Mechanical Loading for Modifying Tissue Water Content and Optical Properties

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ABSTRACT

The majority of the physical properties of tissue depend directly on the interstitial or intracellular concentration of water within the epidermal and dermal layers. The relationship between skin constituent concentrations, such as water and protein, and the mechanical and optical properties of human skin is important to understand its complex nature. Localized mechanical loading has been proven to alter optical properties of tissue, but the mechanisms by which it is accomplished have not been studied in depth.

In this thesis, skin’s complex nature is investigated experimentally and computationally to give us better insight on how localized mechanical loading changes tissues water content and its optical properties. Load-based compression and subsequent increased optical power transmission through tissue is accomplished to explore a relationship between localized mechanical loading and tissue optical and mechanical properties. Using Optical Coherence Tomography (OCT), modification of optical properties, such as refractive index, are observed to deduce water concentration changes in tissue due to mechanical compression. A computational finite element model is developed to correlate applied mechanical force to tissue strain and water transport. Comprehensive understanding of the underlying physical principles governing the optical property changes within skin due to water concentration variation will enable future development of applications in the engineered tissue optics field.
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Chapter 1

Introduction

Tissue optics, which includes light-based therapeutic applications, treatment, and diagnostic techniques, has recently received considerable attention in the scientific and engineering communities. The effectiveness of light-based systems is limited by their inability to deliver optical radiation in deep tissue due to its highly scattering nature. Focused or near-collimated light only penetrates a few millimeters in turbid tissue such as skin before it diverges outward analogous to automobile headlights in fog. Until recently, optical properties of biological tissues were considered fixed. Engineered tissue optics is a new research area that allows reversibly altering light scattering within naturally turbid tissues in a precise and controlled manner.

One of the numerous hypothesized mechanisms to alter optical properties by scattering reduction in skin is dehydration of tissue constituents. By reducing the water content in tissue, light’s path to a desired area is through a more uniform medium. It has been hypothesized that localized mechanical compression causes dehydration of tissue constituents in compression zones, thereby modifying the optical properties. Following that hypothesis, optical property alteration was accomplished in research using mechanical loading, but a quantitative analysis of its effects and the mechanisms behind it have not been studied in depth. The advantage of using mechanical loading for scattering reduction over other methods is its non-invasive application and ability for fast, controlled, and reversible alteration of skin’s properties. By analyzing light transmission and optical properties such as refractive index during and after localized
mechanical compression it is possible to gain insight into the behavior of skin to external loading. Quantifying water transport through tissue during mechanical loading can also help correlate mechanical force to changes in water content, and therefore optical properties of skin.

1.1 Problem Statement and Scope of Thesis

While increased optical clarity and transmission has been accomplished in biological tissue in the past, the mechanisms behind it are not well understood. This thesis will serve as a starting point for the exploration of water transport in the skin’s dermis and skin’s optical properties due to localized mechanical force applied by sapphire compression geometries. In addition to reviewing past successes and methods for increasing optical clarity, this thesis will accomplish four primary goals. 1) An explanation of mechanical loading will introduce a newly developed device to apply localized compression, optical radiation, and surface cooling to tissue. 2) Ex vivo tissue experiments will help characterize the behavior of skin to localized external loading. 3) A simplified computational finite element model will begin to study the mechanical characteristics of skin and quantify water expulsion in the dermis due to localized external loading. 4) Exploration of the simplified finite element model and literature techniques will lead to further suggestions for skin modeling and experimentation.

The thesis is organized as follows. Chapter 2 discusses the complex structure of skin including composition and its optical properties. Light scattering within tissue is discussed and its detrimental effects for light-based treatments and therapies. The history and background behind past optical clearing techniques, such as absorption of hyperosmotic agents, is reviewed followed by the hypothesized mechanisms behind their success. The argument for a shift to other methods such as mechanical loading for optical clearing is presented.

Chapter 3 describes the hypothesized mechanisms behind water movement and subsequent optical clearing due to localized mechanical compression. Past success with mechanical loading is presented followed by the design of a new device. An
explanation of vacuum loading and a basic discussion of the force applied during device application is reviewed. Finally, cooling potential incorporated into the design of the device will be discussed.

Chapter 4 presents experimental results used for quantifying optical power transmission through skin due to localized mechanical compression. Using an optical power meter, light intensity through tissue was gathered for multiple stress and strain states placed on ex vivo skin. The experimental setup is detailed, as well as the loading cycles and conditions applied to the sample. Comparisons are made for different loading states and compression geometries.

Chapter 5 describes the application of vacuum-induced localized mechanical compression for analyzing the refractive index and water content of tissue using Optical Coherence Tomography (OCT). With methods from Sorin and Gray, simultaneous determination of tissue thickness and group refractive index is calculated from OCT images. Using the dynamic refractive index values, the Lorentz-Lorenz rule of mixtures is applied to deduce water concentration change during localized mechanical compression.

Chapter 6 investigates water movement through skin using a computational finite element model developed in ABAQUS. Past skin modeling techniques found in the literature are reviewed followed by the technique decided upon for this research. A simplified tissue representation is developed within ABAQUS where skin is characterized as a mixture of a permeable medium with elastic material properties and a fluid. Results from finite element models used to verify tissue behavior with known solutions are presented. Localized compression is then accomplished with point loading and the mechanical behavior as well as transient water transport through tissue is investigated.

Finally, the thesis concludes with chapter 7, which summarizes the work and results presented in the previous chapters. A discussion of the contribution from this work is included followed by suggestions for future research.
Chapter 2

History and Background

Skin is a large, complex, and multifunctional part of the human body. Its functionality includes, but is not limited to, protecting the body from traumas such as impact, pressure, cutting, friction, and shear [1]. While its complicated structure successfully acts as a protective barrier between the body and external surroundings, it also prevents penetration of light for diagnostic, therapeutic, and treatment purposes. Optical properties of skin have been successfully altered in the past to overcome the limitations of light-based procedures. This section discusses the idea of “Tissue Optical Clearing,” why it is important, and the proposed mechanisms behind it. The past successes of hyperosmotic agents for optical clearing will be presented, followed by the discussion of potentially achieving similar effects due to mechanical loading. This section will also identify potential reasons for using mechanical loading in place of hyperosmotic agents.

2.1 Skin Structure

Human skin varies in thickness, texture, and mechanical strength depending on its location throughout the body. The basic composition and constituents, however, are fairly constant. From the most superficial layer to the most interior, skin is composed of an epidermal layer, a dermal layer, and a subcutis foundation as shown in Figure 2.1. The epidermis serves as the main boundary between the outside world and the human
Composed of mostly keratinocytes, the epidermis averages 0.07-0.12 mm in thickness [2] and protects the body from major water loss to its surroundings. The dermis, which can vary in thickness from 0.3 mm to 3.0 mm depending on location, consists of eccrine and apocrine glands, hair follicles, veins, nerves and the extracellular matrix [3]. Proteins collagen and elastin as well as complex sugars form the extracellular matrix, which provides structural integrity to the skin. More specifically, the dermis is comprised of two layers: the reticular and papillary dermis. The papillary dermis is the more superficial layer and is comprised of loosely arranged collagen fibers. Below the papillary dermis is the reticular dermis. It accounts for the majority of the overall volume of the dermis and is comprised of thick, tightly packed collagen fibers. The reticular dermis provides the skin with strength and elasticity. Beneath the dermis is the subcutaneous layer. Comprised of fat, the subcutaneous layer plays an important role by providing insulation to the body.

Figure 2.1: Skin Schematic. Human skin contains three main layers: the epidermis, dermis, and subcutaneous. http://www.cancer.gov/cancertopics/wyntk/melanoma/page3.
2.1.1 Tissue Optical Properties and Light Scattering

Many light-based treatments, therapies, and optical diagnostic techniques are clinically used today in the medical and research fields. Laser hair removal, cancer ablation, port wine stain treatment, scar/tattoo removal, and tissue imaging are just a few of the applications commonly involving light radiation in skin. Deep penetration of light into tissue is imperative for efficient treatments. However, optical properties such as refractive index, scattering coefficient, direction of scattering, and absorption coefficient affect the potential penetration depth for light delivery. Penetration depth, $\delta$, is inversely proportional to the absorption ($\mu_a$) and scattering coefficient ($\mu_s$) as shown

$$\delta = \frac{1}{\mu_a + \mu_s}. \quad (2.1)$$

Skin acts as a highly scattering medium for visible to near-infrared wavelengths of light due to its complex and nonhomogeneous morphological structure. Light scattering in biological tissues is caused primarily by variation in electronic polarizability at optical frequencies, which may be characterized by variations in the optical index of refraction, $n$. Refractive index is a measure for the amount a medium slows down the speed of light as shown below,

$$n = \frac{c}{v_p}. \quad (2.2)$$

where $c$ is the speed of light through a vacuum and $v_p$ is the phase velocity of a certain wavelength of light in a medium. Tissue constituents such as collagen, lipids, water, cells and their organelles all have slightly different indices of refraction. As light travels through a nonhomogenous medium such as tissue, its velocity will change amplitude at the interface between two constituents. The consequent refractive index variation creates a scattering event. For laser-based techniques, scattering events limit the amount of light delivered to targeted regions, as shown in Figure 2.2.
Figure 2.2: Light Scattering in Tissue. Specular reflection off of the tissue and diffuse reflectance or transmission within the tissue can result in photon redirection away from the targeted region.

Water is a significant contributor to the volume and composition of tissue accounting for 70% of its total weight. Conversely, collagen provides 70% to 80% of the dry weight of skin [4]. Water, with its lower index of refraction ($n=1.33$) than proteins ($n=1.43-1.53$) [5] contributes significantly to the optical index of refraction mismatch, $\Delta n$, giving rise to light scattering.

2.2 Tissue Optical Clearing

Maintaining a collimated or focused beam for selective optical delivery is limited by the highly scattering characteristic of skin. “Tissue optical clearing” permits delivery of near-collimated light deeper into tissue, potentially improving the capabilities of various optical diagnostic, therapeutic, and treatment techniques [6]. Figure 2.3 shows
an example of light distribution through tissue before tissue optical clearing and the desired result after tissue optical clearing has been performed.

Figure 2.3: Optical Clearing. Optical clearing can lead to increased light delivery at a targeted region.

Tissue optical clearing research in the past has focused on using chemical agents such as glycerol and dimethyl sulfoxide to reduce tissue light scattering and increase optical clarity. Numerous technical publications report methods, applications, and potential mechanisms of tissue optical clearing using chemical agents [7-17]. Three hypothesized mechanisms of light scattering reduction induced by chemical agents have been proposed: 1) dehydration or water removal of tissue constituents; 2) replacement of interstitial or intracellular water with an agent that better matches the higher refractive index (n) of proteinaceous structures; and 3) structural modification or dissociation of collagen fibers. The first two mechanisms were first described by Tuchin et al. [8] while the third was mentioned by Yeh et al. [12].

These three, and possibly other unspecified dynamic mechanisms, may be acting synergistically or antagonistically with differing contributions dependent on tissue type and vitality, chemical agent, and delivery method. Optical clearing has been successfully achieved in rat tissue by Vargas et al. by the absorption of the chemical agent glycerol [9]. After injection of the glycerol the rat skin became clearer and previously hidden veins and capillaries became visible.
Because dehydration and refractive index matching of intrinsic structures follow sequentially, the two mechanisms may in fact be coupled. Several investigators have suggested that dehydration alone can reduce scattering in soft tissue by removing water from the space between collagen fibrils, increasing protein and sugar concentrations, and decreasing refractive index mismatch [18-22]. Dehydration of tissue constituents as a sole method for optical property alteration has been explored by Rylander et al. [17].

2.3 Drawbacks of Chemical-based Tissue Optical Clearing

Some of the drawbacks of optical clearing with chemical agents include the slow transport of these agents across the tissue and potential toxicity. Invasive methods such as delivery with a needle can cause pain to the subject. In addition, there is limited selectivity or targeting of the chemical agent, and controlled removal of chemical from the tissue is difficult or impossible.

Because dehydration is an important possible mechanism of optical clearing, it has been hypothesized that other non-chemical techniques for water redistribution, such as applied mechanical force, should also optically clear tissue and alleviate some of the drawbacks of chemical-based optical clearing [6]. Several potential advantages exist for mechanically induced optical clearing as opposed to using chemical methods. Mechanical compression is less invasive and, therefore, safer since no chemical agents are introduced into the tissue and the barrier function of the stratum corneum is maintained. A mechanical clearing technique may potentially provide faster onset, better controllability, and more repeatable optical clearing.
Chapter 3

Mechanical Loading

Mechanical loading for altering optical properties of tissue due to water content modification introduces a non-invasive method that can be coupled with light delivery in one device. This section lays out the proposed methods of water content modification and subsequent optical clearing due to mechanical loading in a well defined flowchart. Past results and devices are reviewed as well as an in depth look at the hypothesized changes in scattering and absorption due to localized dehydration. A new device developed to apply multiple compression points to in vivo and ex vivo skin while permitting use of Optical Coherence Tomography and laser experimentation is presented. The compressive forces induced by vacuum loading, the specific materials of the device, and the geometries of the compression tips are also discussed. Finally, a method for skin cooling through fluid delivery to enhance optical treatments is reviewed.

3.1 Background

As previously mentioned, the proposed importance of dehydration for optical clearing using hyperosmotic agents has lead to other hypothesized techniques for water redistribution in skin such as mechanical loading. It has been hypothesized that the application of mechanical force can induce local water removal from tissue; thereby providing a novel technique for optical clearing. Localized mechanical force is believed to expulse water from underneath localized compression zones, causing dehydration at
those points [6]. Coupled with tissue thickness change, water content modification will alter optical properties of skin at compressed regions. Figure 3.1 shows the hypothesized mechanisms behind localized mechanical loading for achieving increased optical clarity and transmission in tissue. Three separate avenues for delivering more light at targeted regions are outlined.

