Optical Imaging of the Small Airway Mucosa

Biomedical Engineering Design 200/300
Department of Biomedical Engineering
University of Wisconsin-Madison
October 12, 2022

Team Members:

Peter Wawrzyn (Team Leader)
Jade Berget (Communicator)
Andy Slayton (Co-BSAC)
Lily Zahn (BWIG)
Yash Shah (Co-BSAC)
Sofia Castagnozzi (BPAG)

Client:

Dr. Allan Brasier
UW Institute for Clinical and Translational Research, Executive Director

Advisor:

Dr. Filiz Yesilkoy
UW Madison College of Engineering - BME

Abstract

Airway diseases are an increasingly common issue in many humans and the need for effective treatments has never been greater. In order to effectively test these treatments, accurate imaging of the airway mucosa is required. In humans this has been done effectively using Optical Coherence Tomography (OCT) imaging; however, using OCT in mouse test subjects for visualization of the airway mucosa has yet to be demonstrated. The goal of this project is to create and validate a method of imaging the airway mucosa of a mouse test subject in vivo. Specifically, this semester the goal is to create an imaging probe that can access the airway mucosa without damage to the airway and obtain accurate images. The probe must be designed to access the airway of the mouse and allow for stable accurate images. The design team generated three preliminary designs and after evaluation chose the Clear Intubation Catheter. This design uses a clear catheter shell to allow for access and then imaging of the airway mucosa. Multiple materials were considered for design fabrication and silicone was deemed to be the most appropriate. Future work for the team includes manufacturing and testing a prototype of the design based on input from the client and other experts in the field.

I. Introduction

Common airway diseases include asthma, chronic obstructive pulmonary disease (COPD), pulmonary fibrosis, pneumonia, bronchiectasis and lung cancer. These are diseases that most people have been affected by, whether themselves or someone they know. Each of these diseases can affect any gender, race, or age and have varying harmful effects. Asthma affects about 14% of children 5 and under and COPD accounts for about 55% of respiratory diseases in both men and women. In 2017 there were 3,914,196 deaths due to chronic respiratory diseases which accounted for 7% of total deaths globally. This put chronic respiratory diseases as the third leading cause of death in 2017, just behind cardiovascular diseases and neoplasms. These rates have only increased since 1990 [1] and the only way to combat this issue is by developing treatments.

The cells lining the airway play an important role in common airway diseases. The mucosa will change thickness as the body's reaction to certain diseases or infections. When the mucosal lining in the airway thickens it restricts airflow and breathing becomes more difficult [2]. Part of testing treatment effectiveness is to measure that change in thickness of the mucosa.

New therapeutics are being developed for treatment, but a limitation in the work to develop these therapeutics is the difficulty of measuring changes over the course of an experiment in small animal studies. Some imaging techniques being used now are Optical Frequency Domain Imaging (OFDI) and Optical Coherence Tomography (OCT) which are comparable in technique and resolution. Both these options are able to monitor changes in the airway; however, the current probes used with these imaging modalities are too large for small animal testing. B. J. Vakoc et.al. developed and used a balloon stabilization device for OFDI imaging in the distal esophagus of swine [3]. However, a similar approach within a mouse airway would exert excess pressure on the esophagus, potentially harming the mouse. L. P. Hariri et. al developed a custom bronchoscope for use of OFDI imaging in human lungs, however, the device is too large to use in mice and difficult to scale [4]. Due to this problem, the goal of this project is to create and validate an miniaturized OFDI or OCT probe for imaging in the airway of small animals; specifically, creating an imaging probe to be utilized on mice.

II. Background

Client Information and Preliminary Research:

Our client is researching treatments for inflamed and diseased airways. In their research, the lab is testing the effectiveness of treatments on the airways of small animals. The airway mucosa, which consists of epithelial cells lining the esophagus, varies in thickness between healthy and diseased airways. Monitoring the thickness of the airway mucosa can indicate the effectiveness of treatments for lung diseases [5]. Our client is looking to monitor changes in the

thickness of the mucosa of mice while testing the effectiveness of their lung disease treatment. One method of measuring the mucosa thickness is to use optical imaging, specifically OFDI with a probe device.

Optical frequency domain imaging, or OFDI, uses infrared light to generate cross-sectional images of tissue with a resolution between 10 and 20 micrometers [6]. The theory behind OFDI imaging is to direct light waves at tissue and measure the delays of the back-reflected light to estimate the thickness of the tissue [6]. To produce accurate imaging, the device must be stable and able to collect and transmit data to an external imaging device.

