DESIGN AND VALIDATION OF A WEARABLE, CONTINUOUS, AND NON-INVASIVE
HYDRATION MONITOR THAT USES ULTRASONIC PULSES TO
DETECT CHANGES IN TISSUE HYDRATION STATUS

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by
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TITLE: Design and Validation of A Wearable, Continuous, and Non-Invasive Hydration Monitor That Uses Ultrasonic Pulses to Detect Changes in Tissue Hydration Status

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ABSTRACT

Design and Validation of A Wearable, Continuous, and Non-Invasive Hydration Monitor That Uses Ultrasonic Pulses to Detect Changes in Tissue Hydration Status

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Chronic dehydration is an endemic problem for many population groups. Current methods of monitoring hydration status are invasive, time consuming, cannot be performed while exercising, and require lab resources. A proposed solution is a wearable, continuous, and non-invasive device that uses harm-free ultrasonic pulses to detect changes in tissue hydration status over time. Customer and engineering requirements were defined and used to guide the design process. Literature reviews were performed to identify essential information on dehydration, assess current methods, discover state of the art devices, and describe ultrasonic theory. Market research was performed to identify athletes as the target population group. An adjustable elastic nylon bicep band prototype was manufactured and the integration of more advanced components was proposed. The theoretical signal processing method used to detect hydration status was validated through initial tests with a prototype electrical system composed of a Teensy 3.1 board, two 18 kHz piezoceramic disc elements, and an Arduino/LabVIEW interface. Tests with aluminum, rubber, and sponge materials were performed to compare the signal response to propagation through materials with different acoustic properties and water contents. Finally, tests performed with dehydrated bovine muscle tissue revealed a statistically significant difference between hydrated and dehydrated tissue, a promising indication for future device refinement.

Keywords: Ultrasonic, Hypohydration, Euhydration, Dehydration, Tissue Status, Wearable, Non-invasive, Piezoceramic, PZT, Transducers, Signal Processing, Phase Shift, Harmonics

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1. INTRODUCTION

1.1 Problem Statement

Seventy five percent of Americans are chronically dehydrated and care for dehydration results in up to \$5.5 billion per year in preventable hospital costs in the US alone¹. In hospital emergency departments, dehydration is the most common diagnosis in general (7%), early-return (15%), and return-admit (25%) populations². In daily life, the average person turns over 4.58 liters of body water per day and inadequate replenishment of body fluids can lead to significant detrimental effects on overall wellness^{3,4}. Surprisingly, there is no standardized or satisfactory tool for quantification of hydration status despite common recommendation to provide it for athletes, military, geriatric care, pediatric care, prenatal monitoring, firefighters, dialysis patients, and for EMT and general hospital use⁴⁻⁸.

There are many studies that have attempted to develop methods to improve the identification of dehydration and subsequent monitoring policies^{4,6-17}. Despite frequent investigation, there are no satisfactory continuous and non-invasive hydration monitors for hospital use or on the market. Further, the use of subjective assessment measures by physicians and overall lack of awareness of hydration status contributes to dehydration being under-recognized amongst hospitalized, geriatric, and otherwise healthy populations¹⁸.

1.2 Solution

Studies show that the sole act of wearing a fitness device increases a user's awareness of their fitness levels^{19,20}; likewise, wearing a continuous hydration monitor can make users more aware of their hydration and act as a subconscious reminder to stay hydrated. Additionally, the use of a hydration monitor can help prevent dehydration-related illnesses and improve fitness-training methods^{4,21}. The proposed solution is a wearable, continuous, and non-invasive device that uses ultrasonic pulses to detect changes in tissue properties over time. This device provides near real-time monitoring of hydration levels and alerts users of their current hydration status. The near real-time monitoring provides a significant

benefit over other methods that cannot be used while exercising, require constant stops in activity to get feedback, require additional lab equipment, and require significant time resources. This device aims to eliminate invasive, time-consuming, and impractical lab tests to open up a whole new method of patient monitoring and significantly decrease the ineffective subjective treatment of dehydration.

1.3 Purpose

The purpose of this thesis is to design, test, and validate a wearable, continuous, and non-invasive hydration monitor that uses harm-free ultrasonic pulses to detect changes in tissue hydration status.

Within this purpose, there are four subsidiary goals:

- (1) Research current methods of hydration monitoring and identify the benefits of an ultrasonic method.
- (2) Identify an optimal target customer and anatomic location for a hydration monitor.
- (3) Design a device to meet specified customer and engineering requirements.
- (4) Validate the device methods.

2. BACKGROUND

Importantly, there is no standardized test for measuring hydration²². The lack of a standardized test is particularly surprising considering the amount of water in the human body, the importance of water in maintaining homeostasis, the significant physiologic impact of relatively small percentages of body weight water loss, and the prevalence of disruptions of water balance in diseases and ailments commonly treated in hospital settings. In this section, the essentials of dehydration, healthy body water content, the effects of dehydration, clinical presentation of dehydration, suggested hydration strategies, patient populations for a hydration monitoring device, current methods of monitoring hydration status, and state of the art hydration monitors will be discussed.

2.1 Essentials of Dehydration

Water is the most abundant chemical substance in the body, accounting for 60-80% of human body weight²³. The importance of water in the body is derived from five key properties of water: high heat capacity, high heat of vaporization, polar solvent properties, reactivity, and cushioning²³. See Table I for an explanation of each of these properties and their role in maintaining body homeostasis.

Table I: A table of the key properties of water and the role of each property in maintaining homeostasis²³.

Property	Description	Role in Homeostasis
High Heat Capacity	Water absorbs and releases large amounts of heat before changing in temperature	<ul style="list-style-type: none">- Prevents sudden changes in temperature due to external or internal factors- Redistributes heat amongst body tissues
High Heat of Vaporization	Large amounts of heat are required to vaporize water by transforming from liquid to gas via the breaking of molecular hydrogen bonds	<ul style="list-style-type: none">- Allows large amounts of heat to be removed from the body via perspiration- Results in efficient cooling
Polar Solvent Properties	Polarity of water molecules allows for the orientation of slightly negative ends of water towards positive ends of solvents, thus attracting and surrounding other molecules	<ul style="list-style-type: none">- Dissociates ionic compounds and other reactive molecules, allowing for chemical reactions- Allows for the formation of biological colloids by forming hydration layers to shield large charged molecules- Serves as a transport medium to carry

		nutrients, respiratory gases, and metabolic wastes
Reactivity	High reactivity allows water to be a reactant in many chemical reactions	- Allows for hydrolysis reactions and dehydration synthesis, notably in the cases of food digestion and enzymatic activity
Cushioning	Can be used to form a cushion around body organs	- Protects organs, notably the brain and spinal cord, from physical trauma

As seen in Table I, water has a significant role in maintaining proper physiological function. When water homeostasis, known as euhydration, is disrupted, there are substantial side effects²⁴. This disruption can be from inadequate water balance or excessive water balance²³. The following definitions should be considered when investigating hydration status:

- **Hyperhydration:** a state of water excess, or positive water balance²⁴.
- **Euhydration:** a dynamic state of maintaining a proper water balance²⁴.
- **Hypohydration:** a state of insufficient water, or negative water balance²⁴.
- **Dehydration:** the process of losing water from the body²⁴.
- **Rehydration:** the process of gaining body water²⁴.

To understand the consequences of dehydration, it is important to understand the movement of water within the body. See Figure 1 for a diagram of the key water transportation pathways. This diagram shows that total body water resides in two fluid compartments: the intracellular fluid compartment (~66%) and the extracellular fluid compartment (~33%)²³. There are two subcompartments within the extracellular fluid compartment: plasma and interstitial fluid²³. Water moves between compartments along osmotic gradients, which are driven by the concentration of solutes in each compartment²³. Thus, changes in solute concentration result in net water flow into or out of a compartment²³. In euhydration, water flow between compartments is regulated to maintain equal osmolalities between all body fluids²³. To maintain euhydration, increases in the extracellular solute concentration drives water out of cells²³. Conversely, decreases in the extracellular solute concentration drives water into cells²³. It is by this mechanism that

water contributes to the transportation of gases, nutrients, and wastes between the fluid compartments of the body²³.

Figure 1: Exchange of gases, nutrients, water, and wastes between the three fluid compartments of the body²³.

To maintain euhydration, water input must equal water output. Water leaves the body via several routes: vaporized water in expired air, insensible water loss by diffusion through the skin, excretion in feces, and excretion in urine²³. Water output is countered by ingestion of food or liquids and metabolic water produced by cellular metabolism²³. The balance between water output and input is regulated through a thirst mechanism in which an increase in plasma osmolality of 2-3%, presence of angiotensin II, baroreceptor inputs, or other stimuli triggers osmoreceptors in the thalamic thirst center²³. The thirst response is fulfilled soon after drinking water, as the mucosa of the mouth and throat are moistened and stretch receptors in the GI tract are activated²³. Importantly, this quenching response is premature; that is, it allows time for the osmotic gradient changes and prevents over-dilution of body fluids²³.

The premature quench response to hydration does not always provide the body with enough fluids to maintain euhydration²³. For instance, the thirst response can be quenched during demanding athletic activity before adequate water is consumed, the threshold level for a thirst response is highly variable in

people²², elderly may not recognize thirst signals, and, conversely, renal and cardiac patients may be thirsty despite an overload of fluids²³.

Dehydration is essentially when water output exceeds water input over a period of time²⁴. This can occur through two pathways: intracellular or extracellular dehydration²³. Intracellular dehydration is caused by pure water loss from the skin via diffusion and from hypotonic solute losses from sweat²⁵. Conversely, extracellular dehydration occurs when isotonic solute losses accompany body water, such as during cold or high-altitude exposure and in response to secretory diarrhea²⁵. The most significant difference between intracellular dehydration and extracellular dehydration is that intravascular volume contraction is larger in extracellular dehydration, resulting in osmotic and volume differences between the two types of dehydration²⁵. It is important to note that the simplistic definition of dehydration does not specify whether the water balance is disturbed through loss of water with solute in the extracellular fluid compartment or from primary water loss in the intracellular fluid compartment, a factor that must be considered when reviewing literature on dehydration. For example, Ely et al. designed a method to assess extracellular water content²⁵ while a study by Vivanti et al. did not differentiate between intracellular water loss and total body water loss⁶. Considering that both types of dehydration have detrimental physiological effects, that review of literature reveals inconclusive preferences for the importance of monitoring a specific type of dehydration, and that the design of the device in this thesis measures the combined effect of both types of dehydration, any reference to dehydration in this thesis, unless specifically stated as intracellular or extracellular, can be assumed to be whole body dehydration.

2.2 Healthy Body Water Content and Clinical Presentation of Dehydration

Total body water is a function of age, body mass, gender, and relative amount of body fat²³. Typically, infants are 73% water, young males are 60% water, young females are 50% water, and elderly are 45% water²³. The variations in water content among these populations are explained by anatomical differences and tissue-related compositions; for example, infants have low body fat and low bone mass, females have relatively more body fat and less skeletal muscle than males, and greater muscle mass results in higher

water content²³. Further, adipose tissue, typically 20% water, is far less hydrated than skeletal muscle, typically 75% water²³. In fact, studies have found that the absolute water content in muscles varies from 65-76% in people with different total body fat contents²⁶.

Thus, healthy body water content is dependent on individual factors. One person's healthy body water content value cannot be extrapolated for large populations of people¹⁸. To compare extents of fluid loss between people, dehydration is referred in percentage body water weight loss (%BW)⁶. In general, the clinical presentation of dehydration is dependent on the extent of dehydration. Given the high water content of human bodies and the importance of water for maintaining proper homeostasis, loss of even small %BW can have significant consequences^{4,21,23,24}. For every 1% body mass fluid loss, there is an increase in internal body temperature of approximately 0.22°C²⁷. Water loss of only 2% BW results in discomfort, a drop in blood volume, increase in heart rate, and may begin to significantly affect athletic performance and other common bodily functions²³. Loss of more than 3% BW further disturbs physiologic function and increases the risk of heat illness²⁸. Loss of 4% necessitates an increased effort for physical work, 5% causes difficulty concentration, 6% causes impairment in exercise temperature regulation, 10% causes spastic muscles, and 15% causes death⁴. See Figure 2 for a diagram of average body water contents and the effects of percentages of body water weight loss.

Figure 2: Diagram of body water content amounts and the effects of % BW water loss^{4,23,28}.

2.3 Suggested Hydration Strategies

Optimal hydration strategy depends on energy expenditure, environment, and individual needs²⁹. See Table II for the United States Department of Agriculture (USDA) Dietary Reference Intake guidelines for water consumption. As seen in the table, the suggested water consumption for males varies from 2.4 to 3.7 L/day and from 2.1 to 2.7 L/day for females³⁰. Note that these guidelines are for median intakes of generally healthy individuals that are adequately hydrated³⁰. On top of median intake values, the National Athletic Trainer's Association suggests consuming 200-300 mL of fluid on average for every 10-20 minutes during exercise to prevent more than 2% BW water loss per day²⁸. More specifically, a guiding committee led by Casa et al. provided USA Track & Field athletes with a self-testing program for optimum hydration³¹. In this program, it is recommended that a hydration strategy considers the sweat rate, sport dynamics (rest breaks, fluid access), environmental factors, acclimatization state, exercise duration, exercise intensity, and individual preferences³¹. Examination of the suggested hydration strategies revealed a key outcome: the available strategies require complex data logging and must be

recalculated for each activity, climate, environment, and activity intensity^{28–30}. These complexities could be eliminated with the use of a real-time hydration monitor.

Table I: USDA Dietary Reference Intake guidelines for water consumption³⁰. AI = allowable income.

Nutrient	Function	Life Stage Group	AI
Water	Maintains homeostasis in the body and allows for transport of nutrients to cells and removal and excretion of waste products of metabolism.		(L/d)
		Infants	
		0–6 mo	0.7
		7–12 mo	0.8
		Children	
		1–3 y	1.3
		4–8 y	1.7
		Males	
		9–13 y	2.4
		14–18 y	3.3
		19–30 y	3.7
		31–50 y	3.7
		50–70 y	3.7
		> 70 y	3.7
		Females	
		9–13 y	2.1
		14–18 y	2.3
		19–30 y	2.7
		31–50 y	2.7
		50–70 y	2.7
		> 70 y	2.7
		Pregnancy	
		14–18 y	3.0
		19–50 y	3.0
		Lactation	
		14–18 y	3.8
		19–50 y	3.8

2.4 Patient Populations

Aside from use by the general population as a tool for maintaining overall fitness, there are many patient populations that could benefit from a wearable, continuous, and non-invasive hydration monitor. In literature, the most frequently listed groups include athletes, soldiers, geriatric care, pediatric care, prenatal monitoring, firefighters, dialysis patients, and for EMT and general hospital use^{4–8}.

Given the importance of proper hydration for optimal athletic performance, the effects of dehydration and the importance of hydration monitoring for athletes has been widely studied^{10,27,29,31–40}. Regardless of the

method used to assess hydration status in athletes, a common finding among athletic hydration studies is that athletes frequently report to practices or games in an already-dehydrated state. For example, a study by Prentice et al. found that the hydration status of 30 elite rugby participants was consistently lower than recommended levels³², Chia et al. found that 39/40 high school hockey players arrived at a competition already displaying signs of dehydration³³, and Kutlu et al. reported that elite junior taekwondo athletes display indicators of chronic dehydration³⁴. Additionally, athletes commonly train in conditions where heat-related illnesses – like dehydration – are a concern. Football players in preseason summer practices are a particularly at-risk population. Yard et al. reported that since 1995, 31 high school football players have died from heat stroke and that 4.5 per 100,000 athletes suffer from heat-related illnesses, such as dehydration, of which 36.9% miss more than two days of practice³⁵.

Compounding the prevalence of dehydration in athletics are the significant detrimental effects of dehydration on athletic performance. For example, hypohydration has been associated with increased cardiovascular strain and, under severe conditions, cardiac dysrhythmia and arrest may occur⁴¹. Further, dehydration reduces overall heart rate variability and decreases autonomic cardiac stability following exercise⁴¹. The detrimental effects of dehydration are seen particularly in marathon runners, where 15% of runners with exercise-associated collapse (59% of medical tent visits) require intravenous hydration⁴². In total, the athletic population can significantly benefit from monitoring hydration to decrease the detrimental effects of dehydration and improve overall performance.

In the military, soldiers commonly dehydrate 2-5% of BW due to high rates of water loss from environmental exposure and performing stressful physical work⁴. In basic training and active duty, recruits may be sent to areas in which they are not acclimatized to the heat, thus increasing the likelihood of exertional heat illness²⁹. Additionally, of total dehydration-related exertional heat illness cases during basic training, approximately 50% of cases occur in recruits without identifiable risk factors²⁷. Soldiers in active duty may have to be in the field for long periods of time, for which adequate hydration sources must be available. Maughan et al. suggest that soldiers should consider their hydration status before

beginning exercise, the need for fluid, electrolyte, and substrate replacement during exercise, and the need for restoration of water and electrolyte balance after exercise²⁹. In sum, to reduce the water load carried by soldiers, reduce cases of exertional heat illness, and improve cognitive focus during highly stressful military operations, it would be beneficial to have a real-time hydration monitor.

Hydration monitoring for geriatric care is a frequent area of investigation^{6-9,13,15,18,43-45}. Compared with younger populations, the elderly have complications with maintaining euhydration; these complications include a low average body water content, a lack of recognition of thirst signals, and a reduced ability to maintain temperature homeostasis²³. A diagnosis of dehydration in clinical care has been associated with increased morbidity and mortality, and older people 85-99 years of age were reported to be six times more likely to be admitted to the hospital with dehydration than those 65-69 years old⁴⁶. Additionally, the elderly are the largest growing population group and place large demands on medical providers⁶. The difficulty in balancing geriatric physiology with existing measures of dehydration monitoring is shown in a study by Rösler et al., in which bioelectric impedance had less than 44% concordance with clinical judgment of dehydration⁴³. Thus, a more accurate and non-invasive method of monitoring hydration status may significantly reduce the number of elderly hospital remittances, improve geriatric care, and reduce the burden of an aging population on the health system.

Pediatric care and prenatal monitoring can also benefit from a hydration monitor^{4,5,10,27,47-49}. The high water content of infants and children makes proper hydration critical in the prenatal and pediatric stages, specifically for healthy development. For example, failure to increase body water content during pregnancy has been associated with adverse pregnancy outcomes⁵⁰. Additionally, progressive reduction in body water content during gestation has been linked to gestational hypertension and pre-eclampsia, low birth weight, and poor pregnancy outcome⁵⁰. In a pediatric unit, some patients may need hydration status monitoring daily or hourly¹³. While the small size of infants likely requires design modifications, the development of a non-invasive method to monitor hydration is particularly suited for providing ease-of-mind during the prenatal process.