![Figure 3.1: Hypothesized Mechanisms Flowchart](image)

**Figure 3.1: Hypothesized Mechanisms Flowchart.** The flowchart outlines the hypothesized mechanisms that lead to increased light delivery at targeted areas due to localized mechanical force.

Shown by the path of red arrows, localized mechanical force is proposed to dehydrate compression zones and reduce scattering, thereby transmitting a higher percent of the original radiation to a specified region. The green path shows that
reducing tissue thickness or altering the tissue geometry will again deliver more light to the targeted region by decreasing scattering volume. Also, as shown by the black path, altering the tissue thickness/geometry will allow a novel light delivery method through the device at compression tissue zones, increasing light to regions of interest. Although it has been hypothesized that mechanical force will also change optical properties of tissue through blood vessel volume and perfusion, this thesis will deal solely with thickness/geometry and water content modification.

3.1.1 Scattering and Absorption

Locally modifying the water concentration of tissue will change optical properties such as refractive index, absorption coefficient, and scattering coefficient at compression zones. Adjustment of these properties have an overall effect on the local fluence in tissue when a light source is introduced, approximated by,

\[ \Phi(z) = \Phi_0 e^{-\mu_{eff}z} \]  

(3.1)

where \( \Phi_0 \) is the irradiance on the surface, \( z \) is depth of light penetration, and \( \mu_{eff} \) is the effective attenuation coefficient. Light transmission in tissue is attenuated by its highly scattering characteristics as well as its absorptive nature. The effective attenuation coefficient of tissue may be approximated by,

\[ \mu_{eff} = \sqrt{3\mu_a(\mu_a + \mu_s')} \]  

(3.2)

where \( \mu_a \) is the absorption coefficient and \( \mu_s' \) is the reduced scattering coefficient. Brugmans et al. [23] have shown the dependence of the optical absorption coefficient, \( \mu_a \), on the water content in skin,

\[ \mu_a = W_W * \mu_{a,water} + (1 - W_W) * \mu_{a,0} \]  

(3.3)
where \( W_w \) is water weight fraction, \( \mu_{a,\text{water}} \) is the optical absorption coefficient of water at a particular wavelength, and \( \mu_{a,o} \) is the fully dehydrated absorption coefficient in skin. Taking \( \mu_{a,\text{water}} \) as 1.25 cm\(^{-1}\) and \( \mu_{a,o} \) as 0.75 cm\(^{-1}\) at 1,210 nm [24], this relationship illustrates that dehydration causes a decrease in the skin absorption coefficient as shown in Figure 3.2. Decreasing optical absorption in tissue is essential to deliver more light at targeted areas.

![Figure 3.2: Dependence of Absorbance Coefficient on Water Content. A lower mass fraction of water results in a reduced absorbance coefficient.](image)

Scattering reduction by dehydration of skin has been proposed due to intrinsic refractive index matching [6]. The strength and direction of scattered light depends on the size and geometry of the scatterers as well as the index of refraction. In the dermis, scattering may be due to variations of refractive index at the interface between high-index collagen fibers (n \( \sim \) 1.43-1.53) and the surrounding low index water. It has been proposed that skin dehydration may reduce light scattering by increasing the volume fraction of scattering particles such as collagen fibrils.
3.2.1 Past Results

To test the hypothesis of tissue optical clearing through mechanical force and subsequent water redistribution, several device prototypes were designed by Rylander et al. consisting of an array of pins [6]. The prototypes induced spatially localized tissue compression utilizing a 750 mm Hg vacuum pressure source. One device, as shown in Figure 3.3, consisted of an array of pins and a circumscribing brim. The pin array, located within the inner surface of the chamber, was composed of a photoresin which transduced mechanical force to skin. The pins were 1 mm in diameter and 4 mm long with a 20% fill factor (packing density). The device brim interfaced with the skin surface and formed an airtight seal when a vacuum pump was mechanically connected to the device. Vacuum pressure exerted a mechanical transduction force on the skin, causing stretching and compression of tissue between and underneath the pins, respectively.

![Figure 3.3: Device Schematic.](image)

Figure 3.3: Device Schematic. A simplified cross-sectional schematic diagram of the preliminary device applied to the skin surface shows zones for potential optical property modification.

Application of the prototype to *in vivo* human skin resulted in skin stretching and compression beneath the pins and skin stretching between the pins. Skin reddening surrounding the pins was evident and hypothesized to be due to displacement of blood from regions directly underneath the pins into surrounding tissue. Upon removal of the
vacuum loaded device, regions of skin compressed by pins appeared darker, indicating modification of optical properties within. Several minutes after removal of the device, redness of vacuum regions decreased and the thickness of mechanically compressed skin regions rebounded to normal.

Positive preliminary results demonstrated the effectiveness of the device on ex vivo porcine skin using white light photography. Pig tissue samples consisted of a layer of skin approximately 2-3 mm thick and an underlying layer of subcutaneous tissue approximately 1 mm thick. Ex vivo porcine specimens significantly attenuated visible light transmission prior to device application. Following application of the device to the epidermal surface for thirty seconds, both epidermal and dermal views of the porcine skin indicated increased light transmission through locally compressed skin regions [6].

Other researchers have investigated pressure effects on tissue optical properties and reported increased absorption and scattering coefficients due to compression loading [21, 25]. This result is contrary to that of preliminary tests performed by Rylander et al. An important distinction exists, however, between the other research and the experimental methods done by Rylander et al. and those that will be presented in this paper. In previous studies, uniform pressure was delivered across large-area (>100 mm²) ex vivo tissue specimens. The technique and devices used in the preliminary results as well as the experiments in this thesis induce much smaller (~1-15 mm²) compression zones permitting water movement laterally within tissue.

3.2 New Device Platform

Although Rylander et al. created a variety of prototypes for mechanically-induced tissue optical clearing, a new device platform was developed to serve a specific purpose in a collaborative project with Wake Forest University. In the study, prostate cancer cells in the dorsal flank of mice were exposed to laser radiation in order to ablate the cancer cells and study the effect of laser heating. However, excessive heating of the mouse tissue’s surface lead to undesirable results. In addition, heat profiles within the tumor were studied in real time during laser radiation using Magnetic Resonance Imaging
(MRI). To assist localized heating with the intent of reducing surface damage through tissue optical clearing, a new device was designed using non-ferromagnetic materials. Non-ferromagnetic materials are essential when dealing with an MRI due to the intense magnetic field present.

Designed to incorporate vacuum-induced localized mechanical compression, light delivery, and skin surface fluid delivery, the new device has potential to be varied depending on the desired application. Figure 3.4a shows a simplified schematic of the new device platform applied to the skin’s surface. An isometric CAD view of the actual device is shown in Figure 3.4b where the red material denotes sapphire and the clear/grey material denotes plastic.

![Figure 3.4: New Device Platform. a) Simplified cross-sectional schematic diagram of the new device applied to the skin surface. b) CAD drawing of device.](image)

The device consists of a circular chamber containing an array of seven 20-mm long sapphire rods with 3-mm diameter tips arranged with a 33% fill factor (packing density). The array of sapphire rods is held in place with an acrylic frame. During use, the device is placed onto the skin, allowing the sapphire rods and outer ring of the chamber to contact the surface of the tissue. Application of a 750 mmHg vacuum pressure source creates suction inside the device’s chamber, drawing the tissue against
each sapphire rod. Skin compression can also be induced by applying a direct external force onto the device. A separate port was designed into the device for fluid delivery and cooling during application.

3.2.1 Materials

Sapphire rods were specifically chosen as the material for compression and light delivery in the new device platform. Shown in Figure 3.5, sapphire has a rare collection of mechanical and optical properties that makes it desirable for compression and light delivery applications. The Young's modulus and Poisson's ratio (50,000,000 psi and 0.29) of sapphire is comparable to that of iron and steel (30,000,000 psi and 0.21-0.30), making it a rigid tip for compression. The optical and thermal properties of sapphire, however, differ significantly from most ferromagnetic and other similarly mechanically stiff materials. With a thermal conductivity of 35.1 $W/m \cdot K$, sapphire is nearly 10 times as thermally conductive as glass, which has similar optical clarity. Heat release is imperative for a light delivery material as certain wavelengths cause tissue surface damage during radiation.

Figure 3.5: Device Components. a) The main components used in the new device include the sapphire rods, the polypropylene cylinder, and the acrylic discs. b) An exploded view of the assembly shows the components as they are arranged inside the device.
The other two main components of the new device platform are the polypropylene cylinder and the acrylic discs, which form a tight seal on the skin surface and hold the sapphire rods in place, respectively. Chosen mainly for its manufacturing capabilities, the acrylic discs were cut to precision using a 50W CO2 laser cutter made by Universal Laser Systems. Also easy to manufacture, the polypropylene cylinder was chosen as a soft/smooth material so that it would not damage or cause pain to the skin when a seal was created by the vacuum source.

3.2.2 Vacuum Induced Loading

Structural changes of skin induced by vacuum include reduction of skin thickness through stretching of the tissue [26]. Vacuum-induced loading from a 750 mmHg pressure source draws the skin up into the device and against the sapphire rods, causing compression at the skin-sapphire interface. This effect, coupled with the stretching of the tissue, helps accomplish two of the mechanisms discussed earlier leading to potential increase of light at targeted areas. They include compression of the tissue for dehydration as well as reduction of tissue thickness. Since the device was originally designed to incorporate fluid delivery parallel to the direction of the axes of the sapphire rods, one of the two side-holes must be obstructed to solely apply vacuum loading. Using the vacuum port to create negative pressure inside the chamber, the pins produce an equal and opposite reaction force to the vacuum against the skin as shown in Figure 3.6.
**Figure 3.6: Compression by Vacuum Loading.** The resulting force on a single pin due to vacuum pressure loading is equal and opposite.

A force analysis can be done using the known vacuum pressure source to determine the force each rod tip exerts on the tissue. Summing the forces in the y-direction leads to the equation,

$$\sum F_y = F_{\text{vacuum}} + F_{\text{rod}}$$  \hspace{1cm} (3.4)

where $F_{\text{vacuum}}$ is the force applied in the positive y-direction by the vacuum pressure and $F_{\text{pin}}$ is the resulting force applied in the negative y-direction by the pin. Assuming the system is in equilibrium, the force in the y-direction will become zero resulting in,

$$\sum F_y = 0 = F_{\text{vacuum}} - F_{\text{rod}}$$  \hspace{1cm} (3.5)
where the force of the vacuum pressure equals the force exerted by the sapphire rods. With a 750 mmHg vacuum source applied to the skin’s surface inside the device, the resulting compressive force of each sapphire rod is approximately 0.32 lbf or 1.4 N.

### 3.3 Skin Cooling using Fluid Delivery

Laser-based photo-thermal therapies and treatments are limited by their ability to deliver heating in deep tissue while preventing thermal injury to non-targeted healthy tissue. Depending on the wavelength and power of optical radiation, damage and pain can be caused to superficial skin during application. To explore reduction in the thermal effects of optical radiation, a fluid delivery port was included into the design of the new device platform. As shown in Figure 3.7, fluid flow parallel to the center axis of the rods is introduced to develop cross-flow over the sapphire.

**Figure 3.7: Fluid Delivery.** The fluid delivery port produces cross-flow over sapphire rods.
The addition of the fluid delivery accomplishes three separate thermal responses assuming there is a temperature gradient between the skin, sapphire, and fluid. First, similar to a heat exchanger, forced convection occurs between the moving fluid and the sapphire rods. Second, forced convection occurs between the moving fluid and the skin’s surface. Third, conduction occurs between the sapphire rods and the surface of the skin. By passing cool liquid through the device, heat flux is away from the skin. Working either separately or synergistically, these thermal responses should reduce the temperature of the skin’s surface, thereby reducing the probability of damage or pain.

Cooling of the sapphire rods is principal to prevent thermal damage at the skin-rod interface. To determine the potential interaction between the moving fluid and the sapphire, basic heat transfer theory can be utilized. Figure 3.8 shows the necessary dimensions to determine the heat transfer coefficient of a tube bundle with fluid cross-flow. The water is assumed to be 0°C for all calculations.

![Flow over a Tube Array](image)

**Figure 3.8: Flow over a Tube Array.** The diagram defines necessary dimensions for calculating the heat transfer coefficient for flow over a tube array as determined by Incropera and DeWitt [27].
Using the parameters $S_L$, $S_T$, and $D$, the constants $C_1$, $C_2$, and $m$ can be determined to calculate the Nusselt number as shown,

$$\overline{Nu}_D = C_2 \left( 1.13C_1 Re_{D,max}^m Pr^{1/3} \right)$$  \hspace{1cm} (3.6)

where $Pr$ is the Prandtl number. $Re_{D,max}$ is defined as

$$Re_{D,max} = \frac{\rho V_{max} D}{\mu}$$  \hspace{1cm} (3.7)

where $\rho$, $V_{max}$, and $\mu$ are the density, the maximum velocity, and the dynamic viscosity of the fluid, respectively [27]. Using the dimensions shown in Figure 3.9 for the new device platform, the heat transfer coefficient for the sapphire bundle is calculated to be 10,657.6 $W/m^2K$, assuming the moving fluid is water.