Design Specifications:

The goal of this project is to create a probe capable of performing OFDI imaging on the small airway mucosa of mice. The device must be small enough to fit within the mouse airway, which is approximately 1.5 mm in diameter. [7] The device must be kept stable enough to use OFDI to create a 3D map of the airway, measuring the depth of the airway mucosa up to 1 mm and resolution between 5 and 20 micrometers [6] with a Signal to Noise Ratio of at least 80 [8]. The device will be used several times on living mice over several weeks, so it must be reusable and meet our clients lab animal testing policies along with federal regulations [9]. The project has a budget up to $10,000, but that can vary depending on the availability of optical imaging equipment.

III. Preliminary Designs

1. Clear Intubation Catheter:

The clear intubation catheter design contains a clear outer catheter that is designed to hold the mouse airway in place during imaging. The imaging probe, that will be connected to an optical fiber, will be inserted inside this clear catheter. The outer clear catheter will ensure that the airway is not only stable, but also keep the airway safe from potential damage. Figure 1 shows an imaging probe being inserted into a clear catheter which will be in the airway. The diameter of this catheter will be 1.2mm and the length of the catheter will be 15mm to precisely fit into the airway of a mouse.

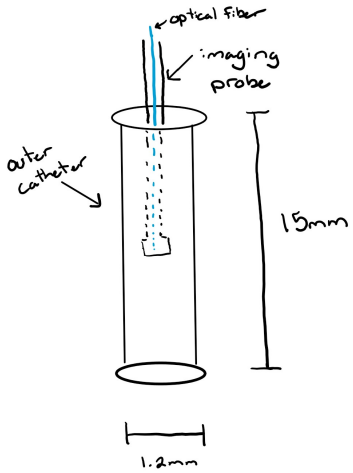


Figure 1: *Clear intubation catheter with probe insertion.*

2. Helical Balloon Catheter:

The helical balloon catheter design contains a small deflated balloon covering on the outside of a helical shaped catheter during insertion. Once the helical catheter has been inserted into the mucosa, the balloon is then inflated in order to stabilize the airway for accurate and safe imaging. The balloon would only cover the body of the catheter, leaving the tip exposed for the imaging probe. With nothing covering the imaging probe, there will be higher resolution with no extra artifacts. The dimensions of this catheter with an inflated balloon would be 1.0mm in diameter and 5.0mm in length. Figure 2 shows an image of the overall design of the catheter during insertion, where the balloon surrounds the body of the catheter waiting to be inflated for imaging. Figure 3 shows how the catheter will look during imaging with the balloon inflated to stabilize the airway.

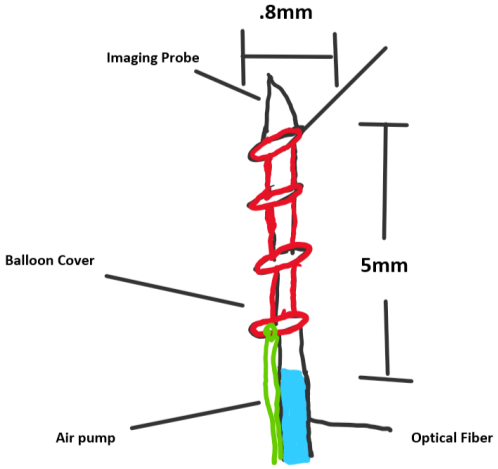


Figure 2: *Helical Balloon Catheter with balloon deflated for insertion.*

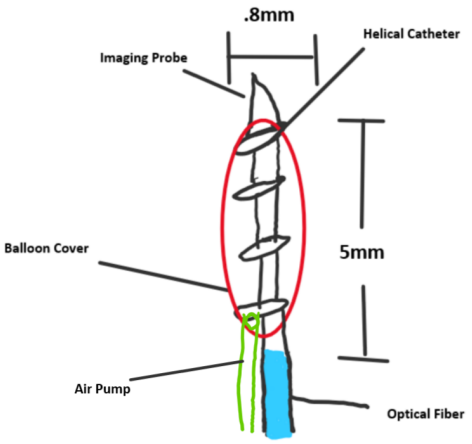


Figure 3: *Helical Balloon Catheter with balloon inflated for stabilization during imaging.*

3. Flexi-Catheter Withdrawal Design:

The Flexi-Catheter Withdrawal design consists of an outer catheter and an imaging probe. The outer catheter acts as a normal catheter but contains the imaging probe housed inside of it. The outer catheter is used as a mechanism to guide the imaging probe into the mouse airway. The outer catheter is then retracted exposing the imaging probe which allows for imaging of the mouse airway mucosa. Figure 4 shows an image of the probe mechanism at the time of imaging. In Figure 4 the outer catheter is retracted exposing the imaging probe to the airway mucosa for imaging.