Firefighters are particularly at risk for dehydration⁵¹⁻⁵³. The heat exposure from fires, stressful work environment, heavy personal equipment, time spent in the field, lack of access to drinking water, and heavy protective equipment contribute to profuse sweating and rapid dehydration⁵². Simulations of fire activities found that dehydrated firefighters demonstrated significantly greater cardiovascular strain than euhydrated firefighters, which emphasizes the importance of proper hydration management prior to arriving on the job⁵². In addition to encouraging firefighters to stay properly hydrated, a hydration monitor could prevent serious instances of dehydration in the field.

A hydration monitor could have significant impact in a hospital environment. For instance, numerous pathologic signs and symptoms, such as significant changes in body fluid retention pre- and post-surgery, digestion problems, high blood pressure, muscle cramps, edemas, or obesity, present in clinical patients with tissue water imbalance⁴. In cardiac applications, fluid overload in patients with acute heart failure has a significant diagnostic and prognostic importance⁸. Thus, a method of easily, quickly, non-invasively, and cost-effectively monitoring hydration can significantly assist diagnostic and therapeutic decision-making for cardiac patients. As a final example, patients on bed rest experience a severe restriction of muscular activity and the equilibrium between intake and loss of electrolytes is markedly affected⁵⁴. Proper hydration strategies can significantly decrease the detrimental effects of bed rest on patients, namely the increase in body dehydration status, excessive mineral loss in urine, and abnormal mineral levels in plasma⁵⁴. In sum, including a test for hydration status on a standard patient intake assessment can increase diagnostic capabilities and provide another metric for monitoring critical patients.

2.5 Current Methods

In the absence of a standardized method to monitor hydration status, there is debate in literature over the best way to measure dehydration^{7,8,14,15,21,55}. In general, there are two classifications of methods to monitor hydration status: qualitative methods and quantitative methods. The use of a particular method depends

on access to equipment, clinical or home setting, diagnostic importance, time available, access to sample-taking resources, and investigator/user familiarity or preference for a particular method.

2.5.1 Qualitative Methods

Qualitative methods for monitoring hydration status include the thirst response, urine color comparison charts, and physician assessment of dehydration-related symptoms. As mentioned in the essentials of dehydration section, the premature quench response to hydration does not always provide the body with enough fluids to maintain homeostasis. For instance, the thirst response can be quenched during demanding athletic activity before adequate water is consumed, the threshold level for a thirst response is highly variable in people²², elderly may not recognize thirst signals, and, conversely, renal and cardiac patients may be thirsty despite an overload of fluids²³. Thus, the thirst response should be avoided as a sole method of monitoring hydration status.

A urine color chart can be used as a visual marker of hydration⁵⁶. See Figure 3 for an example chart in which urine color matching #1-#3 is well hydrated, #4-#6 is dehydrated, and #7 or below requires immediate consumption of fluids. Once a person is aware of the general scale, it does not require any equipment to use a urine color chart. However, urine output may be suppressed in cases of moderate to extreme dehydration and the time delay in onset of the effects of dehydration and output of urine is detrimental to maintaining a healthy hydration status.

Figure 3: Urine color chart used to assess hydration status⁵⁶.

Physician assessment of dehydration-related symptoms uses the presenting state of a patient to evaluate hydration status. See Table III below for an example of a clinical assessment scale for dehydration.

Unfortunately, clinical assessments of dehydration frequently rely on subjective measures performed by physicians that do not regard dehydration as a significant risk factor for hospital monitoring^{2,57}. Therefore, assessments of dehydration can vary significantly based on the physician's experience. For this reason, the use of qualitative methods should be supplemented by the development of a standardized quantitative method and, thus, improve the diagnosis and subsequent treatment strategies for patients with dehydration.

Table III: Dehydration assessment scale⁵⁷.

Sign	Mild (5%)	Moderate (6%-10%)	Severe (>10%)
Skin turgor	Slightly decreased	Decreased	Very decreased
Skin color	Pale	Sallow	Ashen
Oral mucosa	Dry	Very dry	Parched
Tears	±Decreased	Absent	Absent
Fontanel	Normal	Depressed	Sunken
Pulse	±Increased	Increased	Marked tachycardia
Blood pressure	Normal	±Decreased	Decreased
Urine output	Mild oliguria	Oliguria	Anuria
Mental status	Irritable	Lethargic	Unresponsive

2.5.2 Quantitative Methods

Quantitative methods of monitoring hydration level include BW differences, urine output, urine specific gravity, urine osmolality, plasma osmolality, saliva osmolality, dilution methods, and bioimpedance²². Of these methods, bioimpedance and BW differences are the only non-invasive options. BW differences have significant limitations for monitoring hydration status, which will be discussed in the next paragraph. Additionally, due to the problem statement emphasis on non-invasive options for monitoring dehydration, bioimpedance is discussed in detail in the following section. The use of multiple quantitative methods in studies is likely due to the lack of a standardized method for monitoring dehydration, the ability to acquire multiple measures from one sample (of blood, urine, or saliva), and overall confusion on the most accurate or applicable measure of dehydration. Due to the ease of weighing subjects and the relative ease of acquiring urine samples as opposed to blood samples, BW differences and urine measures are the most commonly used and defined quantitative methods for measuring hydration status^{10,33,49}, and will be discussed.

Change in BW can be used to determine net water output and % BW water loss¹⁸. However, this is only accurate in assessing hydration status over a single exercise period and is not an immediate indicator¹⁸. Over long periods of time, BW changes can be attributed to other factors, such as food intake, bowel movement, substrate metabolism, and body composition³⁴. Additionally, relying on pre- and post-exercise changes in BW for hydration status does not allow for changes in hydration strategies during exercise and

can be thrown off by water weight consumed during, or immediately following, the activity⁵⁸. Further, BW water loss does not allow a measure of starting hydration status and only provides a measure of change in body water weight, which can represent different levels of hydration in different people. Regardless, BW water loss as a percentage of starting weight is a commonly used measure of change in hydration status after exercise^{10,18,34,51,59}.

Urine sample measurements for hydration status include specific gravity and osmolality. Urine specific gravity is a measure of the ratio of the density of urine to the density of water³³ and urine osmolality is a measure of the concentration of solutes within the urine²³. A study by Takaki et al. found that urine specific gravity and urine osmolality, measured with a handheld refractometer and osmometer, provide results that are significantly equivalent markers to evaluate hydration status⁶⁰. Kutlu et al. found that there were no significant differences in any of the methods used to monitor urine, which included urine osmolality, specific gravity, and arbitrary units for qualitative color comparisons³⁴. Of the two measures, urine specific gravity has recently gained traction towards being the preferred method of measuring hydration status^{21,22,34,53}.

In terms of an absolute scale, urine specific gravity values are usually between 1.010 and 1.030, with: well hydrated, USG < 1.010; minimal dehydration, USG = 1.010-1.020; significant dehydration, USG = 1.021-1.030; and serious dehydration > 1.030³⁷. Given that urine specific gravity below 1.020 indicates significant dehydration, the NCAA uses urine specific gravity measurements of ≤ 1.020 to identify a state of euhydration prior to wrestling weigh-ins^{10,61}. See Table IV for a table of the percent change in body mass and the specific gravity of urine indicators of hydration status.

Table IV: Indicators of hydration status²⁸.

Condition	% Change in Body Mass	Urine Specific Gravity
Well hydrated	+1 to -1	< 1.010
Minimal dehydration	-1 to -3	1.010 to 1.020
Significant dehydration	-3 to -5	1.021 to 1.030
Serious dehydration	> -5	> 1.030

Despite the common use of urine measurements in hydration studies, there remain significant drawbacks to relying on this method to measure hydration status: it is invasive, it requires time out of exercise to provide a sample, sample acquisition may be impossible in cases of extreme dehydration, additional lab equipment is needed, hygienic acquisition procedures must be followed, and, most importantly, it does not provide real-time feedback of hydration status.

2.5.3 Bioimpedance Devices

The other available non-invasive quantitative method for measuring hydration is with bioimpedance analysis (BIA) devices or through bioelectrical spectroscopy. For this reason, BIA methods for monitoring hydration have been widely investigated to monitor various patient populations^{8,14,16,43,62-68}.

Essentially, BIA devices measure tissue conductivity by using electrodes to acquire processed signals that are used to calculate opposition to the flow of electric current through body tissues⁸. At homeostasis, the conductivity of the tissues is directly proportional to the amount of fluid present. Additionally, variations in the impedance – the frequency-dependent opposition of a conductor to the flow of an administered alternating current – of the tissues cause a phase shift in the applied current. At low frequencies, the impedance of the cell membranes and tissue interfaces is too large for conduction of the current and the current is only conducted through the extracellular fluid. At high frequencies, current flow increases as the cell membranes and tissue interfaces lose their capacitive ability and the intracellular water content can be measured⁶⁸. Thus, by using a combination of low and high frequency currents, BIA is used to determine the extracellular, intracellular, or total body water content.

Hydration studies with BIA devices have several variations in methodology. Different devices, frequency ranges, regression equations, limbs, time frames, target water cavity, and population groups have been investigated^{8,14,16,43,62–68}. Devices used by the investigators of BIA studies include the Bodystat 1500 and dual frequency Bodystat Dualscan devices by Bodystat Ltd^{62,69}, the Hydra 4200 device by Xitron Technologies^{16,63,70}, the BCM Body Composition Monitor by Fresenius Medical Care^{64,71}, the Elektro-Fluid-Graph by AKERN⁴³, the SFB3 analyzer by SEAC⁶⁵, the Bodygram by AKERN⁶⁶, and the Impedimed multifrequency analyzer by Impedimed⁶⁷. See Figure 4 for images of three BIA devices, the Bodystat 1500, XiTRON Hydra 4200, and BCM Body Composition Monitor. The Elektro-Fluid-Graph, SFB3, and Bodygram are no longer available for purchase.

Figure 4: (A) Bodystat 1500 with electrodes⁶⁹. (B) XiTRON Hydra 4200⁷⁰. (C) BCM Body Composition Monitor⁷¹.

The shortcomings of BIA methods include inconclusive results among geriatric populations⁴³, failure to predict changes during short term 2-3% dehydration and rehydration⁶⁵, the need for precise equations dependent on suitable parameters (age-, gender-, and population-specific)¹⁴, inability to be consistently used in patients with water-imbalance diseases (such as liver cirrhosis, renal failure, cardiac disease, and obesity)¹⁴, and variations in reliability and accuracy when monitoring acute changes in hydration status⁴⁰. Importantly, BIA methods can only be used to monitor hydration while the user is inactive, require four electrodes to be adhered to the user, the electrodes/devices have to be calibrated before each measurement,

proper individual-specific regression equations must be used, the electrodes must be placed either on each limb (whole body) or on a single limb (calf), experienced care must be used to attach and operate the electrodes, large wires run from each electrode to the devices, and the devices are large, cumbersome, and cannot be used in patients with amputations, artificial joints, pregnancies, stents, or pacemakers^{8,14,16,43,62–68}. Furthermore, BIA devices are prohibitively expensive; a single unit without the recurring cost of electrodes can run as much as \$1,485⁶².

Two other types of BIA devices are handheld body composition analysis devices and scale body fat monitors. These are more accessible, easier to use, and cheaper options than electrode-based BIA devices. However, due to the more established literature on electrode-based systems, there are few studies on the efficacy of these devices. Regardless, four key limitations exist with the handheld and scale methods: the suggested use of these devices is only when optimally hydrated, each device only measures the composition of the body segment nearest to the device (upper body for handheld, lower body for scale), the devices are only intended for use once a day, and they cannot be used while exercising⁷². Additionally, the most popular handheld model contains documentation excluding use for children of growing stage, elderly people, women after menopause, those having swelling, patients of osteoporosis having very low bone density, people with a fever, bodybuilders or professional athletes, patients undergoing dialysis, and pregnant women⁷² – all of whom could benefit from hydration monitoring. Therefore, handheld body composition device and scale body fat monitors are not suitable for continuous monitoring of hydration status and an alternative method would be beneficial for all target populations.

2.6 State of the Art

There have been recent studies that propose novel devices for monitoring hydration by detecting one of the clinical measures of dehydration. Gunter et al. and Stewart et al. used MEMS-based embedded piezoresistive microcantilever sensors to measure osmolality changes in saliva or urine with 1% accuracy^{7,13}. In these devices, organic or synthetic polymeric materials respond to osmolality changes in a user's saliva by expanding or contracting. See Figure 5 for a diagram of this process. This volumetric change is measured by 300 μm long piezoresistive microcantilevers and was shown to have a non-linear relationship between resistance (ohms) and relative humidity (%)⁷. As of 2007, the researchers were working on larger scale human testing to determine the viability of these sensors as long-term hydration monitoring devices¹³. Regardless of their success in long-term monitoring, there are significant design barriers that must be overcome such as implementation in a device, communication of data, a power source, sensor response to drinking fluids, and clinical studies for different populations, specifically geriatric patients with low salivary response. Furthermore, salivary hydration status sensing is not likely to be continuous.

Figure 5: Operation of an embedded piezoresistive microcantilever device for hydration monitoring⁷.

A MIT-associated company, mc10, Inc., has been investigating small, wearable, and flexible electronic patches to monitor hydration in real-time⁷³⁻⁷⁵. See Figure 6 for an image of an inactive patch on a model's arm. Mc10 claims that the Biopatch has hydration monitoring capabilities, but the company has not released any information on the method they use. See Figure 7 for a concept image of the patch, obtained via a promotional video on mc10's webpage, and a circuitry image from an infographic depicting possible electronic components within the Biopatch. As seen in Figure 7, a Biopatch could contain an LED, photodetectors, a strain gauge, temperature sensors, EEG/EMG sensors, and ECG sensors. The recent article from the Boston Globe revealed that mc10 is developing a commercially available version of the Biostamp for release next year, but specifically stated that the company will not divulge what the Biostamp will monitor⁷⁵.

Figure 6: Image of an inactive flexible electronic patch from mc10⁷⁴.

Further investigation revealed an applied, but **not granted**, patent #WO/2013/033724, ELECTRONICS FOR DETECTION OF A CONDITION OF TISSUE, filed September 4th, 2012⁷⁶. In this patent, mc10 is claiming (in brief summary):

- (1) An apparatus for monitoring a condition of a tissue wherein a measure of the electrical property of the tissue using the apparatus provides an indication of the condition of the tissue⁷⁶.

- (2) The condition of the tissue can be the hydration state, volume of sweat lost, mechanical property of the tissue, or a disease state of the tissue⁷⁶.
- (3) Specifics regarding the components of the apparatus (brace structures, spacer structures, cross-link structures, encapsulated structures, adhesive structures, and encapsulation materials)⁷⁶.
- (4) An apparatus of (1) further containing an ultrasound apparatus, made of an ultrasound generator (piezoelectric crystal) proximate to a first tissue of interest, wherein the ultrasound generator directs ultrasound waves at a portion of the tissue and an ultrasound receiver disposed proximate to a second portion of the tissue of interest provides a measure of ultrasound waves arriving at the second portion of the tissue⁷⁶.
- (5) A method wherein comparing the data to a calibration standard provides an indication of the tissue⁷⁶.
- (6) Further claims on design possibilities, such as curved components, zigzag circuitry, and non-linear components⁷⁶.

Given the information contained in the claims, mc10's electronic patches could use bioimpedance, ultrasonic, capacitive, or any combination of the three measurements to obtain information on hydration status. It is important to note that this patent and the claims within have not been granted; grant of a patent does not guarantee that mc10 will pursue development of hydration monitoring, and no clinical studies from the authors of the patent could be found. While the language within this patent application is concerning in terms of the long-term possibilities of this thesis, consultation with a patent attorney revealed that there is no guarantee that mc10 will be granted all of the claims. The patent attorney also advised that development of an ultrasonic method that does not rely on a known distance between sensors remains a viable patent opportunity. Should mc10 be granted the claims within this patent and no viable patent work-around can be found, there still exists an option of licensing the patent from mc10 to develop a marketable device.

Additionally, a review of mc10's webpage and promotional releases reveals a focus on the Reebok CHECKLIGHT concussion monitor, a different device under development by the 30-person company. The design direction of a patch as opposed to a wearable device is also intriguing; the diminutive size, degradability, lack of hardware components, and ability to wear multiple patches to monitor more metrics is particularly applicable for professional athletes, but may not address a market need for the general population.

Figure 7: Images of: (A) a conceptual hydration monitoring patch by mc10, Inc⁷³ and (B) a diagram of possible electronic positioning within the mc10 patch⁷⁵.

Importantly, a study in 2005 by Sarvazyan et al. proposed a wearable device that uses an ultrasonic method⁴. Sarvazyan, the lead investigator on this study, formed Artann Labs and continued development on this device for two additional studies in 2006 and 2010^{4,9,10}. See Figure 8 for the design progression of the device. Note that despite the different study primary authors, the same investigative group was involved in all three studies. The device was initially designed with a rigid frame, hinge angle sensor, detached portable electronic unit, and for use with the soleus muscle⁴. The rigid frame and hinge angle sensor allow precise detection of the angle between the probes. Then, the angle can be used to determine the distance between the ultrasonic transducer⁴. The theory behind the ultrasonic method used in this study, and further investigated as a part of this thesis, is described in more detail in the upcoming Theory chapter.

Figure 8: Images of the ultrasonic devices used by (A) Sarvazyan et al.⁴, (B) Topchyan et al.⁹, and (C) Utter et al.¹⁰.

A key requirement for the designs by Artann Labs is a *known distance* between the two ultrasonic transducers. This requirement was verified after a patent search revealed that Artann Labs was granted two patents (2006 and 2007) on using an ultrasonic method to monitor hydration status:

- (1) US7033321 B1: Ultrasonic water content monitor and methods for monitoring tissue hydration⁷⁷.
- (2) US7291109 B1: Infant hydration monitor⁷⁸.

The second patent expands on the claims of the first patent, specifically in the design of an infant hydration monitor. In the first patent, Artann Labs is claiming (in brief summary):

- (1) An ultrasonic tissue water content monitor comprising of a first ultrasonic means, second ultrasonic means, and support system to hold the ultrasonic means against tissue at a *known distance* apart. This distance can be determined via a rigid structure or angle sensor.
- (2) A method of evaluating tissue water content using partial or whole propagation of ultrasonic waves through tissue, detection of time-of-flight between a first and second ultrasonic means, calculation of the velocity of ultrasonic waves through tissue, or detection of plurality of velocity measurements through layers of tissue.

The language in the summarized claim (2) is concerning for the long-term possibilities of this thesis, but consultation with a patent attorney revealed that development of an ultrasonic method that does not rely on a known distance between sensors remains a viable patent opportunity. Additionally, a proposed future

direction of this thesis involves a method not discussed in the summarized claim (2). Should no viable patent work-around be found, there still exists an option of licensing the patent from Artann Labs to develop a marketable device.

