. Briefly, 50 µl of the S. aureus culture were plated on Tryptic Soy Agar plates. The devices were placed on a primary coil for wireless powering/heating. The heating temperature was controlled by adjusting the input power of the primary coil using an IFI Sccx100 RF amplifier and a commercial infrared camera.

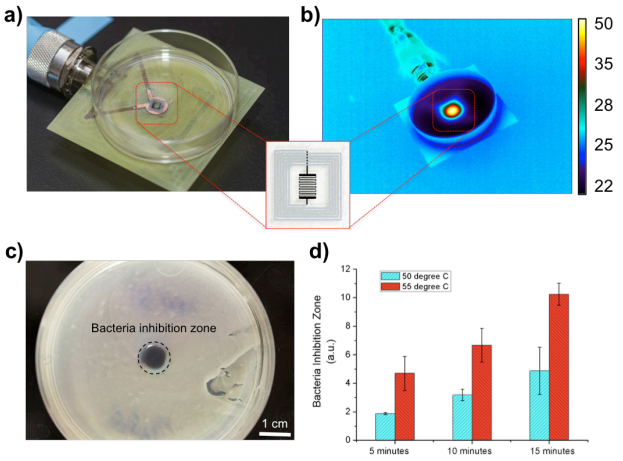

Figure 3: (a) The devices were placed underneath bacterial culture of Staphylococcus aureus grown on an agar plates. (b) The device was wirelessly powered to achieve desired temperature monitored by an IR camera. (c) A clear zone of inhibition, after heat treatment and overnight incubation, was found to correspond to the area of heat treatment application. (d) Increases in power (thus temperature) and duration can both enhance the overall bacterial inhibition effects.

The infrared heat map of the powered device shows a central region with a sharp temperature differential of ~ 28 °C between the core (resistor) and the untreated areas (Fig.3b). Higher temperatures are attainable by adjusting the input RF power in the primary coil accordingly. Once exposed, the bacteria plates (N=6) were immediately placed in a 37 °C incubator and then examined for bactericidal effects the next day. Following incubation and bacterial lawn formation, zones of inhibition were found to correspond to the areas of heat treatment application (Fig.3c). Increases in both power (thus temperature) and duration are feasible and can enhance the overall bacterial inhibition effects (Fig. 3d).

This resorbable device has the potential to be used as an implantable infection mitigation device. To evaluate this, in vivo studies were performed by implanting the devices in BALB/c mice and then infected with a subcutaneous injection of S. aureus at the device

implantation site for surgical site infection mimicking purpose.

All animal experiments were conducted in accordance with Institutional Animal Care and use Committee protocols. Two sets of 10-minute heat treatments were carried out after bacteria injection with an input power of 100 mW and 500 mW at 80 MHz. The corresponding skin temperatures of 42 °C (labelled as low temp) and 49 °C (labelled as high temp) were respectively observed. After 24 hours, infected wounds formed at the site of injection.

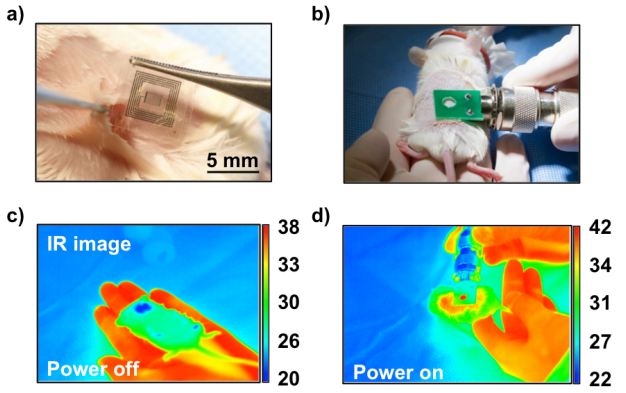

Figure 4: (a)&(b) Optical images of the device before and after implantation. (c)&(d) IR pictures of the implanted device before and after applying the rf power.

The effectiveness of the device was evaluated by excising the infected tissue site and assessing the normalized number of colony forming units (CFU) in the homogenate (n=3) using standard plate counting methods. The tissue shows a clear bacterial count reduction for thermally treated mice (Fig. 5).