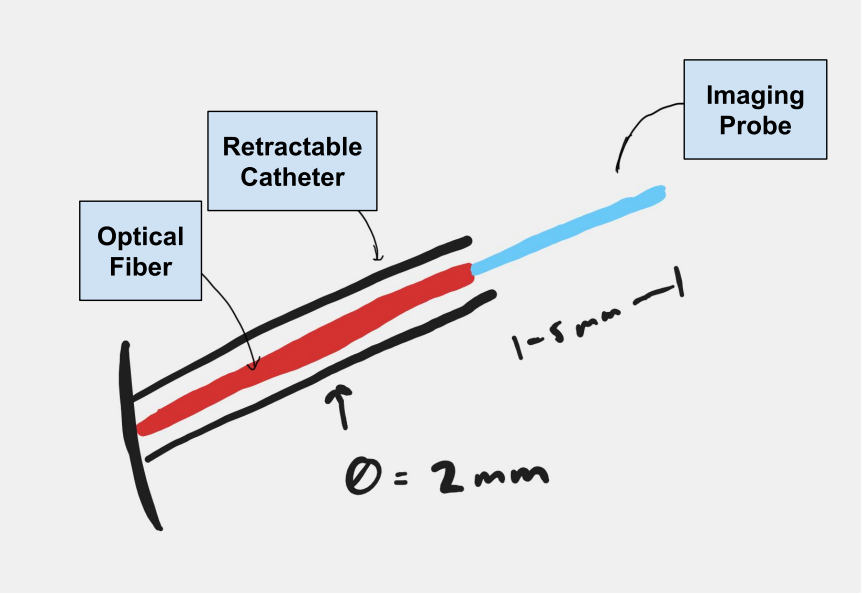


Figure 4: *Flexi-Catheter Withdrawal Design with the outer catheter in the retracted position.*

Figure 5 shows the probe mechanism with the outer catheter advanced forward. This configuration would be used when the probe is being navigated into and out of the mouse airway.

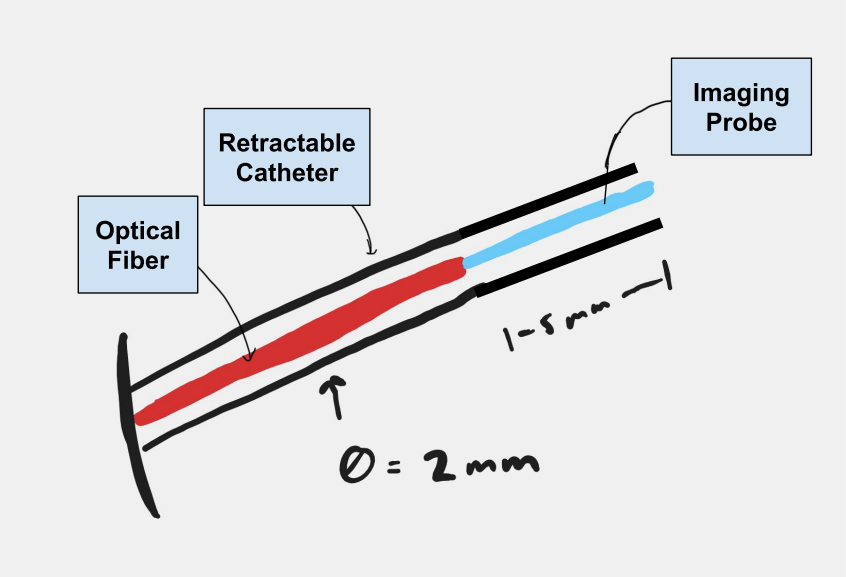


Figure 5: *Flexi-Catheter Withdrawal Design with the outer catheter in the advanced position.***IV.**

Preliminary Design Evaluation

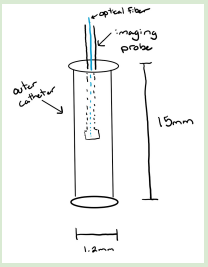
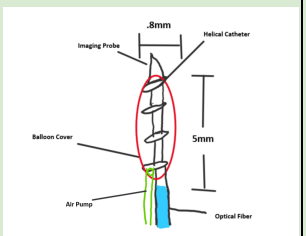
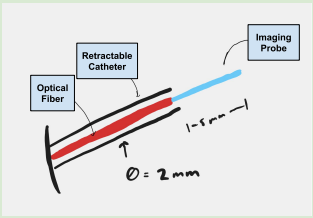
In order to evaluate the three different designs, a design matrix shown in Table 1 was created. All three designs were judged based on their anticipated Stability (30), Manufacturability (25), Accuracy (25), Safety (15), and Cost (5).