Refocusing on the current status of Artann Lab's device, the last published results were from the study in 2010 by Utter et al¹⁰. In 2011, the device was used in an observational clinical trial funded in collaboration with the Bill and Melinda Gates Foundation (NCT01482208)⁷⁹. This trial was specifically for neonatology and pediatric children and infants and/or children up to age 3 years old with diarrhea⁷⁹. In this trial, the investigators used ultrasound velocity measurements through muscle tissues to follow patient hydration status in acute diarrhea and during rehydration therapy⁷⁹. The clinical trial was completed in October 2012, but no results have been published⁷⁹. Although no study results were published, the results must have been promising – On August 1st, 2013, Artann Labs was awarded major funding from the National Institute on Aging/National Institutes of Health for development of the hydration monitor specifically for use in the elderly⁸⁰.

The long-term success of Artann Labs in continued development and acquiring funding support for clinical trials is promising for future investigations. Notably, the devices created by Artann Labs do not allow for continuous monitoring, have large form factors, require a separate operator, and can only be used when the subject is sitting down at rest. Despite these design shortcomings, the ultrasonic method proposed, and later investigated, by the three studies and the clinical trial provides a foundation for the device investigated in this thesis. It is also important to note that the state of the art solutions presented by Gunter et al., Stewart et al., mc10, and Artann Labs are not available on the market, nor have the groups disclosed supporting evidence of a timetable to market.

2.7 Applicable Codes and Standards

Many codes and standards govern the design of an electrical device for use with the human body. Chief among these codes are the regulations set forth by the Food and Drug Administration (FDA). The Center

for Devices and Radiologic Health (CDRH) branch of the FDA oversees the approval and sale of medical devices within the USA. The CDRH is in charge of a risk-based classification system for medical devices and can classify devices as low-risk (class I), medium-risk (class II), or high-risk (III)⁸¹. Depending on classification, a process of approval through the FDA can mean only registering as a manufacturer or it can mean millions of dollars for years of required clinical studies⁸¹. The development process of a non-invasive hydration monitor depends heavily on what classification, if any, it would receive from the FDA. Therefore, it is important to evaluate if the FDA would classify a wearable and non-invasive hydration monitor and, if so, what classification it would receive.

The relevant FDA definition of a medical device is an apparatus that is intended for use in the diagnosis of disease or other conditions, or in the cure, mitigation, treatment, or prevention of disease⁸¹.

Additionally, a medical device data system is a device that is intended to transfer, store, convert, or display medical device data, and would be a class I medical device⁸². Given these two definitions, any electronic monitoring system for use with the body would be regulated. However, there are three confounding factors in the classification of a hydration monitor:

- (1) There are no non-prescription hydration monitors currently regulated by the FDA.
- (2) There are different intended use conditions for a hydration monitor.
- (3) There is ongoing confusion on the role of the FDA in handling health-associated apps^{83,84}.

In factors (1) and (2), preliminary research on the FDA's device database revealed no regulated hydration monitors. Expanding the search for fluid status monitors revealed bioelectric impedance devices for use under the direction of a physician. One such device is the ZOE Fluid Status Monitor by Noninvasive Medical Technologies, Inc⁸⁵. This device is a class II device indicated for use in patients with fluid management problems that are taking diuretic medication, living with heart failure, living with end-stage renal disease, recovering from coronary artery disease or suffering from recurrent dehydration⁸⁵. Given that Artann Labs is currently going through a clinical trial process with their non-invasive ultrasonic method – of unknown classification – and that non-invasive bioimpedance monitors like the ZOE Fluid

Status Monitor are regulated class II devices, it is certain that a hydration monitor for use under physician direction would be regulated by the FDA.

However, as mentioned in factor (2), there are multiple use conditions for a hydration monitor.

Considering the population groups that could benefit from a hydration monitor, a carefully defined intended use condition could avoid or minimize FDA regulation by only intending use as a *monitor* of hydration status for healthy populations. Additionally, as mentioned in factor (3), the confusion over the role of the FDA in handling health-associated apps could allow a hydration monitor device and accompanying app on the market with little or no regulation. For example, a healthcare data management app by Samsung is a class II regulated device⁸⁶ but common heart rate detection and fitness apps are available for download without FDA regulation⁸⁴. Regardless of the path to market and the intended use conditions, FDA approval would allow a hydration monitor to be used on the elderly, pediatric, and other general hospital patients. In which case, the codes and standards in CFR 21 would apply. These include requirements for:

- Risk-based classification (21 CFR 860)⁸⁷
- General hospital and personal use devices (21 CFR 880)⁸⁷
- Labeling (21 CFR 801)⁸⁷
- Medical Device Reporting (21 CFR 803)⁸⁷
- Registration (21 CFR 807)⁸⁷
- Quality systems (21 CFR 820)⁸⁷
- Performance standards for electronic products (21 CFR 1010)⁸⁷
- Pre-market approval (21 CFR 814)⁸⁷
- Post-market oversight (21 CFR 822)⁸⁷

Beyond the FDA, ASTM International has electronics standards for specifying, evaluating, and testing the performance requirements of materials and accessories used in electronic devices⁸⁸. These standards include contamination control, metallic materials, computerized systems, data analysis, information capture, testing, and documentation⁸⁸. In addition, there are ISO standards on piezoelectric devices, integrated circuits, and electronic component assemblies⁸⁹. The use of ultrasonic technology has specific codes as set by the NCRP in their Exposure Criteria for Medical Diagnostic Ultrasound standards⁹⁰.

Within this document is information on the safety, criteria, dose models, and standards associated with ultrasound in medical diagnostics⁹⁰. Finally, any device using wireless communication must adhere to the standards and testing of FCC, as contained in CFR 47⁹¹.

3. THEORY: ULTRASONIC AND PIEZOCERAMIC BASICS

Piezoceramic materials are used in ultrasonic transducers to convert an electrical energy stimulus into transmitted mechanical energy in the form of an ultrasonic wave⁹². Conversely, the same piezoceramic transducers can receive a mechanical signal and convert it into a relative electrical signal. The generated ultrasonic wave is affected by the material properties of the tissues through which it propagates; portions of the wave energy are reflected back toward the transmitter, attenuated by the tissue, or propagated through the tissue⁹². These characteristics of piezoceramic transducers have been utilized to diagnose, treat, or navigate using ultrasonic methods⁹². There are many benefits of ultrasonic imaging, such as the non-invasive nature, radiation free imaging, relative inexpensiveness, portability, real-time imaging, and tomographic views⁹³. These benefits have led to ultrasound imaging being one of the most widely used imaging technologies in modern medicine.

3.1 Historical Perspective

The Curie brothers discovered piezoelectric behavior in the 1880's when they noticed electrification of certain crystals under mechanical pressure⁹⁴. A year later, the brothers, using Lippmann's work in thermodynamics, then confirmed the existence of converse piezoelectric effects (deformation/stress under applied electric fields) in 1881⁹⁴. The first application of piezoelectric technology was by Langevin, who developed the first SONAR out of a quartz transmitter and receiver system for underwater sound⁹⁵.

Piezoelectricity remained a laboratory curiosity until the 1950s, when the invention of piezoelectric ceramics led to significant implementation in many industrial fields⁹⁴. This rise of piezoceramics led to a corresponding increase in the portability and technologic capabilities of ultrasounds. Similarly, the most widely used piezoceramic, PZT, was discovered in 1952 by Shirane et al⁹⁴. The complex nature of the high piezoelectric response in PZT remains obscure, and studies suggesting modifications to the material continue to this day^{93,94}.

3.2 Typical Ultrasonic Transduction Mode

There are several methods of ultrasonic transduction. See Figure 9 for a diagram of the general principle guiding ultrasonic transduction: acoustic reflection at tissue interfaces. The transduction method differs based on the desired application, the designed number of transducers, and the desired outcome (image, wave velocity calculation, phase shift, etc.)⁹⁶. In all methods, a sensor emitter sends a pulse of several cycles of a signal – usually sinusoidal – which travels through the medium, bounces off reflector surfaces, and is captured by the sensor receiver⁹⁶. The received signal depends on the experimental setup; in a single (echo/reflection) transducer setup, the same transducer both emits the signal and receives the reflected wave⁹⁶. In a two-transducer setup (transmission), a receiving transducer, placed on the opposite side of the tested area from the sending transducer, receives the remnants of the original wave after it fully propagates through the limb⁹⁶. See Figure 10 for an example transducer reading of pulse-echo transduction.

Figure 9: Diagram of the principle behind acoustic reflection at tissue interfaces⁹⁷.

Figure 10: An example transducer reading from pulse-echo transduction. Green = sent signal, red = received signal⁹⁶.

3.3 Tissue Material Factors That Affect Ultrasonic Transduction

The overall outcome of an ultrasonic technique is dependent on the material through which the ultrasound wave is propagating. Specifically, the ultrasound propagation is affected by the elasticity, density, acoustic velocity, acoustic impedance, and attenuation of the targeted tissues⁹⁷. As the wave propagates through the tested area, the material properties of the specific tissue types (skin, bone, muscle, etc.) determine the extent to which the wave is absorbed, reflected, transmitted, or diverted⁹⁷. The elasticity, or the ability of the tissue to return to its original shape after a load, and the density, or mass per volume, of the probed tissue determine the extent to which the molecules of the tissue deform under the wave⁹⁷. Similarly, the compressibility, or the fractional decrease in volume as pressure is applied, affects the speed of the wave propagation⁹⁷. The acoustic velocity of a tissue is the combined affect of density and compressibility and is expressed by the relationship in **Equation 1**⁹⁷. The acoustic impedance, a measurement of a tissue's resistance to the wave speed, is a relationship between the tissue density and acoustic velocity. See **Equation 2**⁹⁷.

$$\text{EQ. 1} \quad c = \frac{1}{\sqrt{K\rho}}$$

Where:

c = acoustic velocity
 K = compressibility
 ρ = density

$$\text{EQ. 2} \quad Z = \rho c$$

Where:

Z = acoustic impedance
 c = acoustic velocity
 ρ = density

Essentially, higher density materials have higher impedances and thus pose larger obstacles for the propagation of the ultrasound. However, the most important material property for the generation of an ultrasound image is not a property of the tissues themselves, rather, it is of the impedance mismatching between two adjacent tissues⁹⁷. Similar to the reflection and refraction of light, an ultrasonic wave propagating through the interface of two tissues with the same acoustic impedance will not be reflected⁹⁸. Conversely, propagation through an interface of two materials with different acoustic impedances results in the reflection of the wave, thus allowing visualization of the tissues via the ultrasonic transducers⁹⁸. See Figure 11 for a basic diagram of this principle, called Snell's law.

Figure 11: Diagram of Snell's law for the propagation of an incident wave, reflected wave, and transmitted wave between two mediums of different acoustic impedances⁹⁸.

$$\text{EQ. 3} \quad RF = \frac{(Z_2 - Z_1)}{(Z_2 + Z_1)}$$

Where:

RF = Reflection Factor
 Z_1 = impedance of tissue 1
 Z_2 = impedance of tissue 2

Figure 12: Amplitude reflection factor coefficients for various tissues relative to blood⁹².

In a way, acoustic impedance mismatch behaves like sonar – it takes a difference in impedances to bounce a portion of the ultrasound wave back towards the sensor for visualization. Another method of describing the acoustic impedance mismatch between two tissues is by the amplitude reflection factor coefficients, as shown in **Equation 3**⁹². Visual representation of how ultrasound reflections can be used to identify tissue is seen in Figure 12, where the amplitude reflection factors (presented relative to blood) of different tissues are shown. Conceptually, a larger difference between the bar lengths of two tissues would result in more reflection when an ultrasound wave travels from one tissue to the other⁹².

Finally, the extent to which the specific material absorbs or propagates the ultrasonic wave is a material property known as attenuation⁹⁷. Attenuation is caused by three main factors: absorption, reflection, and scattering⁹⁷. Materials of high attenuation more strongly impede the depth that the ultrasound can propagate⁹⁷. See Table V for a table detailing the attenuation coefficients for selected body tissues at 1

MHz. As seen the in the table, ultrasonic waves are not significantly attenuated by water, aqueous humor, vitreous humor, or blood, but are highly attenuated by lung (air) and bone.

Specific information on how changes in the tissue water content of muscle can be detected by ultrasonic methods appears in the Using Ultrasonic Methods to Monitor Hydration section.

Table II: Attenuation of various human tissues at 1 MHz⁹⁷.

Material	dB/cm
Blood	0.18
Fat	0.6
Kidney	1.0
Muscle (across fibers)	3.3
Muscle (along fibers)	1.2
Brain	0.85
Liver	0.9
Lung	40.0
Skull	20.0
Lens	2.0
Aqueous humor	0.022
Vitreous humor	0.13
Water	0.0022
Castor oil	0.95
Lucite	2.0

3.4 Wave Factors That Affect Ultrasonic Transduction

Ultrasonic transduction applications can be categorized in two ways: imaging and non-imaging⁹³. Imaging applications rely on the real-time display or collection of information to diagnose, navigate, or treat the targeted area⁹³. Non-imaging applications rely on the comparison of collected information over time to track tissue status and diagnose ailments⁹³. For example, fetal ultrasonography is an imaging application and the use of ultrasonic probes to distinguish between calcified and healthy regions of arteries is a non-imaging application⁹⁹. Due to the difference in the type of outcome between the categories, imaging and non-imaging applications rely more heavily on different design variables⁹³. For instance, imaging applications are more focused on the resolution of the generated image while non-imaging applications are more focused on the characteristics of the wave propagation through the target tissues⁹³.

An ultrasonic wave is defined by its frequency (Hz), wavelength (mm), and amplitude (dB)⁹³. Ultrasonic wave frequencies range from 20 kHz, the upper limit of human hearing, to 20 MHz for intensive medical applications⁹⁸. Given that the wavelength and frequency of ultrasonic waves are inversely related, high frequency waves (with short wavelengths) are used to generate high-resolution images⁹³. However, high frequency ultrasonic waves are more attenuated by the target tissue than low frequency waves, thus limiting the penetration of the wave and reducing the effective imaging to more superficial structures⁹³. See Figure 13 for a reference chart of how resolution and penetration change with changes in ultrasonic transducer frequency. For example, high frequency transducers (10-15 MHz) are used to image deep stellate ganglion blocks and low frequency transducers (2-5 MHz) are used to image the more superficial lumbar neuraxial structures⁹³. Thus, the selection of an ultrasonic transducer is a significant design decision that differs based on the intended application⁹³. See Table VI for commonly used frequency, wavelength, attenuation coefficients, and depth values in diagnostic ultrasounds.

Table III: A table of common values for diagnostic ultrasound quantities⁹⁶.

Frequency (MHz)	Wavelength (mm)	Attenuation Coefficient (dB/cm)	Imaging Depth (cm)
2.0	0.77	1.0	30
3.5	0.44	1.8	17
5.0	0.31	2.5	12
7.5	0.21	3.8	8
10.0	0.15	5.0	6

Figure 13: A comparison chart of the resolution and penetration of ultrasonic waves at different transducer frequencies⁹³.

Alternatively, in non-imaging applications the main investigated outcome is a calculation of changes in the ultrasonic pulse velocity. In these cases, the velocity of the transduced ultrasound wave is dependent on the operating frequency of the transducer and the attenuation of the surrounding tissues⁹⁶. See the upcoming Using Ultrasonic Methods to Monitor Hydration section for more information on the use of non-imaging ultrasonic waves.

3.5 Piezoceramic Materials and Their Implementation in Ultrasonic Transducers

The most common piezoceramic used in ultrasonic transducers is a lead-based solid solution: lead zirconate titanate ($\text{Pb}(\text{Zr}, \text{Ti})\text{O}_3$) or PZT⁹⁴. PZT has a perovskite crystal structure, as seen in Figure 14⁹⁴. It can be tailored for specific design constraints via doping to alter the piezoelectric response of the material⁹⁴. The specific piezoelectric response is induced in the material by heating it within a large ($>2000 \text{ V/mm}$) electric field to distort the structure and induce permanent polarization⁹⁴. Applied fields or stresses will induce shape changes or voltage rises, respectively⁹⁴.

Figure 14: Figures of (A) PZT perovskite crystal structure and (B) under a phase transformation along {001}⁹⁴.

The coupling coefficient, k , is the extent to which the PZT material is piezoelectric⁹⁴. In other words, the coupling coefficient is an indication of how efficiently PZT can convert electrical energy to mechanical energy and vice versa. See **Equation 4** for the coupling coefficient of a PZT poled ceramic⁹⁴.

$$\text{EQ. 4} \quad k^2 = \frac{Em^2}{\epsilon_0 \epsilon_r}$$

Where:

- k = coupling coefficient
- E = Young's modulus
- m = piezoelectric motor coefficient
- ϵ_0 = permittivity of free space
- ϵ_r = relative permittivity of the material

The efficiency of PZT (0.7 compared to 0.1 of a quartz crystal) directly explains why PZT is so widely used in ultrasonic applications where power consumption, inexpensiveness, and portability are key design features⁹⁴.

The most common implementation of PZT in ultrasonic methods is via a single-element transducer⁹⁴.

There are four main components of a single-element transducer: a matching layer/lens, electrodes, piezoelectric element, and backing⁹⁴. See Figure 15 for a diagram of a single-element transducer. Since the piezoelectric element has a high acoustic impedance compared to biological tissues, the additional polymer-based layers are added to the active layer to increase the axial resolution of the transducer⁹⁴. The backing material is used to dampen the transducer resonance and increase resolution⁹⁴. This material is selected based on the application; the closer the acoustic impedance is to that of the piezoelectric element, the more dampening the backing applies to the transducer⁹⁴. Lastly, the matching layer is used to increase the sensitivity of the transducer and the lens is used to focus the ultrasonic beam at a desired focusing distance⁹⁴. This distance can be manipulated by changing the radius of curvature of the lens⁹⁴. See Figure 16 for a diagram of the radiation pattern of single-element transducers with a focusing lens.

Figure 15: Diagram of a single-element ultrasonic transducer⁹⁴.

Figure 16: Radiation pattern of a single-element transducer with a focusing lens⁹⁴.

3.6 Medical Applications of Ultrasonic Methods

The most common application of piezoceramic ultrasonic transducers is for ultrasonography⁹⁴.

Ultrasonography is the use of an array of ultrasonic transducers in a delay line setup to image the body in

real-time⁹⁴. In addition, there are other lesser-known medical applications of ultrasonic methods like healing bone analysis, speeding fresh fracture repair, and analysis of material structures.

In healing bone analysis, the clinical assessment of fracture healing process is a subjective assessment that greatly depends on the physician experience, deficient radiographic images, and limited manual manipulation of the fracture site. These limitations can lead to incorrect medical treatment and delayed responses to improper fracture healing. Ultrasonic methods are used as a non-invasive and quantitative method in the testing of healing bone by detecting the changes in attenuation over time at the site of the fracture as four stages of bone fracture healing occur^{23,99}. Since the materials at the fracture interface are constantly changing, the attenuation at the site of the fracture differs a significant amount based on the stage of fracture healing. Thus, ultrasonic probing of the area over time can give an indication of how the repair is progressing. Specifically, both echo/reflection and transmission testing methods can be used to determine the thickness of a bone callus layer and estimate the bone healing level by analyzing variations in the sent vs. received ultrasonic waves⁹⁹.