**Figure 3.9: Device Dimensions.** The dimensions (inches) of the sapphire rod array provide the necessary information for calculating heat transfer coefficient.
Adding sapphire rods to the cross-flow significantly increases the overall heat transfer rate out of the tissue during cooled fluid delivery. To determine the heat transfer rate without rods present, the skin was assumed to be a plate with flow parallel to the surface. Following this assumption, the heat transfer coefficient can be determined using water as the moving fluid. The Nusselt number is calculated as,

\[ \overline{Nu_x} = 0.664 Re_x^{1/2} Pr^{1/3} \]  \hspace{1cm} (3.8)

which can be used to calculate the heat transfer coefficient,

\[ \overline{h}_x = \frac{\overline{Nu}_x k}{x}. \]  \hspace{1cm} (3.9)

Using the heat transfer coefficient, the overall heat transfer rate can be determined using,

\[ q = \overline{h}_x A(T_{skin} - T_{water}) \]  \hspace{1cm} (3.10)

where A is the area of tissue in contact with the water. The skin’s surface is assumed to be room temperature for this calculation. The resulting heat transfer rate without sapphire rods is determined to be 3.5 W. Comparatively, the heat transfer rate out of the skin with sapphire rods assumed as fins with adiabatic tips is calculated to be 12.03 W. This simple calculation shows that the pins increase the heat transfer rate out of the tissue by nearly 3.5 fold.

Assuming the sapphire rods as fins is a liberal estimate of the actual heat transfer rate out of the tissue. Therefore, a surface resistance between the sapphire rods and tissue was applied to reduce the efficiency of the overall heat transfer rate. Using three separate surface resistances found from a heat transfer text book for 1) vacuum interface, 2) interfacial fluid, and 3) solid/solid interfaces, the overall heat
transfer was calculated with each surface resistance. The resulting values of 3.55 W, 7.02 W, and 10.31 W showed that a decreasing surface resistance resulted in an increased heat transfer rate. It should be noted that a very high contact resistance resulted in a similar heat transfer rate as the no-pin circumstance.

3.3.1 Cooling System

To effectively move water through the device while maintaining suction on the skin’s surface, a system was developed using a pool water pump, a fluid reservoir, and an aspirator. An aspirator, also known as a venturi pump, utilizes the venturi effect to produce a vacuum. As a fluid flows through a restricted area of tubing, the velocity increases due to the conservation of mass. The velocity increase causes a pressure drop due to the conservation of energy. As shown in Figure 3.10, the pressure drop, which produces a vacuum when open to air, is used to draw fluid out of the reservoir and through the device. As long as the brim of the device is making a seal with the skin, a vacuum is sustained.

![Figure 3.10: Cooling System.](image)

A pool water pump is used to increase flow rate through the aspirator, creating a vacuum in the device. The vacuum can be used to draw skin against sapphire rods, and/or to draw fluid through the device.
3.3.2 Experimental Results

Cooling during laser therapy is essential to optimize treatment. Damage to superficial layers of tissue can prevent elevated power during treatment, thereby delivering a reduced dose with reduced results. To determine the effect of the designed cooling system, thermal imaging during ex vivo laser heating of porcine skin was performed. Using 5 watts (1064 nm), laser radiation was applied to the surface of an ex vivo pig sample through the device for 45 seconds. After 45 seconds of radiation, thermal images were taken with a FLIR THERMOVISION A40 thermal camera. This experiment was repeated three times for three different device applications.

First, radiation was applied through the device’s sapphire rods while being held in light contact with the skin sample for 45 seconds. Second, vacuum was applied through the device while laser radiation was accomplished. Finally, the cooling system was utilized to pass ~32 °F water through the device during laser radiation. A thermal image of the first application with the device held lightly against the skin sample is shown in Figure 3.11. The surface temperature of the skin at the radiation zone is significantly higher than other locations. The maximum temperature achieved is 303.1 °K.

![Figure 3.11: Thermal Image of Heated Tissue. Increased surface temperature was accomplished while the device was held lightly against the tissue sample.](image-url)
During vacuum, the skin exhibited an increase in the maximum superficial temperature as shown in Figure 3.12. Vacuum was applied during all 45 seconds of laser radiation. The maximum surface temperature reached 304.7 °K during radiation and vacuum. Also apparent during vacuum application was a variation in the surface heating for pin compression zones and vacuum regions.

![Thermal Image of Heated Tissue with Vacuum](image)

**Figure 3.12: Thermal Image of Heated Tissue with Vacuum.** Vacuum application showed an increase in the maximum temperature.

Laser radiation during cooling showed a noticeable decrease in the maximum superficial temperature after 45 seconds. Skin surface temperature maintained approximately 289 °K during radiation. Radiation points corresponding with sapphire rod compression zones reached a surface temperature of approximately 292 °K. As shown in image Figure 3.13, untreated areas of the tissue sample maintained a surface temperature close to 295 °K.
Figure 3.13: Thermal Image of Heated Tissue with Cooling. The skin surface was cooled significantly during fluid delivery.
Chapter 4

Light Intensity through Tissue with Mechanical Loading

To further explore the effects of tissue loading, a simple experiment was developed to apply sapphire rod compression to *ex vivo* animal skin. Determining light transmission through tissue before, during, and after mechanical compression can provide insight into the correlation between time, force, tissue strain, loading geometry, and the optical properties of tissue. This section describes experimental materials and methods used to apply localized compression to *ex vivo* animal skin while simultaneously measuring optical power through the tissue. Using a BOSE ElectroForce and two different compression tip geometries (rounded and flat) for loading, strain and stress measurements with corresponding optical power values are presented and discussed. The specifics such as the loading cycles, loading rates, and maximum force used to investigate variations in transient tissue response are also discussed. The results of overall light power increase and its correlation to strain and stress are shown for this novel non-invasive localized mechanical compression experiment.

4.1 Experimental Overview

Laser beam profiles and light intensity change through tissue in response to mechanical compression has been studied in the past [28]. However, those experiments consisted of uniform compression between two glass slides over a large area of tissue
(5x5 cm²) with static measurements of light intensity. The compressive force applied to the tissue was unknown as the loading was displacement based.

As we believe that localized compression laterally displaces water out of compression zones, subsequently changing optical properties of tissue, dynamic measurements of optical power through tissue due to point loading by a sapphire rod is investigated. Point loading allows water to move from the compression region, whereas uniform compression over a large area limits lateral expulsion in the permeable tissue. In a novel experiment using a BOSE ElectroForce 3100 mechanical testing system (Figure 4.1), load-based (0.5 N – 8 N) localized compression (7-14 mm²) is applied to a 2 mm thick porcine skin sample using two separate compression geometries. The BOSE ElectroForce has a resolution of 0.001 mm (1 micron) and 0.001 N (1 mN), which makes it a desirable test system for dealing with sensitive material such as animal skin.

![Figure 4.1: BOSE ElectroForce.](image)

Testing was setup for displacement-based and load-based compression. The data acquisition system was also programmed to gather simultaneous data from the light power meter.
4.1.1 Materials, Setup and Tissue

The BOSE ElectroForce has the capabilities for compressive or tensile loading. For this application, the compression load cell and displacement cell were configured to deliver both load-based and displacement-based motion towards the center of the ElectroForce. A setup was developed as shown in Figure 4.2 to create localized compression to *ex vivo* animal skin between a sapphire tip and a glass slide. Housed in a polycarbonate structure, which is attached directly to the displacement cell of the ElectroForce, the sapphire rod is held rigidly by a high strength epoxy. Also attached to the polycarbonate housing is the 670 nm laser, which is configured to radiate light through the sapphire rod and onto the *ex vivo* animal tissue.

![Compression Experimental Components](image)

**Figure 4.2: Compression Experimental Components.** Optical power measurements through animal skin as well as the force and tissue strain were recorded simultaneously.
The combination of the polycarbonate housing, the laser, and the sapphire rod moves in unison with the actuated displacement cell towards the load cell for compression. The sapphire rod, protruding farthest down from the displacement cell, is the first material to make contact with the bottom portion of the setup. The skin is placed on a glass sheet, which sits atop a section of aluminum square tubing attached directly to the load cell. To begin compression, the displacement cell and all attached materials move in the negative y-direction until contact with the skin’s surface is made. During compression, 670-nm laser irradiation is applied through the sapphire rod and skin and the transmitted light is measured by a Newport Optical Power Meter (model number 1830). A small hole in the aluminum square tubing allows light to penetrate through the tissue and glass slide, and onto the power meter.

Two sapphire rods with different tip geometries were used to apply force with separate compression areas and geometries. One tip was a polished half-sphere, as shown in the device outlined in Chapter 3. The other was a polished flat-tip with a chamfered brim. The chamfer was created so that the skin would not be damaged by the sharp brim of the sapphire cylinder.

Both *ex vivo* porcine and *ex vivo* dog skin was used in the compression experiments. Samples were approximately 4 cm² and 2 mm thick. For these animals, 2 mm thick tissue contained the epidermis, dermis, and a very thin layer of the subcutaneous. Although the goal was to isolate the epidermis and dermis, the skin samples’ connective tissue was stringy, making it difficult to cut with precision. The porcine skin sample was gathered from the belly of the pig, while the dog tissue was taken from the back of the animal. Both samples were acquired in agreement with an approved protocol from the IACUC and used fresh or kept in a 5° Celsius refrigerator until experiments were performed.

### 4.1.2 Loading Cycles

Time-dependent measurement of optical power through tissue due to localized compression provides insight into the correlation between forces applied and light
transmission through skin. A variable loading cycle was developed to study the light intensity during three separate compression states; 1) during constant displacement rate tissue loading, 2) while force is held at its maximum value, and 3) during relaxation of the tissue. One loading routine was designed and implemented for 45 runs with each sapphire tip (round and flat). The base loading cycle for all 90 runs consisted of four separate commands:

- Load-based compression from 0 N to maximum load at a rate of \( X \) mm/s
- Dwell at maximum load of \( Y \) N for \( Z \) seconds
- Ramp down to 0 N
- Dwell at 0 N for 60 seconds

The variables \( X \), \( Y \), and \( Z \) represent the displacement rate during loading, the maximum load applied to tissue, and the dwell time at the maximum load, respectively. Values used for each variable are shown below in Figure 4.3.

![Figure 4.3: Loading Cycle Variables.](image)

- Displacement Rates (\( X \))
  - 0.1 mm/s
  - 0.05 mm/s
  - 0.025 mm/s

- Maximum Load (\( Y \))
  - 0.5 N
  - 1.0 N
  - 2.0 N
  - 4.0 N
  - 8.0 N

- Dwell Time at Max Load (\( Z \))
  - 0 seconds
  - 30 seconds
  - 60 seconds

The displacement rate to reach maximum load, the maximum load, and the dwell time at maximum load were all varied to complete 45 loading sets for each sapphire rod tip (round and flat).
Prior to experimentation, tests were conducted to set upper and lower bounds of each variable. The minimum force of 0.5 N is comparable to the load applied during a weak vacuum. Vacuum loading with the new device discussed in chapter 3 applies approximately 1 N - 1.5 N of force through each pin. For determining the maximum load, tissue became damaged and therefore unusable when loaded with more than 8 N. It is believed that time-dependent water transport induced by localized loading will have a significant influence on the light transmitted through tissue. Therefore, the dwell time at maximum load is utilized to determine the transient response of the tissue at a constant load. A maximum dwell of 60 seconds is used to view significant changes in transient tissue response.

4.2 Experimental Results

Force applied to the tissue by the sapphire rod, sapphire rod displacement, and light intensity measured by the optical power meter was recorded instantaneously during skin compression by the BOSE data acquisition system. Because each run began with the sapphire rod contacting the top surface of the ex vivo skin, tissue strain can be calculated from the sapphire rod displacement. Using the tissue’s initial thickness, $t_{\text{skin}}$, and the displacement of the sapphire rod, $\Delta y_{\text{rod}}$, skin strain, $e_{\text{skin}}$, is determined by

\[ e_{\text{skin}} = \frac{\Delta y_{\text{rod}}}{t_{\text{skin}}}. \quad (4.1) \]

4.2.1 Curved Tip

Using a 1.5 mm radius hemisphere-tipped sapphire rod for tissue compression, 45 separate loading cases were accomplished. Using the three separate loading rates, five separate values for maximum force, and three separate dwell times, time-dependent optical power, force, and tissue strain data was acquired. Sensitivity of loading parameters to increased optical power is important to gain insight into the response of
skin. Results for several loading conditions with the hemispherical-tipped sapphire rod compression are highlighted below.

For the first plot shown (Figure 4.4), force was ramped up from 0 N to 1 N at a rate of 0.025 mm/sec. When the maximum load of 1 N was reached, the displacement cell released, causing a rapid decrease in force down to 0 N. Data is only shown from the start of the experiment until maximum load was reached. The overall measured optical power increased approximately 263% during compression from 0.004 V to 0.0145 V. The tissue’s thickness was reduced 45% during loading.

![Force vs Tissue Strain](image)

**Figure 4.4. Curved Tip: 1 N, 0.025 mm/s, 0 Dwell.** Loading was accomplished until a maximum load of 1 N was reached.

To investigate the time-dependent response of compressed tissue on light transmission, a dwell of 60 seconds was utilized at the same maximum load (1 N) and loading rate (0.025 mm/s), shown in Figure 4.5. As pointed out by the red line on the graph, tissue force reached a level of 1 N and stopped increasing at a tissue strain of
about 0.32. An intriguing tissue response is observed when the force is held constant at 1 N for 60 seconds. Although no displacement of the cell is specified, tissue strain increased to approximately 0.37 during this time. To hold the force applied to the tissue at 1 N, the displacement cell had to continue to travel. This is commonly referred to as creep, a behavior common to viscoelastic materials. During the 60 seconds of 1 N constant force, the measured optical power increased from about 0.005 V to 0.007 V. The overall transmitted optical power increased approximately 250% during compression from 0.002 V to 0.007 V. The tissue thickness was reduced approximately 37% during compressive loading.

Figure 4.5. Curved Tip: 1 N, 0.025 mm/s, 60 second Dwell. Loading was accomplished until a maximum load of 1 N was reached and then held for 60 seconds.

Figure 4.6 shows tissue response and optical power for a maximum load of 2 N. Again the rate of the displacement cell was 0.025 mm/s with a dwell at maximum load.
for 60 seconds. The red line indicates when maximum force reached 2 N and stopped increasing at a tissue strain near 0.46. During the 60 seconds of 2 N constant force, the measured optical power increased from about 0.011 V to 0.016 V and the tissue strain increased to approximately 0.49. The overall optical power increased approximately 300% during compression from 0.004 V to 0.0016 V. The thickness of the tissue was reduced to ~51% of its initial thickness during the experiment.