The stability of the probe and airway was our highest weighted category at 30 because keeping the airway open and stable while imaging is a key for accurate imaging. The Clear Intubation Catheter scored 5/5 because with the outer catheter in place the airway will be completely open during the entire imaging process with the probe free to move and rotate inside. The Helical Balloon Catheter scored 4/5 because the entire airway does not stay open during imaging but the balloon can add stability around the tip of the probe. The Flexi-Catheter Withdrawal Design scored the lowest at 2/5 because there is no outer component that guarantees the airway to stay open around the probe.

Manufacturability of the probe mechanism was tied for our second highest weighted criteria. This category measured the ease at which the probe mechanism could be manufactured. The Clear Intubation Catheter scored the highest with a 5/5 due to the simplicity of the design. The Flexi-Catheter Withdrawal Design scored second highest with a 4/5 due to the potential difficulty manufacturing the mechanism needed to withdraw the outer catheter. The lowest scoring design was the Helical Balloon Catheter with a score of 3/5 due to the complex nature of manufacturing a balloon small enough for a mouse airway and incorporating that into a helical catheter.

Table 1: *The Design Matrix used to rank the preliminary designs across five weighted criteria. Green highlights are the winner of that section*

Probe Mechanism

Design Criteria	Clear Intubation Catheter 		Helical Balloon Catheter 		Flexi-Catheter Withdrawal Design 	
Stability (30)	**5/5**	**30**	4/5	24	2/5	12
Manufactuability (25)	**5/5**	**25**	3/5	15	4/5	20
Accuracy (25)	3/5	15	**5/5**	**25**	**5/5**	**25**
Safety (15)	**5/5**	**15**	1/5	3	3/5	12
Cost (5)	**5/5**	**5**	4/5	4	**5/5**	**5**
Total (100)	**90**		71		74	

The accuracy that the probe mechanism allows for was tied for our second highest weighted criteria at 25. This category intended to measure the accuracy and resolution that the probe design would allow for an OCT scanning system. The Flexi-Catheter Withdrawal Design and Helical Balloon Catheter both scored a 5/5 for this category because they both allowed for unobstructed visualization of the airway mucosa during imaging. The Clear Intubation Catheter scored a 4/5 on this category because it would require imaging through a clear catheter which could lead to artifacts in imaging and would lead to increased complexity in obtaining accurate imaging.

Safety of the probe was weighted at 15 due to the use with live mouse subjects. This category intended to judge how safely the device could be inserted into a mouse airway. During in vivo imaging of a mouse subject it is important to not damage the airway for proper testing of airway mucosa medications. During the rating of designs the possibility of potentially harmful parts and materials was considered. The Clear Intubation Catheter scored 5/5 due to the lack of complicated parts that could lead to injury. The Flexi-Catheter Withdrawal Design scored 3/5 due

to the possibility of injury during the outer catheter withdrawal. The Helical Balloon Catheter scored a 1/5 due to potential for injury from inflating a high pressure balloon in the airway.

Cost was the lowest weighted criteria at 5 due to the high budget for the project. Additionally, the probe mechanism is a small portion of the project cost compared to the imaging component. Due to this all designs scored highly with, the Clear Intubation Catheter and the Flexi-Catheter Withdrawal Design both scoring 5/5 and the Helical Balloon Catheter scoring ⅘, due to added expense of the balloon mechanism.

The Clear Intubation Catheter scored the highest with 90 total points. This was largely due to the simplicity of the design which leads to easier manufacturability and increased stability compared to other designs.

IV. Fabrication/Development Process

Materials:

For the creation of the imaging probe, our team focused on the outer material that would encase the OFDI technology. To evaluate possible materials, a design matrix shown in Table 2 was created. All three materials were ranked based on their anticipated Moldability and Manufacturability (30), Reusability (25), Safety (20), Shelf Life (15) and Cost (10).