In ultrasonic methods of speeding fresh fracture repair, pulsed low-power ultrasound waves can be used daily to speed the repair of fresh fractures. For example, one study found that daily ultrasound therapy reduced the healing time of broken arm and shin bones by 35-45%²³. The more rapid healing rate is caused by several mechanisms, mainly by the ultrasonic waves stimulating chondrocytes to more rapidly form a fibrocartilage callus¹⁰⁰.

As an example of the use of ultrasonic techniques to diagnose ailments, IntraVascular Ultrasound Imaging (IVUS Imaging) is an imaging method that uses an ultrasonic probe mounted on the distal end of a catheter to image the endothelium of blood vessels. It is commonly used to assess the amount of atherosclerotic plaque in the arteries of the heart. An IVUS image is composed by rotating the ultrasonic emitter inside the vessel while the piezoelectric transducer emits ultrasonic waves and receives echoed signals. Due to the proximity of the probe to imaged area, high frequency ultrasonic waves can be used to

generate high-resolution images. The result is a 360° tomographic view of the vessel endothelium⁹⁹. See Figure 17 for an example IVUS image.

Figure 17: An example of an IVUS image from an atherosclerotic artery⁹⁹.

As outlined in discussion of the tissue material factors and wave factors introduction sections, there are many factors to consider when selecting and using piezoceramic ultrasonic transducers. Unfortunately, not all challenges with using ultrasonic methods can be mitigated by proper design selections. The two largest challenges, speckle and propagation limitations, are discussed below.

Ultrasound images, like many other visualization techniques, are corrupted by the presence of unintended noise. This noise, appearing as a granular pattern called speckle, is caused by reflectors within the tissue¹⁰¹. Like static on a television screen, speckle reduces the overall resolution of the received image and can be problematic for proper diagnoses that depend on ultrasound clarity. However, some studies propose that speckle has information about tissue acoustic properties that can be used for diagnostic purposes⁹⁴. The hypotheses around this method of speckle interpretation rely on speckle being indicative of pathological differences, such as an increase in stiffness or surface roughness, in portions of the same tissue type. For example, the density and pattern of speckle could be used to characterize the echomorphology of atherosclerotic plaque within blood vessels⁹⁹.

The therapeutic and imaging capabilities of ultrasonic methods depend largely on the ability of the ultrasound to propagate through the target tissue. However, materials with high attenuation coefficients hinder ultrasonic waves and limit the applications in which ultrasonic techniques can be used. For example, ultrasonography is of little value for looking at air-filled structures, like the lungs, or those

Figure 18: Ultrasound velocity as a function of water content in animal muscle tissue for (A) different animal types and (B) across or along fibers⁴.

surrounded by bone, such as the brain and spinal cord²³.

3.7 Using Ultrasonic Methods to Monitor Hydration

Studies have shown that muscle tissue properties at hypohydrated, euhydrated, and hyperhydrated levels vary a detectable amount^{4,9,10}. Given that most soft tissues contain about 70-80% water, with the remaining 20-30% consisting of major molecular components, a shift of the molecular concentration causes a change in the velocity due to the contributions of each molecular component on the velocity⁴. Additionally, as described earlier, tissues with lower water levels have higher attenuation coefficients. Assuming other tissue variables remain constant, the change in wave velocity is a direct indication of a change in tissue hydration status^{4,9,10}. This method has been used by the Artann Labs group to measure muscle dehydration with 99% accuracy⁴. Sarvazyan et al.'s 2005 study found an approximately linear relationship between ultrasound velocity and tissue water content: 3.0 ± 0.2 m/s per 1%, which held

constant for up to 10% water loss⁴. See Figure 18 for graphs showing the experimental results for ultrasound velocity of bovine, porcine, and chicken tissues with changing water content.

Additionally, the study found that the molecular composition determines the velocity of ultrasound, while structural and intracellular interactions have less influence⁴. The effect of structural components was described to be only 1.5% of the change in the ultrasound velocity with the decrease in tissue water content⁴. However, while the results of the study were promising, there are confounding factors in adapting this laboratory method for field use. These factors include individual differences in overall muscle structure, short-term changes during physiological activity of muscle, individual variations in the anatomy of the tested subjects, temperature changes in the tested muscle, variable blood flow, and variation in concentration of molecular components of muscle other than water⁴. Specifically, the investigators reported that ultrasound velocity increases up to 3 m/s with contraction due to the increase in blood flow to the tissue and that temperature can have a significant effect on the accuracy of the method⁴. Regardless, the group's 2010 study found that changes in ultrasonic velocity reflect the changes of urine specific gravity, urine osmolality, and body mass during acute dehydration and rehydration¹⁰.

The research of Artann Labs focuses on the change in velocity of an ultrasonic wave through muscle tissue over time. To monitor this change, the investigators require a distance value for each measurement. However, this places undesirable constraints on a device since the transducers must either be kept at a known distance or there must be an accessory structure that reports the distance between the transducers. Additionally, the confounding factors listed by the investigators pose challenges for integration in a continuous real-time device. Thus, it is desirable to find an alternative process for using ultrasonic transduction in the detection of changes in tissue status.

One alternative process is using the phase shift from a sent signal to a received signal. Considering that ultrasonic vibrations travel in the form of a wave, a simplified model of a sent signal can be seen in

Figure 19. This signal is an analog sine wave defined by the wavelength (λ), frequency (f), and period (T) of a complete cycle¹⁰². See **Equation 5** for the relationship between frequency and period; **Equation 6** for the relationship between wavelength, frequency, and acoustic velocity; **Equation 7** for the relationship between wavelength, period, and acoustic velocity; and **Equation 8** for the relationship between material thickness, acoustic velocity, and time of flight¹⁰².

Figure 19: Simplified model of a sent ultrasonic signal in the form of a sine wave¹⁰².

EQ. 5 $f = \frac{1}{T}$

Where:

f = frequency (Hz)
 T = period

EQ. 6 $\lambda = \frac{c}{f}$

Where:

λ = wavelength
 c = acoustic velocity
 f = frequency (Hz)

EQ. 7 $\lambda = cT$

Where:

λ = wavelength
 c = acoustic velocity
 T = period

EQ. 8 $\delta = \frac{ct}{2}$

Where:

δ = material thickness
 c = acoustic velocity
 t = time of flight

Shown in **Equations 5-7** is a direct relationship between the acoustic velocity of a medium and the definition of the wave. **Equation 8** shows the method used by Artann Labs, where a known material thickness and sensed time of flight are used to calculate changes in the acoustic velocity. Recalling that

changes in tissue hydration affect the acoustic velocity of the medium, a change in hydration will result in a detectable phase shift between a sent signal and a received signal, as seen in Figure 20¹⁰³.

Figure 20: Diagram of a phase shift¹⁰³.

In theory, individualized measurements are possible by performing a calibration step that establishes a baseline phase shift at a euhydrated state. Over time, deviations from this baseline shift indicate hypohydration, maintenance of euhydration, or hyperhydration. Accurate implementation of this method will require future investigation of the effect of other physiological variables on the phase shift of the received signal. The sensitivity of the sensors, frequency of measurements, and predictable effect on the phase shift will allow for a device to overcome these effects or even deduct other metrics from the phase shift of the received signal. However, such investigations are beyond the scope of this thesis and are discussed in the Future Functionality section of the Conclusion chapter.

4. OBJECTIVES AND SPECIFICATIONS

The design of a wearable, continuous, and non-invasive device to monitor hydration status can be directed towards two main use conditions: use by a medical professional in a hospital setting or use by an end-user in daily-life situations. These two conditions have widely different design requirements associated with them, such as the level of expertise of the user, restrictions on the size of the device, restrictions on the weight of the device, the likelihood of misuse, the necessity for ergonomic design, the necessity of ease of use, and the overall conditions in which the device would be used. Therefore, it was necessary to address the design of the device toward one specific use condition.

4.1 Market Research

Market research was performed to determine which use condition to address. First, the market size of the populations of interest was characterized^{104–110}. See Figure 21 for a chart of top five populations. As seen in Figure 21, elderly and pediatric populations have the largest market size in the US. However, the market size for athletes is likely underrepresented since only high school and college athletes were included in official census records.

Figure 21: Chart of USA market size for populations of interest.

Considering the large market size of athletes and the information presented in the Codes and Standards section, an athletic market was chosen for several reasons:

- It likely would not need or have minimal FDA approval, resulting in a significantly shorter and less expensive path to market.
- Coding and reimbursement processes would not need to be pursued.
- The athletic market is enthusiastic about purchasing fitness/wearable devices¹¹¹.
- Future integration with other fitness/wearable devices is an exciting possibility.

The commercial potential of the smart wearables market for athletes is large and rapidly expanding. A 2013 IHS report on wearable technologies stated that the world market for wearable technology reached \$8.5 billion in revenues in 2012, shipping 96 million devices in that year¹¹¹. By 2018, unit shipments are expected to reach 210 million, resulting in \$30 billion in revenue¹¹¹. Separately, the top 20 sports and wellness apps had 230 million installs¹¹¹. Overall, there is a significantly growing market for fitness devices, and a hydration sensing technology can meet an available market need.

Additionally, it was necessary to consider a target price for the device before entering the design process. Given that dedicated ultrasonic transducers can cost over \$100, setting a target price significantly guided design decisions by helping to optimize the trade-off between features and cost effectiveness. Commonly available fitness devices range in price from \$50 to over \$450, as seen in Figure 22. Feedback received from surveys of athletes and coaches (cross-fit, ultimate, triathlons, marathoners, college, and a former professional football player), consideration of the components necessary for the device, and reviews of hydration-associated products on Kickstarter led to a target price of under \$100. See Appendix A: Survey Responses and Appendix B: Kickstarter Review Table for more information on the surveys and Kickstarter products.

Figure 22: Chart of target price compared to other available fitness devices.

4.2 Customer Requirements

The following customer requirements were compiled with the intention of addressing the average athlete.

- Alerts to a change in hydration status
- Allows users to track hydration over time
- Easy to use
- Fashionable/sleek appearance
- Lightweight
- Long battery life
- Non-obtrusive design
- Provides accurate feedback on hydration status
- Provides continuous feedback on hydration status
- Provides quick indication of hydration status
- Wearable in the rain/with sweat

4.3 Engineering Requirements

Each customer requirement is addressed by the following engineering requirements.

- Contains sensory feedback that is activated by changes in hydration status
- Can store information of at least three days of activity, with transfer of information to a computer or smartphone
- Can perform desired functions within three seconds
- 80% of surveyed users like the overall aesthetics of the design
- Maximum weight of 0.5 lb
- Minimum battery life for three days of average use
- Maximum thickness of 0.5"
- Detects hydration changes of 1% BW

- Acquires data once per minute
- Contains sensory feedback without manual interaction with the device
- Does not malfunction in the presence of sweat and is splash proof

See Table VII for a summary of the customer requirements, engineering requirements, and a brief introduction to how the success of the proposed solution will be assessed for each requirement. Note that the Final Prototype Testing Results section provides an in-depth description of each analysis.

Table VII: Summary table of customer requirements, engineering requirements, and the corresponding analyses to measure the relative success of the solution.

Customer Requirement	Engineering Requirement	Applicable Analysis
Alerts to a change in hydration status	Contains sensory feedback that is activated by changes in hydration status	Device contains visual, auditory, and/or vibration feedback when hydration status changes
Allows users to track hydration over time	Can store information of at least three days of activity, with transfer of information to a computer or smartphone	Device on-board memory is sufficient for at least three day's worth of data Device contains data transfer process
Easy to use	Can perform desired functions within three seconds	User functions on device can be performed within three seconds
Fashionable/sleek appearance	80% of surveyed users like the overall aesthetics of the design	Survey intended end-users for satisfaction with device aesthetics
Lightweight	Maximum weight of 0.5 lb	Weigh final device
Long battery life	Minimum three days of battery life in average use conditions	Device has enough battery life for three days of calculated power draw
Non-obtrusive design	Maximum thickness of 0.5"	Measure device thickness
Provides accurate feedback on hydration status	Detects hydration changes of 1% BW	Validate accuracy of device
Provides continuous feedback on hydration status	Acquires data once per minute	Verify that device is acquiring new values each minute
Provides quick indication of hydration status	Contains sensory feedback without manual interaction with the device	Device includes visual indication of hydration status
Wearable in the rain/with sweat	Does not malfunction in the presence of sweat and is splash proof	Device is designed to respond to the presence of sweat Electronics are housed in sealed containers

Rationale for each engineering requirement is found in Table VIII.

Table VIII: Explanation of rationale for each engineering requirement specification.

Engineering Requirement	Rationale
Contains sensory feedback that is activated by changes in hydration status	Including sensory feedback will increase user interaction with the device and more easily allow users to adjust hydration strategies.
Can store data from at least three days of activity, with transfer of information to a computer or smartphone	Given that marathon runners and triathletes are key targets for hydration monitoring within the athletic market and that some events take place over multiple days at remote locales, including on-board tracking of at least three days allows for weekend excursions. Transfer of information to a computer or smartphone will allow for long-term collection of data, visualization of trends, and more processing-heavy calculations.
Can perform desired actions within three seconds	Minimizing the amount of time required to perform desired actions will increase user satisfaction with the device. Three seconds was selected so athletes can perform actions without having to stop activity.
80% of surveyed users like the overall aesthetics of the design	Designing a device that appeals to the most users possible will increase the likelihood of market success and user satisfaction.
Maximum weight of 0.5 lb	Athletes commonly run or train with smartphones in bicep bands, a habit verified by over 15,000 phone armbands available on Amazon ¹¹² . Given that the average smartphone is 0.3 lb ¹¹³ , a maximum of 0.5 lb was specified to stay within an established user comfort zone.
Minimum three days of battery life in average use conditions	On-board battery life of at least three days will allow for continuous tracking during weekend excursions. At the same time, maximization of battery life will limit the number of interruptions when users have to stop monitoring to charge the device.
Maximum thickness of 0.5"	A maximum thickness of 0.5" will decrease the device bulkiness while allowing room for the internal electrical and processing components of the device.
Detects hydration changes of 1% BW	Given that changes of 2% BW result in discomfort, a drop in blood volume, increase in heart rate, and may begin to significantly affect athletic performance and other common bodily functions ²³ , detection within 1% BW will increase the efficacy of the device.
Acquires hydration status data once per minute	Although ingested water requires five minutes to be present in plasma and red blood cells ³ , acquiring hydration status data once per minute will provide more data for future investigation into the detection of trends in hydration status and integration with other sensors (such as accelerometers, heart rate monitors, sweat rate monitors, etc.).
Contains sensory feedback without manual interaction with the device	Access to hydration status at a glance will reduce complexity in using the device and allow users to get instant feedback on hydration status.
Does not malfunction in the presence of sweat and is splash proof	Given that long distance runners can sweat a significant amount more than sedentary people ¹¹⁴ , the device must be able to provide accurate data in the presence of sweat. Additionally, a splash proof design will decrease the likelihood of malfunction in sweaty or rainy conditions and provide a key selling feature.

5. DESIGN DEVELOPMENT

5.1 Approach and Design Progression

The step-by-step approach for the development of the hydration monitor was as follows:

1. Identification of the problem:
 - i. Researched existing methods of monitoring hydration.
 - ii. Researched existing devices on the market.
 - iii. Performed market research to validate a market need.
2. Identification of a solution:
 - i. Identified problems with the existing monitoring methods.
 - ii. Reviewed literature to identify a possible solution.
 - iii. Investigated validity of the proposed solution method.
3. Design Process:
 - i. Brainstormed and selected the top three possible body locations.
 - ii. Brainstormed and created conceptual sketches of designs.
 - iii. Identified the top design choice.
 - iv. Identified components required for the top design.
 - v. Obtained components and materials for preliminary tests.
4. Sensor Selection:
 - i. Researched possible sensor options.
 - ii. Optimized sensor selection for design needs.
 - iii. Tested and modified sensors to identify the best option.
5. Method Validation:
 - i. Created testing code and method.
 - ii. Evaluated sensor selection.

- iii. Tested for validation.
6. Design Review:
- i. Identified areas of future improvement.
 - ii. Created CAD models of the final prototype and future design direction.
 - iii. Prepared all documentation, testing processes, and prototypes for review.
 - iv. Created the final thesis report.

5.2 Design Selection

After identifying the athletic market as the target customer for the device, the next step was identifying device location options. Based on athletic form factors and common locations of fitness trackers, nine anatomic locations were brainstormed. See Table IX for a list of the locations, sketches of a device at each location, the positive and negative features of each location, and identification of the top three location options. The top three options were selected through qualitative weighting of the positive and negative features of each location, comparison with the customer and engineering requirements, and with consideration to the skill, cost, and time constraints of the project.

Table IX: Possible anatomic locations, representative sketches at each location, positive and negative features of each location, and identification of top three location options.

Location	Positive Features	Negative Features	Top III
Ankle Band	<ul style="list-style-type: none"> + Unique location + Possible future integration with shoes, socks 	<ul style="list-style-type: none"> - Different shoe lengths may interfere with device - Sock interference - High bone impedance - Inaccessible at a glance - Pain with ankle bone impact - May be confused with a convict ankle monitor 	X
Bicep Band	<ul style="list-style-type: none"> + Familiar location + Ample size for a device + Large muscle mass with small bone impedance + Possible future integration with clothes, football jersey sleeves + Easy glance at status 	<ul style="list-style-type: none"> - Tight feeling when flexed - May interfere with sleeves 	✓
Calf Band	<ul style="list-style-type: none"> + Ample size for a device + Large muscle mass with small bone impedance + Possible future integration with shin guards, high socks 	<ul style="list-style-type: none"> - Inaccessible at a glance - Unfamiliar location - May interfere with capri style pants 	✓
Chest Band	<ul style="list-style-type: none"> + Familiar location + Possible future integration with heart rate monitors, clothing 	<ul style="list-style-type: none"> - Inaccessible at a glance - May interfere with chest-based heart rate monitors - High lung impedance - Large band size required - Uncomfortable long-term wear 	X
Finger Ring	<ul style="list-style-type: none"> + Unique location + Easy glance at status 	<ul style="list-style-type: none"> - Smallest size constraints - Not allowed in athletic competitions - Interferes with grip-associated activities - More likely to get snagged - High bone impedance - Finger sizing issues 	X
Forearm Band	<ul style="list-style-type: none"> + Easy glance at status + Ample size for a device 	<ul style="list-style-type: none"> - Unfamiliar location - Tendency to slide down arm - Interferes with sleeves - High bone impedance 	X
Phone Attachment	<ul style="list-style-type: none"> + Reduced processing, memory, and battery constraints on device + Easier integration with 	<ul style="list-style-type: none"> - Wired connection - Complicated development process - Cannot function without 	X

social networks, other fitness tracking apps	<ul style="list-style-type: none"> - Requires a different accessory band for each phone 	
Upper Leg	<ul style="list-style-type: none"> + Ample size for a device 	<ul style="list-style-type: none"> - Unfamiliar location - Large muscle mass and femur bone likely exceed measurement capability - Inaccessible at a glance - Likely to be uncomfortable when running 	X
Wristband	<ul style="list-style-type: none"> + Familiar location + Easy glance at status + Possible future integration with existing Smartwatch and fitness tracking devices 	<ul style="list-style-type: none"> - Smaller size constraint - Crowded marketplace - May not have enough muscle for accurate measurement - May interfere with smart watch devices 	✓

After preliminary qualitative comparisons with the customer requirements, the ankle, chest, finger, phone attachment, and upper leg locations were immediately eliminated due to their negative aspects significantly outweighing the positive aspects. The remaining locations were further assessed through visual analysis of possible signal transduction routes through the limb. Recalling that muscle tissue is the intended measurand of the ultrasonic signal, visual analysis focused on a linear path of least impedance through muscle tissue. See Table X for the resulting sensor location options.