**Figure 4.6. Curved Tip: 2 N, 0.025 mm/s, 60 second Dwell.** Loading was accomplished until a maximum load of 2 N was reached and then held for 60 seconds.

### 4.2.2 Flat Tip

Using a polished flat-tipped sapphire rod with a chamfered brim for tissue compression, 45 separate loading cases were accomplished. Identical to the experiments with the hemisphere-tipped rod, maximum load, loading rate, and dwell at
maximum load were varied. Results for several of the experiment runs are highlighted below.

For the plot shown below (Figure 4.7), loading of 1 N at a rate of 0.025 mm/s was accomplished. Once the maximum load was reached, the displacement cell was released, resulting in a rapid decrease in the force down to 0 N. The overall measured optical power increased approximately 100% during compression from 0.004 V to 0.008 V. The tissue’s thickness was reduced nearly 75% during loading.

![Graph showing Force (N) vs. Tissue Strain and Optical Power (V) vs. Tissue Strain]

**Figure 4.7. Flat Tip: 1 N, 0.025 mm/s, 0 Dwell.** Loading was accomplished until 1 N was reached.

Maximum loading of 2 N at a rate of 0.025 mm/s was accomplished in Figure 4.8 below. The overall measured optical power increased approximately 250% during compression from 0.004 V to 0.014 V. The tissue’s thickness was reduced to approximately 50% of its original thickness.
Figure 4.4. Flat Tip: 2 N, 0.025 mm/s, 0 Dwell. Loading was accomplished until 2 N was reached.

4.3 Discussion and Conclusions

Skin loading accomplished by the device and vacuum in chapter three creates an effective compressive force of approximately 1.5 N by each sapphire rod. *In vivo* measurements of tissue strain and light transmission due to localized loading are extremely difficult, if not impossible to attain. In order to gather light measurements through tissue, a mechanical testing device was used to simultaneously measure force, tissue strain, and optical power through tissue. The resulting compression experiment with one sapphire rod expanded the possibilities for exploring light intensity increase due to tissue strain, loading force, loading rate, and dwell time.

Results of measured optical power increase due to loads of 1 and 2 N give insight into the response of tissue under 1.5 N vacuum-induced loading. Also, vacuum loading per the optical clearing device in chapter 3 is accomplished at a maximum load
for an extended period of time. Therefore, results that included maximum loads of 1 N and 2 N with constant force dwell times were of special interest. Results showed that the amplitude of localized loading and extended dwell time significantly increased the optical transmission of skin. Loading rate did not appear to have a significant influence on the optical light transmission through tissue.

Variations in skin samples and initial conditions make it difficult to gather conclusions from the presented experimental data. Initial values of optical power varied significantly depending on the tissue sample and tissue strain. Although *ex vivo* samples were carefully handled and cut to similar thicknesses, exact measurement was not accomplished. In addition to optical power measurements, overall strain due to identical specified forces varied from sample to sample. It has been suggested that preconditioning of skin under compressive loading is important to obtain repeatable mechanical response [29].

With an elastic material, a prescribed load would result in a known and constant strain value. However, the anisotropic material properties and non-linear response of tissue involves a transient response due to a constant force, as shown in the experimental results above. When a certain load was held constant, tissue strain and optical transmission continued to grow. Referred to as viscoelastic behavior, mechanical properties exhibited time-dependent changes. This is a common and accepted method for modeling the mechanical response of tissue. This response has a large effect on the optical transmission properties of tissue as well as the strain. It is believed that the viscoelastic behavior in tissue compression is the result of fluid movement over time. It should be noted that viscoelastic behavior of skin under tension is believed to be due to association of collagen fibers [30], and not fluid movement. We believe that the movement of fluid (water) over time has a significant impact on the mechanical and optical properties of skin.
Chapter 5

Optical Coherence Tomography Experiments

To calculate changes in tissue optical properties and water content due to mechanical loading, optical coherence tomography (OCT) was used to image skin before, during, and after device application. Optical coherence tomography is a method of acquiring micron resolution images from a sample of interest using backscattered light measurements. This section reviews the theory and methods behind OCT as well as preliminary images of human skin in vivo before and during compression. An experiment performed with compression on fresh ex vivo porcine skin samples is explained and the resulting images are presented. Using the technique of Sorin and Gray, refractive index and thickness of the ex vivo tissue is calculated from the OCT data. These results lead to water content determination using a biphasic assumption and the Lorentz-Lorenz rule of mixtures. Final results and conclusions are made on deduced water content from OCT images due to mechanical loading.

5.1 What is Optical Coherence Tomography?

Optical Coherence Tomography (OCT) is an imaging technique capable of measuring backreflected light intensity as a function of optical depth (path length) in a specimen [31]. OCT is comparable to ultrasound, with photons acting as the moving energy instead of pressure waves. Based on the principles of Michelson interferometry, coherently backscattered light traveling the same optical path length in reference and
sample arms creates an interference signal. Mapping the depth-wise reflections of near-infrared light from the sample leads to formation of cross sectional images of its features at the micrometer scale [32]. Figure 5.1 shows a schematic of the OCT system. Imaging with OCT can be achieved at a maximum depth of 2 mm in tissue with a spatial resolution ranging from 1 to 10 µm [33]. For investigative purposes, it is necessary to have micron resolution in order to view minimal changes within the sample of interest.

Figure 5.1: Swept Source Optical Coherence Tomography.

5.1.1 In Vivo Compression Experiment

At a depth of about 50-150 µm, near the epidermal/dermal junction in most skin, water content is approximately 70%. Water, having a relatively high permeability and low viscosity, is a likely constituent to be displaced due to localized mechanical compression. The displacement of water out of compressed regions of tissue should have an effect on the overall characteristics including the optical properties. To view changes in backscattered light and optical properties of tissue due to mechanical compression, OCT imaging can be utilized.
In vivo images were acquired through a 3 mm hemisphere-tipped glass rod from multiple parts of the forearm and hand of a 24 year old caucasian male. A recorded OCT image (Figure 5.2a, $\lambda=1310$ nm) with the glass rod lightly touching palm skin clearly showed the epidermal-dermal junction and a backscattering optical depth of approximately 1200 $\mu$m. Following the static image seen in Figure 5.2a, 1 minute of skin compression was accomplished by pushing the palm tissue against the sapphire rod. After 1 minute of compression, OCT image (Figure 5.2b, $\lambda=1310$ nm) showed increased overall light intensity and backreflectance from superficial tissue regions down to depths of 2000 $\mu$m. Compression also showed a significant decrease in the amplitude of surface reflection as well as a reduced epidermal thickness.

**Figure 5.2:** In Vivo OCT Images. *In vivo* a) OCT image ($\lambda=1310$ nm) with the rod lightly touching human skin. b) OCT image after 1 minute of rod compression.

Similar experiments were performed by Rylander et al. using their device prototype described in chapter 3. Using 1 mm pins as the compression geometry, the device prototype was applied with vacuum loading to the volar forearm of a caucasian male. Removal of the device revealed large contrast differences between tissue regions directly below and adjacent to pin indentations. Increased light penetration was observed to correlate directly with skin indentations produced by the pins. Indentations
were approximately 200 μm deep. Light penetration depth under the pins was enhanced 2-3 fold over peripheral tissue regions.

![Image of OCT scan](image)

**Figure 5.3: In Vivo OCT Image by Rylander et al.** *In vivo* human skin OCT image (λ=820 nm) following application of the first device by Rylander et al.

Taken together, results of our studies suggest that point compression laterally displaces interstitial water below the pins, reduces tissue thickness, and modifies optical properties. Absorption and scattering are two tissue properties thought to be altered during compression. Optical absorption may be reduced by displacement of light-absorbing chromophores such as water. Scattering may be locally reduced by decrease in refractive index mismatch between water and proteinaceous structures such as collagen. Further exploration is necessary to determine what optical properties are changing and by what amount.

### 5.2 Ex Vivo Tissue Compression

Quantitative measurements of optical properties and subsequent water content of tissue due to mechanical loading can be accomplished using OCT. To investigate time-dependent optical property changes in tissue due to localized mechanical compression, ~0.65 mm thick *ex vivo* porcine skin was used during device application.
with 3 mm diameter ball lenses as the compression geometry. A simplified schematic of the experimental setup is shown in Figure 5.4. *Ex vivo* porcine skin specimens were obtained from a local abattoir and stored at 5°C until experiments were performed.

![Figure 5.4: Simplified Schematic of Experimental Setup. A vacuum pressure source caused the localized compression of the tissue against the ball lens.](image)

First, OCT imaging was accomplished through a single ball lens, as shown in Figure 5.5. Analyzing the OCT image without skin present is necessary to determine the initial optical location of the mechanical transducer.

![Figure 5.5: OCT Image of Ball Lens. Initial OCT image of the device’s ball lens.](image)
After the image of the ball lens was recorded, porcine skin was placed on top as shown in Figure 5.6. To gather time-dependent images of the skin during compression, dynamic OCT imaging was started at this time.

![Figure 5.6: OCT image of Porcine Skin and Ball Lens.](image)

**Figure 5.6: OCT image of Porcine Skin and Ball Lens.** OCT image of *ex vivo* porcine skin and ball lens prior to application of suction.

After ~3 seconds of data acquisition, the vacuum was turned on, drawing the porcine sample against the ball lens (Figure 5.7). Vacuum was sustained for approximately 15 seconds and then terminated. Initial examination of the compressed sample shows a significant decrease in tissue thickness.

![Figure 5.7: OCT Image of Compressed Porcine Skin.](image)

**Figure 5.7: OCT Image of Compressed Porcine Skin.** OCT image of pig skin pressed against the ball lens while suction is applied. This image was recorded after 10 seconds of suction.
5.2.1 Simultaneous Measurements with Sorin and Gray

Simultaneous dynamic refractive index and thickness of the skin sample undergoing mechanical loading is calculated using OCT images following the technique of Sorin and Gray [34]. To ascertain refractive index of the skin sample over time, optical path distances determined by OCT scans were used. By monitoring the changes in these optical path lengths during application of mechanical forces, such as vacuum-induced stretching and compression, the refractive index and thickness of the skin can be monitored dynamically. Figure 5.8 shows the change in optical path location of the mechanical transducer when the skin sample is placed in the OCT’s imaging path. Each intensity peak coincides with a surface of the actual sample or the mechanical transducer. Optical path location of the transducer changes due to refractive index of the skin sample.

![Figure 5.8: Optical Path Measurements with OCT.](image)

Optical path location of the mechanical transducer changes when a sample is placed between the OCT and the transducer.
The original location of the ball lens prior to skin placement, $y_{\text{lens}}(0)$, was initially measured. At a single x-coordinate corresponding with the crown of the ball lens, the optical path length y-coordinate was measured corresponding with the top of the porcine skin ($y_{\text{top}}$), the bottom of the skin ($y_{\text{bottom}}$), and the top of the ball lens ($y_{\text{lens}}$) as a function of time. Physical thickness of the sample was then found by measuring optical path distances for both the sample thickness as well as the ball lens location as shown [34]

$$\text{thickness}(t) = [y_{\text{top}}(t) - y_{\text{bottom}}(t)] - [y_{\text{lens}}(t) - y_{\text{lens}}(0)].$$  

(5.1)

The refractive index is then found by,

$$n(t) = \frac{(y_{\text{top}}(t)-y_{\text{bottom}}(t))}{\text{thickness}(t)}.$$  

(5.2)

### 5.2.2 Thickness and Refractive Index Results

Using the equations stated above and the OCT images acquired before and during device application, time-dependent porcine skin refractive index is determined. Figure 5.9 shows the pig skin’s physical thickness and refractive index over the length of the experiment. Prior to suction, refractive index of the skin was found to be approximately 1.36. During application of suction, the compressed skin’s refractive index rose nearly 8% to 1.46. Although a significant increase in refractive index occurred at the time of the compression (~3 seconds), a more subtle increase occurred over the final 14 seconds of the experiment. As seen with the previous experiment, viscoelastic behavior (creep) is most likely responsible. This implies that although the skin was compressed suddenly, mass transport inside the tissue continued to take place during the constant compression state, consequently changing its refractive index. Skin thickness, initially ~0.65 mm, was reduced by approximately 38% to ~0.4 mm.
Figure 5.9: Skin Thickness and Refractive Index. Preliminary results showed a significant decrease and increase in thickness and refractive index based on ex vivo OCT data, respectively.

5.3 Determining Water Content from Refractive Index

Mixture and biphasic approaches have been developed extensively in the past to describe the behavior of biological tissues, including skin [2]. Noting that skin is principally water, it is assumed to be represented as a two part mixture consisting of a low refractive index water (1.33) and a high refractive index protein (n=1.53) [5]. Following this assumption, the Lorentz-Lorenz rule of mixtures [35] can be used to relate measured refractive index of skin to its chemical composition (water and protein weight fraction, $\phi_{H_2O}$ and $\phi_p$) as shown in equation 5.4 below,

$$\frac{(n_{skin}^2 - 1)}{(n_{skin}^2 + 2)} = \frac{(n_{H_2O}^2 - 1)}{(n_{H_2O}^2 + 2)} \phi_{H_2O} + \frac{(n_p^2 - 1)}{(n_p^2 + 2)} \phi_p \quad (5.4)$$
where $n_{\text{skin}}$, $n_{H_2O}$, and $n_p$ are the refractive indices of the skin sample, water, and protein respectively. Assuming the skin is a fully saturated medium, the weight fraction of the water and protein can be related as shown in equation 5.5

$$\phi_{H_2O} + \phi_p = 1.$$  \hfill (5.5)

With calculated values of skin refractive index, these equations give a dynamic solution of the water weight fraction during mechanical compression found from the OCT data. Figure 5.10 below shows the water weight fraction during compression decreases significantly from $\sim$0.75 to 0.3. OCT images can be further analyzed to provide water content as a function of lateral (x) position. Water transport can therefore be deduced from this spatiotemporal data set. One limitation to this technique is that the refractive index is averaged across the full thickness of the specimen in the y-direction; therefore measurements are insensitive to water concentration gradients in depth.

