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Design and Application of a Deep Oral Irrigator for Treatment of Halitosis

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Design and Application of a Deep Oral Irrigator for Treatment of Halitosis

THESIS

submitted in partial satisfaction of the requirements
for the degree of

MASTER OF SCIENCE

in Biomedical Engineering

by

Daryl Nguyen

Thesis Committee:
Professor Arash Kheradvar, Chair
Professor Hamid Djalilian
Professor Michelle Khine

2022

DEDICATION

To my late younger sister, Kayleen.

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ABSTRACT OF THE THESIS

Design and Application of a Deep Oral Irrigator for Treatment of Halitosis

by

Daryl Nguyen
Master of Science in Biomedical Engineering
University of California, Irvine, 2022
Professor Arash Kheradvar, Chair

There is a need for an oral irrigation device for reducing odor causing bacteria built up in the throat, preventing tonsil stone formation, and clearing existing tonsil stones. This thesis examined the efficacy of novel shower heads, a deep oral irrigation device, and an oral rinse for cleaning the tonsillar crypts. The custom shower heads tailored for reaching the tonsils were examined with an existing oral irrigation device. Once an effective shower head has been finalized, a deep oral irrigation and tonsil cleaning device was developed based on user requirements from patients affected by halitosis. The novel device has demonstrated the ability to reach the tonsillar fossa behind the tonsillar pillar, dislodge trapped tonsil stones, and rinse the area without spilling liquid back onto the hand of the user. A zinc-based mouth rinse was examined when conducting a minimal inhibitory concentration experiment against *S. aureus*, a model oral pathogen responsible for a wide variety of diseases including tonsillitis. The mouth rinse was effective at inhibiting *S. aureus* growth with a minimum inhibitory concentration for 50% growth (MIC50) of 10% (v/v). Both the mouthwash itself and zinc acetate were successful in inhibiting the growth of *S. aureus*. A minimum inhibitory concentration for 90% growth (MIC90) was achieved at 100 µg/mL which did not change in 10% mouthwash. Overall, the combination of mouth rinse and zinc acetate has shown promising antibacterial activity with great potential to be used as an oral wash for future studies.

CHAPTER 1

Introduction

1.1. Halitosis

Halitosis describes the condition of having persistent unpleasant odor from the mouth. The exhaling of this odor is known as commonly known as “bad breath”. Varying minimum levels of bacteria and volatile molecules found in the oral region are responsible for the unpleasant odors that arise from halitosis. Over 80% of all halitosis cases are due to poor oral hygiene, gingival and periodontal diseases, tongue and gum biofilm, retention of food debris, blockage of tonsillar crypts, and oral infections.[1,2] Different types of bacteria are sourced from diverse origins, for example, may range from eating, the conditions of the surrounding environment, to direct contact with the hands or any given object. Although it is not ever likely to be able to completely eliminate the oral bacteria present in the mouth, good oral hygiene starts with the ability to adequately control the growth of harmful microbes. Harmful oral bacteria thrive in conditions where food debris and sugars are present in the warm and damp environment of the mouth. Occurrences of gingival and periodontal diseases, decaying oral health, and dental caries are catalyzed from the acidic byproducts secreted from these oral bacteria. Food debris are often trapped between teeth, inside tonsillar crypts, and other oral areas that are difficult to detect and remove. As these trapped food debris decay in these inconvenient areas, sometimes unbeknownst to individuals, the higher levels of acidity contribute to the decline of oral health and the rise of halitosis.[2]

The National Institute of Dental Research estimates about 65 million Americans suffer from halitosis or bad breath at a given point in their lives.[3] Behind dental caries and periodontal

disease, halitosis may be the next common cause for a patient to visit an appointment with their dentist. However, the quality of care to address the concerns from halitosis is often lackadaisical. The underlying causes of halitosis are not yet fully understood. Chemical agents and mechanical appliances partially reduce the biofilm and debris load found on the tongue, gums, and oral cavity. The US Federal Drug Administration (FDA) classifies halitosis to be a cosmetic issue.[4] Traditionally, oral rinses were used as an over-the-counter oral antiseptic drug to decrease the chance of infection in wounds in the mouth.[5] Today, oral rinses are used in efforts to cleanse the mouth and to mask the unpleasant odors from halitosis. However, these oral rinses do not solve the root cause of halitosis. Instead, oral rinses act as a deodorant for the oral cavity that must be applied on a frequent basis.

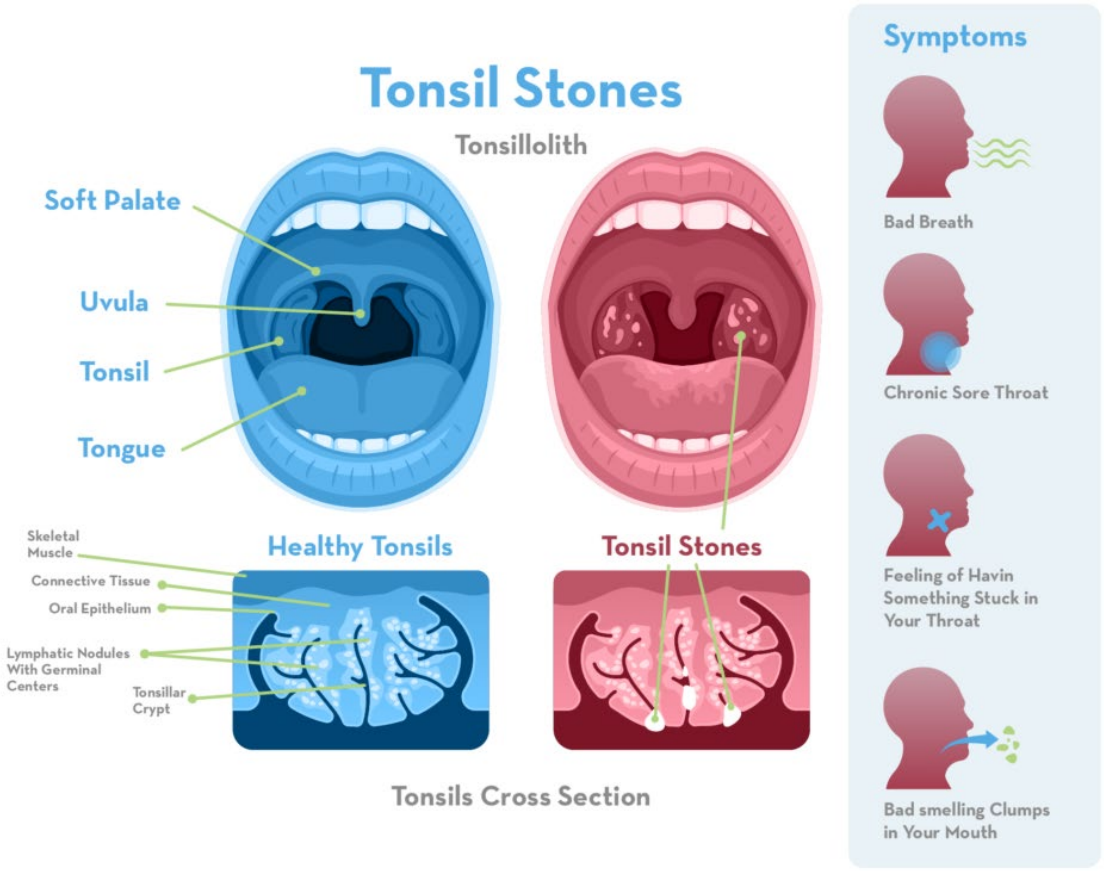

Figure 1.1 The areas and symptoms related to tonsil stones. Accumulation of the sulfur-producing bacteria and debris that are deposited in the tonsils are the main cause of bad breath. Image from Jefferson Dental Clinics, 2020.[6]

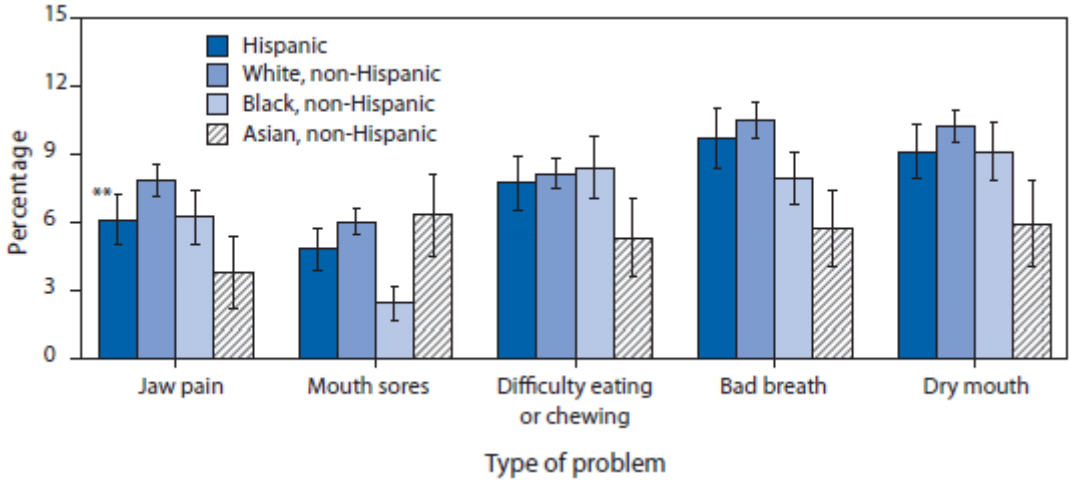

Figure 1.2 The percentage of US adults aged 18–64 years who have had problems involving the mouth. Taken from Centers for Disease Control and Prevention, 2011.[7]

1.2. Halitosis Treatment Options

Due to undesirable experiences from conventional self-treatments, patients that are affected by halitosis are often nervous and skeptical of any treatment. The diagnosis of halitosis begins with completing an extensive examination of the oral cavity.[8] The regions of interest for determining the possible source of halitosis include tight areas between the gum and bone, tongue biofilm, in the back of the mouth, or tonsillar crypts entrapped with food debris. A thorough examination contributes to an accurate diagnosis of halitosis for the individual to be treated effectively. Common available treatment options can be categorized as masking, chemical reduction or neutralization, or mechanical reduction.