Table X: Transverse views of selected sensor location options¹¹⁵.

As seen in Table IX and with respect to the transverse views shown in Table X, the top three location options were determined to be the bicep, the calf, and the wrist. One key difference stands out between the calf and the bicep or wrist options: the calf location is not easily accessible. Given that three of the eleven customer requirements (easy to use, non-obtrusive design, and quick indication of hydration status) govern ease of access and use, the calf location was eliminated as a design option. Thus, the remaining location options were the bicep and the wrist. As seen in Table X, the wrist signal path does not cross a large muscle. However, the possibility of future integration with existing smartwatches and fitness devices was chosen to take precedence over the efficacy unknowns associated with a lack of muscle tissue. Future investigation into the ability to detect signal changes in other tissues could reveal a novel method of detecting hydration status through characteristic changes in blood plasma or tendon tissue. To this end, the superficial front wrist contains a high-density region of tendons as well as superficial veins and deep arteries¹¹⁵, as seen in Figure 23.

Figure 23: Anatomic diagrams showing (A) tendons, (B) superficial veins, and (C) deep arteries of the front wrist¹¹⁵. Blue boxes and arrows indicate regions of interest.

Considering that existing literature on ultrasonic methods for hydration monitoring relies on differences in muscle tissue^{4,9,10}, it was determined that validation of the device method will focus on implementation at a muscular location. Therefore, the bicep location was chosen for the device design. Future design and testing could be performed to probe the validity of the wrist location. See the Future Wrist Location section for more information on adopting a bicep band for a wrist location.

5.3 Design Concept

The selected design was created with an athletic form factor. See Figure 24 for an annotated sketch of the design concept, Figure 25 for a Solidworks model of the prototype band, and Figure 26 for a Solidworks model of a future, more refined, flexible rubber band with embedded electronics.

Figure 23: An annotated sketch of the selected design concept.

Figure 25: An annotated Solidworks model of the prototype band.

Figure 26: An annotated Solidworks model of a future, refined, prototype band.

6. COMPONENTS

Summary Table XI has a list of the components used in the bicep band prototype design. Images, descriptions, and supporting decisions for each component can be found in the subsections that follow Table XI.

Table XI: Summary table of the components for the bicep band prototype design.

Item	Purpose
Piezoceramic Disc Elements	Ultrasonic transducers
Plasti Dip Rubber Coating	Waterproofing and increased durability for the transducers
Teensy 3.1 Microcontroller	Processor and microcontroller for the device
Blue Tactile LED Button	On/Off button
Multi-color LEDs	Status indication lights
Vibration Motor	Vibration feedback
Polymer Lithium Ion Battery	Battery power
Bluetooth LE Module	Wireless communication
ABS Plastic Case	Housing for electronics
Elastic Nylon Hoop and Loop Cinch	Adjustable strap to hold device on the bicep

6.1 18 kHz Piezoceramic Disc Elements

As opposed to dedicated single-element transducers, a piezoceramic disc with a natural frequency near the ultrasonic range can effectively be used as an ultrasonic transducer. Using a disc element instead of a transducer significantly reduces the cost, design complexity, and size of the transducer; however, there is a corresponding drop in signal quality and signal manipulability due to the lack of the polymer backing layers, matching layer, and lens. Three main factors contributed to the decision to use a disc element:

- Purchasing an ultrasonic transducer is cost-prohibitive and requires a custom order from a manufacturer.
- The desired transducer materials, dimensions, and signal properties are unknown at this early prototyping stage.
- The small size and power requirements of a disc element make it appealing for use in a wearable device.

There are varieties of natural frequency piezos with different surface areas. However, the most important technical feature of a piezo disc element is that it has optimal performance at or near its natural frequency. Therefore, a disc with an 18 kHz natural frequency was chosen because it was the highest natural frequency piezo element available for purchase without a custom order¹¹⁶. See the Validation chapter for the outcome of the piezo element tests and Figure C1 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for a drawing of the 18 kHz disc element.

6.2 Plasti Dip Rubber Coating

Plasti Dip rubber coating was used to coat the transducers for improved water resistance, shock resistance, and durability. It was chosen for the ease of application, availability, relatively quick setting time, different application methods, and flexibility¹¹⁷. Tests were performed to determine the effect of a coating on the strength of the signal and select the optimal number of coatings. See the Validation chapter for the tests and Appendix D: Plasti Dip MSDS Sheet for the material safety and data sheet.

6.3 Teensy 3.1 Microcontroller USB Development Board

The Teensy 3.1 is an Arduino compatible microcontroller board that is programmable via USB cable¹¹⁸. The Teensy 3.1 was chosen primarily for its compatibility with Arduino, small size, fast CPU, breadboard compatibility, availability, concurrent measuring of two analog signals, and 3.3V analog pins. Given the infeasibility of custom manufacturing a board at an early prototype stage, using the Teensy 3.1 with the Teensyduino add-on for the open-source Arduino IDE provided access to online Arduino libraries while staying within the cost, time, and technical knowledge limits of this thesis. See Table XII for the technical specifications of the Teensy 3.1 and Figure C2 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for a pinout diagram.

Table XII: Technical specifications of the PJRC Teensy 3.1 microcontroller USB development board¹¹⁸.

Technical Specification		Value
Price		\$19.80
Dimensions		1.4 x 0.7"
Processor: MK20DX256VLH7	Core	Cortex-M4
	Rated Speed	72 MHz
	Overclockable	96 MHz
Flash Memory: 256 kbytes	Bandwidth	192 Mbytes/sec
	Cache	256 bytes
RAM		64 kbytes
EEPROM		2 kbytes
Direct Memory Access		16 Channels
Digital I/O: 34 Pins	Voltage Output	3.3 Volts
	Voltage Input	5 V Tolerant
Analog Input: 21 Pins	Converters	2
	Touch Sensing	12 Pins
	Comparators	3
Analog Output		1 Pin
Timers		12 Total
Communication		USB, Serial, SPI, I2C, CAN Bus, I2S Audio

Figure 27: Newark 18 kHz piezoceramic disc element¹¹⁶.

Figure 28: McMaster-Carr Plasti Dip Rubber Coating¹¹⁷.

Figure 29: PJRC Teensy 3.1 Microcontroller USB Development Board¹¹⁸.

6.4 Blue Tactile LED Button

A tactile LED button was chosen as an On/Off button to provide users with both tactile and visual feedback about the status of the device. A blue tactile LED was chosen for its size, color compatibility, and availability¹¹⁹. See Figure C3 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for a drawing of the button.

6.5 Status Indication LEDs

Multicolor LEDs were chosen as indications of hydration status due to their inexpensiveness, ease of implementing in code, small power draw, and the availability of different colors to indicate different ranges of hydration¹²⁰. See Figure C4 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for an info sheet of the LEDs.

6.6 Vibration Motor

A vibration motor was chosen to provide users with noticeable tactile feedback on changes in hydration status, alert users that interaction is necessary, or affirm the selection of an interaction. The use of vibration was chosen over audible alerts because of its discrete nature and because athletes often listen to music or are in loud environments while training. This vibration motor was selected for its small power draw, relatively high vibration output, compatibility with the Teensy 3.1 board, and small size¹²¹. See Figure C5 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for a datasheet of the vibration motor.

Figure 30: SparkFun blue tactile LED button¹¹⁹.

Figure 31: SparkFun multicolor LEDs¹²⁰.

Figure 32: SparkFun vibration Motor¹²¹.

6.7 400 mAh Polymer Lithium Ion Battery

A polymer lithium ion battery was selected as a power source due to its availability, small size, reduction in user recurring costs (vs. repeatedly buying coin cell, AA, or AAA batteries), compatibility with USB recharging methods, relatively long battery life, and compatibility with the Teensy 3.1 board¹²². A 400 mAh battery was selected primarily for its availability without a custom order. A review of other fitness tracking devices with battery lives of 3-7 days revealed batteries ranging from 50-300 mAh, indicating that 400 mAh with a well designed power saving system is more than sufficient to meet the engineering requirement of three days of intermittent (once per minute) use. Regardless, future calculation of the power requirements for a prototype with additional functionality (as discussed in the Future chapter) will lead to a more informed battery selection. See Figure C6 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for an image of the internal components of the 400 mAh battery.

6.8 Bluetooth LE Module

A Bluetooth LE (BLE) module was selected to provide wireless communication for the device. BLE is the lowest power Bluetooth protocol available and allows for interaction with any 4.0 Bluetooth enabled device¹²³. Additionally, the use of BLE has become an industry standard in wearable devices¹²³. See Figure C7 of Appendix C: Manufacturer Supplied Component Datasheets, Diagrams, and Drawings for a drawing of the module.

6.9 ABS Plastic Case

ABS plastic was selected as the electronics housing material due to its availability, inexpensiveness, common use in electronics, color options, low density, and ease of machining¹²⁴. The plastic case holds the electronic components and allows for increased water resistance if the appropriate seals are used on any holes. Future iterations of the device could use custom-molded ABS shapes to minimize the overall size and increase the durability of the device.

6.10 Hook and Loop Cinch with an Elastic Nylon Strap

A hook and loop cinch with an elastic nylon strap was chosen for the bicep band material due to its flexibility, availability, inexpensiveness, and ability to maintain a pressured contact surface between the bicep and the transducers¹²⁵. The cinch system allows for custom sizing with small increments for fitting a large variety of bicep sizes. With proper sizing, this band is sturdy enough to hold the electronic components without sliding down the arm of a user. Additionally, elastic and nylon are commonly used in available athletic bicep bands and have desirable sweat wicking properties.

Figure 33: SparkFun 400 mAh polymer lithium ion battery¹²².

Figure 34: Digi-Key Bluetooth 4.0 Module¹²³.

Figure 35: McMaster-Carr ABS plastic sheets¹²⁴.

Figure 36: McMaster-Carr hook and loop cinch with elastic nylon strap¹²⁵.

6.11 Band Prototype

See Figure 37 and Figure 38 for images of the prototype band. Unfortunately, unforeseen complications in the development process put significant time constraints on the development and testing opportunities of a fully functional wireless prototype. Due to the time constraints, it was not possible to adopt the wired breadboard prototype into a mounted wireless version. As is, there are two main components to the device:

- (1) An elastic nylon bicep band with an adjustable cinch, Velcro attachments, and nylon tubes that hold the coated piezo disc elements in place. The tubes run the wires from the piezo disc elements, through an opening in the band, and into an interior pocket that would hold the

electronic components. There is a reflective patch covering the location of the interior pocket to indicate where LEDs would appear in a final prototype.

- (2) An electronics breadboard prototype with the Teensy 3.1, uncoated piezo disc elements, and micro-USB connection to a computer for power and data acquisition.

Since it was not possible to assemble and test a wireless version of the device, the main focus of this thesis is on using the electronics breadboard prototype to validate the method of detecting dehydration. All of the testing performed in the Validation chapter was performed using the breadboard prototype. Regardless, the prototyped band shows how the sensors and electronics can be integrated in a flexible, lightweight, and wearable device. See the Validation chapter for more information on the electronics breadboard prototype and the Future Functionality section of the Conclusions chapter for a discussion on possible future directions for an integrated prototype.

Figure 37: Images of the prototype band.

Figure 38: Detail views of the prototype band features.

6.12 Components and Prototype Cost Analysis

See Table XIII for a summary table of the components used in the prototype band. Note that this is not a compilation of all expenses in this thesis, nor does it include the cost of taxes and shipping. Items without vendors were found or provided locally at no expense, but an estimated cost is provided for calculation of the total cost of the prototype. As seen in Table XIII, the total cost for the prototype band is \$22.52.

Future additions of battery power and Bluetooth LE communication would add an additional \$27.43 for a total cost of \$49.95 per band.

Table XIII: Summary table of the suppliers, number, and cost of the components used in the prototype device.

Component	Vendor	Vendor Item #	Quantity	Cost (\$)
Teensy 3.1 USB Board	PJRC	TEENSY31	1	19.80
Multicolor LEDs	SparkFun	COM-12062	4	0.59
18 kHz Piezo Disc Element	Newark	25R0595	2	0.54
Blue Plasti Dip Coating	McMaster-Carr	9560T7	~0.45 oz	0.31
Electrical Wire	-	-	~12"	0.20
Solder Wire	-	-	~0.125"	0.05
1" Elastic Fabric	McMaster-Carr	88225K67	~12"	0.23
0.75" Velcro	-	-	~3"	0.10
Spandex Material	-	-	~24 x 1"	0.20
1" Plastic Cinch	-	-	1	0.50
Total				\$22.52
400mAh Polymer Lithium Ion Battery	SparkFun	PRT-10718	1	6.95
Bluetooth 4.0 Module	Digi-Key	1446-1028-1-ND	1	20.48
Wireless & Battery Powered Total				\$49.95

7. VALIDATION

7.1 Safety Calculations

The safe implementation of the ultrasonic method was investigated through theoretical calculations. The relationship between the intensity (in Watts/cm) and irradiation time of an ultrasonic process determines the safe exposure. See Figure 39 for a plot of this relationship, as given by the NCRP standards for exposure to ultrasonic mechanisms. In addition to an increasingly non-linear response within the tissue, at high intensities ultrasonic waves can produce enough heat to rapidly melt solids or cause cavitation – a phenomenon that occurs when the stresses generated in the ultrasonic waves produce free chemical radicals and promote certain chemical reactions, leading to the formation of cavities – in liquids⁹⁸. While cavitation caused by high intensity ultrasound is beneficial in many applications like ultrasonic cleaning, sonochemistry, transdermal drug delivery, hyperthermia for cancer treatment, and ultrasonic ablation, it is extremely important to avoid unintentional cavitation⁹⁸. For this reason, the theoretical intensity of the selected disc element was calculated. See Figure 40 for the equations and results of this calculation process. Note that this calculation is for a worst case scenario: the highest intensity region of the 6-mm diameter piezo element on wet skin with a reduced resistance of $1000\ \Omega$ ¹²⁶.

Figure 39: Intensity of ultrasound vs. irradiation time for safe exposure to medical diagnostic ultrasounds⁹⁰.

$$Power = \frac{V_p^2}{R}$$

For $V_p = 3.3 \text{ V}$ and $R = 1000 \ \Omega$:

$$Power = \frac{(3.3 \text{ V})^2}{1000 \ \Omega}$$

$$Power = 0.01089 \text{ Watts}$$

$$Intensity = \frac{Power}{Area}$$

For $A = \pi r^2$, $r = 0.3 \text{ cm}$:

$$Intensity = \frac{0.01089 \text{ Watts}}{\pi(0.3 \text{ cm})^2}$$

$$Intensity = 0.0385 \frac{\text{Watts}}{\text{cm}^2}$$

Figure 40: Calculations for the worst-case intensity of the selected 18 kHz disc elements.

In comparison with the safe exposure chart, the calculated worst-case intensity of 0.0385 Watts/cm² does not have an effect at any irradiation time. Theoretically, the selected piezo element could be in contact with wet skin while transmitting ultrasonic waves non-stop at the highest intensity the Teensy 3.1 board can output with no adverse effects on a user.

7.2 Frequency and Signal Detection Test

Tests and calculations were performed to determine the desired frequency and shape of the ultrasonic signal. The following factors were considered in the selection of the frequency:

- The lower limit of the ultrasonic range is 20 kHz.
- Piezo disc elements have optimal performance at or near their natural frequency.
- Power draw increases with increasing frequency.
- The signal processing capabilities are limited by the Teensy 3.1 specifications.

In sum, minimizing the frequency minimizes the processing and power requirements while maximizing the performance of the 18 kHz piezo disc element. Thus, a signal frequency of 20 kHz was chosen.

The sent wave can be a sinusoidal wave or a square wave. While the Theory chapter discussed the use of sinusoidal signals, square signals demonstrate the same shifts but can be easier to process because there are more distinct changes within the signal. Thus, preliminary testing was performed to visually identify which wave shape has a more noticeable change between the sent signal and the received signal. In this testing, an oscilloscope and a function generator were connected to a piezo disc element and used to generate $10\text{ V}_{\text{p-p}}$ sinusoidal and square waves. Another piezo disc element was connected to the second input of the oscilloscope and used to receive the signal. The piezo elements were placed on either side of various limbs to visually identify if the 20 kHz signals could be detected and, if detected, which wave shape showed the most identifiable differences between the sent and received waves. See Figure 41 for a diagram of this setup with a $10\text{ V}_{\text{p-p}}$ square wave through a male forearm.

Figure 41: Image of the oscilloscope and function generator test setup.

As seen in Figure 31, the received signal from the $10\text{ V}_{\text{p-p}}$ square wave is easily detected, well defined, and noticeably shifted from the sent signal. Given this behavior, the ability to detect signals from piezo disc elements was validated and a square wave shape was chosen for use in the prototype.

7.3 Code Development

Through a collaborative process, Ross Gregoriev (MS MATE '14, Cal Poly SLO) generously helped create the validation code by adopting his previous work in analog signals for the purposes of this thesis.

Three programs were used to compile the code for the validation process. See Table XIV for a list of the programs and their role in the validation code.

Table XIV: Summary table of the programs used to generate the validation code.

Program	Role
Teensyduino	Plug-in interface between PJRC's Teensy 3.1 board and the Arduino IDE. Upon installation, Teensyduino added the Teensy 3.1 board to the Arduino IDE board options list. After any changes to the code within the Arduino IDE, Teensyduino served as an intermediary for loading code onto the Teensy 3.1.
Arduino IDE	Development environment used to program the Teensy 3.1 for the analog output and an analog input signal. The finished Arduino code was loaded onto the Teensy 3.1 once. Subsequent control of the Teensy 3.1 was performed via an Arduino IDE interface in LabVIEW.
LabVIEW	Interface used to run the Teensy 3.1, troubleshoot the code, process the signals, and acquire data on the processed signals. LabVIEW is advantageous over the Arduino IDE because it allows for graphic coding using block diagrams and shows live views of the acquired signals via graphs on the front panel.

See Figure 42 for example windows of the Teensyduino, Arduino IDE, and LabVIEW programs.

Figure 42: Example windows of the programs used to generate the validation code; (A) Teensyduino, (B) Arduino IDE, (C) LabVIEW (left) Front Panel and (right) Block Diagram.