Figure 5.10: Water Weight Fraction, Skin Thickness and Refractive Index. Water weight fraction decreases significantly during compression.
5.5 Discussion and Conclusions

Refractive index of a tissue sample can infer chemical composition within. The high percentages of skin’s weight due to water and collagen as well as the large variation of refractive indices for each component lend to a biphasic assumption for material simplification. Localized mechanical compression is believed to displace water under compression zones and subsequently modify optical properties of skin. OCT is a valuable tool for bridging the gap between mechanical loading of tissue and subsequent optical property changes and water content determination.

Skin imaging was accomplished in vivo and on ex vivo tissue with OCT. However, due to its limited penetration depth and tissue’s complex structure, in vivo measurements of refractive index are difficult to obtain. It is also difficult to obtain physical measurements of in vivo tissue such as thickness for experiment validation. Ex vivo tissue samples can be measured easily with a caliper and cut thin enough so that OCT can image through the entire structure. Using a well known method, refractive index and thickness of ex vivo pig skin was determined from OCT images.

A transient response in tissue was evident in thickness and refractive index measurements, and therefore water content measurements, during vacuum loading. Although the initial force was applied almost instantaneously, changes were evident for the next 15 seconds of data acquisition. During this time, a constant load was applied to the tissue’s surface. As previously mentioned, this behavior is commonly referred to as creep. Creep will be discussed more thoroughly in the next chapter. A similar response was witnessed during constant loading in the experiment outlined in Chapter 4. Additional exploration into tissue’s response due to localized loading is essential to understand skin’s complex nature.
Chapter 6

Finite Element Model of Tissue Compression

A finite element model has been developed to further investigate the proposed water content modification and tissue mechanical response due to mechanical loading and to validate the experimental data. This section reviews past models from the literature used to represent tissue and its complex nature. A biphasic mixture model developed by Oomens et al. is reviewed in depth. An ABAQUS model developed to couple stress and fluid flow analysis is then explored including material properties, loading, boundary conditions, SOILS solver, elements, and meshing. Confined compression of pore-fluid elements is explored to characterize deformation due to loading and compare it to elastic material, plane stress/plane strain deformation. Validation for pore pressure characterization within the model results from comparison between a solution found in Oomens’ article and the developed ABAQUS code for 2D pore pressure. Finally, results of skin mechanical behavior and water content modification due to loading by a hemisphere-tipped compression geometry will be presented.

6.1 How Should Skin Be Modeled?

Future development of effective mechanically-based tissue clearing protocols for applications such as optical therapeutics or treatments require a comprehensive understanding of the underlying physical principles governing optical clearing of tissue.
A computational finite element model to correlate applied mechanical force, tissue water transport, modified optical and thermal properties, and resulting light and heat transport is the ultimate goal. This model could serve as a tool for optimizing design parameters of devices and mechanical loading conditions as well as verifying experimental and analytical results. As a first step, a simplified finite element model is developed in this thesis to correlate applied mechanical force with tissue water transport and tissue strain.

As skin is a complex material, variations of physical models describing the mechanical properties have been developed in the past. To correctly model tissue displacement and water transport due to localized compression, both the solid and liquid phases of skin must be accounted for. A biphasic approach is therefore developed based on findings and values of material properties and tissue behavior from past literature models.

### 6.1.1 Past Literature Models

Characterization of skin mechanical properties has been an ongoing research topic for decades. In 1949, Kirk et al. investigated the elastic response of skin and subcutaneous tissue due to indentation [36]. Gibson et al. studied the behavior of skin and more specifically the dynamic arrangement of dermal collagen in response to skin stretching in 1965 [37]. Their findings suggested that the collagen fibers realign in parallel to the direction of stretch. In 1973, Fung [38] noted the importance of identifying a certain stress when defining a Young’s modulus to explain mechanical characteristics of soft tissues. Combined with collaborators Lanir and Tong [39, 40], Fung also studied the mechanical properties of rabbit skin, including its stress-strain relationships.

As interest for skin characterization through mechanical loading was growing, Bader et al. [41] pointed out in 1983 that the identification of elastic or viscoelastic constants in tissue due to indentation had not been attempted. His studies resulted in the calculation of a stiffness moduli as well as a viscoelastic parameter due to indentation. In 2003, Wu et al. stated that although the collagen fiber arrangement alone creates a very stiff medium in stretch, most of the loading due to compressive forces is carried by the
tissue matrix. Recently, numerous publications have identified material constants for the identification of skin mechanical properties. However, the amount of research put forth into determining mechanical properties of skin due to indentation is much less than that of torsion, tension, or vacuum. A thorough review of the literature revealed a wide variety in modeling and representing the mechanical properties of skin.

**Isotropic, Linearly Elastic Model**

Although it is known that skin’s mechanical characteristics do not behave in an isotropic, linear manner, it has been noted that for some applications a simple description of the mechanical properties is very useful [42]. Defined by Hooke’s law, linear elastic materials behave that a certain extension (strain) of the material is directly proportional to the stress applied. A well known linear equation relating stress and strain consists of Young’s modulus, $E$, as shown

\[
\sigma = E\epsilon
\]  

(6.1)

where $\sigma$ is the stress and $\epsilon$ is the strain of the material. One reason why linear elasticity is usually not used for skin modeling is that it does not correctly account for mechanical properties of materials undergoing large strains.

Using indentation, suction, tension, and torsion, the elastic modulus and Poisson’s ratio have been determined for skin and its subcutaneous layer by multiple authors. Although some material properties are identified in the literature, Diridollou et al. mentioned that much of the data collected for stress-strain relationships in tissue are mostly descriptive [43]. In 2000, Diridollou et al. [44] pointed out that values of Young’s modulus that are published can vary by a factor of 3000 due to the experimental method. They therefore proposed to account for external conditions by preloading *in vivo* skin during suction tests to determine a Young’s modulus and describe its elastic mechanical behavior. Using inverse analysis combined with an indentation test performed by Pailler-Mattei [45], Delalleau et al. predicted an elastic modulus and Poisson’s ratio for human
skin *in vivo*. They noted, however, that the anisotropy and viscoelastic behavior of skin needs to be further characterized as well as the specific behavior of multiple layers. More recently, in 2008, Pailler-Mattei et al. studied the contributions from both the muscle and subcutaneous, as well as the dermis, en route to defining Young’s modulus for each layer due to *in vivo* indentation test. Some of the overall material properties for skin are defined in the table below.

**Table 6.1: Material Properties of Skin from the Literature.** Young’s modulus determined by multiple experimemtal methods [46], [47], [41], [44], [48].

<table>
<thead>
<tr>
<th>Young’s Modulus</th>
<th>Pailler-Mattei et al.</th>
<th>Delalleau et al.</th>
<th>Bader et al.</th>
<th>Diridollou et al.</th>
<th>Sanders</th>
</tr>
</thead>
<tbody>
<tr>
<td>Method</td>
<td>Indention</td>
<td>Indention</td>
<td>Indention</td>
<td>Suction</td>
<td>Torsion</td>
</tr>
</tbody>
</table>

|                  | 35 kPa                | 5.67 kPa         | 1.51 kPa     | 129±88 kPa        | 0.1-0.02 MPa |

**Non-linear and Hyperelastic Models**

More complex theories behind the mechanical characteristics of skin describe the non-linear stress-strain response due to loading and attempt to identify parameters of the material using hyperelastic material models. Non-linear elastic models provide a means to describe observed material behavior that is not satisfied by the linear elastic relationship between stress and strain. Commonly used to model rubber’s non-linear, isotropic, and incompressible behavior, application of hyperelastic theory for biological tissues exhibiting similar characteristic behavior was a logical path [49]. It should be noted, unlike most rubbers, soft tissues are not completely incompressible.

In 2003, Hendriks et al. pointed out that many of the literature values for elastic properties of skin such as Young’s modulus are affected by the various factors such as severity of deformation, skin hydration, tissue thickness, and experimental technique. Assuming the skin was isotropic and incompressible, they utilized extended Mooney material behavior to account for the non-linear stress-strain relationship of skin during suction experiments. In a later publication, Hendriks et al. also studied the effects of
variations in suction diameter on skin’s behavior [50]. Both of the papers presented calculated parameters for extended Mooney material behavior. In 2006, Shergold et al. [51] performed both uniaxial compression and uniaxial tension experiments on pig skin in order to characterize the stress versus stretch ratio response of the tissue. They identified parameters for one term Ogden material behavior as being a good fit for the experimental data and stated that Mooney-Rivlin model was unable to describe the skin’s behavior.

Two recent papers by Wu et al. [52] and Delalleau et al. [4] described the nonlinear response of skin due to compression tests and suction tests as having two separate mechanical stiffness’s during loading. Wu et al. used an unconfined compression test on pig skin to separately identify the stress-strain properties of skin and subcutaneous tissue. They noted that the transition period from low to high stiffness associated with skin and subcutaneous tissue was at nominal strains around 0.3, and 0.4, respectively. This transition corresponded to a severe increase in the slope on the nominal stress-strain curve. Delalleau et al. performed suction experiments to gather stress-strain data on *in vivo* human skin. Agreeing with past literature, they identified two separate slopes on the stress-strain curve for skin under suction. The small slope during the initial straining of the tissue is due to the collagen fibers orienting in the direction of the stress, whereas the high slope at larger strains is due to the collagen stiffness once it has been arranged parallel to the stretch [37]. With the two separate slopes, they identified a Young’s modulus for each and the strain that corresponded to the transition period.

**Viscoelastic Model**

An added dimension commonly used to describe the behavior of biological tissues relates to its time-dependent nature when stressed. Termed viscoelastic, a material exhibiting this type of behavior reacts both viscously and elastically when undergoing deformation [30]. Common properties of viscoelasticity include hysteresis, creep, and stress relaxation. Creep refers to a material with a constant stress applied
that undergoes increasing strain. Stress relaxation refers to a material with a constant strain applied that undergoes decreasing stress. Hysteresis refers to the difference in the stress-strain relationship during cyclic loading and unloading [30]. Skin is often referred to as a viscoelastic material as it behaves with the characteristics of elastic solids as well as viscous liquids.

In 1965, Tregear et al. investigated the viscous material behavior in both human and rat skin [53] due to compression. Applying a constant force, compression over time was measured and compared to an experiment with a viscous liquid being compressed between parallel plates. In 1986, Larrabee thoroughly discussed the viscoelastic behavior of skin and commented on the affect of elastin fibers as well as the arrangement of the collagen fibers during straining [30]. In 1995, Edwards et al. [1] described the creep behavior skin exhibited during in vitro tensile testing and how it was affected by age, making it hard to characterize.

In two separate publications, Wu et al. [54, 55] investigated the viscoelastic characteristics of skin in compression. First, to develop a model consistent with physiological loading conditions, they aimed to account for the coupled non-linear elasticity and viscoelastic behavior of skin under confined and unconfined compression. Their results gave parameters for a three-term Ogden fit for the elastic properties of the tissue and a two term-Prony series fit for the stress-relaxation defining two relaxation time coefficients [54]. Secondly, creep and relaxation of both skin and subcutaneous tissue was observed during unconfined compression tests. Parameters were identified for both creep and relaxation of the two tissues [55].

Non-linear Mixture Approach

As we are concerned with not only the mechanical properties of skin due to compressive loading but also the water transport and subsequent constituent concentration changes, a mixture model was investigated. Civil engineering commonly uses a mixture approach to model the behavior of soil, with the two constituents being ground water and the solid soil. The theory behind mixture rules are discussed in the
work of Atkin and Craine [56, 57]. In 1984, Mow et al. applied the theory of mixtures to articular cartilage to investigate fluid transport and mechanical properties [58]. Following that work, in 1987 Oomens et al. [2] used a biphasic non-linear mixture theory to represent the solid and fluid phase of skin. Assuming an incompressible elastic solid and an incompressible fluid, they reduced the field equations to those used in soil analysis.

Oomens et al. again pointed out that the fiber network (collagen and elastin) is most likely responsible for the stiffness and non-linear behavior of skin during stretching. They note that in compression, however, it is the groundsubstance that most likely plays a major role in the mechanical response and that the time dependent properties can be explained when incorporating the fluid movement [2]. It is this theory that matches well with the hypothesis that localized mechanical loading expelses water from beneath compression zones, thereby changing the constituent concentrations within. As the material is compressed, fluid will be forced away. The viscosity of the fluid will provide a resistance to flow, resulting in time-dependent behavior of the solid material. A confined compression test as well as experimental results of in vitro compression of tissue on a rigid boundary done by Oomens et al. provides a bases and analytical check for potential finite element modeling of tissue compression.

6.2 ABAQUS Model

With knowledge from the literature in mind, a mixture approach seemed to best fit the needs for explaining both the solid and liquid phases of tissue. Fortunately, ABAQUS contains a built-in method for analyzing a biphasic material such as soil. By adopting the biphasic modeling method and applying tissue material constants found from the literature, a skin model was developed within ABAQUS. Using multi-scale finite element methods (FEM) in ABAQUS, a model representing external localized compression of tissue and resulting water transport was explored. The model predicted the force distribution throughout the tissue resulting from compression, the deformation of the tissue, and the directional movement of water out of regions of high stress.
6.2.1 SOILS Solver Analysis

Coupled pore fluid diffusion and stress analysis (SOILS analysis) in ABAQUS couples the deformation of a permeable elastic solid with fluid movement. The solver, referred to as SOILS, was designed to approach complex problems in soils in terms of pore pressure changes, fluid content, solid deformation, and stress analysis. The SOILS analysis models a material by considering the medium contains multiple phases. In a biphasic representation, an effective stress principle is developed to describe the coupled behavior of the solid material with the fluid, or “wetting liquid,” behavior. Saturation of the medium is defined within the analysis. When fully saturated, all voids in the material are filled with the “wetting liquid.” The mesh is attached to the solid phase of the material, allowing the fluid to flow through it. Both transient and steady-state solutions are available.