Biofilm removal from the gum, cheeks, and tongue is typically the first line of action to control halitosis.[8,9] Tongue brushing or scraping have demonstrated to be potentially successful in reducing biofilm and halitosis.[8,10] The application of a toothbrush to remove biofilm from the tongue reduces approximately 45% of volatile sulfur compounds (VSCs). Unlike toothbrushes, tongue scrapers are ergonomically designed to remove VSCs more effectively and reduces 75% of VSCs.[11] According to a randomized control trial for mouth odor reduction through tongue

cleaning techniques, study groups between adults affected by halitosis demonstrated a small but statistically significant difference in VSC level reductions when using toothbrushes compared to tongue scrapers.[12] However, there is currently no clinically significant evidence for treating halitosis through dietary modifications, tongue brushing, xylitol chewing gum, or scraping the tongue.[13]

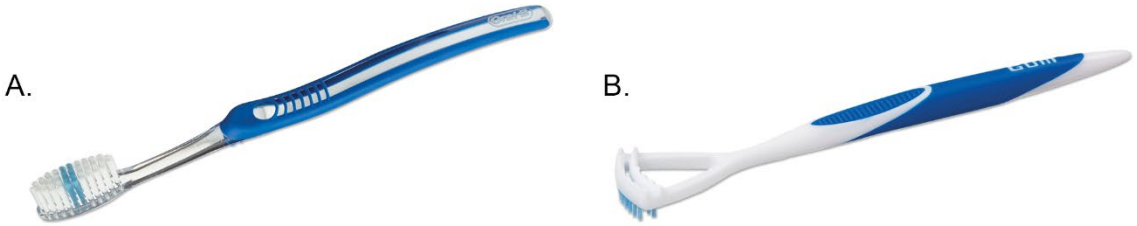

Figure 1.3 Two different types of commercial consumer oral hygiene tools. A) The Procter & Gamble Oral-B® Indicator toothbrush. B) The GUM Dual Action Tongue Cleaner.

Antibacterial agents in mouth rinses commonly include ethyl alcohol and chlorhexidine (CHX) for off-the-shelf and over-the-counter uses, respectively. Cetylpyridinium chloride (CPC) and triclosan are also commonly used agents for reducing odor-producing bacteria.[1,14] According to a study for comparing mouth rinses against halitosis, mouth rinses consisting of zinc and chloride dioxide demonstrate the neutralization of the VSCs while mouth rinses consisting of antibacterial agents CHX and CPC demonstrate the inhibition of VSC production.[15] The current gold standard antibacterial agent in mouth rinses for treating halitosis is CHX. According to a randomized, double–blind, cross–over study design, a mouth rinse containing CHX in combination with CPC demonstrated the greatest reduction in bacteria and VSC levels.[16] In a different mouth rinse study to evaluate the synergy between the combination of CHX and zinc, the mouth rinse also demonstrated a significant drop in bacteria and VSC levels.[17] However, common reports of teeth staining and a displeasing aftertaste from patients after using CHX may discourage a long-term routine.[18]

Commercial mouth rinses such as Listerine demonstrated adequate reduction of sulfur-

producing bacteria in healthy individuals affected by halitosis.[19] Mouth rinses that include triclosan demonstrate reduction of halitosis and gingivitis.[20] A significant reduction of VSC levels were achieved through the use of a triclosan toothpaste and tongue scraper.[20] Similar results were achieved through a 3 week randomized double blind trial study using a triclosan formula in reducing VSC, oral bacteria, and halitosis.[21]

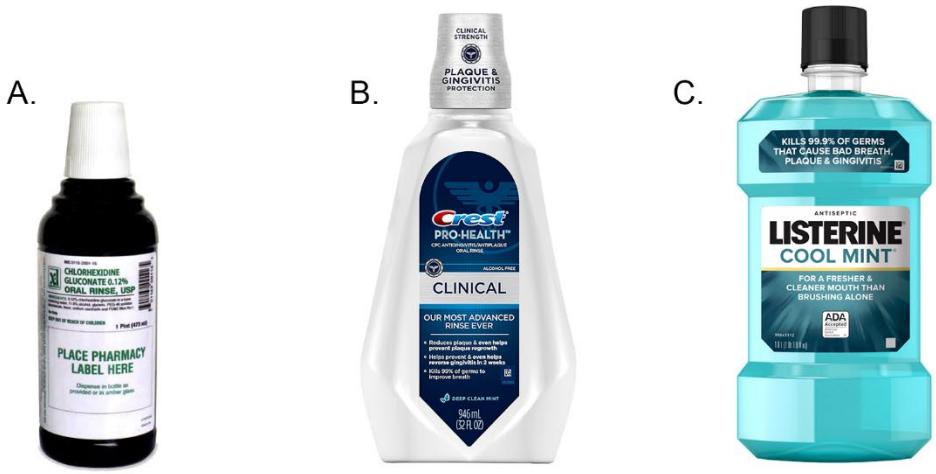

Figure 1.4 Three different types of commercial mouth rinses. A) The Xttrium Laboratories Prescription CHX Gluconate Mouthwash Oral Rinse. B) The Crest Pro-Health Clinical Rinse with CPC. C) The Listerine Cool Mint Antiseptic Mouthwash.

Chlorine dioxide is used as a sulfur-oxidizing agent where 29% of subjects had a reduction of bad breath after 4 hours.[22] Chlorine dioxide reacts with sulfur compounds by the metal ions binding with the sulfur radical ions; thus, inhibiting the production of VSCs.[22,23] A study involving the daily consumption of probiotic tablets containing *Lactobacillus salivarius* WB 21 demonstrated control of halitosis.[24] Tea tree oil may inhibit the growth of halitosis bacteria as a combination product with alpha-bisolol.[25] Photodynamic therapy is a two-stage treatment that combines a photosensitizer drug with light exposure to reduce the concentrations of VSCs. Though commonly used for treating acne and cancer cells, photodynamic therapy shows promising results as an alternative treatment for halitosis.[26]

Chewing gum, oral rinsing products, fluoride toothpastes, and mints are masking agents used for a short-term remediating effect.[27] Peppermint tablets and oil can temporarily remediate halitosis by promoting saliva production to counter dry mouth.[28] Propolis, a resin-like substance produced by bees, has demonstrated to be effective in the reduction of pathogenic organisms for periodontal diseases and halitosis.[29,30]

However, the mechanical methods of oral hygiene alone are not adequate for treating halitosis due to the lack of access to some areas.[31] Chemical mouthwashes and masking agents lack of proper penetration to inaccessible areas deep in the oral cavity.[32]

1.3. Oral Irrigation

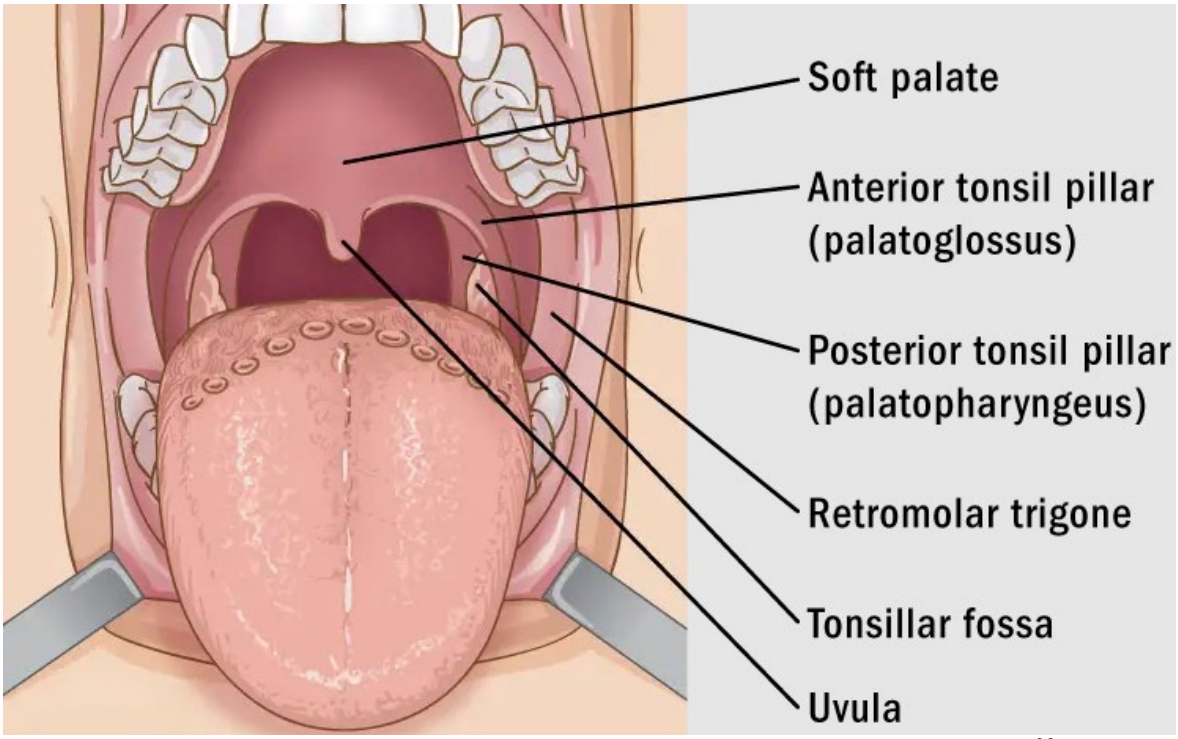

Figure 1.5 Structures related to the tonsils. Taken from THANC Foundation, 2022.[33]