The general process of the code development and subsequent data acquisition was as follows:

- Optimal signal acquisition timing calculated.
- Timing variables defined in the Arduino Teensy_Acquisition and Analog_Acquire code.
- Arduino code loaded onto the Teensy 3.1 via the Arduino IDE and Teensyduino.
- LabVIEW interface with Arduino created in the Block Diagram panel.
- Signal processing loops created in the LabVIEW Block Diagram panel.
- Desired output graphs generated in the LabVIEW Front panel.
- *Run test.*
- Export data from the desired output graphs in the LabVIEW Front panel to Excel.
- Generate charts from the data in Excel.

The analog signal generated by the Arduino/Teensy 3.1 code is characterized by the number of samples in each period, the voltage of the waveform, and the length of the period. The Teensy 3.1 processor datasheet and the provided Freescale ADC calculator were used to calculate the optimal number of samples per period at 15 samples. The voltage of the waveform is 3.3 V, as dictated by the Teensy 3.1 analog pin outputs. Finally, using **Equation 5** (as shown in Figure 43), the desired frequency of 20 kHz was used to calculate the period of the wave at 50 μ s.

$$f = \frac{1}{T}$$

$$T = \frac{1}{f}$$

$$T = \frac{1}{20,000 \text{ Hz}}$$

$$T = 50 \mu\text{s}$$

Figure 43: Calculation of the period for a 20 kHz wave.

See Figure 44 for a flowchart of the validation code, Figure 45 for a pinout diagram of the Teensy 3.1, Figure 46 for an image of the prototype circuitry, and Figure 47 for identification of the modules on the Front Panel. The full Arduino codes can be found in Appendix E: Arduino Code – Teensy Acquisition and Appendix F: Arduino Code – Analog Acquire. The full LabVIEW Block Diagram and Front Panel can be found in Appendix G: LabVIEW Block Diagram Panel and Appendix H: LabVIEW Front Panel.

Figure 44: Flowchart of the validation code.

Figure 45: Pinout diagram of the Teensy 3.1 with the validation code.

Figure 46: Prototype Teensy 3.1 running the validation code via a breadboard and two 18 kHz piezo disc elements.

Figure 47: Identification of the modules within the LabVIEW Front Panel. Note that the 5th harmonics plot is not used for gathering or analyzing data.

Essentially, this code is generating a specific analog signal to send through one piezo element, receiving an analog signal at the other piezo element, converting the received signal to digital data, processing the data, and printing graphs of the processed received signal. An example diagram of the sent, received, and an overlay of the sent/received signals is shown in Figure 48. As seen in Figure 48, the received signal carries a certain amount of *distortion* compared to the sent signal. Recall the goal of determining hydration status from the magnitude of a phase shift between a calibrated baseline signal and an acquired signal; likewise, the amount of distortion can be used to compare the two signals. The distortion in the received signal is due to the attenuation, or capacitive effect, of the material through which the signal is propagating. Theoretically, a decrease in tissue water content leads to more attenuation, which causes more distortion, thus indicating dehydration. In the case of this validation code, distortion is shown as a plot of the harmonics of the sent signal. While the ultimate goal for the code in the hydration monitor is to have a comparator numeric value from the calculation of the phase shift between two signals, the time constraints of this thesis restrict the validation to visual analysis of two signals.

Figure 48: Comparison diagram of the sent and received signals for the 3.3V, 20 kHz square waves.

7.4 Distortion Validation

To validate attenuation as the theoretical cause of distortion, the signals received through two materials of similar thickness and different attenuation properties were measured. See Table XV for the materials, images of the samples, thickness of the samples, and the calculated average distortion amplitude. Note that attenuation coefficient values could not be found for the materials at 20 kHz, but the attenuation of solid materials varies with frequency by an approximately linear relationship, so the ratio between the two attenuation coefficients is similar at 20 kHz¹²⁷. Raw data from these tests can be found in Appendix I: Accessories to the Distortion Validation Tests. As seen in the aluminum sample images, there are two holes extending through the sample. Care was taken to keep the transducers on the middle solid portion of the aluminum sample.

Table XV: Sample properties for the distortion validation test.

Material	Images of Sample	Thickness of Sample	Attenuation Coefficient (at 10 Hz) ¹²⁷	Calculated Average Distortion Amplitude
Aluminum	<div>Side: </div> <div>Top: </div>	9.0 mm	0.4 neper/m	7614.5 dB
Synthetic Rubber	<div>Side: </div> <div>Top: </div>	10 mm	15 neper/m	66.6 dB

See Figure 49 and Figure 50 for the harmonic plots of the two material samples.

Figure 49: Harmonics plot for the aluminum sample.

Figure 50: Harmonics plot for the rubber sample.

See Figure 51 for a combined plot of the two samples. As seen in the plots, the amplitudes of the harmonics are very different, indicating that the material with a higher attenuation coefficient has lower harmonics amplitudes.

Figure 51: Combined harmonics plot for the aluminum and rubber samples.

Interestingly, the attenuation coefficients of the materials and the average distortion values have similar percent differences. The percent difference between the attenuation coefficients is 190% and the percent difference between the average distortion values is 197%. See Figure I1 in the Appendix I: Accessories to the Distortion Validation Tests for the calculation of the percent differences. The similarity in percent differences may indicate a minimum threshold sensing value in which material attenuation differences within a certain range cannot be detected using the processing in this validation code. However, this possibility requires future investigation with additional materials to compare a wide variety of attenuation coefficients. Regardless, the main outcome of this test – a lower amplitude from a higher attenuating material – supports the theory that a plot of harmonics will reflect differences in the attenuation coefficients of the material through which the signal is propagating.

7.5 Sponge Water Content Validation Test

The ability to detect water content in a material with a constant thickness was investigated using a sponge wetted with incremental amounts of water. See Figure 52 for an image of the setup for this test. First, the vulnerable electrical components of the prototype were protected from moisture and secured to the breadboard via insulating electrical tape. Since these piezos do not have a Plasti Dip coating, each piezo element was secured to a rubber eraser to make them easier to manipulate, reduce the conductive effect of touching the elements while tests are being run, prevent the solder points from contacting water, and minimize the effect of manually applied pressure on the driving elements of the piezos. A new sponge was removed from its packaging, weighed, and placed in a Ziploc bag. The harmonics of the dry sponge were measured at three locations. Then, the Ziploc bag was opened and one tablespoon of lukewarm water was carefully spread over the top of the sponge. The weighing and signal acquisition procedures were repeated at the same three locations for up to six tablespoons of water. See Appendix J: Accessories to the Sponge Water Content Validation Test for the raw and averaged data from this test. Additionally, see Figure 53 for a plot of the averaged acquired data from the Dry to 2T levels.

(A)

(B)

(C)

(D)

Figure 52: Prototype setup for the sponge water validation test. (A) Protected breadboard connections and connection of the piezo elements to the rubber backing, (B) Placement of sensors and measurement points on the sponge, (C) Side view of the experimental setup with a constant material thickness, and (D) Method of adding water to the sponge.

Figure 53: Harmonics and trendlines for the sponge test.

As seen in Figure 53 and consistent with the results from the distortion validation test, an increase in the water level of the sponge corresponds to a decrease in the harmonic amplitude. However, the reduction in harmonic amplitude is not consistent with an incremental increase in the amount of water within the sponge. In other words, increasing the water content by a second tablespoon did not cause the same drop in harmonic amplitude as adding the first tablespoon of water to the dry sponge. This observation was noted again when a third tablespoon of water was added. As seen in Figure 54, the average harmonic amplitude from the third tablespoon to the sixth tablespoon did not change by more than 0.5 dB. This plateau of the average amplitude plot can be explained by three factors:

- (1) The sponge, once saturated, does not change in attenuation.
- (2) The geometry of the sponge, namely the coarse green layer, may cause the water to settle at the bottom of the sponge and affect the detection of increasing sponge water content.
- (3) The inhomogeneity of the sponge macrostructure does not provide consistent attenuation behavior.

A future, more precise, investigation on the behavior of the harmonics amplitude with samples of increasing water content can be performed by using homogenous blocks of agarose gels with various water contents. Regardless, the ability of the prototype to detect the initial increase in water content contributes to the validation of the method.

Figure 54: Average harmonic amplitude values for the sponge test.

7.6 Meat Dehydration Test

The distortion validation and sponge water content tests both showed an ability to detect a change in the acoustic properties of a material. However, the acoustic properties of tissues with various hydration states cannot be extrapolated from behavior in aluminum, rubber, and sponge materials. Further, it is unknown how the harmonic amplitude relates to the percentage dehydration of a tissue. For this reason, a final test was performed to assess if the prototype code can detect percent changes caused by the dehydration of muscle tissue. See Table XVI for the stepwise process, images, and a description of each action performed in the meat dehydration test.

Table XVI: Stepwise process, images, and description of actions performed in the meat dehydration test.

Step	Image	Description
1		<ul style="list-style-type: none"> - Obtained six slices of thin cut beef eye of round steak. - Placed slices in individual bags for identification and safe handling of raw meat. - The orientations of the samples within the bags were noted.
2		<ul style="list-style-type: none"> - Used an acrylic sheet with 20 mm hashes to mark three equally spaced measurement locations for each sample. - The apparent tissue component for the measurement locations (muscle fiber or adipose tissue) was visually noted, but not discriminated in the harmonics measurements.
3		<ul style="list-style-type: none"> - Each sample was individually weighed with respect to the 6-gram Ziploc bag. - Note that the scale has ± 2 gram tolerances.
4		<ul style="list-style-type: none"> - An infrared thermometer was used to sense the temperature of each sample prior to harmonics measurements. - Note that all measurements were taken at 71 ± 1.8 °F.

5		<ul style="list-style-type: none"> - The same prototype set up from the sponge water content test was used to obtain the harmonics measurements. - Three measurements were obtained for each sample.
6		<ul style="list-style-type: none"> - The samples were equally spaced on the top plate of a Nesco American Harvest Food Dehydrator & Jerky maker. - Three samples were dehydrated at a time to expedite the test process though uniform drying and staggered data acquisition. - Note that the thickness of the dehydrator plates limited the sample thicknesses to less than 1". - The samples were dehydrated at 95 °F for 20-minute increments.
7		<ul style="list-style-type: none"> - After a 20-minute dehydrating increment, the samples were placed back in their Ziploc bags and allowed to cool to room temperature. - The Ziplocs were left open during the cooling process to avoid condensation within the Ziploc. - Five data sets were obtained for a total dehydrating time of ~1 hour for each sample.
8		<ul style="list-style-type: none"> - Data for all the samples was compiled. - Average harmonics amplitudes were calculated for each location. - The three average values per sample were averaged again to get a single average harmonics amplitude for each sample at each set point. - The percent dehydration at each set point was calculated by finding the percent difference between the starting weight of the sample and the weight at the particular set point. - The relationship between the average harmonics amplitude and the percent dehydration was plotted and analyzed. - Statistical significance analysis was performed in JMP.

See Table XVII for a summary of the average harmonic amplitude for each sample and Table XVIII for a summary table used in the statistical analysis. Additionally, see Appendix K: Accessories to the Muscle Dehydration Test for the data table used to compile averages during testing and for a summary figure of the statistical analysis that was performed in JMP.

Table XVII: Summary table of the weight, % dehydration, and average harmonics amplitude for each of the samples at four set points. Note that the first set point is the baseline value used to determine percent differences and is not included in trend analysis.

Sample	Set	Weight (g)	% Dehydration	Average Harmonics Amplitude (dB)
1	1	52	0%	15.31
	2	50	2%	18.41
	3	44	8%	15.69
	4	38	16%	4.59
2	1	56	0%	19.81
	2	52	4%	13.74
	3	46	10%	13.36
	4	42	14%	4.57
3	1	70	0%	24.94
	2	66	3%	14.11
	3	58	9%	13.11
	4	52	15%	4.6
4	1	70	0%	18.16
	2	66	3%	13.87
	3	60	8%	13.57
	4	50	17%	4.62
5	1	68	0%	13.49
	2	66	1%	14.99
	3	60	6%	15.8
	4	48	17%	8.47
6	1	76	0%	14.58
	2	74	1%	14.72
	3	70	4%	26.89
	4	58	13%	8.78

Table XVIII: Summary table of the average harmonic amplitudes at each test time point. The data points in bold were used for statistical analysis of a more hydrated state compared to a dehydrated state.

Sample	Location	Set 1 Amplitude	Set 2 Amplitude	Set 3 Amplitude	Set 4 Amplitude
1	1	12.12	20.8	16.11	4.48
1	2	14.35	18.77	16.41	4.48
1	3	19.48	15.65	14.56	4.82
2	1	19.06	13.85	13.75	4.35
2	2	21.39	14.68	12.76	4.76
2	3	18.99	12.71	13.56	4.61
3	1	23.37	13	12.49	4.6
3	2	28.76	14.11	13.07	4.99
3	3	22.68	15.22	13.78	4.2
4	1	25.81	13.92	14.73	4.53
4	2	14.21	14.35	13.38	4.69
4	3	14.46	13.34	12.58	4.64
5	1	13.66	14.73	16.67	8.37
5	2	14.19	14.99	15.24	8.28
5	3	12.62	15.24	15.51	8.75
6	1	17.83	14.6	16.54	9.06
6	2	14.02	14.23	32.03	8.5
6	3	11.89	15.34	32.1	8.78

See Figure 55 for a plot showing the relationship between the average harmonics amplitude and percent dehydration at all sample locations for all set points. Note that this plot does not contain the initial 0% dehydration values because they were used to normalize the dehydration percentages for the other set points. Additionally, one outlier (> 2 SD from mean, Set 6: 4%, 26.89 dB) value was excluded from the trend analysis.

Figure 55: Relationship between the average harmonics amplitude and percent dehydration for all sample locations.

Statistical analysis in the form of a paired t-test between the average values from Set 2 and Set 4 ($n=18$) showed a statistically significant difference ($p < 0.0001$) between the harmonics amplitude at a more hydrated state compared to a dehydrated state. Overall, there is a linear trend between decreases in tissue water content and decreases in the average harmonics amplitude ($R^2 = 0.756$). The lack of a stronger R^2 value can be explained by the experimental limitations and errors:

- The meat samples could not be more than 1” thick to fit in the Nesco dehydrator, a limitation that likely resulted in less tissue hydration at the start of the test. A higher starting hydration would allow for easier measurements of the small increments of hydration percentages. Additionally, a thicker sample is a more similar representation of a muscle that the device would monitor.
- The scale used to weigh the samples – and calculate subsequent dehydration percentages – could only measure ± 2 grams. Considering that some weight changes were 2 or 4 grams, this is a significant experimental limitation and source of experimental error.

- The Nesco dehydrator could not run continuously. To minimize the amount of time required for the test, all of the samples were measured after the same amount of time dehydrating. Thus, it was difficult to obtain a wider range of percent dehydration values.

Future use of thicker samples, a more precise scale, and a more advanced dehydrator can significantly reduce the experimental error in the test system and allow for further investigation of the signals at lower levels of dehydration (0%-5%). Overall, the trend between average harmonics amplitude and percent dehydration is a promising indication that the method used in the prototype can detect changes in the hydration status of muscle tissue.

8. CONCLUSION

It was determined that the average harmonic amplitude of a received ultrasonic signal can be used to detect differences in muscle tissue hydration status. This outcome is particularly promising given that a 20 kHz – the lowest limit of the ultrasonic range – 3.3 V square wave output through a readily available processor and inexpensive 18 kHz piezo disc element was sufficient for detection. This setup has exciting future possibilities for implementation in a light wearable device with low power consumption.

8.1 Limitations

The limitations of the meat dehydration study were discussed as possible sources of experimental error. In addition to those limitations, it should be noted that the validation presented in this thesis is a first step toward the development of a continuous wearable hydration monitor. Future investigation into the optimum number of samples to average, how many measurements to acquire in a given time period, the time required for this method to detect a change in hydration status (how many samples to store in the memory for direct comparison), and the range of acceptable values to remain within a hydration status range will allow for real-time monitoring of hydration status. See the upcoming Future Functionality section for more discussion on how to test for and overcome these limitations.

8.2 Future Functionality

Refining the signal processing capabilities of the device will help increase the precision of the measurements. This refinement could mean adding additional filters during the signal acquisition process, adding signal amplification, or custom designing the piezo disc elements. Beyond adjustments to the code and transducers, the most immediate future step is to add wireless capabilities to the device. Prior to adding the Bluetooth module, it is important to develop a data storage system that uses the onboard memory by optimizing the number of samples taken, samples averaged, and the frequency of the signal acquisition process. Then, a method of selecting the appropriate data to leave, transfer, or overwrite must

be designed. Finally, the frequency of data “push” to a device must be optimized to the power requirements of the BLE method.

Future integration of other metrics like heart rate, sweat rate, BMI, accelerometer data, skin temperature, and anthropometrics could improve the accuracy of the device. Theoretically, individual tests could be performed with concurrent data from one of the other metrics. Then, the effect – if any – of each metric on the processed signal can be isolated and used to adjust the signal prior to comparison with the established baseline. See Figure 56 for a basic diagram of this process.

Figure 56: Flowchart for future adjustments with advanced metrics.

After determining the most important metrics to track in conjunction with the acquired signal, the appropriate inputs and/or sensors can be added to the device. For example, if anthropometrics or BMI has a significant effect on the signal, a user can add measurements during the calibration process. Likewise, if motion has a predictable effect on the acquired signal, an accelerometer can be added for signal adjustment.

8.3 Future Wrist Location

Future investigation on the ability of the proposed method to detect changes in hydration status at a wrist location will allow for integration with other commonly available wearable devices. Changes in the harmonic amplitude of blood plasma or tendons could be detected by using a piezo transducer designed to probe at the specific depths of the desired tissue. See Figure 57 for a Solidworks model of a wrist concept device. Overall, this thesis showed promising indications for the future refinement of a wearable, continuous, and non-invasive ultrasonic method for monitoring body hydration level.

Figure 57: Images of a wrist concept model.

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APPENDICES

Appendix A: Survey Responses

Athletes and coaches were surveyed via email for currently owned devices, knowledge of dehydration, desired design considerations, and interest in a hydration monitor. Athletes were asked the following questions:

1. Are you aware of the importance of proper hydration? What do you know about proper hydration?
2. How do you assess how much water you need to drink throughout the day and while training?
3. Would you use a wearable, continuous, and non-invasive hydration monitor while exercising?
4. Would you use the hydration monitor during the day (while not exercising)?
5. If you would use the hydration monitor, what are the biggest design requirements you have for such a device?
6. What sports / fitness activities are you regularly involved in?
7. What fitness monitoring devices do you currently use (heart rate monitors, apps, GPS watches, etc.)?
8. Any comments regarding the functionality of the device (what would you want it to do?), hydration testing, or general comments would be greatly appreciated!

Coaches were asked the following questions:

1. What sport / fitness activity do you coach?
2. Do you use health status monitoring of any sort (weight, BMI, heart rate, fitness tests, etc.) for your team?
3. Would you want your athletes to wear health status monitors during exercise, training, or competition?
4. Do you think proper hydration is an important part of a training regimen?
5. Would you want your athletes to use a wearable, continuous, and non-invasive hydration monitor during exercise, training, or competition?