Table 2: *The Design Matrix used to rank the probe materials across five weighted criteria. Green highlights are the winner of that section*

Probe Material

Design Criteria	Polycarbonate		Polypropylene Copolymer (PPCO)		Silicone Rubber	
Moldability & Manufacturability (30)	4/5	24	2/5	12	**5/5**	**30**
Reusability (25)	**5/5**	**25**	4/5	20	**5/5**	**25**
Safety (20)	**5/5**	**20**	**5/5**	**20**	**5/5**	**20**
Shelf Life (15)	4/5	12	**5/5**	**15**	**5/5**	**15**
Cost (10)	**5/5**	**10**	2/5	4	4/5	8
Total (100)	91		71		**98**	

To fabricate the probe, our team will use the silicone rubber as the outer material for the imaging technology as it ranked the highest in total for our criteria, and has many properties that make it favorable for biomedical applications. It is a firm and flexible material that can withstand a wide range of temperatures, chemicals, and UV exposure, making it ideal for sterilization conditions. It is also readily available, easy to manufacture inert, nontoxic, and nonbiodegradable, making it a suitable option for invivo biotechnology. [10]

Methods:

1) Design Clear Intubation Catheter on Solidworks
2) A model of the probe will be created using the Formlabs Form 3 printer at the UW Madison Makerspace. The material that will be used to print the probe is the Formlabs flexible resin
    a) Flexible resin will be used to resemble the silicone material used on the prototype
3) Model will be tested and modified for final dimensions
4) The team will send finalized design to a biotechnology company for custom silicone molding

V. Testing/Future Work

Moving forward, the team will construct a prototype probe following the winning design and material evaluated in the design matrices. Despite not having the probe attached to the rest of an imaging machine, there are still relevant tests our probe needs to undergo. The most important of these tests is to measure the force imparted on the airway by our probe. This will be done by measuring the coefficient of friction between the silicone catheter and a biologically similar material. The team can also test the silicone as a testing medium by using an existing OCT device; our research suggests silicone wouldn't interfere with imaging, but our fabrication process could affect this.

VI. Discussion

The probe must be minimally invasive in order to protect the test subjects and to ensure the highest accuracy in the images produced. If the probe itself is contributing to the inflammation response in the mucosa then it will skew if not invalidate the results. Aspects like these will be assessed if Dr. Brasier tries to take a drug into clinical trials, therefore the final machine must limit physical contact as much as possible. As mentioned previously, testing will be conducted to ensure the probe's safety. Specific figures will be discussed with the client once the final design has been fabricated. There are also animal care standards and regulations relevant to our design, however, the restraints due to data accuracy require a much higher degree of animal protection than is required by regulation[9][11].

VII. Conclusions

Imaging tools are intricate and expensive machines that provide scientists and medical professionals an essential insight into biological organisms. Our team will design and manufacture a probe to help afford this insight to our client, Dr. Brasier, in his research using lab mice. Specifically, Dr. Brasier intended to measure the inflammation of the airway mucosa in mice, an indicator or deteriorating health to assess the efficacy of novel drugs. For this task, optical coherence tomography (OCT) is well suited to produce high resolution volumetric images to completely image the mucosa. We learned from our professional advisors that to make a device like this would traditionally take a team of graduate students and much more than our budget. In spite of this, we believe that through several courses of BME design, a completed OCT imaging device can be delivered to Dr. Brasier.

Our team will continue with the intubation tube design, using silicone rubber. The intubation tube will be the simplest and most effective way to stabilize the source of light in the airway. We will conduct imaging tests on the silicone sheath that we fabricate in order to ensure that the optical properties do not interfere with the imaging process. We will focus our research on the manufacturing process of small, precision instruments. Fabrication at this scale is challenging because there is a lot of precision required to successfully rotate an imaging fiber steadily and avoid damage to the airway. When the prototype is completed, we will test the probe's impact on a simulated airway to ensure that it does not contribute to any perturbation recorded by the probe.

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IX. Appendix

A: Product Design Specification

Function:
The goal of this project is to create and validate an optical frequency domain imaging (OFDI) probe for imaging in the airway of small animals. The cells lining the airway play an important role in common airway diseases and new therapeutics are being developed for treatment. A limitation in the work to develop these therapeutics is the difficulty of measuring changes over the course of an experiment. Some imaging techniques, including OFDI, are able to monitor changes in the airway; however, they are too large for small animal testing. Due to this problem, the aim of this project is to create an effective miniaturized OFDI probe for the use in animal testing.

