CHARACTERIZATION OF FRICTION AND MOISTURE OF PORCINE LINGUAL TISSUE IN VITRO IN RESPONSE TO ARTIFICIAL SALIVA AND MOUTHWASH SOLUTIONS

by

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STATEMENT OF THESIS APPROVAL

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ABSTRACT

Traditionally, oral tribology research involved the use of polydimethylsiloxane (PDMS) as a substitute for lingual tissue. This allowed researchers to construct custom surfaces with different topographies and varying moduli of elasticity. Although PDMS surfaces have been commonly used in oral tribology research, in vitro porcine tongues were used during this research due the similarities with the human tongue. This research focuses on the frictional and moisture effects produced by eleven mouthwash formulations, real and artificial saliva, and moisture variability when applied to porcine tongue tissue samples. Friction was measured using a Butterfly Haptics 6-axis magnetic levitation haptics device equipped with a custom tactor designed to mimic human skin and controlled by a hybrid force/position controller. Moisture was measured with a meter using a relative scale of 0-99 where 0 represents the minimum amount of moisture and 99 the maximum amount.

A comparison of the effects of stimulated human saliva vs. artificial saliva demonstrated the human stimulated saliva had a greater friction coefficient and overall moisture content. Preliminary experiments demonstrated a decrease in friction determined by the amount of moisture present in the surface of the tongue. The friction coefficient and moisture content were discovered to reach average minimum values of approximately 0.35 and 10 respectively. These initial findings were confirmed by expanding the experiment to include a greater number of data points. The eleven mouthwash solutions were tested using 110 porcine tongue tissue samples and produced a lower friction coefficient than the artificial saliva while retaining the approximately the same amount of moisture.

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ACKNOWLEDGMENTS

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CHAPTER 1

INTRODUCTION

Friction and moisture are important metrics when developing a product which is placed in the oral cavity. This research was performed to give the sponsor, Colgate-Palmolive, quantitative data regarding the friction of oral care products. This data can then be used to determine if any correlations exist between the quantitative data found in this research and consumer studies performed by Colgate-Palmolive.

The measurement of friction and moisture within the oral cavity falls under the field known as oral tribology. Traditionally, oral tribology research involved the use of polydimethylsiloxane (PDMS) as a substitute for lingual tissue. This allowed researchers to construct custom surfaces with different topographies and varying moduli of elasticity. Although PDMS surfaces have been commonly used in oral tribology research, in vitro porcine tongues were used during this research due to their similarities to the human tongue.

Friction was measured using a Butterfly Haptics 6-axis magnetic levitation haptics device equipped with a custom tactor designed to mimic human skin and controlled by a hybrid force/position controller. Moisture was measured with a meter using a relative scale of 0-99, where 0 represents the minimum amount of moisture and 99 the maximum amount.

The goals of this research include:

- Testing tongue samples using unstimulated human saliva and artificial saliva and providing a comparison of the formulas.
- Determining the effects of moisture on the friction coefficient by testing a designated number of tongue samples exposed to ambient conditions.
- Providing a statistical analysis of eleven mouthwash formulas by testing 110 tongue samples.

1.1 Background

Oral tribology is the study of friction and lubrication as it pertains to the elements of the oral cavity which are responsible for mastication and transportation of food to the rear of the oral cavity. These elements include the teeth, the temporomandibular joints, the hard and soft palates, and the tongue [1].

In the study of tribology, there are three types of lubrication regimes, which are referred to as boundary lubrication, mixed lubrication, and hydrodynamic lubrication. Boundary lubrication occurs when the interacting surfaces are in contact, and is characterized by having a higher friction coefficient than the other two regimes. The mixed lubrication regime is the transition between boundary and hydrodynamic lubrication, and has characteristics of both. Hydrodynamic lubrication occurs when the load is carried by the lubricating medium and the friction coefficient is dictated by the viscosity of the fluid rather than material surface characteristics [1], [2]. While most engineered systems are designed for use in the hydrodynamic regime in order to decrease wear, oral tribology focuses on the effects of friction in boundary and mixed lubrication regimes [3], [4].

Although oral tribology research focuses primarily on studying the friction coefficient, the methods and goals of researchers are diverse. One study mathematically defined thick, smooth, and slippery as functions of friction in order to develop predictive equations as an aid in evaluating the texture of orally consumable products [5]. Others used panels of individuals to taste and rate the texture of various food products [6], [7]. However, most research tends to focus on using a tribometer to obtain a coefficient of friction.

Since the friction coefficient measured in the boundary and the mixed lubrication regimes depend on the structure of the surfaces in contact, some researchers constructed textured surfaces from polydimethylsiloxane (PDMS) in order to study how surface topography would affect friction. The textured surfaces consisted of hemispherical pins of varied height and density on the scale of micrometers in order to mimic the filliform papillae which increase the friction between the tongue and the food [8].

A different approach to determining the effects of surface topography was used by Bongaerts et al. Instead of constructing textured surfaces from hemispherical pins, the PDMS was cast against sandblasted steel and glass plates which resulted in a textured pattern transfer to the silicone [9]. Dresselhuis et al. used smooth PDMS surfaces with different moduli of elasticity. The surfaces were coated with either an oil and water emulsion or unstimulated human saliva [4]. In addition to PDMS being used as a test surface, Joyner et al. also used uncoated whey protein isolate (WPI) and high density polyethylene (HDPE) coated in an oil and water emulsion [10].

Although PDMS surfaces have been commonly used in oral tribology research, others have elected to use porcine tongues because they closely approximate human tongues. Each have anterior sections which are covered by fungiform papillae scattered between filiform papillae.

Fungiform papillae are mushroom-shaped and contain taste buds. Filiform papillae are the most numerous papillae and increase the friction between the tongue and food. The filiform papillae of human and pig tongues have the same shape and the kertinisation processes are similar [11].

Ranc et. al used in vitro porcine tongues to determine the influence of the salivary layer on the friction coefficient of the lingual mucosa. The specimens were cut so only the anterior one-third remained. Half of these samples were placed in a ventilation box in order to dry, and the remaining half were soaked in human saliva. The tongues soaked in human saliva were found to have lower friction coefficients than those which were uncoated and dried [11]. De Hoog et. al tested a variety of surfaces, including porcine tongue and esophagus, using oil-based emulsions and commercial dairy products [12]. The porcine esophagus was used as the tactor with a normal force ranging from approximately 0.2-2.0 N. With an approximately 0.2 N load, the average friction coefficient was approximately 0.3 with an applied emulsion consisting of 40% oil. Dresselhuis et al. also used porcine tongues coated in an oil and water emulsion as a substitute for human tongues [13].

In addition to the various surfaces tested, a variety of machines have also been used to obtain the friction coefficient. Ranc et al. used a sliding tribometer which moved a steel ball across the surface to be tested [8], [11]. Dresselhuis et al. used a configuration called an optical tribological configuration (OTC). This machine used confocal laser scanning microscopy to optically measure the forces exerted on the surface being tested [2], [13].

DeHoog et al. used a machine which consisted of a rotating shaft with a cantilevered beam. One end of the beam was placed above the rotating shaft and the other end of the beam was attached to a load cell. Tubular samples of material to be tested were placed on the rotating shaft. While the shaft rotated, weights were used to increase the normal load on the free end of the cantilevered beam until it touched the material on the rotating shaft. As the beam touched the material a force was exerted on the load cell and the friction coefficient was calculated from this force [12]. Bonagaerts et al. used a more traditional device which consisted of a spinning plate and a pin with sphere on the end. As the plate started to spin, the pin was lowered onto the plate. The spinning motion of the plate caused the sphere to spin once contact had been made and the force exerted on the sphere was measured with a force transducer [9].

De Wijk and Prinz tested the lubricating qualities of vanilla custard and mayonnaise with a machine which consisted of a rubber band, an electric motor, and a load cell. One end of the rubber band was looped around a cylinder attached to the electric motor while the opposite end was connected to a load cell. The rubber band was coated with the substance being tested. When the machine was switched on, friction between the cylinder and the rubber band produced a force measured by the load cell [14].

Rheometers measure the flow of liquid in response to an applied force and are commonplace in many food laboratories. In order to mitigate the cost of buying a tribometer, Goh et al. adapted a rheometer to be used as a tribometer [15].

The research described in this thesis implements a magnetic levitation haptic device with a custom tactor designed to mimic human skin. The machine is able to provide reciprocating motion at a constant velocity while being able to maintain a constant normal force. This device was used in previous research, also funded by Colgate-Palmolive, to quantitatively characterize human skin in vivo after being exposed to various shower gel products. The goal of the project was to determine if any correlations existed between the quantitative data and the qualitative data obtained from consumer studies [16].

In the research, Yardley used a PIV hybrid force/position algorithm to control the magnetic levitation haptic device. The setup was used to measure the friction, dynamic skin stretch, and viscous damping produced by eight body wash products after being applied to 32 test subjects. The first two of these metrics were measured after the body wash had been applied, rinsed off, and finally when the skin had dried. Viscous damping was measured only after the body wash had been applied to the subject's skin.

Friction was determined by moving the tactor across the subject's forearm while maintaining a constant force 0.1 N and velocity of 2 mm/second. Dynamic skin stretch consisted of using system identification techniques to determine the mass, stiffness, and damping constant of the skin. During testing, a series of 26 frequencies were used to change the desired position and a 0.5 N normal force was exerted on the tactor while moving a distance of 0.5 mm. The viscous damping produced by the various body wash products was measured after a selected product had been applied to the subject's skin. A 0.1 N normal force was applied to the tactor with an initial velocity of 4 mm/second over a distance of 16 mm. This process was repeated twice, after which the velocity was doubled up to 16 mm/second.