Out of respect of maintaining anonymity for the survey responders, individual survey answers are not included in this thesis. See Table A1 for a summary of the responses.

Table XIX: Summary table of survey responses.

Athletes	# Responses	10
	Sports / Activities	Swimming, Triathlons (x3), CrossFit (x2), Ultimate Frisbee (x2), Volleyball, Football
	Levels Represented	NCAA D1, Adult Fitness, Professional Leagues
Coaches	# Responses	9
	Sports / Activities	Track, Cross Country, Men's Soccer, Women's Soccer (x2), Wrestling, Beachbody, Ultimate Frisbee, CrossFit
	Levels Represented	High School, NCAA D1, Adult Fitness, Professional Leagues

Appendix B: Kickstarter Review Table

Table XX: Goal amounts and total amounts raised for hydration-related products on Kickstarter.

Product*	Tech	Goal (\$)*	Raised (\$)*
Dash	Tracking	260,000	3,390,551
EMOTIV	EEG	100,000	1,643,117
Omate	Watch	100,000	1,032,352
AGENT	Watch	100,000	1,012,742
Bia	Tracking	400,000	408,160
ALPHA	HR	100,000	321,314
Melon	EEG	100,000	290,941
Nava	Water Bottle	50,000	261,677
LUMO	Back Posture	100,000	200,504
Flyfit	Tracking	90,000	123,817
PROMiXX	Water Bottle	30,000	58,898
ILUMI	Water Bottle	10,000	35,608

* Information gathered via searching on kickstarter.com.

Figure 58: 18 kHz piezoceramic disc element drawing¹¹⁶.

TSD1265

Figure 60: Blue tactile LED button drawing¹¹⁹.

LED MIXED PACK

sparkfun.com

COLOR	BRIGHTNESS	WAVELENGTH	VDROP
BASIC RED	150 - 200mcd	620 - 625nm	2.0 - 2.4 V
BASIC YELLOW	150 - 200mcd	587 - 591nm	2.0 - 2.4 V
BASIC BLUE	300 - 400mcd	465 - 467.5nm	3.0 - 3.4 V
BASIC GREEN	150 - 200mcd	570 - 575nm	2.0 - 2.4 V

Figure 61: Multicolor LEDs infosheet¹²⁰.

Specification	Value
Voltage [V]	3
Frame Diameter [mm]	10
Body Length [mm]	3.4
Weight [g]	1.2
Voltage Range [V]	2.5~3.8
Rated Speed [rpm]	12000
Rated Current [mA]	75
Start Voltage [V]	2.3
Start Current [mA]	85
Terminal Resistance [Ohm]	75
Vibration Amplitude [G]	0.8

www.precisionmicrodrives.com

Tel: +44 (0) 1932 252482

Fax: +44 (0) 1932 325353

Email: sales@precisionmicrodrives.com

Figure 62: Vibration motor datasheet and drawing¹²¹.

Figure 63: Polymer lithium ion battery internal components diagram¹²².

Figure 24: Bluetooth LE module drawing¹²³.

Appendix D: Plasti Dip MSDS Sheet

*Estimate
N/A-Not Applicable
N/R-Not Restricted
N/E-Not Established

MATERIAL SAFETY DATA SHEET

NOTE: BLANK SPACES ARE NOT PERMITTED. IF ANY ITEM IS NOT APPLICABLE, THE SPACE MUST BE MARKED TO INDICATE THAT.					
IDENTITY (As shown on Label or package) 14.5 Fluid Ounce PLASTI DIP			PART NO. IF APPLICABLE		
SECTION I					
MANUFACTURER'S NAME Plasti Dip International, Inc.			EMERGENCY PHONE No. 1-800-424-9300		
ADDRESS (NUMBER, STREET, CITY, STATE AND ZIP CODE) 3920 Pheasant Ridge Drive			REVISION #		
Blaine, MN 55449			MANUFACTURER'S PHONE No. FOR INFORMATION 1-763-785-2156		
			DATE MSDS WAS PREPARED December 29, 2011		
SECTION II - HAZARDOUS INGREDIENTS INFORMATION. All Health Hazards which comprise 1% or greater of the composition and all carcinogens if 0.1% of the composition or greater.					
HAZARDOUS COMPONENTS CHEMICAL and IDENTITY AND COMMON NAME (S)	% Wt. (OPTIONAL)	CAS NO.	OSHA PEL	ACGIH TLV	OTHER LIMITS RECOMMEND
VM&P Naphtha	32-38	64742-89-8	300 ppm	300 ppm	None
Hexane	16-19	110-54-3	500 ppm	50 ppm	None
Toluene	13-16	108-88-3	200 ppm	50 ppm	None
Methyl Ethyl Ketone	3-7	78-93-3	200 ppm	200 ppm	None
Resins	25-27	N/A	N/A	N/A	None
SECTION III - PHYSICAL / CHEMICAL CHARACTERISTICS					
BOILING POINT 149 - 285°F	SPECIFIC GRAVITY (H₂O =1) 0.79 - 0.83		APPROXIMATE WEIGHT PER GALLON (LBS) 6.60-690		
VAPOR PRESSURE 125 mmHg @ 20°C	VAPOR DENSITY (AIR = 1) Heavier than air		EVAPORATION RATE (BUTYL ACETATE =1) > 1.0		
SOLUBILITY IN WATER Insoluble	% VOLATILE 72-75% VOC LBS./GAL 4.8-5.2		OTHER (IF ANY) None		
APPEARANCE AND ODOR Various Colors, Honey Like Substance - Characteristic Odor					
SECTION IV-FIRE AND EXPLOSION HAZARD DATA					
FLASH POINT (METHOD USED) -10.0°F TCC		FLAMMABLE LIMITS 0.9 - 11.5	LEL 0.9	UEL 11.5	
EXTINGUISHING MEDIA Carbon Dioxide, Dry Chemical, or Foam					
SPECIAL FIRE FIGHTING PROCEDURES Self contained breathing apparatus with a full face piece, operated in pressure demand or other positive pressure mode.					
UNUSUAL FIRE AND EXPLOSION HAZARDS This material is flammable and may be ignited by heat, sparks, flame or static electricity.					
HAZARDOUS PRODUCTS FORMED BY FIRE OR THERMAL DECOMPOSITION Carbon Dioxide and/or Carbon Monoxide.					
EXPLOSIVE LIMITS (% BY VOLUME IN AIR) 0.9 - 11.5					
SECTION V - OPTIONAL HAZARD RATINGS IDENTIFICATION					
HAZARD RATING 4-EXTREME 3-HIGH 2-MODERATE 1-SLIGHT 0-INSIGNIFICANT **SEE SECTION IV			National Fire Protection Association (NFPA) FIRE 3 REACTIVITY 0 HEALTH 2 SPECIAL HAZARDS None		

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(Appendix D: Plasti Dip MSDS continued)

SECTION VI - REACTIVITY AND STABILITY DATA						
STABILITY		UNSTABLE	STABLE X	CONDITIONS TO AVOID	None	
INCOMPATIBILITY (Materials to Avoid) Strong acids, bases, oxidizing agents, selected amines with alkali metals and halogens.						
HAZARDOUS DECOMPOSITION OR BY PRODUCTS Carbon Monoxide, Carbon Dioxide						
HAZARDOUS POLYMERIZATION		MAY OCCUR		CONDITIONS TO AVOID		
		WILL NOT OCCUR X		None		
SECTION VII - HEALTH HAZARD DATA						
ROUTES OF ENTRY		INHALATION? Yes		SKIN? Yes	INGESTION? Yes EYES? Yes	
HEALTH HAZARDS		ACUTE X	See Signs and Symptoms of Exposure below.			
		CHRONIC X	Brain and Nervous System Damage (Referred to as solvents or painters syndrome). Drying or Cracking skin.			
CARCINOGENICITY: No						
SIGNS AND SYMPTOMS OF EXPOSURE						
Headache, Dizziness, Drowsiness, Fatigue, Irregular Heartbeat, Skin and Eye Irritation.						
Target Organs: CNS, CVS, PNS, Liver, Kidneys, Lungs, Respiratory System, Skin.						
MEDICAL CONDITIONS GENERALLY AGGRAVATED BY EXPOSURE Pre-existing Heart, Liver, Kidney and Lung disorders.						
EMERGENCY AND FIRST AID PROCEDURES		Ingestion: Contact Physician or Poison Control Immediately.				
		Inhalation: Remove to fresh air. Administer Oxygen or Artificial Respiration if Necessary.				
		Eye Contact: Flush with large amounts of water. If irritation persists, contact Physician.				
		Skin: Wash with soap and water.				
SECTION VIII - PRECAUTIONS FOR SAFE HANDLING AND USE						
STEPS TO BE TAKEN IN CASE MATERIAL IS RELEASED OR SPILLED		Wipe up with floor absorbent. Transfer to hood. Prevent run-off to sewers.				
		Eliminate all sources of ignition. Ventilate to maintain exposure below PEL's. Use sand or other material to dam or contain spills. If large spill, notify appropriate state and local agencies.				
WASTE DISPOSAL METHODS		Dispose of product in accordance with local, county, state and federal regulations.				
PRECAUTIONS TO BE TAKEN IN HANDLING AND STORAGE		Keep away from sparks, flame and heat sources. Do not store above 120°F and use adequate ventilation.				
		Avoid inhalation of vapors and contact with liquid product. Use good personal hygiene.				
OTHER PRECAUTIONS		Keep Container Closed When Not In Use. Containers should be disposed of in an environmentally safe manner in accordance with Governmental Regulations.				
SECTION IX - CONTROL MEASURES						
RESPIRATORY PROTECTION (SPECIFY TYPE) Depending on the Airborne concentration, use a Respirator with appropriate NIOSH approved cartridge or supplied air equipment.				PROTECTIVE GLOVES		
				Impervious		
VENTILATION		LOCAL EXHAUST Supplemental (if needed)		SPECIAL None		
		MECHANICAL (GENERAL) To maintain exposure below PEL's		OTHER None		
EYE PROTECTION				OTHER PROTECTIVE CLOTHING OR EQUIPMENT		
Chemical splash goggles or approved eye protection.				Impervious Clothing/Boots as needed.		
WORK HYGIENIC PRACTICES Wash thoroughly after handling.						
SECTION X - TRANSPORTATION INFORMATION (Optional)						
D.O.T. PROPER SHIPPING NAME Coating Solution (UN 1139)				D.O.T. HAZARD CLASS 3, PG II		
IATA PROPER SHIPPING NAME Coating Solution (UN 1139)				IATA HAZARD CLASS 3, PG II		
DOT EXCEPTION See DOT 49CFR 172.102, New Special Provision 149 AND DOT 49CFR 171.8 FOR CONSUMER COMMODITY, ORM-D						
SECTION XI - 313 SUPPLIER NOTIFICATION						
THIS PRODUCT CONTAINS THE FOLLOWING CHEMICALS SUBJECT TO REPORTING REQUIREMENTS OF SECTION 313 OF THE EMERGENCY PLANNING AND COMMUNITY RIGHT -TO-KNOW ACT OF 1986, 40 CFR 372, (see table on page 1 for CAS # and percent by weight).						
Hexane, Toluene and Methyl Ethyl Ketone						
WARNING: THIS PRODUCT CONTAINS A CHEMICAL KNOWN TO THE STATE OF CALIFORNIA TO CAUSE CANCER AND BIRTH DEFECTS, OR OTHER REPRODUCTIVE HARM.						

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Prepared By: Mark Kenow

THE INFORMATION PROVIDED HEREIN IS BASED ON DATA CONSIDERED ACCURATE. NO WARRANTY IS EXPRESSED OR IMPLIED REGARDING THE ACCURACY OF THE DATA OR THE RESULTS OBTAINED FROM ITS USE. SINCE THE INFORMATION CONTAINED HEREIN MAY BE APPLIED UNDER CONDITIONS BEYOND THE VENDORS CONTROL AND SINCE SUBSEQUENT DATA MAY SUGGEST MODIFICATION OF THE INFORMATION, VENDOR ASSUMES NO RESPONSIBILITY FOR THE RESULTS OF ITS USE.

Appendix E: Arduino Code – Teensy Acquisition

```
//Teensy Acquisition V1_0
//Written by Ross Gregoriev for use in Zoie Engman's Thesis Project
//Last Edit: 4 June 2014
//Use with Analog_Acquire, Teensy 3.1, and LabVIEW basic serial.vi
//
//Sends and Receives analog signals from the Teensy 3.1,
//sent frequency, length of samples, and delay are defined within.

#define TEST_LENGTH_SAMPLES 512//34 waveforms
#define PosAnalogInPinA10
#define NegAnalogInPinA11
#define ExcitationPinA0
#define ExcitationFrequency 20000//20khz
#define LEDPIN 13
#define MS_DELAY 100//delay 100 ms

typedef struct ultrasonic
{
    uint16_t rawdata[TEST_LENGTH_SAMPLES];//raw data input
    uint16_t Reference[TEST_LENGTH_SAMPLES];//reference data input
} ultrasonic;

ultrasonic DataIn;
byte readComplete = 0;
void setup() {
    // put your setup code here, to run once:
    Serial.begin(115200);
    //configure input pins
    pinMode(PosAnalogInPin,INPUT);
    pinMode(NegAnalogInPin,INPUT);
    //configure output pins DAC capable
    pinMode(NegAnalogInPin,OUTPUT);
    pinMode(LEDPIN, OUTPUT);
}

void loop() {
    AnalogAquireInit();
    tone(ExcitationPin, ExcitationFrequency);
    while(1){

        // idleing code
        if( Serial.available()>0){
            //read incoming command
            byte in =Serial.read();
        }
        if(readComplete){
            for( int i=0;i<TEST_LENGTH_SAMPLES;i++){
                Serial.println(DataIn.rawdata[i]);
            }
            readComplete=0;
            // restart DAC or PWM
        }
    }
}
```

```

    //RESTART analog dma
    DMA_ERQ |= 0x01; //reset dma
    NVIC_ENABLE_IRQ(IRQ_DMA_CH0);
    DMA_INT= 0x01; //enable DMA

}
delay (MS_DELAY);
}
}

void dma_ch0_isr(void)
{
    DMA_CINT = 0; //disable interrupt
    //insert what to do with code here
    readComplete = 1 ;

    static byte state = 0;
    state = !state;
    digitalWrite(LEDPIN, state);
}

```

Appendix F: Arduino Code – Analog Acquire

```
//Analog_Aquire
//Written by Ross Gregoriev for use in Zoie Engman's Thesis Project
//Last Edit: 4 June 2014
//Use with Teensy_Aquisition_V1_0, Teensy 3.1, and LabVIEW basic serial.vi
//
//Low level drivers Hydrex, ADC Conversion

void AnalogAquireInit(void){
//ADC conversion setup: setup to be as long as possible
ADC0_CFG1 = ADC_CFG1_ADIV(2) | ADC_CFG1_ADLSMP | ADC_CFG1_MODE(3) | ADC_CFG1_ADICLK(1);
ADC0_CFG2 = ADC_CFG2_ADHSC | ADC_CFG2_ADLSTS(1) ;// High speed configuration, long samp

ADC0_SC2 = ADC_SC2_DMAEN ;//00000100 DMA ENABLE
ADC0_SC3 = ADC_SC3_ADC0 ;//001000 CONTINUOUS CONVERSION

//DMA channel #0 previously set up=
// enable clocks to the DMA ccontroller and DMAMUX
SIM_SCGC7 |= SIM_SCGC7_DMA;
SIM_SCGC6 |= SIM_SCGC6_DMAMUX;
DMA_CR = 0;
DMA_ERQ = 0;

// DMA channel #0 writes audio data to rawdata[512]
DMA_TCD0_SADDR = &ADC0_RA ;
DMA_TCD0_SOFF = 0;
DMA_TCD0_ATTR = DMA_TCD_ATTR_SSIZE(DMA_TCD_ATTR_SIZE_16BIT) | DMA_TCD_ATTR_DSIZE(DMA_TCD
DMA_TCD0_NBYTES_MLNO = 2;
DMA_TCD0_NBYTES_MLNO = 2;
DMA_TCD0_SLAST = 0;
DMA_TCD0_DADDR = &DataIn.rawdata[0];
DMA_TCD0_DOFF = 2;
DMA_TCD0_CITER_ELINKNO = TEST_LENGTH_SAMPLES;
DMA_TCD0_DLASTSGA = -(TEST_LENGTH_SAMPLES * 2 );
DMA_TCD0_CSR = DMA_TCD_CSR_INTMAJOR | DMA_TCD_CSR_DREQ;
DMA_TCD0_BITER_ELINKNO = TEST_LENGTH_SAMPLES;

DMAMUX0_CHCFG0 = 0;
DMAMUX0_CHCFG0 = DMAMUX_SOURCE_ADC0 | DMAMUX_ENABLE;
NVIC_ENABLE_IRQ(IRQ_DMA_CH0); //enable interrupt
ADC0_SC1A = ADC_SC1_ADCH(0x00); // pin A10 for + and A11 for - if in differential/dac
DMA_ERQ |= 0x01; //enable dma
}
```

Appendix G: LabVIEW Block Diagram Panel

Appendix H: LabVIEW Front Panel

Appendix I: Accessories to the Distortion Validation Tests

$$\% \text{ Difference} = \frac{|\Delta V|}{\frac{V_1 + V_2}{2}}$$

$$\% \text{ Diff}_{\text{Attenuation Coeff}} = \frac{|15.0 - 0.4|}{\frac{15.0 + 0.4}{2}} * 100$$

$$\% \text{ Diff}_{\text{Attenuation Coeff}} = 190\%$$

$$\% \text{ Diff}_{\text{Average Distortion}} = \frac{|7614.5 - 66.6|}{\frac{7614.5 + 66.6}{2}}$$

$$\% \text{ Diff}_{\text{Average Distortion}} = 197\%$$

Figure 65: Calculation of the percent difference between the attenuation coefficients and the average distortion value for each signal.

Table XXI: Raw data for the distortion validation tests.