The ideal treatment to remove sophisticated sources of halitosis is an oral irrigator that provides access to complex oral regions such as the crypts of the tonsillar fossa and behind the anterior tonsillar pillars, a shower head for biofilm removal and proper penetration of the deep

oral areas, and complete ease of use as a household oral hygiene appliance. Recently, water flossers, also known as water jets, are becoming a popular type of oral irrigation device (OID) for removing debris and plaque in areas that are difficult to access with toothbrushes. The water jet pulsations and adjustable water pressure are the primary mechanisms of OIDs that provide cleaning in challenging areas. Biofilm and bacterial debris are removed from the shearing forces generated by the water pressure. For clinical efficacy, an OID should provide a minimum of 1200-1400 pulses per minute at 60 pounds per square inch (PSI). Even pressures up to 160 PSI for 30 seconds can be achieved without producing irreversible damage to the gingiva and is safe for gingival and junctional epithelium attachments.[34,35] Applying a water jet for 3 seconds onto a surface also demonstrated that the OID can eliminate 99.9% of biofilms present on that surface.[36] OIDs are commonly used several times a day for patients facing gingival conditions or conditions that worsen with the use of a traditional toothbrush or floss. OIDs can also use antiseptics or antibacterial mouth rinses in place of water. A water jet using a CHX mouth rinse has demonstrated to be clinically effective.[37] However, a primary concern with the use of the water jet for treating halitosis is the possible discomfort and damage to the more delicate tissues in the back of the oral cavity, especially areas of the tonsillar fossa and tonsil pillars where biofilm and trapped debris are often overlooked and untreated.

By using the oral irrigator designed for the deeper areas of the oral cavity, tonsil stones accumulated in the tonsillar fossa can be easily removed. Additionally, the continuous use of the OID can keep the tonsillar fossa clean, helping prevent the accumulation of dead bacteria and food debris, and resolving the cause of halitosis from tonsil stones.

1.4. Outline of the Thesis

The main objective of this research is to develop a novel oral irrigation device to treat potential sources of halitosis. An irrigating mechanism to jet a zinc formulated mouth rinse into the deep crevices of the tonsils will promote the removal of trapped debris, biofilm, and tonsil stones. The development of this deep oral irrigating and tonsil cleaning device could potentially address cases of halitosis. Different shower heads were compared for comfort based on their output, ease of use, and the accessibility of challenging areas in the oral cavity. Commercial oral irrigators were initially used during the development of a custom shower heads tailored to reach the tonsillar fossa in order to rapidly test designs before the development of a new irrigation system. Miniature diaphragm pumps and their pressure outputs were studied for comfort. Batteries and other electronics were investigated to meet the design requirements of a new irrigation device. Chapter 2 elaborates this design process in detail along with the circuit design of a custom irrigation device.

After a set of shower head designs were chosen, multiple sets were manufactured using certified biocompatible materials. Chapter 3 describes the entire manufacturing process. Each set of the shower heads were sanitized in alcohol and cured under ultraviolet light. After post-processing, shower heads were used orally as a method to study the feasibility and efficacy of the design.

The antibacterial studies for a zinc-based alcohol-free mouthwash are described in Chapter 4. The bacterium chosen for antibacterial studies were considered based on relevance and availability. *S. aureus* can be isolated from the oral cavity and was selected to be tested against the zinc-based mouth rinse. Suspended liquid cultures and streaked culture plates were used for the antibacterial testing of the mouthwash. Minimum inhibitory concentration studies

were conducted with varying concentrations of the zinc acetate in the mouthwash. A concentration of the zinc acetate in the mouthwash was determined to be effective to inhibit bacterial growth.

The shower head design and mouthwash recipe may be optimized to improve the overall ability to eradicate tonsil stones and halitosis through an on-going pilot study at the University of California, Irvine Medical Center for the treatment of halitosis. Chapter 5 discusses the future works of the patient studies, shower head designs, and potential use of the mouthwash formula.

CHAPTER 2

Deep Oral Irrigation Device Design

2.1. Background

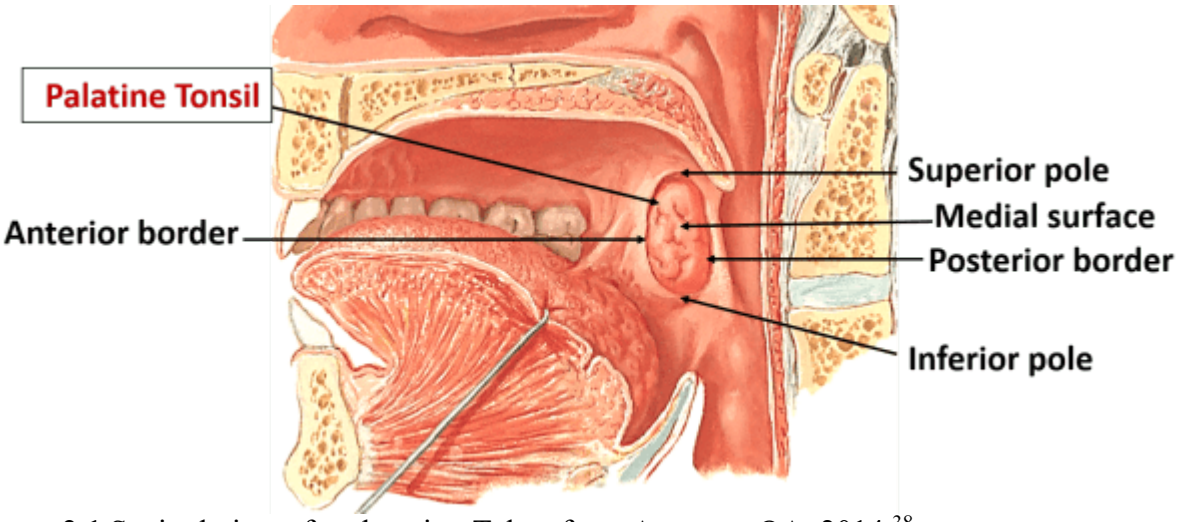

Figure 2.1 Sagittal view of oral cavity. Taken from Anatomy QA, 2014.[38]

Designing an oral irrigation device to deep clean and remove tonsil stones from the oral cavity presents many challenges. It is considered that tonsil stones are made of trapped debris and bacteria accumulated in the crypts of the palatine tonsils.[39] The groove shape of the palatine tonsil bring difficulty in the removal of tonsil stones with the use of conventional oral hygiene instruments. Currently, patients who wish to remove tonsil stones have tried to remove them themselves using a variety of apparatuses or have requested their healthcare providers to have them removed at a hospital or clinic. Individuals who perform the removal process at home often use their fingers or toothpick-like instruments to push out the larger tonsil stones within the crypts. Dentists and healthcare professionals often use a professional air or water syringe to

dislodge and flush out the tonsil stones.[40] Despite these efforts, the deeply grooved shape of the palatine tonsils prevent the complete removal of tonsil stones in the tonsillar crypts even in professional settings. In serious cases, the tonsils of the affected individual are removed surgically through tonsillectomy. However, removal of the tonsils at an early age may contribute to a higher risk of heart disease.[41]

A tool specifically made with tonsil rinsing and cleaning features is a much needed alternative. A Waterpik™ flosser is a water jet device with a shower head designed to remove debris between the teeth and gums. This water jet method has shown promising results in removing bacteria, debris, and biofilm.[36,42,43]. With a shower head specifically designed to reach the tonsillar crypts and pressure settings configured to effectively wash the tonsil stones, designing a deep oral irrigation and tonsil cleaning device may be achievable.

2.2. Design

User Requirements	Design Input / Metrics	Design Output
Allow for access to back of throat and tonsillar crypts.	Shower Head Length	114 millimeters
	Shower Head Diameter	8 millimeters
Prevent spillage onto hand or device during use.	Device Distance from Mouth	76 millimeters
	Angle of Shower Head from Device	135 degrees
Cleaning of delicate oral tissues is easily controllable and comfortable.	Water Pressure	13 PSI
	Button / Switch	Momentary Push Button Switch for On/Off Pump
Can be used without power outlet.	Lithium Battery	3.5 hour battery life

Table 2.1 Device design requirements for a deep oral irrigation and tonsil cleaning device. Design inputs and outputs are based on user requirements received by otolaryngologists and their patients.