Yardely demonstrated the magnetic levitation haptic device could be used to determine the friction produced before and after a medium was applied to human skin. Since this research uses in vitro tissue samples and liquid products, dynamic skin stretch and viscous damping are not relevant. As a result the primary focus is on determining the frictional and moisture effects produced by various mouthwash solutions.

CHAPTER 2

METHODOLOGY

2.1 Experimental Setup

2.1.1 Magnetic Levitation Haptic Device

The tribometers used in oral tribology research have usually operated using circular movement relative to a polydimethylsiloxane (PDMS) surface in place of tongue tissue. Although this setup has traditionally been used, it has been argued that a machine which produces a sliding motion more closely mimics the mechanical action of the tongue within the oral cavity [4], [13].

The setup used in this research uses a Butterfly Haptics Magnetic Levitation Haptic Device (MLHD) and custom tactor (Figure 2.1). This setup was used for a previous research project which measured the haptic response of in vivo human skin after various body wash products had been applied [16].

The MLHD is a six degree-of-freedom (6-DOF) device which uses six Lorentz actuator coils to produce rotational and linear forces on a central component known as the floater. While in operation the floater of the MLHD has no physical contact with the rest of the device, and as a result there is no friction or backlash produced. The MLHD is capable of moving the floater in a 24mm spherical diameter with a position resolution of <2.0 microns and a position bandwidth of 140 Hz. The MLHD is also capable of producing a maximum force of 40 N at a bandwidth of >2000 Hz with a resolution of 20 mN.

Since the MLHD does not come equipped with a force/torque sensor, the floater of the MLHD was modified to accept an ATI Nano-17 6-axis force/torque sensor. The sensor was fastened between two aluminum plates approximately 968 mm^2, with the tactor bolted to both aluminum plates and a brass coupler secured to the bottom plate (Figure 2.2). The tactor consists of a hemispherical cap nut with a 15.875 mm diameter and a 1 mm coating of NuSil Med 4014 silicone to mimic the texture of human skin [16].

To protect the internal electronics of the MLHD from the liquid solutions applied to the tongue samples, a 0.127 mm thick sheet of flexible transparent plastic was placed over the MLHD. A hole was cut in the center of the plastic to allow the sensor and the tactor to pass through and make contact with the surface of the tongue sample being tested.

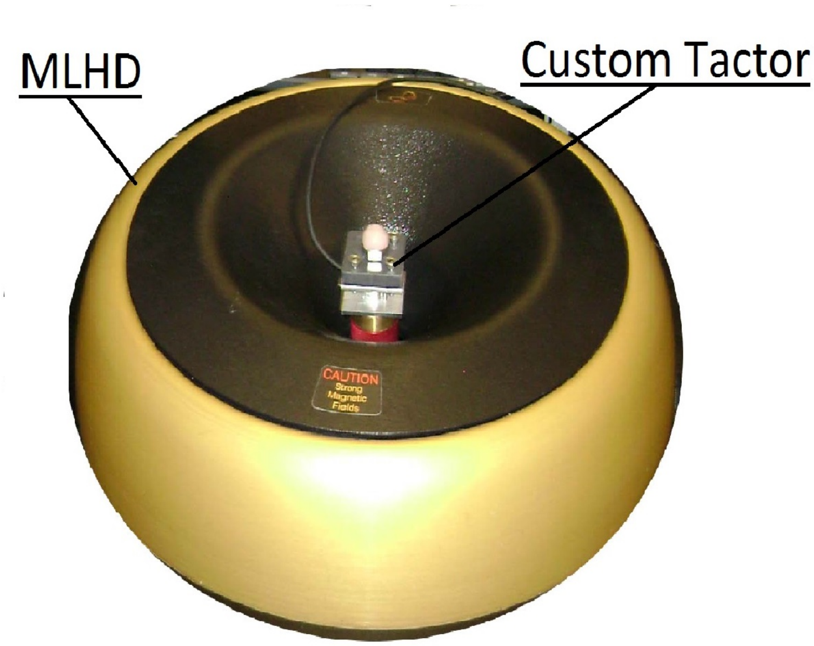


Figure 2.1. Butterfly Haptics 6-DOF Magnetic Levitation Haptic Device with custom tactor. The device uses six Lorentz coils to produce rotational and linear forces on the floater. While in operation the floater has no physical contact with the device. As a result no friction or backlash is produced.

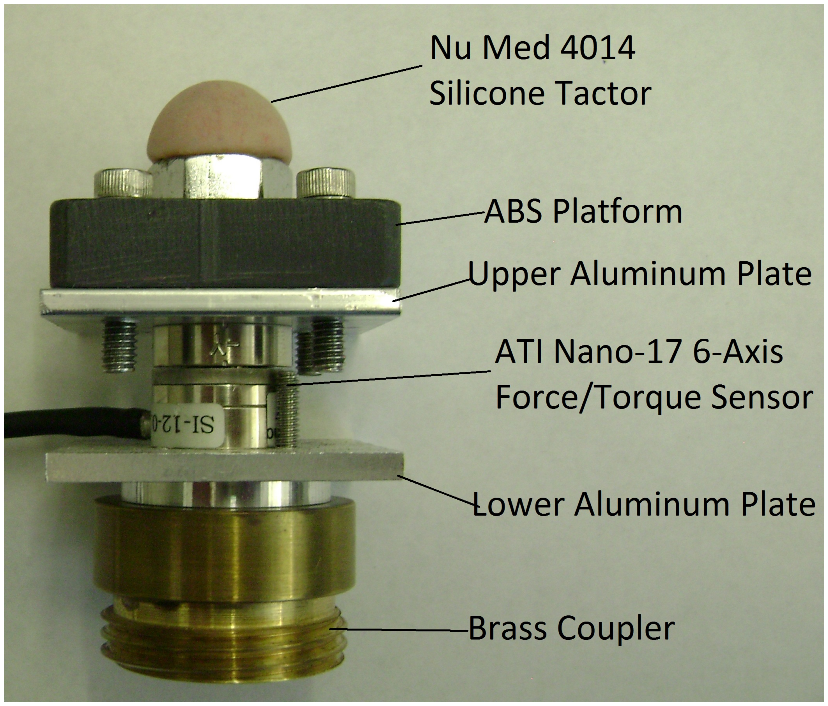


Figure 2.2. Custom silicone tactor attached to an ATI Nano-16 force/torque sensor. The tactor consists of a hemispherical cap nut with a 15.875 mm diameter and a 1 mm coating of NuSil Med 4014 silicone to mimic the texture of human skin.

In order to test in vitro porcine tongues with the MLHD, a platform was constructed to hold the tongue sample and allow the tactor to make contact with it. The platform was made from a 762 mm X 203 mm X 3.2 mm aluminum plate with a 38.1 mm X 19 mm slot milled in the center to expose a section of the tongue sample to be tested (Figure 2.3). The dimensions of the slot were experimentally determined. The platform was secured to a pair of laboratory jacks by fitting two screws into each jack. The screws were placed through a pair of corresponding holes located at each end of the platform, and the position of the jacks was maintained by using masking tape to mark two square areas on the desk housing the MLHD (Figure 2.4). It was determined through trial and error that the optimum height of the platform is 114 mm.

Once a tongue sample was secured to the platform, the control algorithm was activated and the tactor moved across the surface of the tongue sample. The tactor moved a length of 22 mm ten times while maintaining a constant normal force of 0.1 N and a velocity of 1 mm/second. Since the length of the MLHD workspace is 24 mm along the x-axis, the 22 mm stroking distance allows a maximum amount of data to be collected while maintaining a safe distance from the edge of the workspace. Prior work by Ranc et al. had used a sliding tribometer with a 0.1 N normal force and a 1 mm/second velocity on in vitro porcine tongues without any damage occurring to the tongue [11].

A PIV hybrid force/position control algorithm was implemented to maintain constant velocity and force while minimizing error. Hybrid force/position algorithms are used to control either force or position of an axis. Since the MLHD can produce linear or rotational movement about each axis, there are six degrees of freedom which must be specified as force or position control.

In the typical block diagram force and position are controlled by a selection matrix (Figure 2.5). A selection matrix is a diagonal matrix which either contains a one or a zero. Each element of the diagonal represents one of the degrees of freedom. The matrix is multiplied by a six element column vector representing position. If an element in the diagonal of the selection matrix is zero, the result of the matrix multiplication negates position control for that degree of freedom. Since the selection matrix is used for position control, an identity matrix is subtracted from the selection matrix and the result is used for the force controller.

Since the control algorithm was programmed in object oriented C++, a vector of six boolean values was used in place of the selection matrix. The six boolean values were set to either true or false. The six values represent one of the MLHD's six degrees of freedom. If a value is true, position control is enabled. If a value is false, force control is enabled. The hybrid position/force controller has two matrices of gain values.

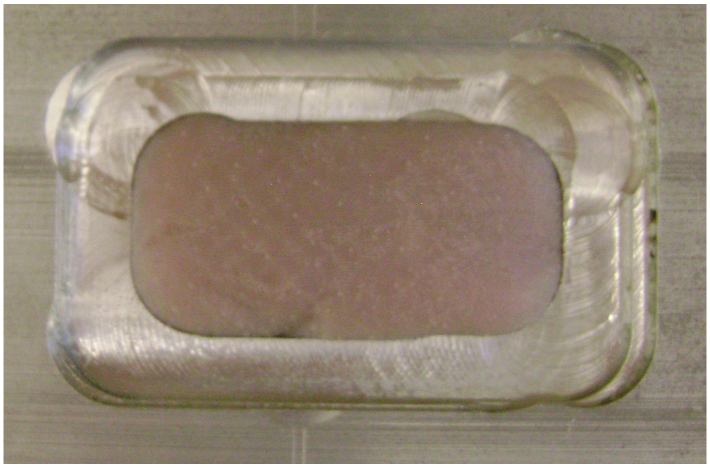


Figure 2.3. Test area of a tongue sample. The 38.1 mm X 19mm slot was milled in the center of a 762 mm X 203 mm X 3.2 mm aluminum plate. The tongue sample is fastened to the aluminum plate and the slot exposes the section to be tested.