Using the SOILS analysis to represent skin during loading, the tissue was characterized as a fluid permeable medium with linear elastic material properties for the solid. The fluid was identified as water with the solid representing the collagenous groundsubstance in the dermis. For localized compression, a pin was modeled as a non-porous linear elastic material representing the sapphire compression tip. Flow for a wetting liquid through porous media is defined by Darcy’s law [59, 60], as shown:

\[
\frac{du_w}{dx} = -\frac{k}{\gamma_w} \left( \frac{\partial \mu_w}{\partial x} - \rho_w g \right) \tag{6.2}
\]

where \( f \) is the volumetric flow rate of wetting liquid per unit area of porous media, \( k \) is permeability, \( \partial \mu_w/\partial x \) is the change in pore pressure over change in position, and \( \gamma_w \), \( \rho_w \), and \( g \) are the specific weight and density of the wetting fluid and gravitational acceleration, respectively. The model assumed the media is fully saturated with the wetting liquid; therefore the void volume in tissue is equal to the volume of the fluid (water).
6.2.2 Material Definitions

Within the SOILS solver, material definitions for both the solid and liquid phase of the analysis must be defined. Void ratio, $e$, commonly defined as the volume of voids to the volume of solid, is identified in ABAQUS as

$$ e = \frac{dV_v}{(dV_g + dV_t)} $$

(6.3)

where $dV_v$ is the void volume in the medium, $dV_g$ is the volume of the grains of solid material in the medium, and $dV_t$ is the volume of trapped wetting liquid in the medium. For skin’s representation, the volume of the trapped wetting liquid in the medium is assumed to be zero. Void ratio is directly related to porosity, $n$, defined as the volume of voids over the total volume of material as shown,

$$ n = \frac{dV_v}{dV} $$

(6.4)

where $dV$ is the total, or bulk volume of the medium. ABAQUS defines a direct relationship between void ratio and porosity

$$ e = \frac{n}{1-n}. $$

(6.5)

Saturation is defined by the following relationship,

$$ s = \frac{dV_w}{dV_v} $$

(6.6)

so that a saturation of 1 indicates the volume of wetting liquid, $dV_w$, is equal to that of the volume of voids.
In addition to void ratio, the density, initial pore pressure, permeability, and mechanical properties of the solid phase of the material must be defined. Due to limited information regarding directional flow in tissue, an isotropic assumption for the water permeability through skin was adopted. It should be noted that permeability in ABAQUS, \( \bar{k} \), is commonly referred to as “hydraulic conductivity” by other authors (units of LT\(^{-1}\)). The other common definition of permeability, \( \bar{R} \), is shown below,

\[
\bar{R} = \frac{v}{g} \bar{k}
\]

(6.7)

where \( v \) is the kinematic viscosity of the wetting liquid and \( g \) is the magnitude of the gravitational acceleration. Permeability as defined in ABAQUS therefore has the viscosity of the wetting liquid already factored in. Values in the literature define a wide range of permeability for whole skin as well as human dermis. Galey et al. [61] defined permeability for whole skin as \( 4.4 \pm 1.7 \times 10^{-7} \) cm/sec and \( 611 \pm 83 \times 10^{-7} \) cm/sec for the human dermis using tritiated water. They noted that the value for whole skin they found was nearly 10 times greater than that found by Scheuplein et al [62]. Comparatively, Lesch et al. [63] found a value of \( 44 \pm 4 \times 10^{-7} \) cm/min for skin. Dick et al. found permeability of \textit{in vitro} human abdominal skin using water to be \( 0.0008 \) cm/hr [64].

It should be noted that although viscoelasticity is a dominant property of skin, it has been suggested that rapid loading can cause the skin to behave with non-viscous mechanical properties due to the lack of time for fluid to displace. This has been accomplished in unconfined compression tests in the past to measure the instantaneous elastic properties before the tissue has no time to relax [52]. With vacuum induced loading, as accomplished by the optical clearing device, the load is applied almost instantaneously to the skin. The solid material properties can therefore add another degree of complexity to the model depending on the mechanical properties assigned. To simplify the analysis of compressed tissue in the ABAQUS model in order to better understand its behavior, linear elastic material properties were assigned to the solid. The material properties used in the simplified ABAQUS model are defined below in
Table 6.2. It should be noted that an averaged Young’s modulus of elasticity was used from the literature. A quasi-incompressible assumption was made for the skin yielding a Poisson’s ratio equal to 0.45 [4].

Table 6.2: Material Properties of Skin in ABAQUS. The material properties used to define skin in ABAQUS is based on a simplified model.

<table>
<thead>
<tr>
<th>Skin Density</th>
<th>Poisson’s Ratio</th>
<th>Young’s Modulus</th>
<th>Permeability</th>
<th>Void Ratio</th>
<th>Specific Weight of Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0403 lbf/in³</td>
<td>0.45</td>
<td>20 psi</td>
<td>2.887x10⁻⁸ in/sec</td>
<td>2.333</td>
<td>0.03605 lb/in³</td>
</tr>
</tbody>
</table>

6.3 ABAQUS Model Characterization

When using a computational finite element method, it is important to verify results of the models and check that they are in line with analytical or known solutions. For an extremely complex problem such as fluid flow through pores due to deformation of an elastic medium, a simple, all-inclusive analytical check is not easily attainable. To verify both the linear elastic behavior of the solid and the pore/fluid behavior of the medium in the ABAQUS model, two separate validation models were developed.

6.3.1 Deformation in Confined Compression

Deformation of the solid in the SOILS analysis is coupled with pore pressure and therefore fluid flow throughout. By defining linear elastic material properties to the solid phase of the computational model (Young’s modulus and Poisson’s ratio) the deformation of the model under a specified stress can be easily determined. Analysis of fluid flow is available for 2D plane strain, axisymmetric, and 3D problems in ABAQUS. A 2D plane strain and axisymmetric model was developed in ABAQUS using the continuum pore fluid/stress elements defined by the program [60]. To verify the deformation of these elements under compressive stress, a simple confined compression test was developed.
As shown in Figure 6.1, a pressure force of 4 psi was applied to the top surface of a slender column developed in ABAQUS. The bottom of the part was pinned in the x and y direction while the sides of the model were prescribed a sliding boundary condition (no movement in x). Linear elastic material properties as specified in section 6.2.2 were assigned to the solid phase. The bottom was prescribed a pore pressure of zero to allow built up pressure due to deformation to leak out. A steady-state analysis was then run with the pressure force ramping up over the length of the analysis.

![Figure 6.1: Boundary Conditions and Loading for Confined Compression.](image)

Pressure was applied to the top surface of the material to deform the elements.

To compare results of the model to simple known solutions, plane strain and plain stress calculations as specified by Hooke’s Law were completed for determining the resulting strains [65]. For plane stress ($\sigma_{zz} = \sigma_{xz} = \sigma_{yz} = 0$), equations are shown below with resulting cancellations due to the boundary conditions,

$$\sigma_{xx} = \frac{E}{1-\nu^2} (0 + \nu \epsilon_{yy})$$  \hspace{1cm} (6.8)
\[ \sigma_{yy} = \frac{E}{1-v^2} (\nu \sigma_{xx} + \epsilon_{yy}) \]  
(6.9)

\[ \sigma_{xy} = \frac{E}{1+v} \epsilon_{xy} \]  
(6.10)

where \( \sigma_{xx} \) and \( \sigma_{yy} \) are the stresses in the x and y directions, and \( \epsilon_{xx} \) and \( \epsilon_{yy} \) are the strains in the x and y directions, respectively. For plane strain (\( \epsilon_{xz} = \epsilon_{zx} = \epsilon_{yz} = 0 \)), the equations are as shown below.

\[ \sigma_{xx} = \frac{E}{(1+v)(1-2v)} \left[ (1-v)\epsilon_{xx} + v \epsilon_{yy} \right] \]  
(6.11)

\[ \sigma_{yy} = \frac{E}{(1+v)(1-2v)} \left[ v \epsilon_{xx} + (1-v) \epsilon_{yy} \right] \]  
(6.11)

\[ \sigma_{zz} = \frac{E}{(1+v)(1-2v)} [ \epsilon_{xx} + \epsilon_{yy} ] \]  
(6.12)

\[ \sigma_{xy} = \frac{E}{(1+v)} \epsilon_{xy} \]  
(6.13)

Results from the ABAQUS model and the plane stress and plane strain calculations for a 4 psi applied pressure force are shown in Table 6.3.

**Table 6.3: Stress and Strain Results.** Comparisons from plane stress, plane strain, and ABAQUS model.

<table>
<thead>
<tr>
<th></th>
<th>( \sigma_{xx} )</th>
<th>( \sigma_{yy} )</th>
<th>( \sigma_{xy} )</th>
<th>( \sigma_{zz} )</th>
<th>( \epsilon_{xx} )</th>
<th>( \epsilon_{yy} )</th>
<th>( \epsilon_{xy} )</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Plane Stress</strong></td>
<td>-1.8</td>
<td>-4</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>-0.1595</td>
<td>0</td>
</tr>
<tr>
<td><strong>Plane Strain</strong></td>
<td>-3.2727</td>
<td>-4</td>
<td>0</td>
<td>-3.2727</td>
<td>0</td>
<td>-0.05272</td>
<td>0</td>
</tr>
<tr>
<td><strong>ABAQUS</strong></td>
<td>-3.273</td>
<td>-4</td>
<td>0</td>
<td>-3.273</td>
<td>0</td>
<td>-0.05273</td>
<td>0</td>
</tr>
</tbody>
</table>

Results show a match between the plane strain calculations and the pore fluid/stress elements in ABAQUS for the confined compression test. This verification
allows us to characterize the deformation behavior in ABAQUS while neglecting the pore pressure aspect of the medium.

### 6.3.2 Pore Pressure Comparison with Known Results

To account for pore pressure in the SOILS analysis and characterize its behavior, a comparison between a solution reviewed by Oomens et al. and a simple ABAQUS model is explored. In 1987, Oomens et al. [2] developed a biphasic mixture representation to approach the mechanics of skin. The simplified model consisted of two tissue constituents; an elastic solid serving as the fibre network of skin within a colloid-rich groundsubstance, and a colloid-poor Newtonian fluid. In their work, comparisons between the behavior of the mixture and two known solutions to a pressure-leakage test were investigated. The transient test represented a compression test of a column of a porous solid, initially 100% saturated with fluid. The column rests atop a porous filter while a compressive load compacts the sample at time $T>0$. The filter has a high permeability compared to the sample, allowing the column to deform in the negative $y$-direction as fluid is expelled.

The boundary conditions stated that at time $T=0$, the pressure throughout the column ($0 \leq Y \leq 1$) was known to be $P=1$, where pressure and time are non-dimensionalized. At time $T>0$, the pressure was $P=0$ at $Y=0$, which represents free drainage at the bottom of the column. There is also no flow through the top of the column, represented by the boundary condition $dP/dY = 0$ at $Y=1$ (Figure 6.3). Two methods were reviewed in Oomens’ paper calculate the varying pressure in the column over time during compression.

The first solution reviewed in the literature is a finite difference method to determine change in pressure over time throughout the column for confined compression. Pressure is related to time, $t$, and column height, $y$, by

$$
\frac{\partial p}{\partial t} = HK \frac{\partial^2 p}{\partial y^2}
$$

(6.14)
where $H$ is a confined compression modulus or aggregate elastic modulus, and $K$ is one-dimensional permeability. The finite difference method is essentially solved as a 1D heat transfer problem, with pressure replacing temperature, and the product of confined compression modulus and the one-dimensional permeability acting as the thermal diffusivity.

The other solution provided in the literature comes from Carlslaw and Yaeger’s work in 1947 on diffusing media. They related pressure to time and column height by,

$$P = \sum_{i=0}^{\infty} \frac{2}{N} (\sin NY) e^{-NT}$$

where $P$, $T$, and $Y$ are the non-dimensionalized pressure, time, and height, respectively, with

$$N = \frac{\pi}{2} (2i + 1).$$

The non-dimensionalized variables are defined as

$$Y = \frac{y}{L}$$
$$T = \frac{HK}{L^2} t$$
$$P = \frac{A}{\pi} p$$

where $y$ is the column height, $t$ is time, $p$ is pressure, and $A$ is the cross-section of the sample. Results from the solution methods are shown in a non-dimensionalized plot in Figure 6.2. Solutions for $T=0.025$, $T=0.22$, and $T=0.57$ are shown to compare our calculated solutions to those presented in Oomens’ paper.
Figure 6.2: Pore Pressure throughout the Column over Time. This figure shows the non-dimensionalized pressure versus height for both the Carlslaw and Jaeger and the finite difference solutions.

In order to characterize the pore pressure analysis in the ABAQUS model, a comparison between the SOILS analysis and the known pressure solutions was conducted. A column was therefore modeled in ABAQUS to verify the results shown in Figure 6.2. Since the porous solid is considered 100% saturated, the void ratio within the material was equal to the fluid ratio. Boundary conditions were applied to the model that prescribed an initial pore pressure to the entire column and allowed the bottom pore pressure to freely drain after time $T=0$. The bottom of the model was held in the lateral direction ($x$-direction), and an axisymmetric boundary condition was used on the left side. Using the SOILS solver, a transient solution was completed until the pore pressure throughout the body had decreased to 0. The boundary conditions for time $T>0$ are shown in Figure 6.3.
Figure 6.3: Boundary Conditions for Pore Pressure Leakage. The initial body pore pressure dispersed through the bottom of the column \((P=0)\) for \(T>0\).