Figure 2.2 Design stages of a custom shower head.

The base of the shower head design was reverse engineered from a Waterpik™ shower head design to allow a custom shower head to be rapidly tested with a Waterpik™ device. Calipers were used to evaluate the dimensions of the base of the original shower head design. The 2D sketches of the custom shower head design were drawn in SOLIDWORKS™, a solid modeling computer-aided design (CAD) and computer-aided engineering computer program. The design outputs were generated based on the user requirements of the design requirements (Table 2.1). The 2D sketches were then generated as a 3D model using the revolved boss extrusion feature of the CAD software. The 3D model is then exported to a Standard Triangle Language (STL) file, the industry's standard data transmission format for 3D models. This format approximates the surfaces of a solid model with triangles. Once converted and exported as an STL format, the 3D model can be opened with popular and industry software for CAD modeling and additive manufacturing, such as 3D printing software. The details of this 3D printing additive manufacturing processes are described in Chapter 3.

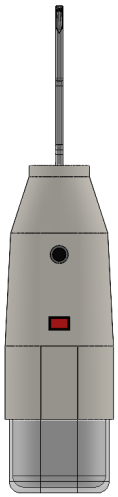

Figure 2.3 3D CAD model of a tonsil cleaning device.

The deep oral irrigation and tonsil cleaning device consists of a rigid shower head with a long neck for reaching the behind the tonsillar pillars. A liquid reservoir containing 200mL of volume is used for water or an oral rinse to cleanse the tonsillar areas over a period of 30 seconds. The electrical design includes a 12-voltage (V) 1450 milliamp hour (mAh) rechargeable battery pack, miniature pump, microcontroller, a voltage regular, power switch, and momentary push button switch to control the shower head through a proposed electrical design (Figure 2.4).

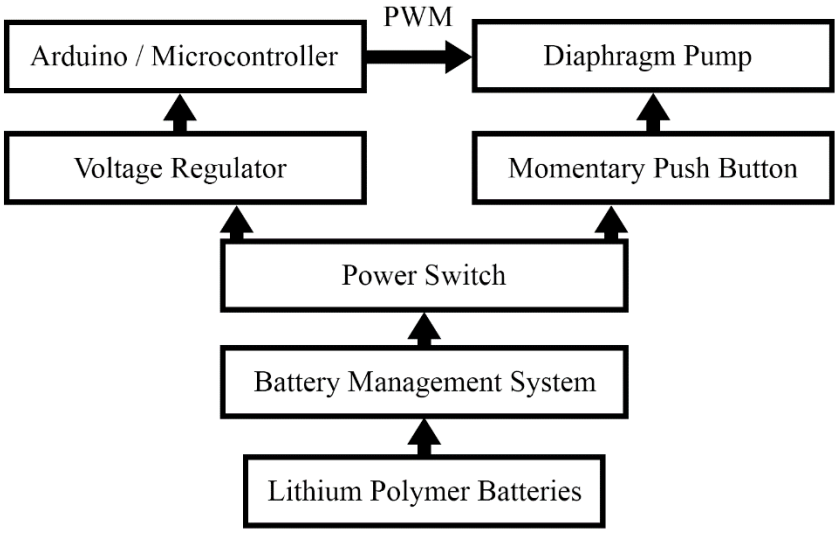

Figure 2.4 Circuit design of deep oral irrigation and tonsil cleaning device.

2.3. Materials Investigated

Component	Description	Brand
Battery	1450mAh 3.7V Lithium Polymer Battery	Jauch Quartz™
Battery Management System	3S 12V 10A Lithium Battery Protection Board	Comidox™
Power Switch	Mini Power Rocker Switch	Philmore™
Voltage Regulator	5V 1.5A Linear Voltage Regulator	STMicroelectronics™
Microcontroller	Arduino Uno	Arduino™
Diaphragm Pump	12V Diaphragm Pump with PWM Speed Control	Kamoer™
Momentary Push Button Switch	Momentary Push Button Switch	Philmore™

Table 2.2 Selected electronic components after material investigation.

The investigated materials are summarized in Table 2.2. The battery was selected to be a lithium battery. Lithium batteries comprise of a lithium-based oxide and a carbon material as the positive electrode active material and the negative electrode active material, respectively.[44] Common types of lithium batteries are generally categorized as a polymer electrolyte battery or liquid electrolyte battery. Batteries using polymer electrolytes are lithium polymer batteries and batteries using liquid electrolytes are called lithium ion batteries. Lithium polymer batteries are becoming an attractive option due to their flexibility, compact size, and high electrochemical performance.[45] Since lithium polymer batteries are made of flexible materials, the shape of the batteries are commonly prismatic cells, pouches, and can be curved. Prismatic lithium polymer batteries use square aluminum containers and pouch batteries use pouch containers made from thin metal plates. In contrast, lithium ion batteries are typically manufactured as a cylindrical cell enclosed in cylindrical aluminum containers. The lithium polymer battery is a favorable choice for reducing the size and weight of electronic devices. Compared to lithium ion batteries, lithium

polymer batteries are slim, lightweight, and exceptionally stable.[46] The typical nominal operating voltage of a lithium battery is 3.7 V. Due to their high energy density per unit mass, the operating voltage of lithium batteries is three times higher than the widely used nickel-cadmium battery, such as the nickel-metal batteries from Duracell™.

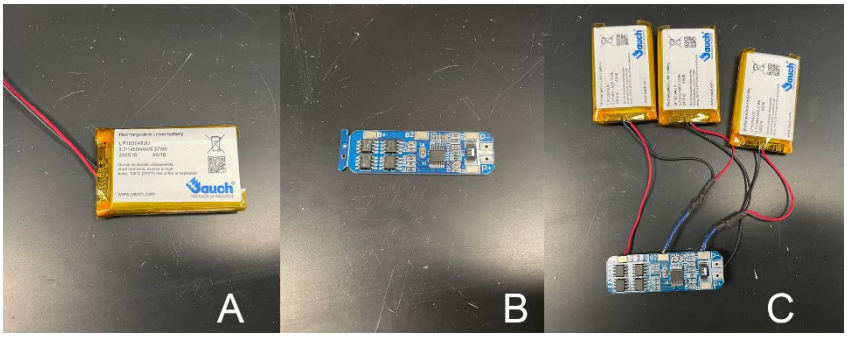

Figure 2.5 Components of the 12V battery pack. A) Jauch Quartz™ LP103048JU lithium polymer battery. B) Comidox™ HX-3S-01 Lithium Battery Protection Module. C) Components of the 12V battery pack soldered together as a single circuit.

The lithium polymer battery was selected from Jauch Quartz™. The lithium polymer battery pack is wired in a 3-series (3S) to provide a 11.1V rating (Figure 2.6). The battery pack is the soldered to a battery management system (BMS). The lithium battery protection board was selected from Comidox™ to safely discharge and charge the lithium polymer battery pack, help prevent overcharge, and effectively power the miniature diaphragm pump at 12V. The battery management system includes primary and secondary protection circuits to avoid the lithium battery pack from exploding. The battery pack and BMS is wired with a separate socket that can be connected to a standard power supply to recharge the device.

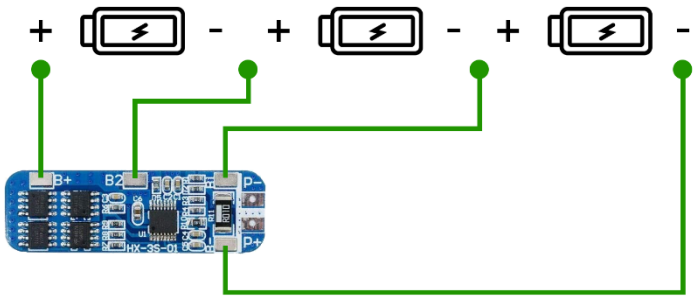

Figure 2.6 Wiring diagram for 3S lithium battery and battery management system.

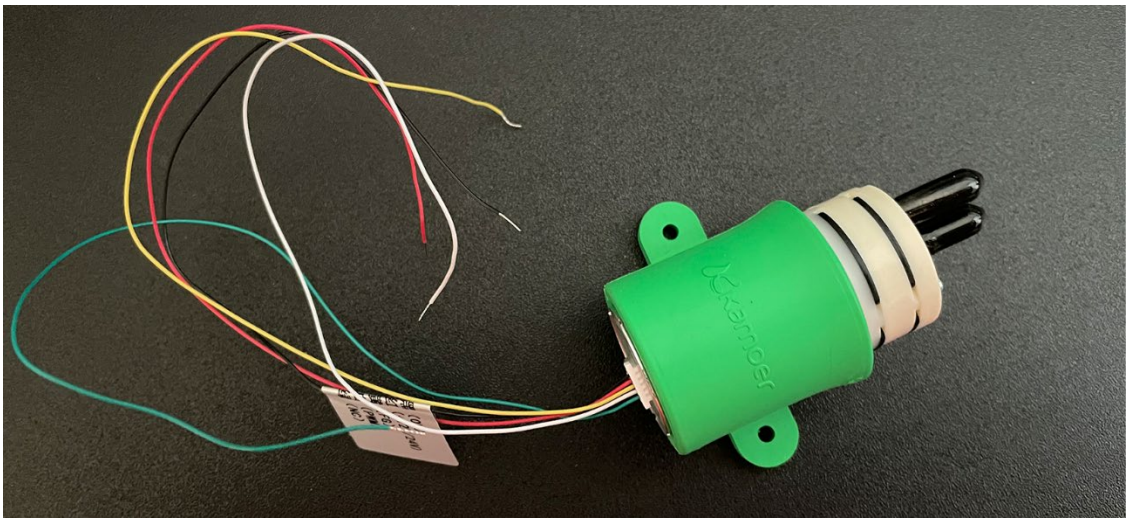

Figure 2.7 Kamoer™ KVP04 liquid diaphragm pump.