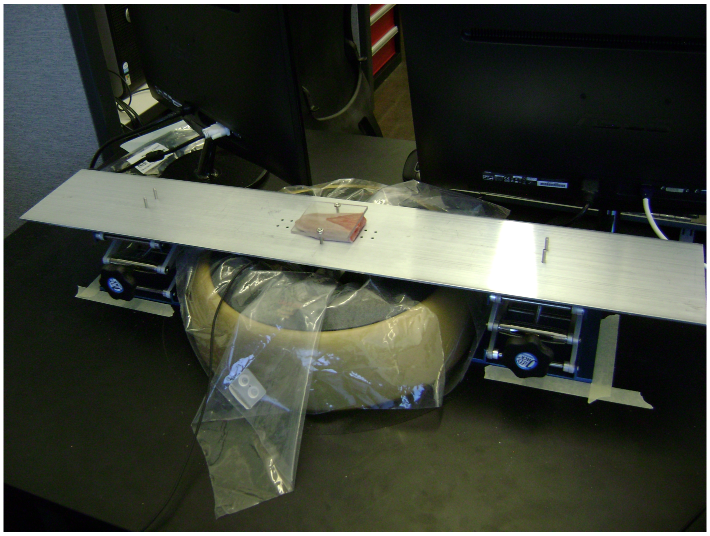


Figure 2.4. Experimental setup. The experimental setup consists of the MLHD, custom tactor (not seen), jack stands, and aluminum platform. The tongue sample is secured to the center of the aluminum platform. A slot in the center of the platform allows the tactor to make contact with the tongue sample. The jack stands are fitted with two screws which fit through corresponding holes in the platform. The screws secure the platform to the jack stands and masking tape was used to mark proper placement of the jack stands.

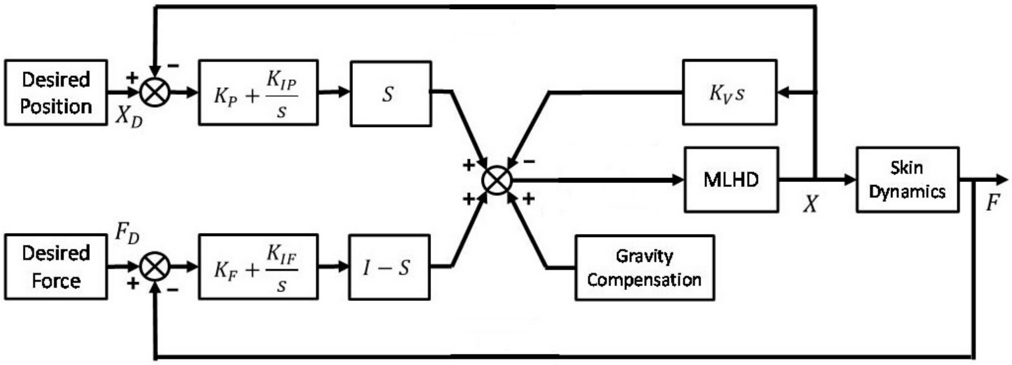


Figure 2.5. Block diagram of PIV hybrid force/position control algorithm.

One matrix corresponds to position control and the other to force control. The values for the gains were tuned experimentally. The hybrid position/force controller was programmed in C++ on a client computer running Linux. The client computer is connected via an Ethernet connection to a server computer running QNX which is connected to the MLHD. Data from the force sensor are acquired by a Sensoray 626 data acquisition card and stored on the client computer.

2.1.2 MoistSense II Skin Moisture Meter

A MoistSense II skin moisture meter was used to obtain the moisture content of the tongue samples. The meter is manufactured by Moritex (Figure 2.6) and operates by using an interdigital capacitor (IDC) as the moisture sensor (Figure 2.7). Each of the lines in Figure 2.7 corresponds to a plate in a capacitor and is either negatively or positively charged. The IDC is separated from the skin by an insulating layer of plastic, which protects it from damage as well as preventing a conductive connection to the skin. When the capacitor is charged, a scatterfield is produced at the edges of the plate, and if a surface containing water is brought into the scatterfield, the capacitance is increased. The depth to which the scatterfield can penetrate is dictated by the thickness of the insulating layer and the spacing between the conductive material of the IDC [17]. The MoistSense II moisture meter is activated by pressing it against the skin and waiting for an audible beep. Once the meter beeps, an integer in the range of 0-99 is displayed representing the moisture content. Zero represents no moisture and 99 the maximum amount of moisture. The average moisture content of the tongue sample was acquired by taking three separate readings over the test area.

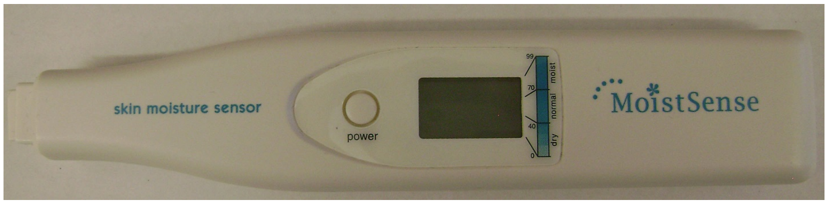


Figure 2.6. MoistSense II skin moisture meter manufactured by Moritex.

2.2 Experimental Procedures

2.2.1 Preparation of Materials

Porcine tongues closely approximate human tongues. Each have anterior sections which are covered by fungiform papillae scattered between filiform papillae. Fungiform papillae are mushroom shaped and contain taste buds. Filiform papillae are the most numerous papillae and increase friction between the tongue and the food. The filiform papillae of the human and porcine tongues also have the same shape and similar keratinisation processes, and the interpapillary epithelium is parakeratotic [11].

Porcine tongues from animals 6-9 months old were purchased by Colgate-Palmolive and shipped overnight in a foam cooler containing ice. Once received, the tongues were individually packaged with a food grade vacuum sealer, and stored in a freezer at approximately −20 °C.

In order to prepare the tongues for testing, each one was thawed, rinsed with cold tap water, patted dry with a paper towel, cut approximately between 38 mm and 51 mm from the back of the tongue, and the remaining portion was cut in half (Figures 2.8). Each half of the tongue constitutes one tongue sample. Each tongue sample was placed on the aluminum platform with a plastic plate bolted on top to secure the sample (Figure 2.9). The aluminum platform was subsequently placed with the center of the exposed region over the tactor. The MLHD was set to ten strokes with a length of 22 mm and a normal force of 0.1 N. Each tongue sample was tested in ambient temperature and humidity.

In addition to porcine tongues, Colgate-Palmolive provided artificial saliva and eleven different mouthwash products. The artificial saliva consists of one part mucin and one part buffer solution. Each mouthwash product was labeled by Colgate-Palmolive, and it was not disclosed which products, if any, were competitors' brands, and which were produced by Colgate-Palmolive.

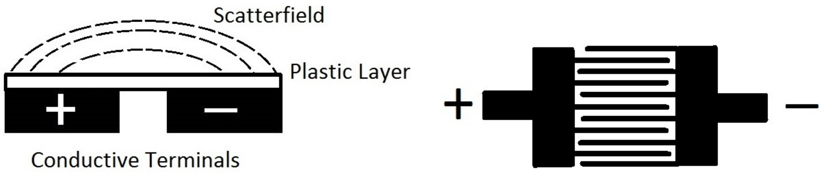


Figure 2.7. Side and top views of the interdigital capacitor (IDC).

2.2.2 Comparing Real and Artificial Saliva

Approximately 15 mL of human saliva was collected from a male donor 39 years of age. In order to separate the saliva from any solid material, a test tube containing the saliva was placed in a centrifuge for 5 min at 5000 RPM. Twenty tongue samples were chosen at random and split into two groups of ten. Each tongue sample was treated with 0.5 mL of artificial saliva or 0.5 mL of human saliva. This resulted in having ten tongue samples tested using artificial saliva and ten tongue samples tested using human saliva. The solutions were applied using a small paintbrush to ensure even distribution. Once the coatings were applied, tests were performed to measure moisture and friction.

2.2.3 Preliminary Experiments

2.2.3.1 Artificial Saliva and Moisture Variability

A pilot study was conducted with two groups of tongue samples. One group was used to determine the frictional effects of moisture variability and the other group was used to determine the effects of artificial saliva. Four tongue samples were selected randomly and exposed to ambient conditions in 15 minute intervals over a 45 minute period in order to reduce the skin moisture content. The friction coefficient of each tongue sample was tested at the end of each 15 minute interval. The remaining four tongue samples were soaked in 0.5 mL of artificial saliva for 15 minutes, with an additional 0.5 mL added at each test interval. The MoistSense II skin moisture meter had not been acquired at the time of this test, and as a result moisture content was not measured.