Client requirements:

- The imaging device must be able to visualize the airway mucosa of mice in vivo.
- The device must be able to be maneuvered through a mouse airway for imaging.
- The project's budget is up to $10,000 depending on availability of already purchased resources

Design requirements:

1. Physical and Operational Characteristics

a. Performance Requirements: The imaging device must be able to image the airway mucosa of a murine test subject. The device should be able to measure up to 1 mm in depth of the airway mucosa and should have a resolution of 5 - 20 μm, which is comparable to existing OCT systems [1]. The device should not harm the mouse when sedated to allow for testing throughout a drug testing protocol.

b. Safety: The device must come with clear and concise instructions for device usage and must only be used by a trained operator with animal subjects training [2]. The device material must not cause biological harm to the mouse or the user and no sharp edges should be exposed as part of the device that could cause internal injury to the mouse subject during imaging.

c. Accuracy and Reliability: The device must have a Signal to Noise Ratio (SNR) of 80 or more, which is a baseline for imaging biological targets [3]. The

imaging device must also demonstrate significant correlation ($p < 0.01$) between calculated airway wall layer areas using histology, ex-vivo, and in-vivo approaches [4].

d. Life in Service: The imaging probe should be reusable on different subjects, operating at least 10 minutes per use, averaging 2 minutes per data set with OFDI technology, [5] or 4 minutes with OCT technology [6]. The material of the probe will be sterilized by autoclaving.

e. Shelf Life: The shelf life will be dependent on quality of materials, e.g. fiber optic cable, biocompatible finish.

f. Operating Environment: The imaging probe must withstand temperatures between 20°C (68 °F) and 135°C (275 °F) for storage and sterilization conditions. The probe must also avoid corrosion from in-vivo testing and sterilization chemicals.

g. Ergonomics: The imaging probe must be made out of a material that can be used safely inside an organism with no reaction and can be sterilized. It must be able to maneuver the cartilage rings in the upper part of the trachea and measure within the airway mucosa [7].

h. Size: The probe must be able to fit inside the airway of a mouse. Its diameter must be less than 1.5mm, the diameter of a mouse trachea [8].

i. Weight: The probe must be light enough to be hand maneuvered by the same hand and weigh no more than 5.1 pounds [9].

j. Materials: Potential materials that may be included are fiber-optic cables, silicone rubber, and a camera and lens. The materials will all be biocompatible and autoclavable if the design is made to be reusable.

k. Aesthetics, Appearance, and Finish: The finish must be smooth to limit physical interference of the sample. The finish must be biocompatible. The appearance and aethstetic of the device does not contribute meaningfully to its efficacy .

2. Production Characteristics:

a. Quantity: We will manufacture one final design and test it in the small airway

mucosa of a small mouse.

b. Target Product Cost: The manufacturing cost may be more expensive due to the specialized style of optical imaging necessary. The total cost should be approximately $5,000-$10,000. Although this cost is high, the total cost can be made lower based on materials already readily available to our team.

3. Miscellaneous

a. Standards and Specifications: Must avoid or minimize discomfort, distress or pain consistent with sound scientific practices [10]. Animals that are subject to prolonged discomfort or distress must be given proper sedation [10]. Animals must be humanely and safely handled, treated and transported [11].

b. Customer: The customer is asking for a device which can image the mucosa in lab mice to record the effects of trial drugs. Mice are sedated prior to imaging. Perturbation of the mucosa and other tissue by the probe would negatively impact the accuracy of the data taken. The customer would like a feature of the probe to indicate the depth of the probe in the mouse.

c. Subject-related concerns: The materials and maneuverability of the probe must ensure the mouse is unharmed and procedures follow lab animal safety protocol while using the product.

d. Competition: Hariri et. al. published a study in 2012 recording the use of OFDI on human lung imaging. The study described two methods, one of bronchoscopy airway-centered imaging and one of parenchymal imaging. A custom-built bronchoscope was used with a diameter of 0.8 - 1.7 mm. The OFDI methods were only performed on human lungs [12].

Templin et. al. published a study in 2010 that successfully used OFDI for stent healing evaluation in vevo on pigs. The study described using a Terumo-OFDI catheter on a 0.014-inch guidewire. The study successfully used OFDI on the animals 1, 3, 10, 14, and 28 days after the stents were implanted [13].

Vakoc et. al. published a study in 2006 where OFDI was successfully performed on the distal esophagus of two swine using a 4.5 cm long inflatable balloon. The study successfully acquired cross sectional imaging of the mucosal vascular network within two living swine [14].

No patents were found for a product that could successfully use OFDI in the airways of any animals smaller than pigs.

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