The liquid diaphragm pump was selected from Kamoer™. The device requires the shower head to supply fluid at a relatively high pressure to a certain area in the oral cavity to remove biofilm, clean the area, and dislodge tonsil stones. Diaphragm pumps operate without leaks and can still operate with air mixed into the system. Compared to ordinary piston pumps, diaphragm pumps do not contain a member deteriorating over time that contaminates the fluid to be delivered by the pump. Additionally, the diaphragm in a diaphragm pump is not driven mechanically, but indirectly by a fluid pressure medium.[47] The diaphragm forms a boundary between the fluid and the pressure medium. The diaphragm pump in this designed in powered by the lithium polymer battery pack and is activated when the individual presses and holds their finger on a designated button. This button was selected to be a momentary push button switch from Philmore™ to achieve this activating function. The controllable water pressure of this pump is set by the output of the microcontroller.

The microcontroller was selected to be an Arduino Uno from Arduino™. In general, microcontrollers are small computers on a single integrated chip. They often consist of analog-to-digital and digital-to-analog converters, central processing units (CPU) for executing tasks,

programmable peripherals for interacting or sending data from external devices, memory, a system bus to carry data between the CPU and the memory, internal or external sensors, and transducers. The Arduino Uno has been programmed to control the pulse-width modulation (PWM) feature of the diaphragm pump. Proper footprints for a phase-correct PWM mode have been implemented in the code in case controlling PWM with mechanical a dial or potentiometer is to be performed in the future.

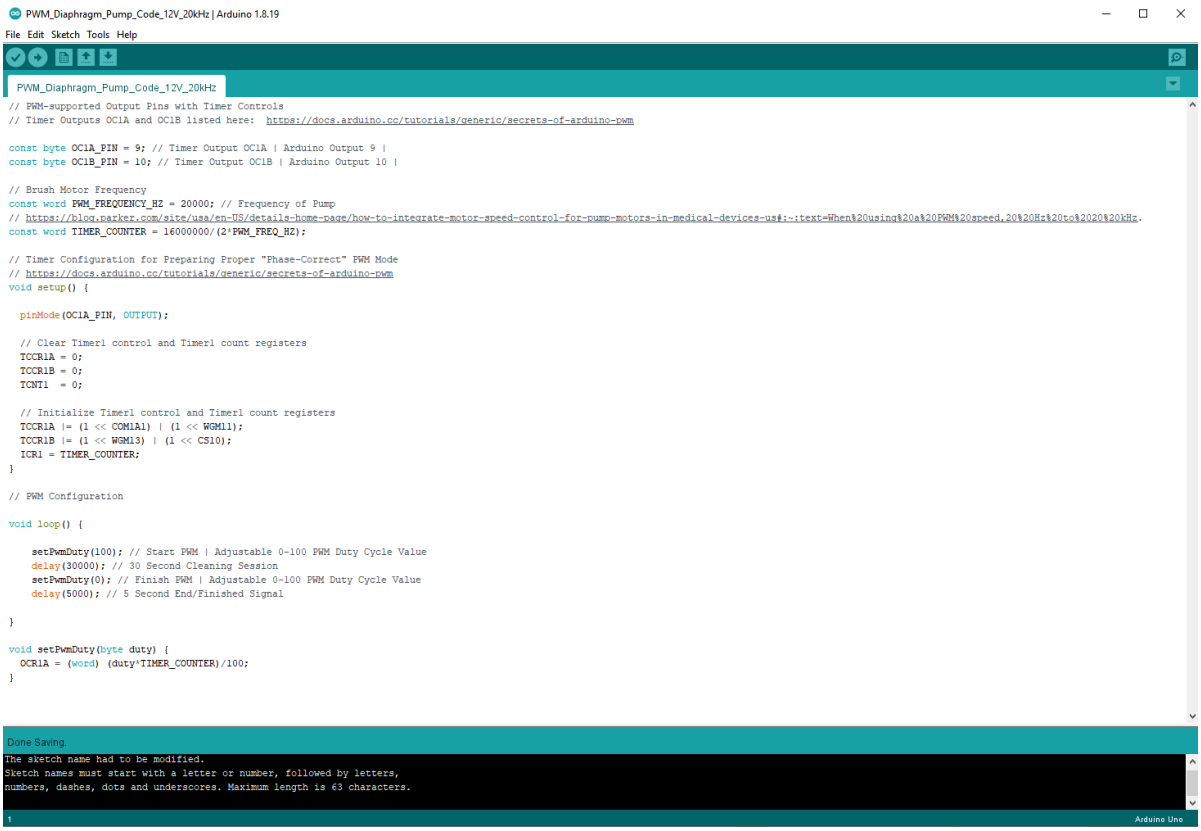

Figure 2.8 Programming code to control the pressure of the diaphragm pump.

CHAPTER 3

Manufacturing of Oral Irrigation Device

3.1. Background

Additive fabrication is a manufacturing process to build objects through layers of solidification. Specifically, each layer is formed and adheres to either a previously formed layer or a platform upon which the object is built on. Each layer represents a 2D slice of the object. Since these slices are the cross-sections of the object, the entire object is formed once every following slice has been processed. Common additive fabrication methods include selective or fused deposition modeling (FDM), stereolithography (SLA), and laser sintering.[48]

FDM is the most widely used technology for additive manufacturing and 3D printing.[48] In fused deposition modeling, or FDM 3D printing, layers of heated materials are extruded through a small nozzle and fuse together in a pattern to create an object. The nozzle heats the material up to a melting temperature that allows the material to bind to a platform or existing layers of the material, creating an object layer by layer. Generally, the materials are calibrated with specific 3D printer settings to allow proper cooling of the heated material once extruded to solidify different filaments. In certain FDM 3D printers or settings, the cooling of the material is accelerated through the use of cooling fans attached on the extrusion head to help prevent warping. When importing a 3D object to print, the FDM 3D printing software codes the printer to selectively deposit melted material in a pre-determined path. A variety of parameters can be adjusted for the object to be printed, including the temperature of the nozzle, the temperature of

the build platform, the build speed, the cooling speed, and the layer height of each slice. The entire job can be inspected through a layer-by-layer preview screen before continuing to print. Common materials for FDM 3D printers are thermoplastic polymer filaments, which come in spools ready to be loaded into the printer. Additionally, materials like dissolvable supports, metal, and chocolate are compatible with FDM 3D printers. Before printing, the FDM 3D Printer heats up the nozzle in which the filament would extrude from and the warms up the print head where the first layer of the filament would adhere to. Adhesives are typically applied to the print bed, also known as the build platform, before printing to ensure the adhesion of the first filament layer to the print bed. The FDM 3D printer takes the filament from the spool and squeezes it through the heated hot end of the nozzle, melting the filament to be then deposited in layers on the heated print bed. The layers continue to deposit and repeats until the object is formed. Once the print has been completed, the print bed must cool down. Otherwise, the object becomes more challenging to remove and is prone to damage. A common defect in FDM addictive manufacturing is the warping of objects. Thermal expansion allows the opportunity for the extruded material to reduce in size during the cooling and solidification process. Since subsequent layers of the object cool at different rates than their previous layers, the dimensions of the object also change at different rates. The differences in cooling and shrinking dimensions between layers of the object builds up internal compression forces, which may the pull up areas from the very first layer as a response. The fine-tuning of print settings, using well-known and tested materials, and quality adhesives between the filaments and build platform can prevent warping.

As another approach to additive fabrication, stereolithography or SLA 3D printing is one of the fastest growing 3D printing technologies due to its ability to produce high-accuracy, precise, airtight prototypes with a variety of advanced resin materials. SLA 3D printing uses the

same principles as FDM 3D printing where the selected material is solidified at each layer until the object forms. In SLA 3D printing, the layers of desired objects are formed by using a curable liquid resin with a laser set at a wavelength to solidify each thin layer of the object.[49] The exposure to the laser beam cures a precisely thin layer of liquid resin, which causes it to harden and adhere to previously cured layers or the bottom surface of the build platform.[50] Similar to FDM 3D printing, SLA typically applies the first layer onto a build platform, then one on top of another and repeats this process until the object is fully formed. Common materials for SLA include biocompatible certified resins, resins filled with glass-shards to produce reinforced objects, and elastic materials that mimic common grades of silicone rubber. Advancements to SLA resin formulations bring desirable mechanical, thermal, and optical material characteristics to tailor the needs of the industry.

Advancements in 3D printing continue to improve the workflow for businesses that rely on receiving rapid feedback on prototypes before full production. Complex and precise parts, or parts that often require a large upfront cost to be produced, can have the costly prototyping processes substituted through the benefits of 3D printing technologies. New business models are being developed due to the available option of having rapid prototypes iterations, faster production workflows, and reduced outsourcing costs. As the accessibility, materials, and affordability of 3D printing progresses, the technology will allow engineers, designers, and industry professionals to integrate 3D printing into business operations and development cycles to meet market opportunities and demands.