2.2.3.2 Mouthwash Solutions

Initial experiments with the mouthwash solutions involved four mouthwash formulations provided by Colgate-Palmolive. Each formulation was tested with three tongue samples. Each tongue sample was dried in ambient conditions for approximately 45 minutes to minimize the moisture content. After 45 minutes the tongue samples were tested to determine moisture content and the friction coefficient. The tongue sample was subsequently removed from the platform, evenly coated with approximately 0.5 mL of artificial saliva and 0.5 mL

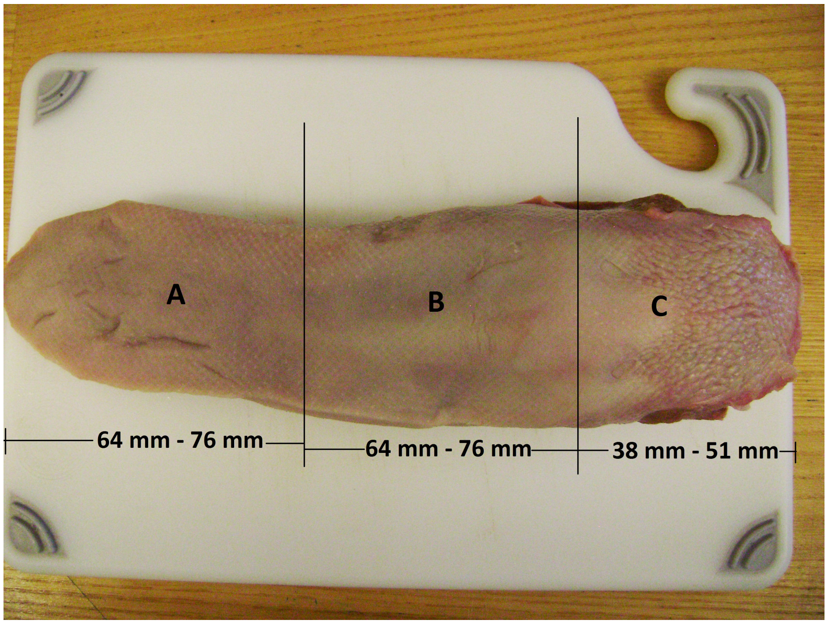


Figure 2.8. Whole porcine tongue. The tongue is cut into three sections. Section C is discarded while A and B are used for testing purposes.

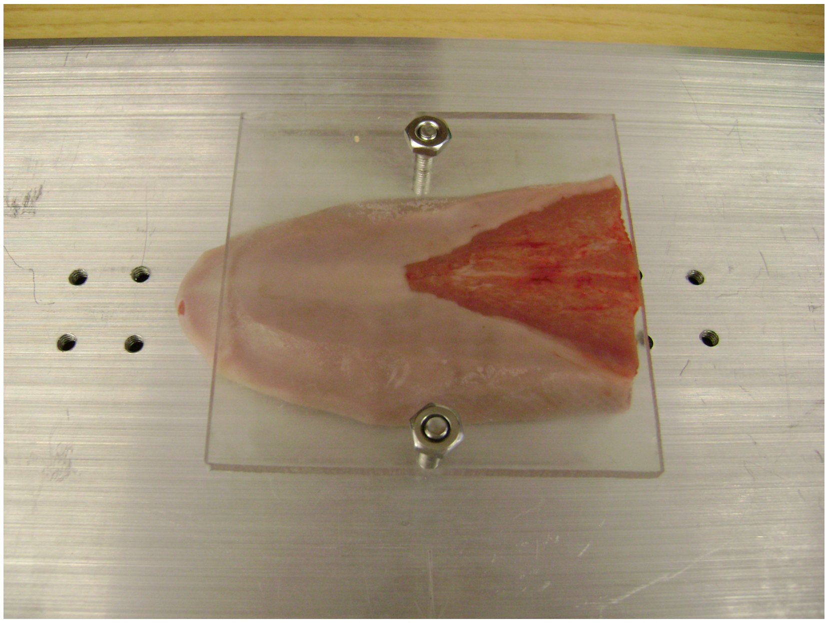


Figure 2.9. A tongue sample secured to the aluminum platform.

of a selected mouthwash. The tongue sample was subsequently tested again to determine the moisture content and the friction coefficient.

2.2.4 Final Experiments

2.2.4.1 Moisture Variability and Artificial Saliva

The preliminary experiment gives a general idea of the frictional effects exhibited by the surface of the tongue after being treated with artificial saliva and after the moisture content is reduced. However, it is limited in scope due to the lack of data concerning moisture content prior to testing the friction coefficient, and the limited number of intervals at which the testing occurred. As a result, the time interval was narrowed to 10 minutes over the course of an hour and an initial test was conducted at 0 minutes. Since the testing occurred at seven different time points, it was decided to test each tongue sample only once in order to eliminate the possibility of decomposition altering the results. Four tongue samples were tested at each time point, giving a total of 28 tongue samples tested in each group.

2.2.4.2 Mouthwash Solutions

It was decided to expand the preliminary mouthwash experiment using 110 tongue samples, and to add seven additional mouthwash solutions. As before, each tongue sample was dried in ambient conditions for 45 minutes, after which the moisture content and the friction coefficient were determined. These tongue samples were not treated with artificial saliva or mouthwash and are referred to as untreated. Once each of the tongue samples had been tested, a coating of 0.5 mL of artificial saliva was evenly applied with a small paintbrush and testing was conducted again. The final test consisted of evenly applying a 0.5 mL coating of a randomly selected mouthwash solution with a different small paintbrush. Ten tongue samples were tested for each of the eleven groups.

2.3 Data Analysis

After testing was completed, a linear piecewise model was constructed from the data using a constrained linear regression add-in for Matlab. Figure 2.10 shows an enlarged segment of the data acquired. The red line represents the linear piecewise model, the green vertical lines mark two regions which will be used to fit the model to the data, and the blue markers represent the raw data. Using this model, an average friction coefficient was calculated.

The horizontal portions of the data represent the measurements taken as the tactor slides across the surface of the tongue sample. Once the data was processed and an average friction coefficient had been obtained, a Lilliefors test was conducted in order to establish that the

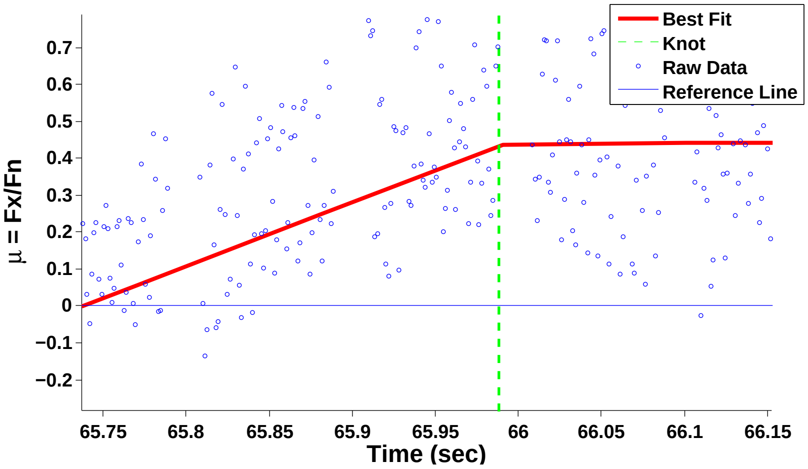


Figure 2.10. Close up view of the linear piecewise model. The linear piecewise model is represented by the red line and the raw data is represented by the blue dots. The horizontal portions represent the tactor sliding across the surface of the tongue sample.

data were normally distributed. After the data were verified to be normally distributed, a one-way Analysis of Variance (ANOVA) test was conducted for a between-subjects design using the statistics toolbox in MatLab. ANOVA is a statistical test which compares the means of two or more groups with normally distributed data to determine if there is at least one group which is statistically significantly different from the rest of the groups [18]. A statistically significant result is obtained when the resulting p-value is less than the significance level [19]. If the difference is determined to be statistically significant, the null hypothesis can be rejected as being true. The null hypothesis states that no difference exists among the groups being tested, and if an ANOVA test rejects it as being true, post hoc testing must be used to determine which groups are different. A significance level of 0.05 was used with the statistical testing described in this thesis.

Since statistical significance can always be obtained by testing a large enough sample size, it only tells part of the story. If a statistical difference is found, it is considered necessary to give an effect size [18], [19], [20]. An effect size essentially describes the magnitude of the difference between the two groups. Two normally distributed data sets with different means will still contain values which are common to both data sets. The Common Language Effect Size (CLES) compares the means and standard deviations of two groups and returns a percentage value which indicates the probability of one of the data sets containing a larger value, assuming the values are chosen at random. If the two groups being compared have mean values which are the same or very close, the value returned will be 50%. This indicates the mean values of the two normally distributed data sets have the same value.

CHAPTER 3

RESULTS

3.1 Comparing Real and Artificial Saliva

The data indicate artificial saliva has a lower average friction coefficient and does not provide as much moisture as real saliva (Figures 3.1 and 3.2). A t-test along with the Common Language Effect Size (CLES) was used to analyze the differences. With regards to the friction coefficient the t-test returned a p-value of 0.189. Since the p-value was not less than the alpha value of 0.05, the null hypothesis cannot be rejected.

Using CLES, the real saliva solution was determined to have 65% probability of having a greater friction coefficient. The same methods were also applied to the different values regarding moisture. This time, the two-sample t-test returned a p-value of 0.02, and the real saliva solution was determined to have 78% probability of having a greater moisture value.