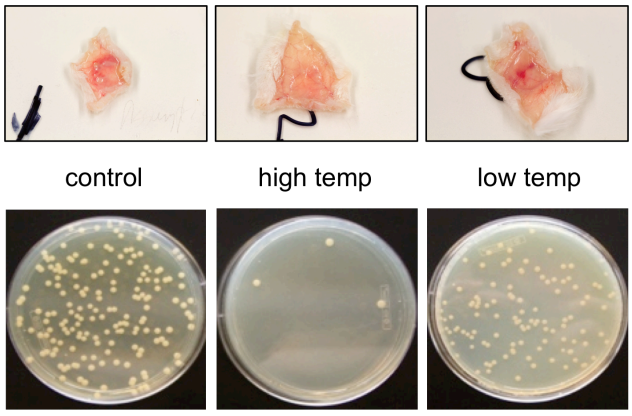

Figure 5: The infected tissues were collected 24 hours after thermal treatments and were assessed by counting the normalized number of colony forming units (CFU) in the homogenates (n=3) using standard plate counting methods.

To further evaluate the degradation process of the device *in vivo*, devices prepared in the same fashion as described previously were implanted and post-operatively wirelessly activated in the sub-dermal region of BALB/c mice. It is noted that the encapsulation was implemented in a way that the devices were able to survive the surgery process and the initial function checking point and started to degrade within a few hours to better access the device's degradation behaviors after finishing its function. Examination showed that the entire device fully degraded after 15 days (Fig. 6).

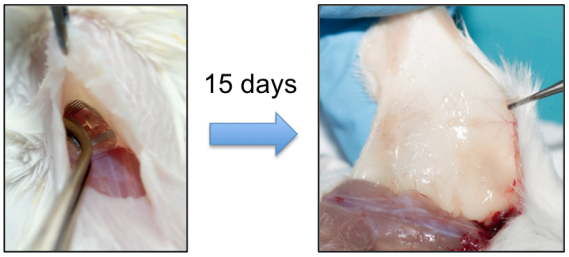

Figure 6: Devices were implanted and examined after 15 days showing no traces of the implanted device.

DISCUSSION

Recent studies demonstrate a class of complete water-soluble and fully resorbable silicon-based components, shedding light on new classes of biodegradable devices with integrated functionalities beyond the sole and more specific function offered by currently available devices (e.g. resorbable sutures, degradable intravascular stents, and matrices for drug release). A disadvantage of the original embodiment of this class of resorbable electronics is the relatively slow dissolution rates of silicon and silicon oxide being used as active elements and the passivation layer respectively, which range from weeks to months, depending strongly on temperature, pH, and thickness[11]. With only Mg and MgO used, devices reported in this work dissolve much faster via hydrolysis and get consumed in several hours (as opposed to weeks) once exposed to water and PBS solution. The rate of dissolution of silk (serving as the material for encapsulating and also mechanical supporting material) can be controlled over a wide range of time (from minutes to months, if not longer). Both lifetime (i.e. device staying functional) and existence time (i.e. device being fully degraded) of the device can both be specifically adjusted, chosen via the crystallinity of the silk.

One of greatest successes in localized thermal therapies in the past decade has been achieved by using biocompatible gold nanoparticles/nanoshells as the near infrared (NIR) absorber (i.e. the thermal coupling agent) for thermal ablative therapy for cancer. The size of those inert particles used is crucial in terms of getting enough high optical activities and precise delivery *in vivo*. Bio-distribution studies revealed that gold nanoparticles of smaller sizes (e.g. less than 50 nm in diameter) showed widespread distribution in nearly all the tissues including blood, liver, lung, heart and even brain, while larger particles tended to get accumulated locally in organs. Detailed studies of chronic toxicity of gold nanoparticles of different sizes, configurations and concentrations, especially long term ones, are greatly needed. The devices reported here operate at radio frequencies with broader operating window (for potentially multi-band selective triggering) and considerably greater penetration depth, which opens up new avenues to novel implantable thermo-therapeutic medical devices that deliver applicable

thermal therapy on demand and then get fully resorbed by metabolic pathways of the organism in the absence of residual side effects.

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