3.2. Materials and Methods

Figure 3.1 Biocompatible SLA Printer. Taken from Formlabs, 2022.[51]

The Form 3B was selected as the manufacturing printer for its Advanced Low Force Stereolithography (LFS)™ technology (Figure 3.1). It uses a flexible resin tank and a custom-designed, user-replaceable Light Processing Unit (LPU) laser system to produce consistent, accurate prints with smooth surface finish and part tolerance precision. The design files were imported into PreForm. PreForm is a software that communicates with the printer and allows adjustments to the print settings for preparation. Once prepared, the print is checked for printing compatibility by verifying the minima and support needed for the material to print successfully. Once approved, the print begins. The BioMed Clear Resin was material selected for the print job. BioMed Clear Resin is a hard, strong material for biocompatible applications requiring long-term skin or mucosal membrane contact. This USP Class VI certified material is suitable for applications that require wear resistance and low water absorption over time. Parts printed with

BioMed Clear Resin are compatible with common sterilization methods. BioMed Clear Resin is manufactured in a facility that follows regulation from the International Organization for Standardization (ISO) and United States Food and Drug Administration (FDA). Specifically, the BioMed Clear Resin is produced in an ISO 13485 facility and is supported with an FDA Device Master File.

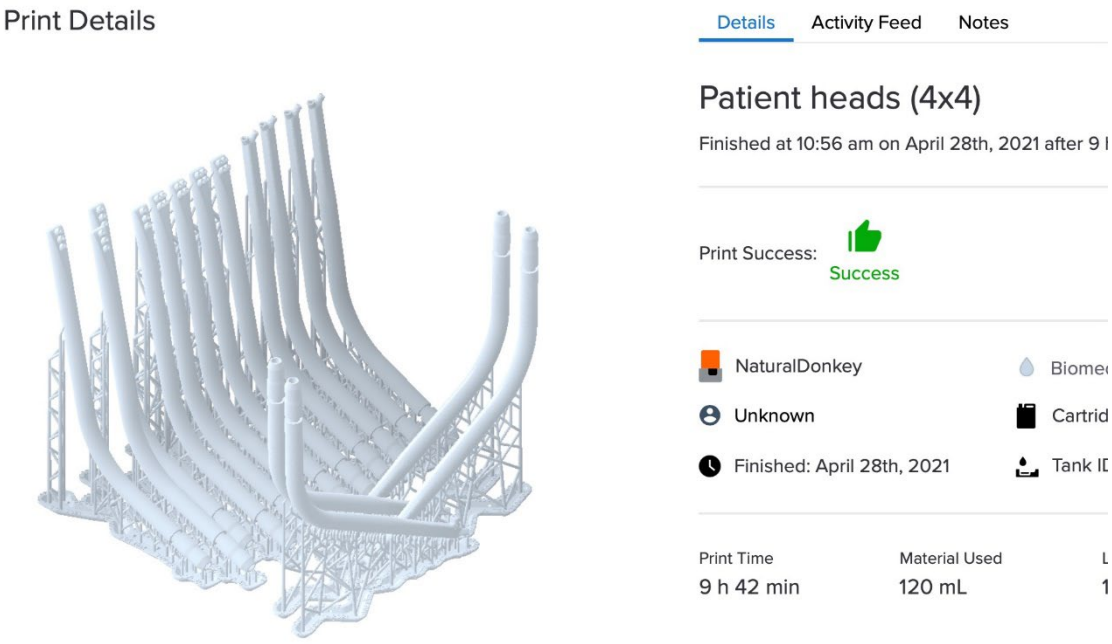

Figure 3.2 Preview of SLA 3D printing software PreForm.

After the printing process is completed, the parts are washed in 99% isopropyl alcohol (IPA) for a 60 minutes to eradicate remaining uncured resin. Although the Form 3B manual states the washing time should be 10 minutes, it is found that 60 minutes is much more effective in removing the stickiness of the uncured resin while producing very smooth surfaces even after removing the supports. The Form Wash machine was selected for its precise cleaning results due to an agitation mechanism embedded in the device. Once the washing is completed, Form Wash automatically lifts parts out of the IPA, avoiding over-soaked, warped prints. The parts air dry and become ready to be cured in the Form Cure.

Figure 3.3 Post-processing machines for SLA 3D printing. Form Wash (left) is an alcohol-washing machine from Formlabs once the print has been completed. Cure (right) is a curing chamber from Formlabs for post-curing processing. Taken from Formlabs, 2022.[52]

The Form Cure is an ultraviolet-radiating chamber to cure the air-dried parts. Form Cure is designed for curing parts printed on the Form 3B. Post-curing maximizes material properties for stereolithography prints, improving the strength and performance. Form Cure precisely combines heat, mirrors, and ultraviolet light to consistently cure the prints. A rotating turntable provides uniform ultraviolet (UV) exposure during the curing process. The following settings were used based on the Formlabs manual to maximize strength of the prints.

Form Cure System	
Degrees	60°C
Minutes	60

Table 3.1 Optimal settings for the post-processing UV-curing machine. BioMed Clear resin requires 60 minutes for a full cure.

3.3. Results

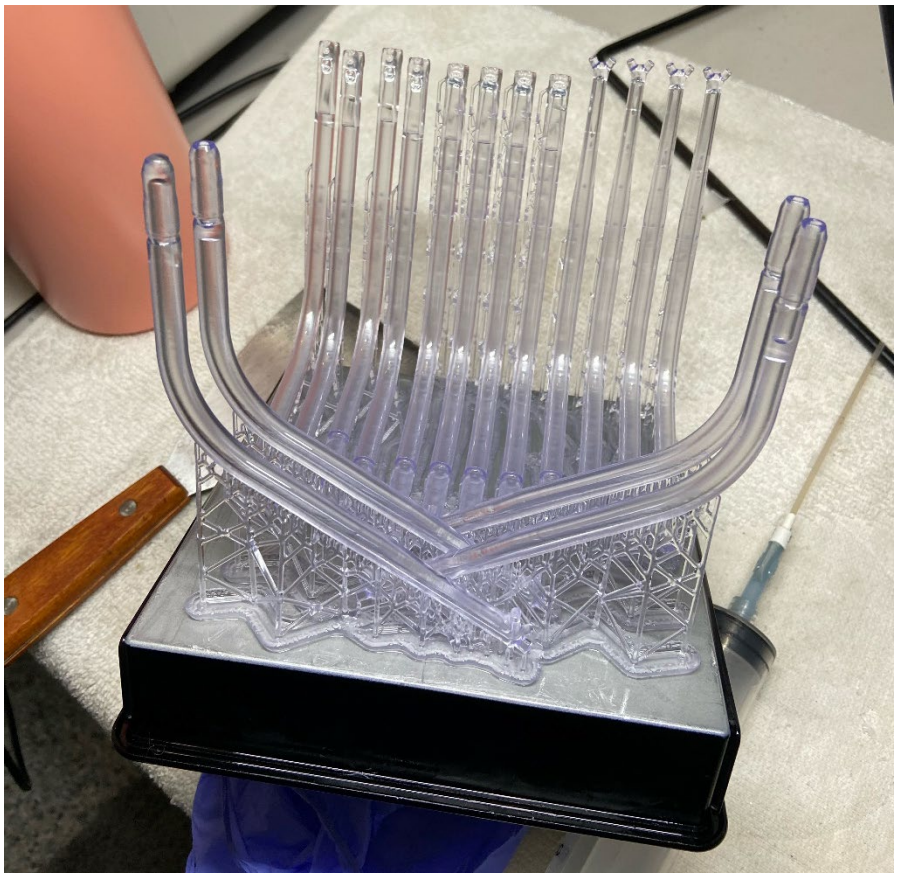

Figure 3.4 A printed set of shower head designs. The shower heads are printed in biocompatible resin using PreForm and the Form 3B.

The shower head designs are printed BioMed Clear Resin over a duration of 10 hours. The shower head designs are arranged in such a way to be able to rapidly produce a maximum amount of sets per print. The orientation and angle of these pieces ensure a high likelihood of successfully printing without errors. The pieces are then washed in 99% IPA for 60 minutes in the Form Wash, trimmed from their supports, polished, then cured in the Form Cure for 60 minutes at 60°C.

Figure 3.5 Demonstration of initial custom shower head design.

The original Waterpik™ shower head has been noted to be painful, uncomfortable, and overwhelming when rinsing the back of the oral cavity even at the lowest setting. The new custom shower head designs are tested with an interchangeable handle to rapidly receive feedback for a comfortable water pressure. The custom shower head design has multiple outlets to distribute the water pressure from the corded model of the Waterpik™. The shower head designs successfully eject liquid at the desired angle of 135° through all three holes (Figure 3.5) and is noticeably more comfortable on the tonsils from self-testing and external feedback.

After designing a functional shower head design and identifying a comfortable water pressure range, the manufacturing of a deep oral irrigation and tonsil cleaning device based on the investigated materials and design requirements begins. The enclosure body of the device is manufactured through the Form 3B SLA 3D printer. The electrical components of the device are wired in a completed circuit (Figure 3.6).