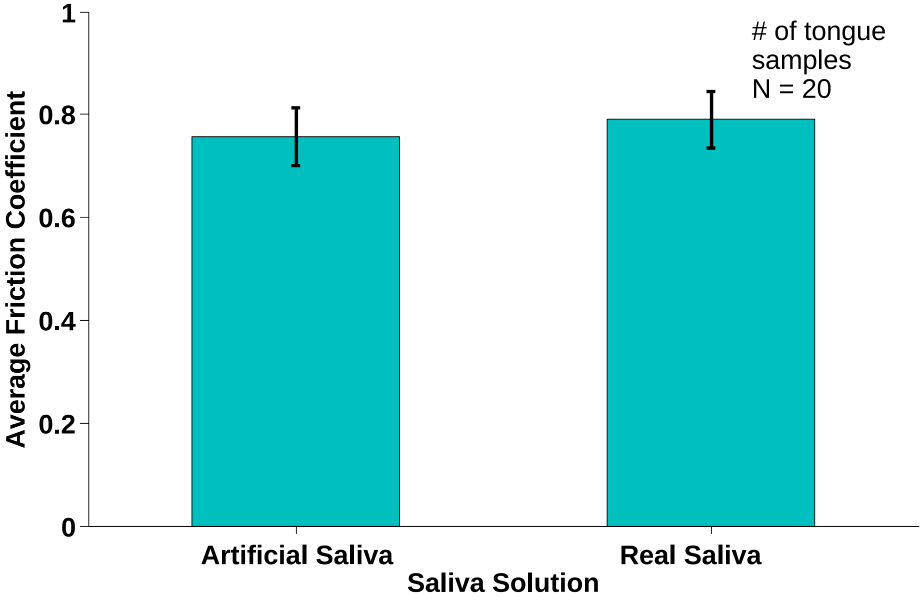


Figure 3.1. The average friction coefficient produced by artificial and real saliva.

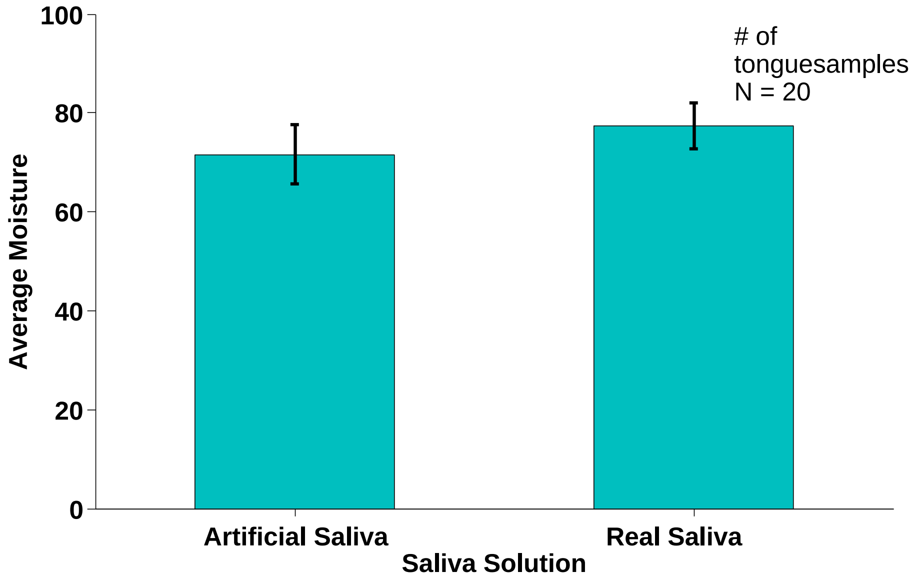


Figure 3.2. The average moisture provided by real and artificial saliva.

3.2 Preliminary Experiments

3.2.1 Moisture Variability and Artificial Saliva

The data indicate that the untreated tongue samples show a greater reduction over time in the friction coefficient vs. the tongue samples treated with artificial saliva (Figure 3.3). The term untreated refers to tongue samples which were not treated with either artificial saliva or mouthwash. The untreated tongue samples were exposed to ambient conditions for the indicated time in order to reduce the skin moisture content. The friction coefficient of the untreated tongue samples decreased from approximately 0.7 to 0.5 during the 15 to 30 minute interval. From 30 to 45 minutes the friction coefficient decreases again, but not as dramatically. The tongue samples treated with artificial saliva also show a decrease in the friction coefficient at each time point. However, the decrease appears almost constant when compared to the untreated tongue samples.

3.2.2 Mouthwash Solutions

A comparison of the data revealed that the average friction coefficient increased after the artificial saliva and mouthwash had been applied to the tongue samples (Figure 3.4). At first glance the differences seem minor. Looking at the data comparing the average moisture content of the two states reveals that the tongue samples contain approximately four times more moisture after being treated with mouthwash and artificial saliva (Figure 3.5).

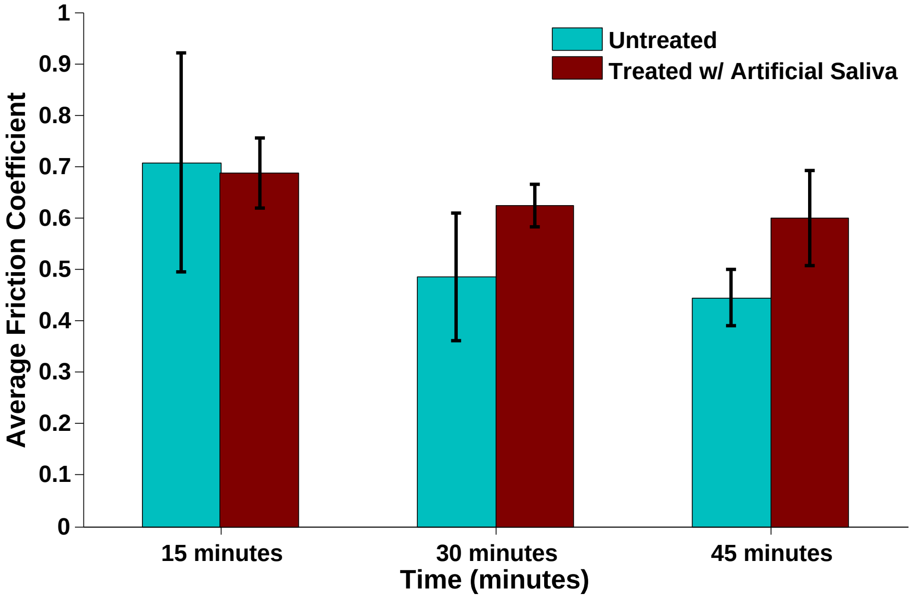


Figure 3.3. Preliminary experiment design: The average friction coefficient produced by moisture variability and artificial saliva with 95% CI bars.

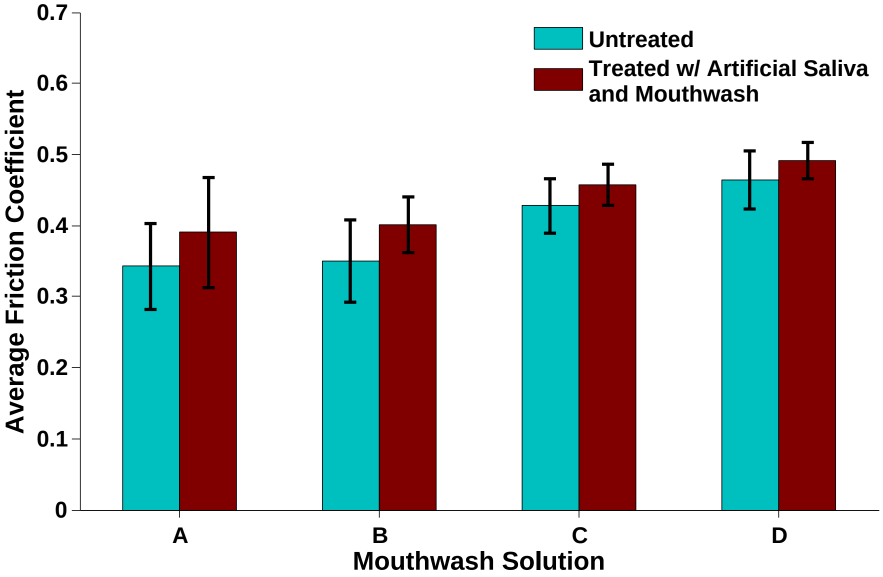


Figure 3.4. Preliminary experiment design: The average friction coefficient produced by four mouthwash solutions with 95% CI bars.

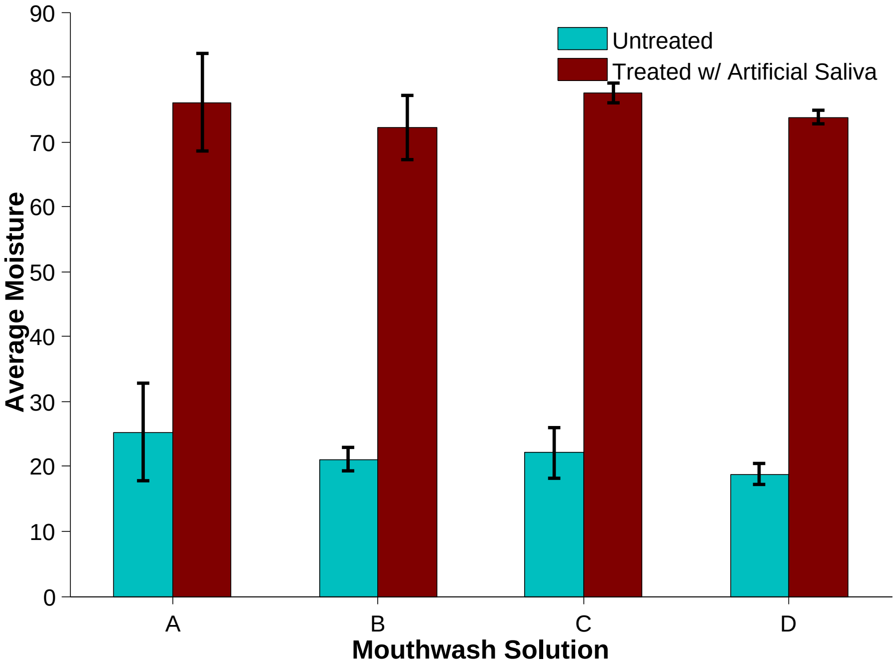


Figure 3.5. Preliminary experiment: The average moisture provided by four mouthwash solutions with 95% CI bars.