Results from the ABAQUS simulation at time increments identical to those displayed in Oomens’ paper \((T=0.025, T=0.22\) and \(T=0.57\)) for the known solutions can be used for comparison. Extracting pore pressures from the meshed nodes throughout the body, as shown in Figure 6.4, allows gathering numerous pore pressures at corresponding column height locations. While completing the analysis and comparing results, it was determined that the time it took for pore pressure to expire from the column was sensitive to permeability as well as Poisson’s ratio (Appendix C). As permeability and material properties were chosen from experiments performed in the literature, uncertainty in these values could greatly affect the results of this simulation. Figure 6.5 shows the results from the ABAQUS simulation at the specified time increments. The results were very consistent to the calculated values of pressure for the material properties used in the ABAQUS model. Characterizing the pore pressure behavior in the ABAQUS model is essential to validate the simulation technique and develop confidence in the computational model for more complex problems.
Figure 6.4: Element Meshing in ABAQUS. Meshing scheme attached to the solid column (1a) and the solution at T=0.1 (1b). The top of the column in 1b still has a pore pressure of 1 (red), but the rest of the column is decreasing as pressure leaks out of the bottom of the model (blue).

Figure 6.5: ABAQUS Pore Pressure Comparison. The results of the simulation show a similar response over time as the known solution.
6.4 ABAQUS Pin Compression Results

To explore the more complex problem of tissue deformation and interstitial fluid movement due to localized loading, an axisymmetric model was developed in ABAQUS with hemisphere geometry to apply compression. The skin was represented as a biphasic soft tissue mixture containing the material properties defined above in section 6.2.2. The compression pin, representing the sapphire rods, was modeled as a linear elastic material with sapphire material properties. The stiffness of the pin due to material definitions was significantly larger than that of the tissue, effectively reducing the pin to a rigid compressor. Numerous geometries were explored for the skin representation; however, the boundary conditions and loading remained constant for all presented results in this section.

An image of the axisymmetric model for realistic tissue geometry as well as the applied boundary conditions is shown below in Figure 6.6. Applied boundary conditions included an axis of symmetry for the inner edge of the tissue (yellow dotted line) as well as a vertically constrained condition for the bottom of the tissue (motion restricted in the y-direction). The outer edge of the tissue was prescribed a vertical floating boundary condition (motion restricted in x-direction only). To model compression as accomplished in the BOSE ElectroForce experiments and to compare to published results from soft tissue compression on a rigid foundation [66], loading included a negative force (red arrow) to the mechanical transducer on a small region of tissue at the center axis of symmetry. This load represents a force similar to the rod’s effective compressive force on skin during device application and vacuum-induced tissue optical clearing. The axisymmetric representation is equivalent to looking at the half-section view of a circular piece of tissue with hemisphere-tipped compression geometry. To recreate realistic skin geometry treated by device application, a region of tissue representing the area between two pins was modeled with a thickness of 2.0 mm.
A transient simulation was performed to realistically apply the compressive load and view a transient response of water movement within the skin over time. Analysis with SOILS solver in ABAQUS consisted of three separate steps. The initial step applied all tissue properties to their respective geometries. This included assigning material properties to the skin and sapphire rod. Step-1 immediately followed the initial step and prescribed all boundary conditions as well as initiated the negative force from the sapphire pin onto the skin. Designed to be completed over a one second time period, step-1 linearly ramped the pin force from 0 lb to 0.15 lb, compressing the tissue as a result. Step-2 included a transient analysis at constant load with no new or modified conditions. During this time, transient water transport due to built-up pore pressure from skin deformation occurred. The analysis was programmed to stop when the change in pore pressure for the current time increment was lower than $1 \times 10^{-10}$. Such an insignificant change in pore pressure is likely an indication of the model approaching steady-state.
6.4.1 Mechanical Behavior of Skin during Compression

Prior to gathering results of transient water movement due to localized compression, it is important to characterize the mechanical behavior of the skin representation during loading. As Oomens et al. noted the viscosity of the fluid in a biphasic representation causes resistance to flow during loading. This resistance creates viscoelastic and non-linear behavior of the bulk material depending on the loading rate. As vacuum loading with the device is accomplished nearly instantaneously, the response of the tissue model was studied for a near-instantaneous loading condition. The skin was loaded with three separate forces over three different time periods to view variations in response to loading rates and magnitudes. Results for loads of 0.05 lbf applied over 0.025 seconds, 0.1 lbf applied over 0.2 seconds, and 0.17 lbf applied over 1 second are shown (Figure 6.7). After the force was applied, it was held constant until skin pore pressure reached quasi-equilibrium ($\Delta u < 1 \times 10^{-10}$).

![Figure 6.7: Pin Force vs. Strain.](image)

This figure shows three separate loading cases to study the viscoelastic behavior. Creep is obvious as the strain increases during a constant force.
For a load of 0.05 lbf applied over 0.025 seconds, the tissue strain reached ~0.18 during loading (red dashed line), but continued to increase to ~0.24 as the force was held constant. For the load of 0.1 lbf applied over 0.2 seconds, the tissue strain reached ~0.3 during loading (blue dashed line), but continued to increase to ~0.345 as the force was held constant. For the load of 0.17 lbf applied over 1 second, the tissue strain reached ~0.44 during loading (green dashed line), and continued to increase to ~0.455 as the force was held constant. This time-dependent material behavior is referred to as creep and is a common viscoelastic response of skin. A similar response of increasing strain during constant stress was viewed in the BOSE ElectroForce experiments in Chapter 4 and the OCT experiment in Chapter 5. It should be noted that the slope of the force/strain curve was nearly linear during loading. This matches well with the hypothesis that rapid loading can isolate skin material properties of the solid as there is not enough time for fluid to displace and the skin to relax [52].

A separate loading condition was explored to study the time-dependent reaction force due to pin displacement and relaxation of the skin in the ABAQUS model. Similar to the experiments performed by the Bose ElectroForce, a displacement was prescribed to the pin at a constant strain rate over a period of multiple seconds. The force experienced by the pin during displacement was then recorded within ABAQUS. A rigid body with identical dimensions replaced the sapphire rod for this model to extract the force experienced by the pin. Two separate cases were explored; a prescribed pin displacement of 0.025 inches over a period of 1 second and 60 seconds. The results show an increased force developed in the pin during displacement when applied over 1 second compared to 60 seconds (Figure 6.8). This implies that during faster pin displacement, water did not have time to disperse from underneath the compression geometry, causing a higher stress to build within the tissue.

Relaxation is another behavior commonly used to describe viscoelastic materials. It refers to a material that, while undergoing a constant strain experiences decreasing stress. Figure 6.9 shows force relaxation over time for a displacement of 0.025 inches applied over 1 second. For the initial displacement of 0.025 inches, the force exerted by
the tissue on the pin was ~0.086 lbf. Over the next 40 seconds, the force dropped approximately 6% to ~0.081 lbf.

**Figure 6.8: Pin Force vs. Pin Displacement.** This figure shows an increased force exerted on the pin during the faster displacement rate (1 second).

**Figure 6.9: Pin Force vs. Time.** Relaxation occurred over a period of ~40 seconds, dropping the force exerted on the pin by the tissue nearly 6% during constant pin displacement.
6.4.2 Pore Pressure and Water Movement

Again utilizing a thin skin layer (2 mm) with a tissue region representing the area between two pins, transient pore pressure increase and subsequent water movement was observed due to localized compression. A load of 0.15 lbf was applied as a linear ramp function over 1 second to the center axis of the sapphire pin to represent an effective compressive force due to device application. Initial solutions (T=0.1 seconds) in ABAQUS showed an increase in pore pressure under the compressed region and a decrease in pore pressure in other regions (Figure 6.10a). Similar results and flow fields were found by Oomens et al. [66] during soft tissue compression on a rigid foundation (shown in Appendix B). However, the magnitude of pore pressures in their study was less due to a different compressive force and compression geometries (D= 15, 40 mm). Figure 6.10b shows the pore pressure at the end of the linearly ramped loading period (T=1 second).

As specified by Darcy’s law, the change in pore pressure is the driving force for fluid flow in a porous media. Increased regions of pore pressure flow to lower regions over time in an attempt to reach equilibrium. Subsequently, fluid flow from regions of initially high to initially low pore pressure is accomplished. As outlined in the previous section, the reduced pore pressure and subsequent flow over time due to the fluid’s
viscosity results in a gradual increased strain at the compressed region until equilibrium is reached within the tissue. Results for water content at two specified times are shown in Figure 6.11. ABAQUS refers to the fluid volume in the material as Fluid Volume Ratio, or FLUVR.

![Figure 6.11: Water Ratio in ABAQUS](image)

Water ratio at two separate transient solutions show a decrease in water ratio under the compression region from (a) 1 second to (b) 60 seconds.

Initially 0.7, results show a decreased water ratio of approximately 2.5% to ~0.682 at time T=1 second at the compression zone and an increase of approximately 1.5% to 0.711 to the right of pin compression. At time T=60 seconds, results show a decrease in water ratio of 7.5% to ~0.647 at the compression zone and an increase of approximately 3.1% to 0.722 to the right of pin compression. Decreased regions of water ratio extended to the base of the tissue below the compression region (2 mm). The most significant change in water ratio occurred within the most superficial regions of the tissue down to 25% of the total thickness. Although the simulation ran for over 150 seconds, most of the transient water transport was accomplished in the first 60 seconds. The final fluid volume ratio at the end of the simulation was only 0.004 (0.647) less than at 60 seconds.
6.5 Discussion and Conclusions

Skin modeling has been accomplished with numerous material models in the past. Some publications report material constants while others simply investigate the overall behavior of skin in response to an experimental method and report their results in a descriptive manner. Whether experimentation is accomplished with compression, torsion, suction, or tension, an all-inclusive set of material parameters does not exist due to skin’s complex nature and structure. However, it is possible to characterize skin’s behavior due to a certain type of loading and geometry. An extensive literature search has resulted in a number of methods for representing skin’s complex mechanical properties and behavior in response to external loading.

Although many of the literature methods correctly describe tissue’s response in a simplified loading case, only the mixture approach incorporates the fluid movement directly corresponding with external loading. ABAQUS includes a coupled analysis which incorporates both the fluid phase as well as the solid phase of a biphasic material. Adopting ABAQUS’ biphasic analysis as a modeling technique to investigate skin behavior and water movement resulted in a complex, yet realistic simulation involving localized tissue compression. Two analytical checks have been discussed throughout this chapter to develop confidence in the values output by the ABAQUS simulation in the more complex skin model. In addition, pore pressure and consequent fluid movement due to quasi-instantaneous localized compression has been investigated. As this research is concerned with the amount of water displaced within skin due to localized compression as well as characterizing the overall behavior of tissue, the mixture approach seems to be an exceptional method to approach the modeling of skin.

Results from the localized compression model displayed a number of positive findings in regard to tissue optical clearing. In accordance with permeability values found from the literature, the majority of fluid movement was accomplished over the initial 60 seconds of localized compression in the simulations. This result implies that optical clearing of tissue due to water content modification can be accomplished in a relatively short amount of time. Also, while including two material phases in its analysis,
the mixture approach considers viscosity of the fluid phase leading to viscoelastic and non-linear bulk material behavior regardless of the solid mechanical properties. Results of both stress-based and strain-based compression simulations revealed the creep and relaxation of skin. Modeling both of these time-dependent characteristics is necessary to describe skin’s realistic behavior. It should be noted that convergence within 1% was accomplished through mesh refinement for presented models and values. Finally, although the amount of water displaced due to localized compression in the current model does not correspond with the experimentally deduced values, it is assumed that adopting a new model for the material properties of the elastic solid will allow for increased deformation and subsequent lower water concentration.
Chapter 7

Conclusions and Future Work

The potential of light-based treatments, therapies, and diagnostics for superficial layers of the body is far reaching with the ability to significantly improve the quality of human life. Optimizing optical techniques and increasing their efficiency can reduce costs, reduce treatment lengths and occurrences, and increase outcomes. Tissue optical clearing through localized mechanical compression is a promising method for improving efficiency of optical systems by increasing light transmission through skin, reducing its thickness, and providing a novel light delivery platform. Before clinical use is possible, however, significant advances must be made in understanding the mechanisms of increased optical clarity through skin.

7.1 Summary of Contributions

Tissue optical clearing has been accomplished in the past with both hyperosmotic agents as well as through localized mechanical compression. Mechanical loading for tissue optical clearing has potential advantages to hyperosmotic agents because of its non-invasive nature. It is thought that mechanical loading for tissue optical clearing can produce faster onset and more controllable results than absorbing or injecting hyperosmotic agents into the skin. Mechanical loading for tissue optical clearing is also completely reversible with no permanent effects witnessed to this point. While preliminary results have been shown for tissue optical clearing through
mechanical compression, the mechanisms behind its success have not been quantified or explored.

A new device was developed to incorporate vacuum-induced mechanical compression, skin surface cooling, and light delivery through 3 mm sapphire rods with hemisphere tips. The device adds yet another design to the plethora of prototypes designed by Rylander et al. for investigating the effects of mechanically-induced optical clearing. Designed to easily adapt depending on the application, the new device will be a key element during future experimentation. Entirely made of plastic and other non-ferromagnetic materials, the device can also be used inside of an MRI for temperature mapping during laser radiation.