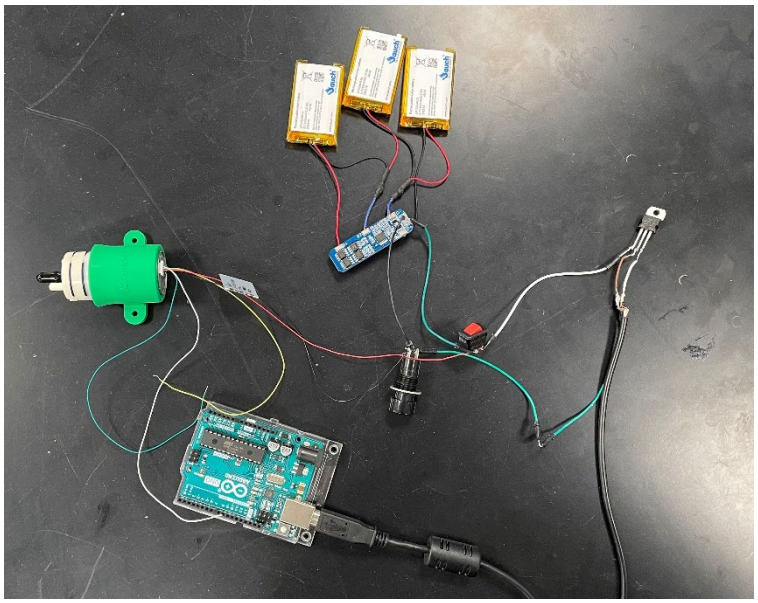

Figure 3.6 Internal electrical design of deep oral irrigation and tonsil cleaning device.

The deep oral irrigation and tonsil cleaning device is then assembled and tested for design validation of the user requirements. The device provides access to the back of the throat and tonsillar crypts, prevent spillage onto the hand during use, cleans the delicate oral tissues with ease in comfort and control, and can be used as a portable device without an outlet. Tonsil stones can be dislodged through the activation of the device once the tonsillar fossa has been accessed behind the tonsillar pillars (Figure 3.7).

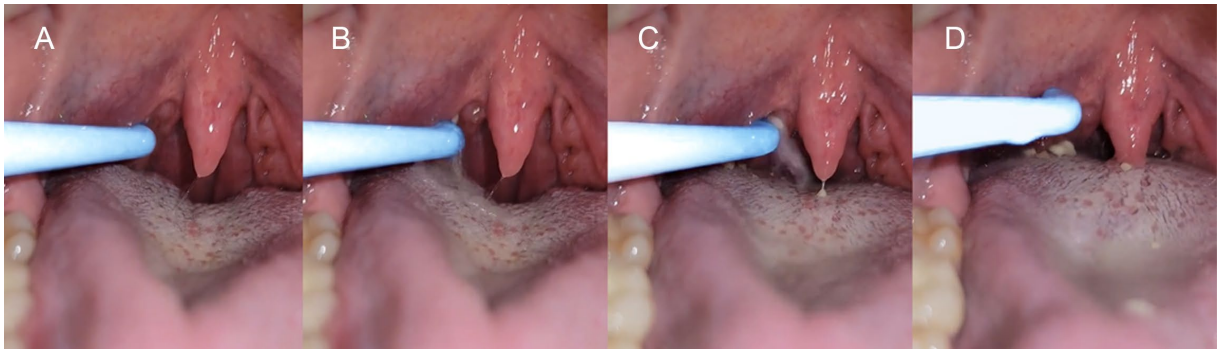

Figure 3.7 Demonstration of dislodging tonsil stones behind the tonsillar pillar. A) Positioning of shower head behind tonsillar pillar. B) Shower head is activated as water streams in and out of tonsillar pillar. C) Mid-ejection of tonsil stone above and around the shower head during activation. D) Dislodged tonsil stones lay below shower head as the user retracts the deactivated device.

CHAPTER 4

Design of a Mouthwash for Antibacterial Activity

4.1. Background

Aerobic bacteria and anaerobes that penetrate into biofilms of the oral cavity are associated with halitosis and oral diseases. For example, *P. gingivalis* and the facultative anaerobic bacterium *S. mutans* harbors and penetrate into hypoxic areas such as the gap between the tongue papilla and mouth, a recognized area of decay and halitosis. Sulfite, methanethiol, butyrate and propionate are the common microbial organic volatile byproducts of decay.[53] Sulfite promotes oral epithelial toxins and are mainly responsible of intra-oral halitosis.[1,54]

There are many commercial oral rinses available in the market to combat halitosis. As covered in Chapter 1, many any of these mouth rinse products essentially consist of CHX, CPC, or ethyl alcohol. Non-alcoholic mouth rinses also contain zinc as zinc has been shown to have antibacterial properties even at lower concentrations which indicate it is safe to be used in oral health products.[55] Zinc is commonly used in dental cements, as a toothpaste ingredient, and has a long and safe history of safe use.[56-58] Zinc is an essential mineral for many human biological functions and is a common mineral found in vitamin supplements. With the intended ability of zinc ions to destroy and/or reduce the malodor of sulfite, an oral rinse containing zinc will be tested against an available bacteria strain found in the oral cavity. Strains of *S. aureus* can be isolated from the oral cavity and was selected to be tested to be the bacterium for this research. An oral rinse solution containing zinc has been tested against *S. aureus* to find the minimal

inhibitory concentration needed to inhibit bacterial growth for this chapter.

4.2. Materials and Methods

A mouthwash solution was diluted to have a concentration of zinc acetate at 0.6%w/w and heated to a temperature of 250˚F (121˚C) for sterilization. *S. aureus* RN4220 were first streaked onto luria broth (LB) agar plates from a 20% (v/v) glycerol:LB frozen stock. Liquid cultures were then grown overnight (~18 h) from single colonies in 3 mL LB, resuspended to a final concentration of 0.1 OD600 in fresh LB (20 g/L), and a 3 mL aliquot sub-cultured for 6 h. Subculture cell suspensions were then diluted to 0.1 OD600 and added to wells of a 96-well plate, followed by filter-sterilized mouthwash/Zn(OAc)2 solutions to bring the final volume to 200 µL. Depending on the experiment, 50 or 100 µL of bacterial suspension were added bringing the final LB concentration to 5 g/L or 10 g/L respectively. The final concentration of cells in each well under all conditions was 0.05 OD600, which corresponds to approximately 8x106 cells/mL.[59] Cultures were grown at 37C for 20 h with OD600 being measured every 5 min with shaking between each read.

4.3. Results

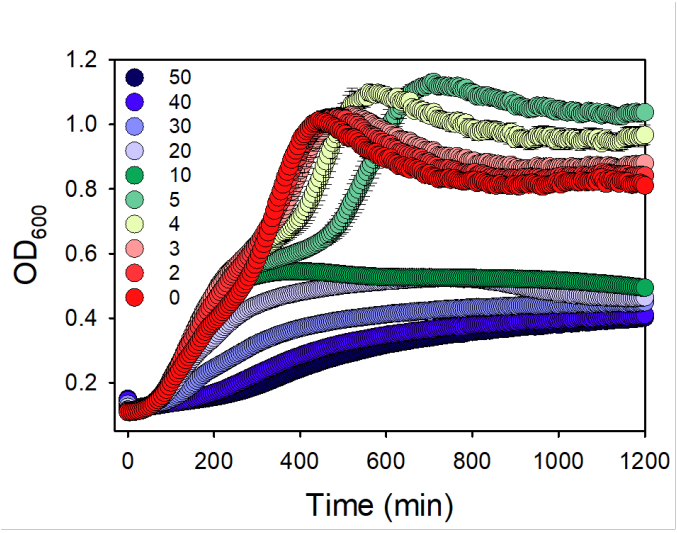

Figure 4.1 Mouthwash solution inhibiting bacterial growth. S. aureus growth profiles in 10 g/L LB with varying concentrations of mouthwash solution. Values in the legend correspond to concentrations of mouthwash solution in % (v/v). Error bars represent standard deviation (n=3).

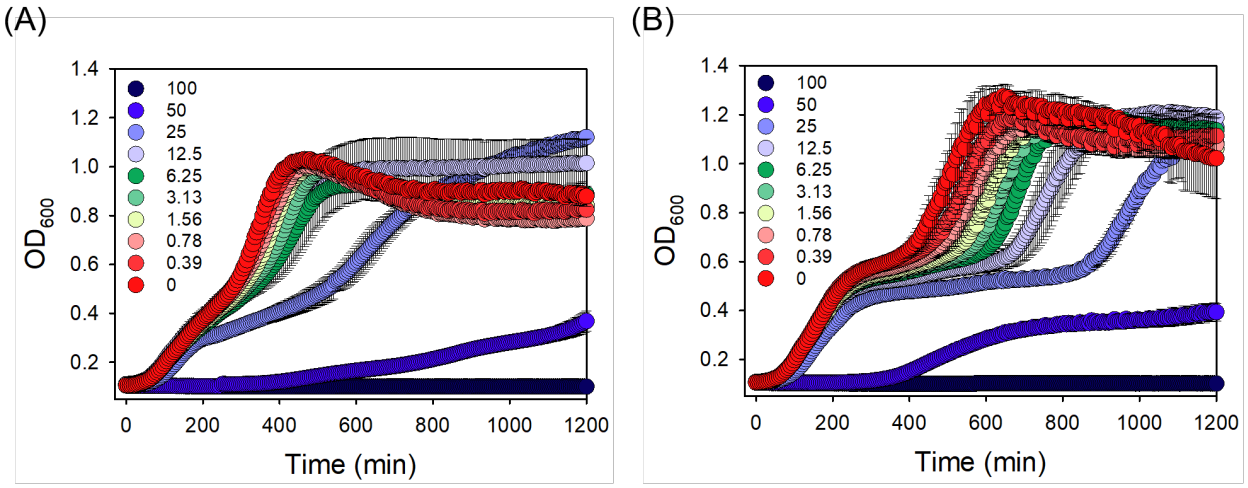

Figure 4.2 Zinc acetate inhibiting bacterial growth. S. aureus growth profiles in (A) LB (10 g/L) or (B) LB (10 g/L) and 10% mouthwash (v/v) with varying concentrations of zinc acetate. Values in the legend correspond to concentrations of zinc acetate in µg/mL. Error bars represent standard deviation (n=3).

