CHEMICAL DISSOLUTION OF ABDOMINAL ADHESIONS

Hanna Barton, Julia Handel, Nathan Richman, Kathryn Hohenwalter, Raven Brenneke

Advisor: Professor Kristyn Masters, **Client**: Dr. Philip Bain, Dean Clinic

Department of Biomedical Engineering, University of Wisconsin-Madison

Abstract

Abdominal adhesions are common byproducts of abdominal surgeries and can oftentimes result in serious complications. The goal of this design project is to decrease the reformation of abdominal adhesions in surgical patients through a minimally invasive method. The team's design solution involves administration of the fibrinolytic agent plasmin to inhibit the adhesion formation process. A PEG-DA hydrogel crosslinked *in vivo* will be used to administer plasmin to adhesion sites during laproscopic surgeries. Testing revealed plasmin's ability to degrade fibrin gels in an *ex vivo* environment. Future work will focus on refining testing conditions to more accurately reflect body levels of plasmin and fibrin.

Background and Problem Definition

Abdominal Adhesions

- Bands of scar-like tissue connecting tissues not normally connected [1]
- Caused by upper and lower abdominal surgeries[1]
- Form after 67-100% of abdominal surgeries [1]
- 15-18% cause further complications (i.e. bowel obstruction, infertility)[1]
- Formation involves fibrin proliferation and maturation into a collagenous matrix[2]

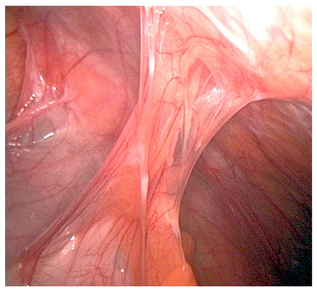

Figure 1. Image of abdominal adhesion

Problem Statement

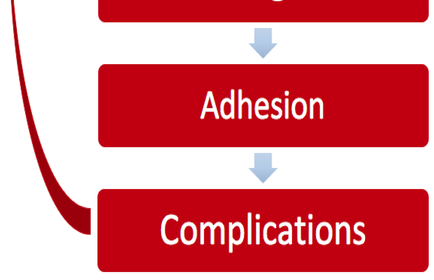

Figure 2. Adhesion formation flow chart[3]

Goal

- Design a solution to decrease recurrent adhesions following abdominal surgery

Product Specifications

Safety:
- Must be FDA approved and sterile (FDA Articles 820.25, 70, 50, 72)[4]

Biocompatibility:
- Maintain normal blood homeostatic conditions: pH 7.4, concentration of fibrinogen 200-400 mg/dL[5]

Ease of Use:
- Should not hinder surgical efficiency
- Should be compatible with current surgical technology

Final Design

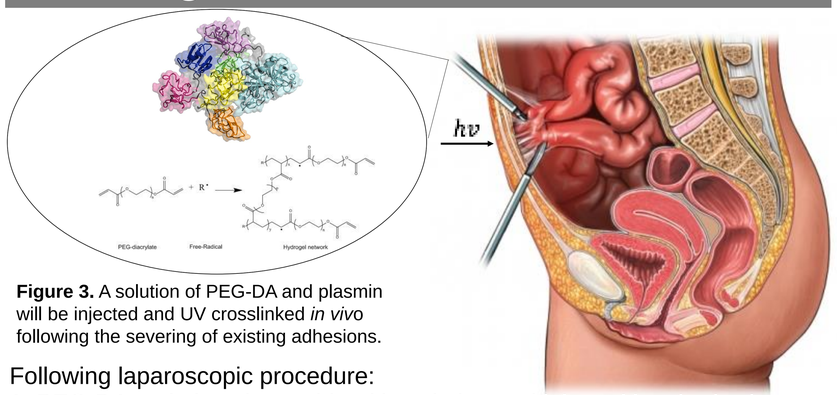

Figure 3. A solution of PEG-DA and plasmin will be injected and UV crosslinked *in vivo* following the severing of existing adhesions.