However, it is well known the friction coefficient of skin is determined by the amount of moisture it contains [1]. Due to this relation, it was initially thought the artificial saliva and mouthwash would increase the friction coefficient more than indicated due to the amount of moisture provided. However, friction increases only if a lubricating medium such as mouthwash or artificial saliva is not present. Since the number of samples used for this experiment do not provide a statistical analysis with adequate power, statistical analysis could not be used to determine the differences.

3.3 Final Experiments

3.3.1 Moisture Variability and Artificial Saliva

While the preliminary experiment verified that the friction coefficient of skin decreases as the skin dehydrates, it was decided to broaden the number of time points and include moisture data. The data show the same trend as the preliminary data, but on a larger scale. The untreated samples initially exhibit a larger friction coefficient, but as time passes, moisture evaporates from the tongue samples, and as a result the friction coefficient decreases to a steady value of approximately 0.35. The data also show that the tongue samples treated with artificial saliva do not exhibit a predictable trend (Figures 3.6 and 3.7). Since the

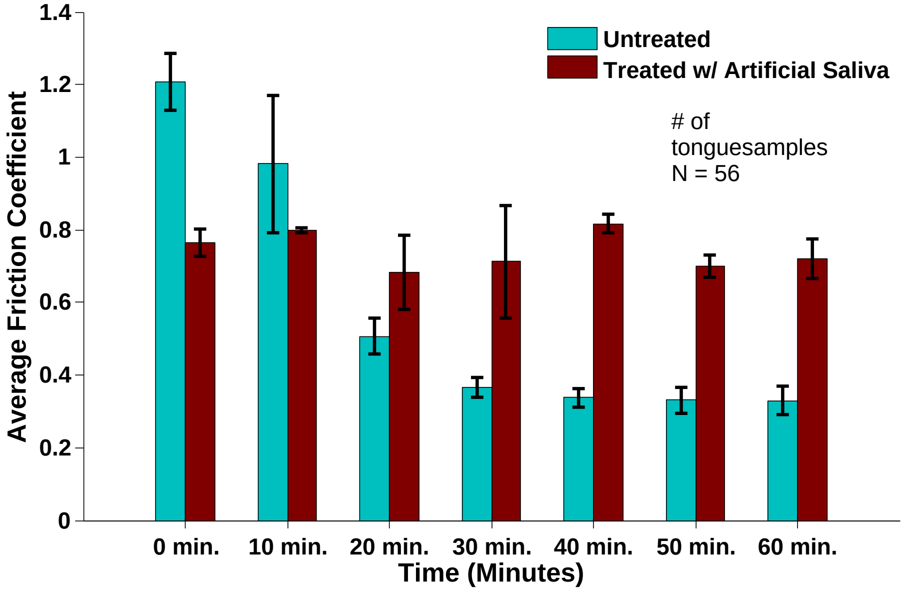


Figure 3.6. Final experiment design: The average friction coefficient produced by moisture variability and artificial saliva with 95% CI bars.

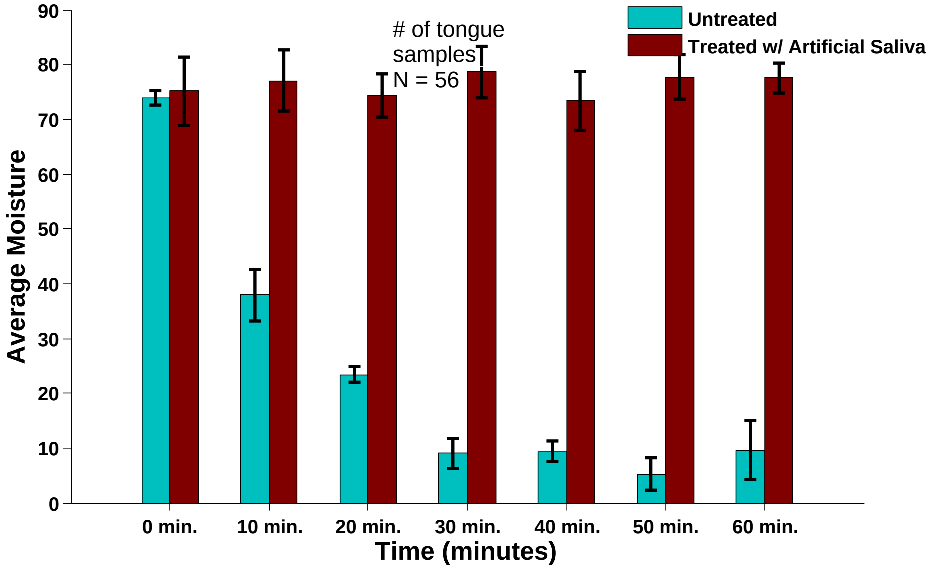


Figure 3.7. Final experiment design: The average moisture produced by artificial saliva and moisture variability with 95% CI bars.

moisture content of the tongue samples is relatively constant when compared with the friction coefficient, it is assumed the variances are in the artificial saliva and testing procedures.

3.3.2 Mouthwash Solutions

The preliminary experiment compared the differences in friction and moisture of a tongue sample before and after a treatment of artificial saliva and mouthwash. Since the prior experiment did not include a test to determine the effects of artificial saliva without mouthwash, the final experiment was designed with this in mind.

The data show the untreated state provides the lowest moisture and friction coefficient. This result is consistent with previous experiments, which demonstrate that the friction coefficient of skin decreases as moisture content decreases. After the artificial saliva was applied, the friction coefficient and moisture content more than doubled. The mouthwash solutions do not appear to affect the moisture provided by the artificial saliva, but a decrease in the friction coefficient is apparent. Figures 3.8 and 3.9 show the mouthwash solutions by descending order of the average friction coefficient. Figure 3.10 shows the relative difference between the friction coefficient produced by artificial saliva and the friction coefficient produced by artificial saliva and mouthwash.

To determine which mouthwash solutions, if any, provide a statistically different friction coefficient, a one-way ANOVA test was used. The ANOVA test reported at least one statistically significant difference among groups tested (F[10, 99]=13.9, $p < 0.01$). A separate ANOVA test was used to determine if any of the groups in Figure 3.10 were statistically different. This was done in order to determine if the changes provided by the mouthwash solutions were statistically different. At least one statistically significant difference was found (F[10, 99]=2.64, $p < 0.01$).

In order to determine which specific groups were statistically different, a Tukey-Kramer post hoc test was performed after both ANOVA tests. The results of the post hoc test and the CLES values are shown in Table 3.1. This table indicates which of the groups treated with artificial saliva and mouthwash are statistically different, as well as the effect size. The table can be read by cross referencing the numbers on the right with those on the top. Table 3.2 contains the CLES values for the ANOVA test comparing the groups in Figure 3.10, and can be read the same way. The boxes contain the color green if the post hoc test found a statistically significant difference between two mouthwash formulas, and red if none was found. The value found in each box represents the probability the formulation listed on the right will have a greater friction coefficient than the formulation listed on the top. For example, a green box containing a value of 88% is found by cross referencing A with D in

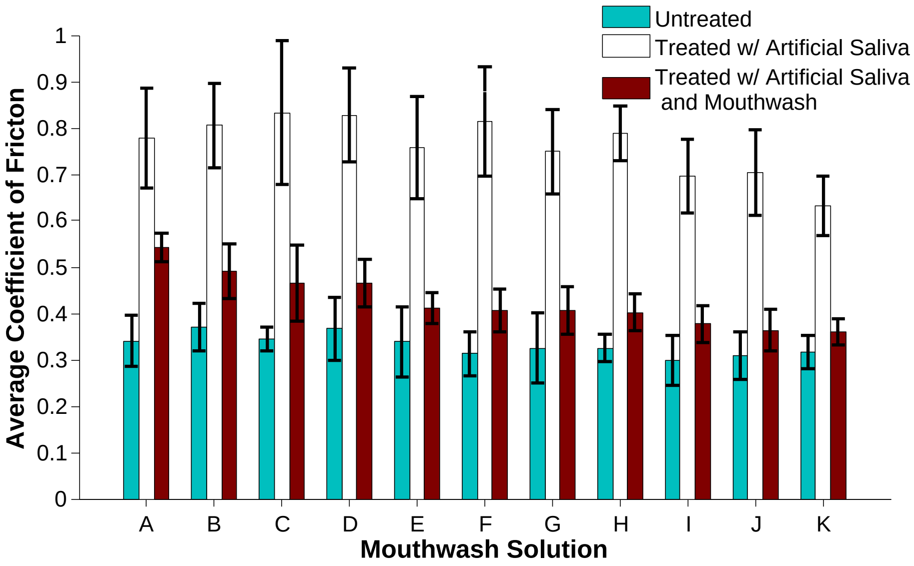


Figure 3.8. Final experiment design: The average friction coefficient produced by eleven mouthwash solutions with 95% CI bars.

Table 3.1. This represents a statistically significant difference with an 88% probability A will have the greater friction coefficient than D. It must also be noted that effect size has no bearing on whether or not a statistically significant difference is found. As a result some of the data will not yield a statistically significant difference, but will have an effect size as large as 88%, which could be the result of variance in the testing procedures.

Since Table 3.2 shows only two statistically significant differences, it would be easy to interpret the results as indicating that the change provided by all but two mouthwash solutions are different. However, it must be kept in mind that the standard deviation is much larger for the tongue samples treated with artificial saliva vs. those treated with artificial saliva and mouthwash (Figure 3.11). This effectively means that any changes provided by the mouthwash solutions are closer to absolute, and not relative changes.