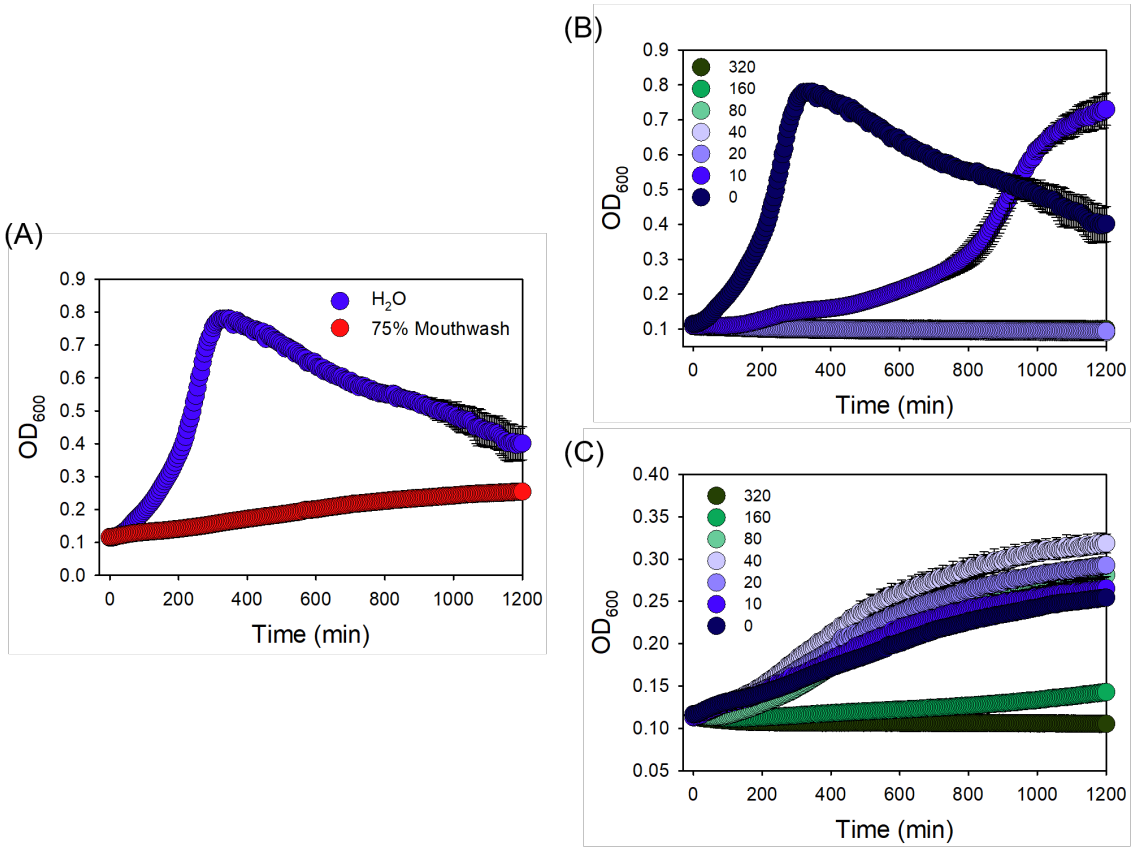

Figure 4.3 Zinc acetate at higher mouthwash concentration inhibiting bacterial growth. (A) Comparison of S. aureus growth in LB (5 g/L) and 75% (v/v) mouthwash or water. Growth in (B) 5 g/L LB or (C) 5 g/L LB and 75% mouthwash at varying concentrations of zinc acetate. Values in the legends correspond to concentrations of zinc acetate in µg/mL. Error bars represent standard deviation (n=3).

Our results show that both mouthwash itself and zinc acetate were successful in inhibiting the growth of S. aureus, a model oral pathogen responsible for a wide variety of diseases including tonsillitis.[60-62] As shown in Figure 4.1, the mouthwash was effective at inhibiting S. aureus growth with a minimum inhibitory concentration for 50% growth (MIC50) of 10% (v/v). To evaluate the antibacterial effect of zinc acetate in the formula, growth was evaluated at varying Zn(OAc)2 concentrations both at the MIC50 of mouthwash and without the presence of mouthwash as shown in Figure 4.2. Under both conditions, the addition of zinc acetate enhanced the inhibitory activity in a concentration-dependent manner. A minimum inhibitory concentration for 90% growth (MIC90) was achieved at 100 µg/mL which did not change in 10% mouthwash.

However, as shown in Figure 4.2B, the 10% mouthwash condition did enhance the growth inhibitory activity as the growth curves exhibited a shift in the onset of stationary phase by ~200 min, and was accompanied by a plateau in the exponential phase. This suggests a synergistic effect from the combination of zinc acetate and mouthwash. These experiments were repeated at elevated (75%) mouthwash concentration to reproduce conditions relevant to practical use. Figure 4.3A compares the growth under this condition with and without 75% mouthwash, where the final concentration of LB is 5 g/L. At this relatively low concentration of LB, a death phase is observed in the control condition as the concentration of nutrients is insufficient to maintain the culture at stationary phase. With 75% mouthwash, a strong inhibitory effect is observed, but not enough to reach an MIC90 similar to that observed at lower concentrations (Figure 4.1). Without mouthwash (Figure 4.3B), zinc acetate addition was able to completely inhibit bacterial growth at much lower concentrations than in more rich media, with an MIC90 of 20 μg/mL. Interestingly, in the presence of 75% mouthwash, this MIC value increased to 320 μg/mL. This is likely due to components of the mouthwash solution such as organic, oxygen or nitrogen-containing molecules coordinating with zinc ions and effectively sequestering them and preventing them from exerting their antibacterial activity. Further studies involving components of the mouthwash solution are required to verify this hypothesis. Although this complicates implementation of this in the proposed device, elevated (1-10 mM) concentrations of zinc acetate can likely achieve the desired growth inhibitory activity without having to reach levels (25-50 mM) that compromise taste.[63] Additionally, viability assays should be performed at these elevated concentrations to establish bactericidal activity at the desired exposure time. Overall, the combination of mouthwash and zinc acetate has shown promising antibacterial activity with great potential to be a future oral wash.

CHAPTER 5

Conclusions

5.1. Conclusions

The experiments for the deep oral irrigation and tonsil cleaning device demonstrated validation of the user requirements. The shower head has been successfully designed to comfortably reach the back of the throat to cleanse the tonsils and nearby regions of interest. The cleansing action of the device was conducted behind the tonsillar pillar and was demonstrated to have the ability to dislodge tonsil stones, a source of halitosis. The bending angle of the shower head demonstrates the ability to rinse the back of the throat without obstructing the user's view and prevented water from spilling back onto the hand. The adjusted pressure force through the PWM module of the pump and design of the tailored shower head received better comfort ratings than a standard oral irrigator at the lowest pressure setting. The momentary push button switch provides control of when these pulsating cycles of fluid can occur. The bacterial studies demonstrated that our zinc-based mouthwash inhibited the growth of bacteria. The mouthwash was effective at inhibiting *S. aureus* growth with a minimum inhibitory concentration for 50% growth (MIC50) of 10% (v/v). The compilation of this data suggests conducting a new study that includes the use of a deep oral irrigation system and zinc-based mouthwash on patients with halitosis. This research has shown benefits for the use of a shower head tailored to the use of rinsing the tonsillar crypts, demonstrated potential for combining use with a zinc-based mouthwash, and has helped lay the groundwork for future studies and devices for treating halitosis.

5.2. Future Work

The compilation of design feedback and antibacterial data shows a great promise for the success of future experiments. Currently, there is an ongoing study at the UCI Medical Center on patients with halitosis for the feedback on the tailored shower heads for their ability to rinse the tonsils and throat without discomfort. The study has recruited 3 patients so far and their follow up visits are pending. There are already planned future studies include an evaluation for using both the shower head and mouthwash for the treatment of halitosis. Other vitamins and ingredients are planned to be included in the mouthwash recipe to enhance the taste and efficacy of the antibacterial properties. The UCI Medical Center has an internal laboratory to rapidly test for minimal inhibition concentrations (MIC) and minimal bactericidal concentrations (MBC) to detect what concentration inhibits the growth of bacteria and kills the bacteria in a certain timeframe, respectively. Thus, there will be another plan for testing a new mouthwash at the UCI Medical Center against bacteria responsible for halitosis and oral disease.

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