Following laparoscopic procedure:

1. PEG-DA and plasmin combined in solution and injected into body via laproscopic probe and UV crosslinked
2. PEG-DA gel swells in the first 24 hrs and begins release of plasmin
3. Plasmin is released via the hydrogel linearly with $\sqrt{t}$ over time (Eq. 1) to prevent fibrin formation in the first 72 hours[6]

$$\frac{\partial c}{\partial t} = \nabla \cdot (D \nabla c), \nabla D = 0$$

Equation 1. c = c(x,y,z,t), D = diffusion coefficient. Model is simplified by assuming D is constant

4. After all plasmin is released, PEG-DA remains *in vivo* and serves as a physical barrier to prevent further adhesion reformation[7]

Methods and Testing

Hydrogel Drug Release Testing

Testing release of PEG-DA hydrogel using fluorescent molecule to model plasmin

- Methods: PEG-DA gels were crosslinked with a fluorescent molecule and supernatant was tested at t=24 and t=48 hours for fluorescence
- Testing was inconclusive, fluorescence measured was 0

Fibrin Degradation Testing

Testing to determine amounts of plasmin that will degrade fibrin

- Methods: Differing amounts of plasmin were added to 100 uL fibrin gels and weighed after 30 mins incubation
- Plasmin testing showed statistical significance between control and plasmin groups, but not between plasmin groups

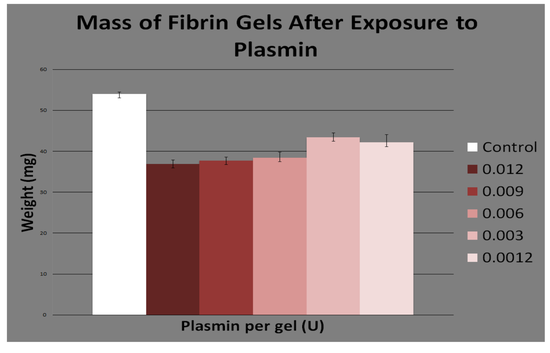

Figure 4. Mass of fibrin gels after 30 minute incubation with varying amounts of plasmin. All plasmin treated groups are statistically significant from the control, but not from eachother.

Plasmin Treatment (U)	Percent Degradation
0.012	31.72
0.009	30.15
0.006	28.82
0.003	19.54
0.0012	21.97

Figure 5. Percent degradation calculated in reference to average control fibrin gel mass. No values are significantly different from the others.

Discussion of Results

- Fibrin testing demonstrated that over a 30 min. incubation period, there was approximately 30% degradation of fibrin
- Potential problems with hydrogel testing
  - PEG-DA gel pore size too small
  - Photobleaching of fluorescent agent caused by UV crosslinking
  - Formation of hydrogel only on circumference of well

Figure 6a. Creation of fibrin gels

Figure 6b. Image of fibrin gel

Future Work

1. Development and Identification of ideal PEG-DA hydrogel
   - Photobleaching of fluorescent testing
     - Use different fluorescent agent to test diffusion rates and crosslinking
   - Develop an accurate method to measure the concentration via volume
2. Use knowledge of the fibrin degredation pathway to test other preventative agents (i.e. tissue plasminogen activator)
   - Identify agent with lowest systemic effect and greatest local effect
3. Future testing to connect fibrin degradation by plasmin *ex vivo* to the prevention of fibrin formation *in vivo*
   - Determine optimal concentration of plasmin in PEG-DA hydrogel to balance prevention of fibrin formation and limit systemic effect

Acknowledgements

- **Dr. Philip Bain**, Dean Clinic
- **Dr. Kristyn Masters**, Department of Biomedical Engineering
- **Dr. John Puccinelli**, Department of Biomedical Engineering
- **Dr. Greg Matske**, SSM Health
- **Andrew Dias**, Murphy Lab
- **Murphy Group**, BioInspired Materials Laboratory

References

1. Buåureanu, Æ. A., & Buåureanu, T. A. S. (2014). Pathophysiology of Adhesions, (3), 293–298
2. R.T. Beyene et al. (2016). Intra-abdominal adhesions : Anatomy , physiology , pathophysiology , and treatment, *52*(2015), 271–319.
3. Brüggmann D, Tchartchian G, Wallwiener M, Münstedt K, Tinneberg HR, Hackethal A: Intra-abdominal adhesions: definition, origin, significance in surgical practice, and treatment options. *Dtsch Arztebl Int* 2010, 107(44):769-775.
4. CDC, "Guideline for Disinfection and sterilization in healthcare facilities, 2008," CDC, 2009. [Online].
5. A. Marucco, I. Fenoglio, F. Turci and B. Fubini, "Interaction of fibrinogen and albumin with titanium dioxide nanoparticles of different crystalline phases", *Iopscience.iop.org*, 2013.
6. F. Bierbrauer, "Hydrogel Drug Delivery: Diffusion Models", *University of Wollongong*: 1-31.
7. Bart W.J Hellebrekers, Trudy C.M Trimbos-Kemper, J.Baptist M.Z Trimbos, Jef J Emeis, Teake Kooistra, Use of fibrinolytic agents in the prevention of postoperative adhesion formation, Fertility and Sterility, Volume 74, Issue 2, August 2000, Pages 203-212, ISSN 0015-0282, http://dx.doi.org/10.1016/S0015-0282(00)00656-7.