A novel experimental technique to couple mechanical force by a sapphire rod and tissue strain with transmitted laser power through \textit{ex vivo} tissue has been developed in an attempt to quantify the optical effects of localized mechanical compression. Using a BOSE ElectroForce mechanical testing system to apply point compression, an optical power meter measured instantaneous transmitted laser power through tissue. Force, loading rate, and dwell time at maximum load was isolated to determine the separate effects of each variable on transmitted optical power. Loads similar to those experienced during vacuum-induced optical clearing were used to relate the effects from the \textit{ex vivo} experiments to \textit{in vivo} device application. In addition, the mechanical properties of tissue were explored during this experiment with viscoelastic effects of tissue evident during compression. This technique opens an avenue for our lab to further characterize the stress-strain behavior of \textit{ex vivo} tissue due to localized compression tests.

After increased transmission was accomplished through point loading of \textit{ex vivo} tissue, optical coherence tomography was utilized to specifically explore the dynamic change in refractive index of a skin sample during vacuum-induced localized compression. The physical thickness of the sample as well as the refractive index was deduced using the techniques of Sorin and Gray during compression. With a simple assumption characterizing the tissue as a biphasic material, a mixture rule was used to deduce water concentration from OCT images. As water content modification is thought
to be one of the main mechanisms of mechanically-induced tissue optical clearing, it is important to quantify the concentration changes in tissue due to loading. Calculated changes in water content during vacuum loading provides validation to assumptions made in the past about water concentration modification due to mechanical compression.

Finally, the mechanical response of skin was explored in an extensive literature review in which stress-strain relationships for multiple loading conditions over the past few decades were identified. The results of the literature review confirmed our expectations that the linear and non-linear response of tissue and the viscoelastic effects due to loading make skin a difficult material to model. In an attempt to verify the OCT results, a simplified computational finite element model has been developed to predict water transport and tissue strain due to localized mechanical loading. By representing skin as a biphasic material, built in viscoelastic properties such as creep and relaxation were represented due to the fluids’ viscosity. Investigating a computational finite element model to effectively represent skin’s characteristic behavior due to compressive loading provides insight into the intrinsic modifications within the dermal layer. It is imperative to quantify and explore the specific behavior of skin under loading to understand the mechanisms behind vacuum-induced optical clearing.

7.2 Future Work

Although significant progress has been made during this research to understand and characterize the optical and mechanical behavior of tissue due to localized loading, there is much room for further development. Experimentation to characterize skin’s behavior to external forces has been an ongoing area of research for nearly a century. However, minimal information has been gathered for skin behavior due to compressive forces in comparison to that done on tensile, torsion, and suction. The work that has been done is primarily descriptive or specific to one type of loading and fails to identify all-inclusive constants for tissue’s behavior due to stress. Modeling skin’s behavior is therefore very difficult as published material constants vary with linear, non-linear, and
viscoelastic responses. Modeling skin’s behavior due to vacuum-induced loading is also very complicated because it includes a compressive force as well as a suction force.

### 7.2.1 Experiments

The optical coherence tomography experiment and transmitted optical power measurements due to localized loading were a sufficient start to identify changes in tissue’s optical and mechanical properties due to pin compression. Both experiments provided data to reinforce the hypothesis that localized loading has an effect on the intrinsic characteristics within tissue. However, additional work needs to be done to effectively characterize skin’s behavior and gather reproducible results.

#### Optical Power Measurements

Optical power measurements through skin during point compression showed a significant increase in the transmitted power. Comparison of overall percent increase in optical power varied depending on the compression geometry, maximum load, and dwell time. The initial values of transmitted optical power, however, varied depending on the tissue sample. More precise techniques must be developed to produce similar tissue samples for results comparison. Although the preliminary results provided sufficient data for an overall comparison between loads and geometries, further experimentation with comparable skin samples must be done to isolate and compare specific loading cases. Poor resolution of the optical power meter also limited the confidence in the overall results, making it difficult for comparisons between experimental runs for overall increased optical power. A second set of experimental data should show an increased resolution for the optical power meter by changing data acquisition settings. The resolution of the load cell could also be improved by placing the entire setup on a floating optics table, thereby reducing any external effects such as vibration.

In addition, the loading rate did not have a significant or obvious effect on the transmitted optical power. This could be contributed to the fact that loading rate was only varied by a factor of 4 (0.025 mm/s to 0.1 mm/s). A larger range of loading rates
could provide more insight into the viscoelastic behavior of skin. Some authors suggest that near-instantaneous loading will reduce the effective viscoelastic properties of the tissue during initial loading. Loading during the experiment discussed in this thesis was accomplished over seconds or minutes. As vacuum-induced loading is applied nearly instantaneously, a much faster loading rate should be explored. Dwell time had a significant effect on the overall transmitted optical power due to tissue’s viscoelastic properties. However, light intensity and tissue strain continued to rise during the entire 60 seconds of dwell time in this set of experimental data. A longer dwell should be instituted to decide an appropriate time for equilibrium to be reached. Reaching equilibrium is imperative for optimizing results for tissue optical clearing.

Using a sensitive mechanical testing system such as the BOSE ElectroForce also allows for investigation into skin mechanical properties. A number of authors have investigated the hyperelastic and viscoelastic properties of tissue with confined and unconfined compression tests. Continued exploration of point loading could lead to characterization of tissue properties due to localized confined or unconfined compression. A hyperelastic model could be fit to the data and those parameters could be used for future modeling of tissue’s compressive nature. Also, viscoelastic constants for creep and relaxation could be identified for future modeling purposes.

**OCT Experiment**

The OCT experiment showed a significant change in the refractive index and thickness of skin due to vacuum loading. A biphasic assumption allowed water content deduction from refractive index data, which showed a significant reduction in water concentration during vacuum loading. However, results were only analyzed for a single experiment for one thickness and layer of skin. Repeated experiments are necessary to verify the results from the data presented in this thesis. Also, different compression geometries (radius of indenter, shape) should be investigated to compare results.

The epidermis and subcutaneous most likely have a large effect on the response of tissue due to loading and therefore should be considered during experimentation in
the future. In addition, optical path distances were gathered from the center of the skin at the apex of the ball lens by averaging over a number of pixels in the OCT images. Refractive index can be gathered at separate lateral locations along the ball lens’ surface for location-dependent measurements. Further examination of these measurements can provide insight into water transport for the spatiotemporal data set.

### 7.2.2 Model

An exhaustive literature search has identified numerous publications that provide linear elastic, non-linear elastic, and viscoelastic constants for skin’s stress-strain and time-dependent behavior. Hyperelastic models, such as Ogden, Mooney-Rivlin, and Neo-Hookean have commonly been used to fit the stress-strain data gathered during experimentation. Non-linear and viscoelastic parameters have been determined to describe the characteristic behavior for confined and unconfined tissue compression. A mixture approach to the mechanics of skin has also been studied by Oomens et al. As this thesis was the first step to exploring the modeling of water transport through tissue due to vacuum-induced loading, many improvements can be made to the simplified model presented for a more realistic representation of skin.

Skin was modeled as a biphasic mixture with the solid portion representing the collagen rich groundsubstance and the water representing the colloid-poor Newtonian fluid. Linear elastic material properties were assigned to the water permeable solid and room temperature material properties were assigned to the water. The biphasic material performed similar to tissue in many aspects as outlined in this thesis. Although no viscoelastic properties were specified, the viscosity of the fluid phase created time-dependent behavior such as creep and relaxation. Along with tissue deformation and tissue strain, fluid (water) volume ratio of the material can be deduced from the computational model, making it a desirable technique for future exploration of mechanically-induced tissue optical clearing. However, the boundary conditions in the simplified model investigated throughout this thesis are not realistic for vacuum loading. In addition, the amount of water moved during loading was less than we expect as well
as that deduced from the OCT experiment. To explore improvements to the model, initial solutions were developed in ABAQUS and subsequent recommendations are made.

Vacuum Loading

Because the majority of the effort for modeling skin was directed towards characterizing the behavior of the bulk material and fluid phase during localized compression, the realistic loading condition experienced during device application and subsequent vacuum loading was not explored. Adding a degree of complexity, vacuum loading incorporates not only an effective compressive force beneath the pin but also a suction force between the pin and circumscribing brim of the device. This loading condition causes tissue to react under a compressive and tensile force simultaneously.

Applying a positive force (positive y-direction) to the top layer of skin in the ABAQUS simulation while restricting motion of the sapphire rod would provide a similar loading condition to that accomplished during *in vivo* device application. However, the rigid foundation at the base of the skin as currently modeled would prevent movement of tissue during suction. To account for this, a moving boundary condition can be prescribed by applying stiffness (elastic foundation) to the bottom boundary. A first-cut expanded model was explored by applying a pressure load to the top surface of the skin. The bottom tissue boundary was prescribed an elastic foundation defined as having similar stiffness to muscle or subcutaneous tissue. Figure 7.1 shows the deformation of the elastic foundation as well as the fluid volume ratio of the tissue during loading. An interesting effect during simulation showed an increased fluid volume ratio above the center axis of the rigid foundation. To realistically replicate the conditions experienced by the tissue during device application, further work must be done in the future.
Figure 7.1: Vacuum Loading. To realistically recreate the loading condition from the device, vacuum loading must be modeled.

Multi-layer Skin

As shown in the previous image, boundary conditions can greatly affect the behavior of a material exposed to loading. The bottom boundary defined in the sapphire rod compression model in chapter 6 allowed no fluid flow and no movement in the y-direction; effectively representing a rigid, non-porous foundation. As skin realistically sits on a foundation of subcutaneous and muscle in most parts of the body, more realistic boundary conditions for the model must be explored. Simultaneous determination of subcutaneous and muscle layer mechanical characteristics have been investigated in the literature during compression and indentation tests [46, 52]. Adding additional layers below the dermis (Figure 7.2) could create a more reasonable bulk material coinciding with more realistic deformations and stress-strain relationships during loading. Future work should include added layers to the current model as well as investigation into the possibility of fluid flow between layers.
Figure 7.2: Device Geometry, Multi-layer Skin. An extended depth of the tissue including multiple layers could more realistically represent the boundary conditions during in vivo skin loading.

New Material Definitions- Inverse Analysis

The biphasic representation of skin using SOILS analysis in ABAQUS has proven to be an exceptional method to study the deformation and water transport in skin due to localized loading. The resulting fluid volume ratio under compression zones during simulation, however, does not match well with experimental data or expected results. Although behavior such as tissue creep and relaxation were adequately represented during simulation, the linear elastic material properties attached to the solid seemed to prohibit accurate bulk material behavior. To justify this assumption, a simple ABAQUS model was created to apply a prescribed pin displacement to skin geometry representative of that in the BOSE ElectroForce experiment. An identical loading condition to that achieved in the experiment was applied including a rod displacement of 0.025 inches (equal to 0.33 tissue strain) over 41.5 seconds. The force experienced by the pin during displacement was then recorded in ABAQUS. Figure 7.3 shows the increasing force and increasing tissue strain during point loading for both the ABAQUS model as well as a BOSE ElectroForce compression experiment.
Figure 7.3: Model Comparison to BOSE Experiment. Results from the model during a prescribed pin displacement shows matched results until a tissue strain of approximately 0.1.

As shown, the linear elastic material definition of the solid, which governs the overall stiffness and deformation of the skin, leads to the tissue only coinciding with the experimental data for the first ~0.1 tissue strain. The behavior of the ex vivo tissue in the BOSE experiment displays a non-linear increasing stiffness with significant strain increase. To account for this behavior, I suggest a non-linear or hyperelastic model be used in the future to describe the solid phase of the skin. ABAQUS contains a feature in which it fits a hyperelastic model to test data that is experimentally obtained. Also, by utilizing experimental data an inverse solution could back out material properties of the skin. Much iteration would have to be accomplished as skin behaves differently under varying strain rates and loading conditions. Wu et al. identified the nonlinear-elastic behavior of the skin due to unconfined compression with 90% confidence intervals [52]. Their results showed a very similar loading curve to the BOSE experimental data with a transition from the low stress region to high stress region occurring around 0.3 tissue strain.

A first-cut simulation utilizing hyperelastic solid material properties from the literature was developed in ABAQUS. Results (Figure 7.4) show a significant decrease in fluid volume ratio at the compression zone and near the rigid foundation as well as
increased tissue strain when compared to the linear elastic model. This is an encouraging result as it shows the potential of another material model to better represent the tissue strain and water content modification. Although the result is encouraging, little confidence should be placed in it as the material properties used in the model were obtained from the literature for a unique loading case. Therefore, an inverse analysis should be performed in the future to accurately acquire material properties from localized compression experimental data for skin modeling.

Figure 7.4: Hyperelastic Model with Ogden Coefficients.
References

22. Tuchin, V.V., "Tissue Optics: Light Scattering Methods and Instruments for Medical Diagnosis". SPIE Tutorial Texts in Optical Engineering TT38. 2000, Bellinham, WA.
Appendix A

Mesh Convergence

To correctly gather information from the ABAQUS simulations element meshing was investigated to ensure results were consistent versus artifacts due to element size. Meshing was accomplished by seeding each edge of the material with a prescribed number of nodes. As shown below in Figure A.1, coarse meshing schemes were investigated as well as fine element meshing.

![Figure A.1: Mesh Convergence in ABAQUS.](image)

Mesh convergence included seeding the edges of the elements with a prescribed number of nodes. Seed edge by size was accomplished over a wide range including (a) 0.01 and (b) 0.0025.

Each meshing scheme was performed with a structured technique and quadrilateral element shapes. Two distinct nodes with separate output values were chosen to compare different meshes. Minimum fluid volume ratio at the compressed region and displacement of the top right corner of the model were compared over several meshing scheme. At a “seed edge by size” value of 0.0015, convergence was achieved with less than a 1% difference in values when compared to 0.002.
Table A.1: Mesh Convergence Results. The displacement at the upper right point of the figure and the overall smallest void ratio were used for mesh convergence. The meshing scheme used was determined by percent difference dropping below 1%.

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Appendix B

Permeability and Poisson’s Ratio in Pore Pressure Column

Figure B.1: Pore Pressure Results for Varying Material Parameters. Pore pressure was sensitive to permeability and Poisson’s ratio.