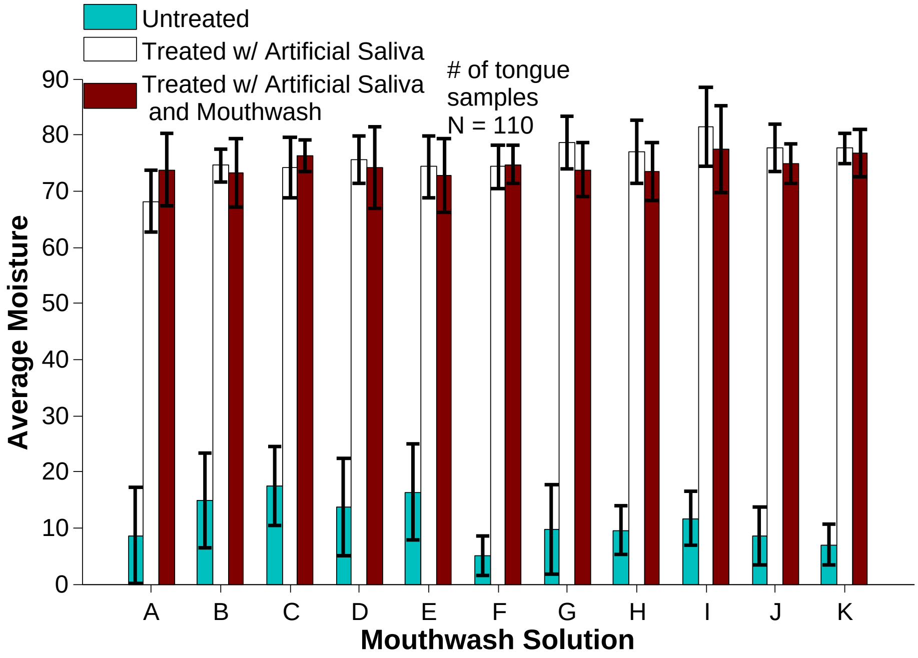


Figure 3.9. Final experiment design: The average moisture provided by four mouthwash solutions with 95% CI bars.

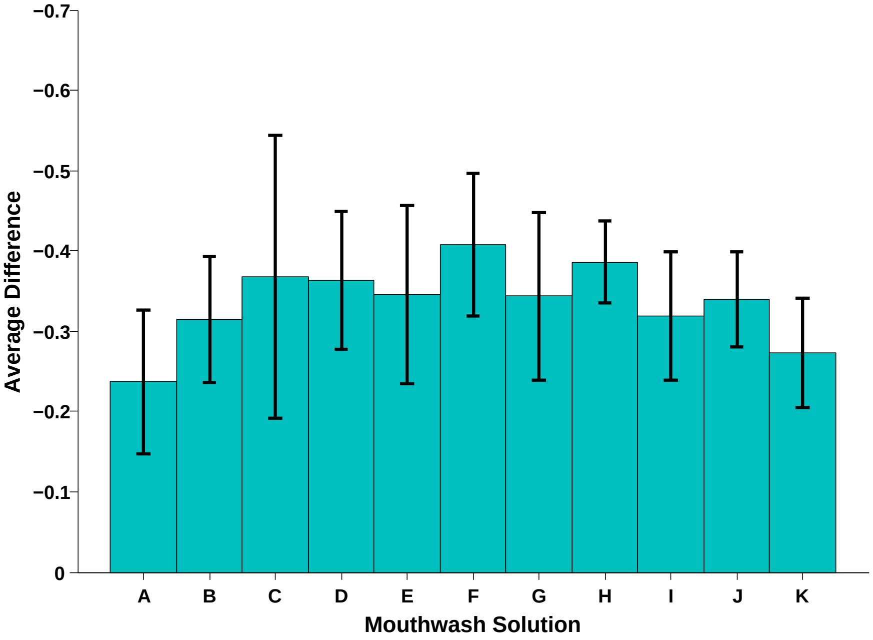


Figure 3.10. The change in friction between the tongue samples treated with artificial saliva and those treated with artificial saliva and mouthwash.

Table 3.1. Common Language Effect Size Values of the Friction Coefficient Provided by Mouthwash and Artificial Saliva.

	A	B	C	D	E	F	G	H	I	J	K
A	50%	77%	79%	88%	100%	99%	99%	100%	100%	100%	100%
B		50%	58%	60%	88%	86%	85%	90%	94%	96%	98%
C			50%	50%	75%	74%	73%	78%	84%	87%	90%
D				50%	84%	81%	80%	86%	92%	95%	100%
E					50%	50%	50%	58%	72%	82%	88%
F						50%	50%	57%	69%	78%	82%
G							50%	56%	68%	77%	80%
H								50%	64%	75%	79%
I									50%	63%	66%
J										50%	50%
K											50%

0.341	0.77905	0.54206	**Total-02**	0.056	0.1075	0.0309	**5**
0.37	0.80544	0.49143	**1*54099-**	0.0514	0.0911	0.05947	**2**
0.344	0.83302	0.46564	**BTN-052**	0.0253	0.1558	0.08189	**1**
0.367	0.82782	0.46507	**DF-0520**	0.068	0.1016	0.05115	**3**
0.339	0.75769	0.41117	**1*48086-**	0.0747	0.11	0.03289	**4**
0.313							
0.320							
0.326							
0.299							
0.308							
0.316							

Table 3.2. Common Language Effect Size Values of the Changes in Friction Provided by the Mouthwash Solutions.

	F	H	C	D	E	G	J	I	B	K	A
F	50%	58%	58%	66%	66%	70%	74%	77%	80%	89%	91%
H		50%	54%	62%	63%	67%	74%	77%	80%	92%	93%
C			50%	52%	54%	56%	56%	60%	62%	70%	74%
D				50%	53%	56%	58%	63%	67%	79%	83%
E					50%	53%	54%	60%	63%	77%	78%
G						50%	50%	56%	59%	71%	71%
J							50%	50%	54%	71%	77%
I								50%	54%	68%	75%
B									50%	65%	72%
K										50%	60%
A											50%

	Total-0	1*5409	BTN-052	DF-052	1*4808	DF-CO-	MH	DF-HSO	MHA	MHH	MJH
tal-0211	50.0%	77.2%	78.8%	87.9%	99.8%	99.0%	98.5%	99.7%	99.9%	100.0%	100.0%
54099-SA		50.0%	57.8%	60.1%	88.1%	85.6%	84.5%	89.6%	93.8%	95.9%	97.6%
-052014-SA			50.0%	50.0%	75.2%	73.8%	73.2%	77.9%	83.9%	87.3%	89.8%
052014-SA				50.0%	83.8%	80.7%	79.5%	85.9%	91.7%	94.7%	99.6%
48086-SA					50.0%	50.0%	50.0%	57.7%	72.1%	81.6%	87.8%
CO-1*64505						50.0%	50.0%	56.5%	68.8%	78.0%	82.1%
MHCA							50.0%	56.1%	67.7%	76.7%	80.2%
SO-1*64760								50.0%	63.9%	74.7%	79.3%
MHACA									50.0%	63.0%	65.9%
MHHCA										50.0%	50.0%
MJHCA											50.0%

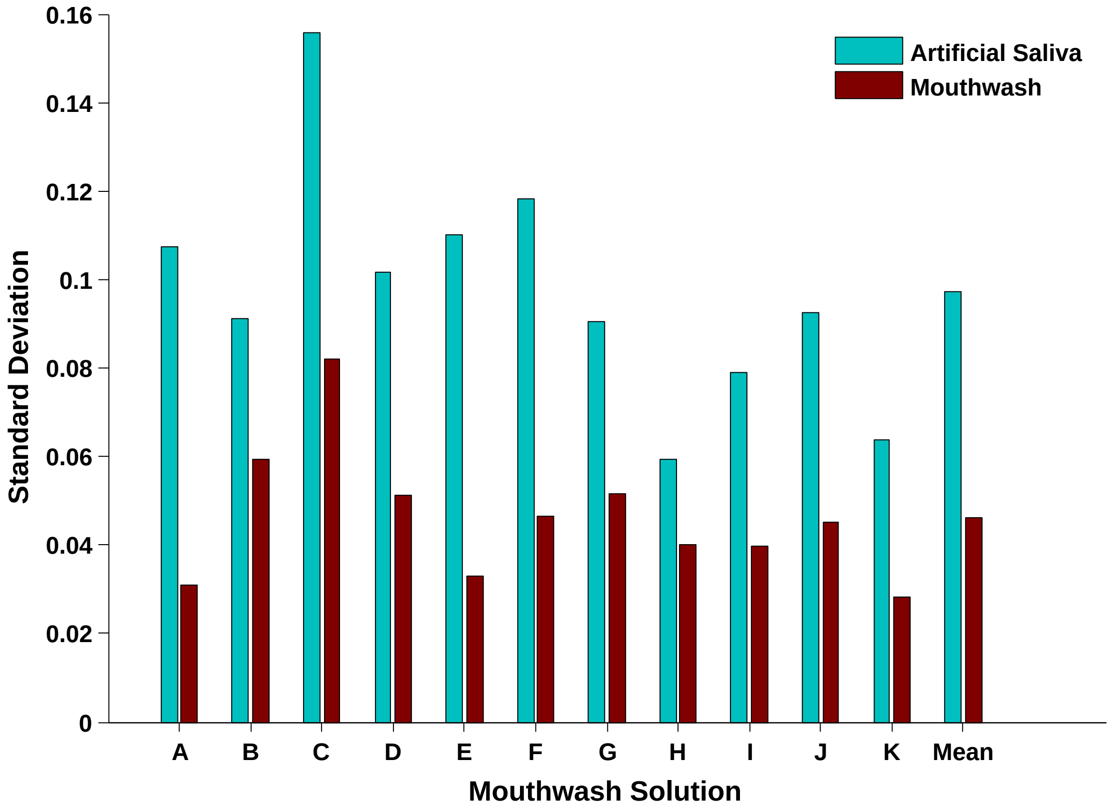


Figure 3.11. The standard deviation of each group after being treated with artificial saliva, and artificial saliva and mouthwash. The last group, labeled "Mean", represents the mean values of the standard deviation.

CHAPTER 4

CONCLUSION

4.1 Overview of Work

In vitro porcine tongues were used during this research to determine the frictional and moisture effects of eleven mouthwash formulations, real and artificial saliva, and moisture variability. Friction was measured using a magnetic levitation haptics device equipped with a custom tactor designed to mimic human skin and controlled by a hybrid force/position controller using PIV control.

The comparison of real and artificial saliva indicates the artificial saliva produces a lower moisture content and friction coefficient. The statistical analysis indicated that the moisture difference between the two groups is statistically significant with 95% confidence, but the friction coefficient fails to meet this criteria. Despite this failure to meet statistical significance at the specified confidence level, the fact that the friction coefficient and moisture content of the artificial saliva is lower may still be relevant, considering the previously mentioned relation between moisture and friction. The lower values might be attributable to the fact that unstimulated saliva has a lower friction coefficient than stimulated saliva [21]. Stimulated saliva is collected after mastication occurs and corresponds to the real saliva used in this research, while the artificial saliva provided by Colgate-Palmolive may closely approximate unstimulated saliva.

In addition to determining the differences between real and artificial saliva, preliminary experiments with a small number of samples provided insight into the general effects produced by moisture variability and artificial saliva. The data indicate the friction coefficient decreased in the untreated samples as the surface moisture in the tongue sample decreased. The friction coefficient of the tongue samples soaked in artificial saliva also decreased over time, but not as quickly as the untreated samples.

Since a moisture meter was not available at the time of the preliminary test, the experiment was repeated after a MoistSense II moisture meter was acquired. The experiment was repeated using a larger sample size and smaller time intervals. The data for this experiment show that the untreated tongue samples initially show a larger friction coefficient when compared with those treated with artificial saliva. However, as the moisture evaporates from the tongue samples, the friction coefficient decreases to a steady value of approximately 0.35. The data also show that the tongue samples treated with artificial saliva do not exhibit a predictable trend. Since the moisture content of these tongue samples is relatively constant when compared with the friction coefficient, it is assumed the variation in the friction coefficient is due to variances in the artificial saliva and testing procedures.

While the tongue samples treated with artificial saliva have an average friction coefficient in the range of 0.70-0.80, the mouthwash solutions produce a lower friction coefficient. Preliminary testing revealed the mouthwash solutions produced an average friction coefficient of 0.39-0.49 but retained the moisture content of the artificial salivary layer. The final experimental design added a test to measure the friction coefficient and moisture content after the artificial saliva had been applied. This test was added in order to have a direct comparison of the effects of the artificial saliva before the application of the mouthwash solution.

The preliminary experiment compared the differences in friction and moisture of a tongue sample before and after a treatment of artificial saliva and mouthwash. Since the prior experiment did not include a test to determine the effects of artificial saliva without mouthwash, the final experiment was designed with this in mind. It was also decided to expand the experiment to include seven additional mouthwash solutions. Each mouthwash solution was tested using ten tongue samples, giving a total of 110 tongue samples tested.

The ANOVA and Tukey-Kramer post hoc test revealed which groups had statistically significant differences in the friction coefficient after being treated with mouthwash and artificial saliva. These tests were also performed on the data in Figure 3.10. The common language effect size (CLES) was used to describe the magnitude of these differences. Table 3.1 shows the magnitude of the differences in the friction coefficients provided by the mouthwash solutions. Comparing the number of differences with the ones listed in Table 3.2, it can be seen that while the mouthwash solutions may have statistically significantly different friction coefficients, the changes provided by the same solutions do necessarily provide the same statistical results. These differences in statistical significance can be attributed to the fact the friction coefficient provided by artificial saliva has a larger standard deviation than the mouthwash solutions. This effectively means that any changes provided by the mouthwash solutions are closer to absolute and not relative changes. A post hoc power analysis of the data in Figure 3.8 and Figure 3.10 reveals the power of tests to be at 1.0 and 0.97 respectively.

Although the magnitude of these differences is described, it is unknown how significant they are in terms of the sensory capabilities of the human tongue. At the time of this writing no studies could be found which describe quantitatively the sensitivity of the human tongue to friction. As a result, it is unknown whether the average human tongue can differentiate between a friction coefficient of 0.54 and one of 0.36.

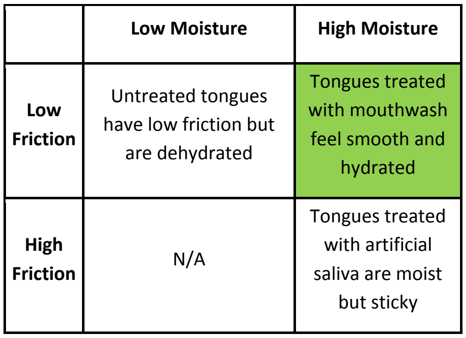
However, Table 4.1 summarizes the hypothesis that a tongue with high moisture and low friction will feel smooth and hydrated, while those with low moisture and low friction may feel smooth and dry, and tongues with high moisture and high friction may feel moist and sticky.

Future work on this subject could be expanded to include a sensory study using a panel of individuals to determine quantitatively the sensitivity of the human tongue to friction. In vitro porcine tongues were used in this study due to the similarity they share with human tongues. This study could also be expanded to perform testing on porcine tongues in conditions that closely mimic the human oral cavity in terms of humidity and temperature.

4.2 Contributions

While other studies have used in vitro porcine tongues [8], [12], this study is believed to be unique in using a Magnetic Levitation Haptic Device combined with a force-feedback control algorithm as a tribometer. In addition, no other oral tribology research could be found which used a tactor which replicated the texture of skin. Also, while most studies merely recorded and compared the mean friction coefficient values produced by lubricating mediums, this study used statistical analysis to determine whether any of the differences measured were significant with a 95% confidence level.

Table 4.1. Hypothesis of the effects moisture and friction.



	Low Moisture	**High Moisture**
Low Friction	Untreated tongues have low friction but are dehydrated	Tongues treated with mouthwash feel smooth and hydrated
High Friction	N/A	Tongues treated with artificial saliva are moist but sticky

APPENDIX

MATLAB SCRIPT USED TO PROCESS DATA

```
%Chop Data
data_begin = find(time < 5.5);
data_begin = data_begin(length(data_begin));
data_end = find(time> 115);
data_end = data_end(1);
uchop = u(data_begin:data_end);
timechop = time(data_begin:data_end);

%Find where the slope changes
d_desp = diff(Desp(:,1));

% Determine the sign of each element
sign_d = sign(d_desp);

%We should be left with 10 non-zero elements
diff_sign_d = diff(sign_d);

%Store values in a vector
vec = find(diff_sign_d  = 0);

%Construct a vector for the knots
%This loop determines where two knots are very close together. Since we
%only want one knot at any one location, this eliminates one of those
%knots and places a zero in the vector.
j = 1;
for i = 1:length(vec)
    if i == length(vec)
```

```
    pvec(j-1) = time(vec(j-1));

elseif abs(vec(i+1)-vec(i)) > 2
    pvec(j) = time(vec(j));
    j = j + 1;

elseif abs(vec(i+1)-vec(i)) <= 1
    j = j+1;
end
end

%Find the non-zero values of the pvec vector
ovec = find(pvec  = 0);
%Contstruct a new vector from the non-zero values of pvec
for i = 1:length(ovec)
    nvec(i) = pvec(ovec(i));
end

%lvec provides a knot value between the diagonal portions of each segment.
%THE VALUE 'A' MAY HAVE TO BE ADJUSTED FOR EACH DATA SET
A = 2.5;
lvec = nvec(2:length(nvec)-1)+A;

%This finds the average the two previous knots. This provides a middle
%point
mvec(1) = 0.5*(nvec(2)+nvec(1));
for i = 2:length(nvec)-1
    mvec(i) = 0.5*(lvec(i-1)+nvec(i+1));
end

%initialize the knot vector with the first three values
kvec=[nvec(1) mvec(1) nvec(2)];

%Fill the knot vector with the appropriate values
k= 3;
```

```
for i = 1:length(lvec)
    kvec(k+1) = lvec(i);
    kvec(k+2) = mvec(i+1);
    kvec(k+3) = nvec(i+2);
    k = k+3;
end

%The knot vector becomes the values previously assigned plus the last value
%of 115.1. This value provides an end point in order to contain the data
%set.
jvec = kvec(end)+A;
hvec = 0.5*(jvec+115.1);
kvec =[kvec jvec hvec 115.1];

%invoke the slmengine. 'pred' is the the vector containing the predicted
%values along the curve and 't' is the corresponding time vector.
[slm,t,pred] = slmengine(timechop,uchop,'degree',1,'plot','on','knots',kvec);

%plot the trendline without the data
figure; plot(t,pred,'r'); axis equal;

%Isolate the slip regions
b = 3; c = 4;
for i = 1:length(nvec)
    if c <= 33
    vec1 = find(t <= kvec(b));
    vec2 = find(t >= kvec(c));
    p1(i) = vec1(end);
    p2(i) = vec2(1);
    b = b + 3;
    c = c + 3;
    end
end

%All the regions where slip is measured are put into a vector called
```

```
%slipvec
slipvec = [pred(1:p1(1)), pred(p2(1):p1(2)), pred(p2(2):p1(3)), ...
          pred(p2(3):p1(4)), pred(p2(4):p1(5)), pred(p2(5):p1(6))...
          pred(p2(6):p1(7)), pred(p2(7):p1(8)), pred(p2(8):p1(9))...
          pred(p2(9):p1(10)), pred(p2(10):p1(end))];

slipvec = abs(slipvec');

%Find the average coefficient of friction. Do not use the last point.
slipavg = mean(slipvec)
```

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