Aluminum	Rubber	Aluminum (cont.)	Rubber (cont.)
----------	--------	------------------	----------------

Time - Collected (Specific Harmonic)	Amplitude - Collected (Specific Harmonic)	Time - Collected (Specific Harmonic)	Amplitude - Collected (Specific Harmonic)	Time - Collected (Specific Harmonic)	Amplitude - Collected (Specific Harmonic)	Time - Collected (Specific Harmonic)	Amplitude - Collected (Specific Harmonic)
04:04.3	9925.3	17:02.9	30.5519	44:20.3	9696.39	03:53.9	30.4286
15:17.3	10231.2	28:14.9	36.0797	52:52.3	4891.4	13:34.9	51.9579
29:40.3	9420.38	42:38.9	56.1258	01:24.3	10110.8	23:49.9	165.259
38:12.3	4307.61	15:32.9	37.49	10:28.3	1508.69	35:47.9	52.5902
47:10.3	11550.1	43:22.9	49.4677	20:20.3	9800.84	47:39.9	65.5677
47:12.3	11544.5	54:45.9	71.0579	30:14.3	3853.49	57:30.9	85.3168
47:15.3	11535.8	06:38.9	158.818	39:26.3	3301.58	09:15.9	180.899
47:17.3	11530	16:30.9	75.8923	49:43.3	9834.38	24:40.9	40.1229
47:20.3	11521	25:02.9	45.4644	59:37.3	10636.3	34:48.9	59.0795
47:44.3	11444.2	36:25.9	142.791	09:40.3	12522.8	02:12.9	86.6381
04:49.3	10845.4	48:22.9	74.9459	18:12.3	12928.7	21:34.9	48.2205
20:09.3	8818.4	59:10.9	65.6857	26:44.3	4113.77	30:19.9	44.3693
29:24.3	5831.91	08:05.9	37.3787	35:16.3	12317.4	50:08.9	54.2834
37:56.3	10130.3	19:45.9	24.4488	43:48.3	3822.91	59:36.9	46.4071
54:27.3	9926.67	31:41.9	32.1407	52:20.3	4547.94	21:18.9	49.1174
04:23.3	5896.12	41:50.9	44.7335	00:52.3	1639.07	38:22.9	81.7093
04:27.3	5904.01	50:22.9	35.1677	09:24.3	7891.06	49:25.9	50.6844
04:29.3	5908.11	59:25.9	21.4034	17:56.3	5753.22	00:56.9	50.2658
04:32.3	5914.48	11:30.9	47.9876	26:28.3	2642.85	12:30.9	77.0507
04:35.3	5921.1	22:57.9	74.9449	35:45.3	7075	21:02.9	80.0747
14:16.3	11673.7	33:02.9	68.8839	45:23.3	4067.36	32:42.9	56.1552
24:02.3	9968.92	41:34.9	37.9355	01:35.3	11330.6	42:16.9	39.7675
34:12.3	13246.2	52:29.9	37.6429	46:41.3	9583.85	54:03.9	73.7149
43:59.3	10258.1	04:32.9	55.7061	57:13.3	2793.65	03:42.9	52.9612
54:03.3	9859.74	15:42.9	40.1672	08:52.3	2993.74	12:14.9	33.1425
01:45.3	5732.3	24:20.9	35.4834	21:04.3	4225.83	23:48.9	55.9535
11:48.3	9043.08	36:07.9	49.1433	30:45.3	5785.48	35:36.9	190.792
20:20.3	8919.31	47:21.9	43.0887	40:43.3	4423.17	46:22.9	61.69
28:52.3	4509.86	58:22.9	29.5843	50:51.3	9844.2	55:36.9	62.0071
38:09.3	11434	07:41.9	46.7181	00:04.3	10654.6	05:22.9	187.565
47:49.3	10846.4	19:30.9	55.1581	08:36.3	12603.9	16:57.9	60.0463
58:02.3	11133.8	30:48.9	41.3086	17:08.3	7307.17	26:59.9	88.0307
07:46.3	11044.7	38:41.9	52.1983	25:40.3	11205.1	37:34.9	80.9194
17:51.3	11767.4	49:34.9	165.658	34:12.3	11671	46:49.9	56.8683
27:27.3	13609.6	59:08.9	50.5038	42:44.3	5149.96	14:25.9	74.5647
37:08.3	12280.3	10:39.9	50.0009	51:16.3	10191	43:36.9	59.5349
45:40.3	9940.48	21:58.9	165.526	00:26.3	10377.8	02:54.9	62.3182

54:12.3	5510.96	31:18.9	52.4977	10:28.3	1702.56	19:30.9	81.7337
02:44.3	1674.22	42:19.9	36.421	20:18.3	4077.1	28:30.9	84.0702
11:30.3	10087	54:22.9	49.2553	30:26.3	3983.87	56:52.9	59.5471
21:21.3	1948.23	06:03.9	71.2102	40:20.3	11745.4	06:56.9	56.5311
31:21.3	4638.7	14:54.9	74.3751	50:12.3	11184.6	18:27.9	59.9559
41:27.3	10628	23:42.9	49.1703	58:18.3	2488.56	28:14.9	84.1507
51:22.3	10412.8	35:49.9	53.7899	08:04.3	9866.56	38:31.9	58.762
01:30.3	10039.1	47:21.9	38.4638	16:36.3	1593.98	50:04.9	59.8272
11:00.3	2860.79	57:34.9	51.0893	25:08.3	3942.02	02:16.9	73.4153
19:32.3	6121.42	07:15.9	37.187	33:40.3	1919.2	10:54.9	59.7051
28:04.3	12383.2	19:08.9	37.0785	42:12.3	2310.88	19:52.9	76.3875
36:36.3	9948.67	30:26.9	77.3406	50:44.3	5671.38	31:36.9	83.924
45:08.3	6001.38	40:14.9	73.0502	59:58.3	8643.1	41:42.9	61.4621
53:40.3	10992.8	48:58.9	50.2766	09:54.3	2203.28	53:02.9	74.3402
03:09.3	5757.79	00:20.9	162.06	20:09.3	7555.48	02:06.9	73.137
13:14.3	10551.6	12:18.9	58.6759	29:09.3	10859.2	13:07.9	61.3309
22:18.3	13378.9	22:54.9	172.932	39:55.3	8342.44	23:02.9	78.4906
30:42.3	5832.14	32:10.9	47.4466	50:23.3	10674.7	36:14.9	62.1188
40:40.3	7800.24	44:01.9	43.6095	59:00.3	10003.2	44:56.9	80.2566
50:48.3	11089.3	55:48.9	34.4108	07:32.3	4993.67	58:58.9	61.7117
00:12.3	4124.73	05:34.9	52.9697	16:04.3	4143.51	10:59.9	76.8942
10:04.3	2406.68	14:06.9	175.434	24:36.3	5187.71	11:03.9	77.0456
19:00.3	10604.6	25:22.9	52.413	33:08.3	8990.24	11:06.9	77.159
27:32.3	11091.1	36:47.9	33.3129	41:44.3	6033.12	11:08.9	77.2346
36:52.3	11244.2	48:14.9	48.92	51:35.3	2897.29	28:33.9	62.9398
46:43.3	1962.37	56:57.9	59.1494	01:16.3	2051.66	42:04.9	63.9211
56:20.3	11002.9	09:04.9	154.057	11:01.3	2418.7	03:39.9	63.1263
06:45.3	6675.02	20:35.9	60.3492	19:36.3	2375.94	15:29.9	60.4023
15:38.3	12283.1	30:54.9	47.5578	30:58.3	2724.04	26:54.9	60.4237
25:52.3	12697.7	40:23.9	66.0939	40:51.3	1750.59	35:42.9	47.6432
35:46.3	10504.6	52:17.9	47.3516	49:56.3	11843.9	35:43.9	47.6806

Table XXII: Summary table for the distortion validation test.

Metric	Aluminum	Rubber
Average	7614.5	66.6
Max	13609.6	190.8
Min	1344.2	21.4
Stdev	3670.4	35.3

Appendix J: Accessories to the Sponge Water Content Validation Test

Table XXIII: Sample weights for the sponge water content test.

Sample	Weight (g)
Dry	24
1T	36
2T	50
3T	60
4T	74
5T	104
6T	124

Table XXIV: Raw data for dry, 1T, and 2T of the sponge water content test. Dry = sponge prior to water added, Samples given in the form of #TL where # = number of tablespoons added and L = location of the measurement.

Dry 1	Dry 2	Dry 3	1T1	1T2	1T3	2T1	2T2	2T3
11.6116	15.2787	8.51108	3.47935	4.14777	5.49769	5.34698	3.44292	3.66312
2.82473	11.7086	5.96021	5.01598	7.95644	1.83152	4.00356	4.94778	2.4842
5.84431	4.72547	6.96679	5.47366	4.67466	3.08421	1.44331	3.40356	3.90317
6.79996	5.42303	3.55741	9.58503	8.29311	4.32393	5.78056	4.41149	5.9978
15.2515	8.13265	4.82126	2.56101	7.14567	4.71867	5.31109	3.25692	8.89031
18.4167	7.32567	4.85874	6.33385	5.34898	6.87699	5.19399	8.26373	4.70799
7.51577	17.569	4.09844	4.55317	5.25846	4.50123	5.10209	8.26653	5.9803
13.4042	24.0276	4.09541	5.72173	6.70004	3.64283	3.46998	6.77382	2.45366
6.2212	16.8997	6.79621	6.87559	4.92018	3.07791	5.33353	3.8319	2.54455
12.9593	5.87366	12.1136	3.8389	7.56584	5.66874	1.92071	6.66652	4.82393
7.82393	8.97018	8.88995	5.39262	5.69511	5.3466	3.55395	5.30268	4.18226
10.6721	6.41658	7.58359	8.26414	8.34357	2.09147	4.26688	4.19891	10.3517
11.4121	9.53099	5.34801	8.15764	11.6232	1.54188	2.83677	2.08519	3.57834
6.55674	5.01274	8.77464	7.75658	7.03634	3.45514	4.90733	3.8576	2.92593
12.475	10.4198	10.1193	7.36221	8.49315	4.00271	3.57733	3.24601	2.50184
7.85241	16.5869	7.82059	5.48135	6.57541	3.42497	5.40216	5.67145	4.28083
9.09899	11.147	5.76761	9.3191	3.26202	3.14877	5.1566	6.29928	9.06294
21.8304	12.8855	5.57123	6.60381	4.19958	4.74761	2.84601	5.42015	7.52296
7.98378	10.0881	9.23785	5.61363	7.69813	2.33926	6.46737	8.57731	5.10656
11.8997	11.0259	4.47447	3.50542	7.34566	7.45266	5.50344	4.20289	6.00657
7.77658	7.40888	5.55014	6.30758	6.66969	5.51027	7.93523	3.37048	3.14973
7.85689	11.451	7.96671	7.79743	8.63159	3.21238	4.27466	3.28719	4.62501
12.5108	11.0932	5.29356	7.61016	7.18033	8.48044	3.97432	5.82308	1.67493

10.2787	5.61694	4.28294	4.02278	7.67584	3.38584	3.52383	4.19633	3.3395
11.7291	9.64156	5.78346	3.62019	7.59601	2.29961	4.32201	2.61726	4.98201
11.8623	14.0852	6.35887	6.38528	7.30646	2.51547	3.17098	3.11246	8.44549
6.42159	11.2316	9.0821	8.01563	6.80716	6.95543	3.41231	4.9063	8.92028
13.0407	8.58482	4.89853	12.5036	7.14829	3.38638	3.68613	2.30886	3.18257
6.07101	9.49728	6.76815	6.56077	6.24846	3.42747	4.81406	1.26826	3.67623
5.43439	8.98164	3.94468	3.97662	3.61754	6.09936	6.49893	3.15768	2.96837
11.577	11.0238	7.01433	4.48735	2.22775	6.91223	6.3872	3.50929	3.94291
6.81476	14.6306	4.54565	5.24587	3.80817	3.84331	5.84026	4.61753	5.05751
10.3563	5.63997	5.77377	4.33519	10.0577	2.78847	4.81898	5.88616	2.27529
14.5173	6.06865	8.63827	7.39359	8.13539	4.83549	5.01316	3.57035	4.44457
13.5423	12.5025	5.82366	7.64006	4.66075	5.79089	3.24535	2.62341	3.29743
16.2078	17.8472	7.04987	6.68307	2.98355	3.38881	3.46815	5.1243	6.36591
8.67746	18.1529	2.37411	7.13008	4.41678	6.65536	2.76664	2.03934	4.44401
11.0526	8.87438	4.28081	3.91928	5.9594	5.41401	4.94103	1.72585	5.24632
15.2864	14.9293	4.1321	10.0754	6.64817	9.37958	6.109	7.86891	5.63044
8.63545	7.60665	4.62427	3.75231	8.04521	3.75776	5.79314	6.16111	3.14691
12.3401	9.38365	9.97428	8.11416	6.20973	2.92523	3.56905	5.32882	2.9517
8.88986	8.83753	6.14893	5.52844	6.69691	4.21573	4.314	2.99672	5.87182
6.77443	12.7189	2.70791	7.43078	7.99637	7.14267	2.46234	5.73408	4.74774
13.458	26.6159	5.4847	5.16981	8.16283	3.41887	4.32809	3.45851	5.09697
8.68899	6.8194	2.12649	4.4344	6.54715	4.90746	3.95105	4.78974	5.2423
10.2089	6.00346	11.7796	9.78473	3.64249	3.94366	2.81022	6.62878	6.60438
7.87041	17.757	9.73606	10.0364	3.70878	4.46126	7.32679	4.45441	5.79895
6.80534	13.5826	2.18593	6.79094	4.18528	1.65956	4.81777	4.8392	3.39028
17.1	10.3847	2.69106	5.12385	5.26924	3.17724	5.97652	5.73323	4.02464
11.0103	22.3816	5.78928	3.96515	9.16278	5.13226	7.19496	5.90591	6.44525
12.4864	10.3088	5.20603	7.80097	3.26315	4.62716	6.95787	4.90996	2.26318
6.63703	5.70359	6.78132	7.71846	6.19216	4.81996	3.74394	7.06779	4.68997
3.40763	11.1063	4.20304	5.66861	4.86991	3.83289	2.45002	7.51684	2.39621
21.9458	23.0592	5.57394	4.18134	7.07335	5.69773	3.05573	4.74133	4.08176
13.1846	9.93643	3.66751	3.85225	4.44221	1.86293	4.15362	4.93655	5.23224
12.6372	8.42846	8.62712	4.60767	8.62033	3.99443	3.00408	7.19562	4.47634
9.59513	4.88728	3.45899	5.69686	5.00489	6.92738	3.78709	4.82678	6.35084
14.5817	9.27483	7.88431	7.17188	3.27592	6.49973	3.4054	8.33746	4.74956
8.30255	8.459	8.59954	9.43961	7.71215	7.45952	4.65668	5.57952	6.40237
9.4805	9.74255	6.74445	6.61585	6.33758	2.8869	6.52527	8.12137	4.40631
11.0481	5.06057	9.14658	7.18484	4.68263	3.88828	3.34675	5.01968	4.79939
11.8812	5.09676	2.83292	5.88376	7.46104	5.45723	3.00835	2.80136	5.01985
15.1504	17.9058	5.96289	8.40766	2.10275	5.15105	7.93268	4.41837	3.42466
16.3533	6.16636	6.92102	5.23671	6.87915	4.96893	4.67915	5.85817	3.37965

7.14201	13.4744	2.28919	6.94575	2.9882	2.55293	3.79109	6.15916	4.40941
9.8792	18.5869	6.35807	5.3597	3.45118	4.64836	9.81358	5.99512	4.47741
5.6235	10.8627	3.74017	9.71725	5.56931	4.7678	6.45386	2.53009	10.6172
7.16606	9.86616	8.71031	8.61444	5.85354	6.94223	3.28832	6.12457	8.19458
3.66388	16.5077	7.40806	4.94301	7.25844	4.58131	4.00909	3.40245	6.2284
10.4222	9.02094	10.7311	4.583	6.72276	7.0253	2.99343	5.03759	2.34613
12.3559	8.48655	10.4447	9.27592	5.93164	6.61473	5.29423	3.64605	5.38355
9.02198	8.368	3.78727	8.44338	2.52061	6.61774	2.42072	3.66527	4.46422
10.9666	10.125	8.77952	5.89909	4.02636	4.33313	4.70975	6.21299	3.17242
7.61786	8.04704	5.44639	2.84586	6.43971	1.90208	2.88114	4.41387	3.26497
7.56769	10.2489	4.76294	9.93861	9.52404	2.28136	8.62762	6.94141	6.28814
14.9735	7.68326	5.4352	4.0227	6.15986	5.00472	7.03809	2.92612	5.34295
8.03708	7.66061	4.17262	6.50853	4.50175	4.00836	2.97837	6.08199	6.28722
12.6897	7.43487	11.4607	6.71345	6.61894	6.20206	4.00907	5.84931	4.80848
14.6444	15.7795	8.00544	6.12633	5.25083	5.38891	4.18912	1.97692	3.75019
9.7816	8.61164	6.80444	8.36323	7.0903	3.83371	4.11951	4.04388	3.34832
11.4917	8.01468	2.35721	5.39507	3.54755	2.46602	3.32111	7.36416	2.826
7.98338	8.45273	9.35013	5.11026	8.97498	7.75358	4.76038	6.20435	7.43618
8.34854	6.08537	5.62912	7.37184	4.28947	4.49289	5.19256	3.00721	4.23544
14.2692	10.3886	5.751	6.97195	4.91457	7.15174	4.14411	3.62217	5.11997
14.6433	17.3608	6.97558	8.96852	3.63016	6.33985	3.18428	6.26739	5.7505
9.28155	7.76652	3.44148	7.0961	5.95032	6.94509	2.37484	3.30661	3.96106
17.966	9.50899	3.92823	10.3875	4.77295	2.02654	4.51113	4.56591	4.42661
10.1892	14.8041	4.43829	4.76726	3.70067	1.73924	2.99468	4.54683	5.77878
10.066	10.4936	4.18005	3.57986	6.613	2.96945	2.56558	3.60621	5.05046
7.81417	8.88737	3.67514	3.81717	9.25317	6.16626	2.99878	4.155	2.98277
15.0811	9.57705	8.00406	5.89553	3.45803	3.046	5.84985	2.98954	10.9857
13.1612	9.51374	5.16361	4.979	2.8	2.91716	4.7486	6.73248	4.01268
15.4475	23.6505	2.11667	5.79446	4.66838	7.24247	3.97157	2.46682	2.7126
8.06709	21.3551	5.35918	5.79051	7.13732	4.68424	5.34928	5.92331	7.01554
9.05266	11.7776	4.62538	5.78765	8.04684	4.17481	4.8781	4.81644	7.48442
12.0274	10.3333	4.73983	5.78401	5.58097	4.02548	5.92232	1.97278	3.7697
10.1309	5.32038	6.21682	5.78227	8.97931	3.99847	2.60335	1.95332	4.21522
8.05848	5.43211	5.76167	5.77974	5.8503	5.20417	2.32847	2.83355	4.2423
8.58838	13.5759	6.7237	5.88056	6.39775	2.6133	2.78814	4.49181	5.12823
10.8746	11.2384	5.83399	6.11215	8.89847	4.49179	4.2646	7.71151	6.19512

Table XXV: Raw data for 3T, 4T, 5T, and 6T of the sponge water content test. Dry = sponge prior to water added, Samples given in the form of #TL where # = number of tablespoons added and L = location of the measurement.

3T1	3T2	3T3	4T1	4T2	4T3	5T1	5T2	5T3	6T1	6T2	6T3
3.0204	5.02196	7.90254	6.2054	3.60657	4.71766	4.16724	5.68188	6.14851	4.62654	5.85004	7.31352
4.47052	4.11445	4.24304	5.49116	3.9416	1.96059	6.34006	4.91619	3.93688	3.67555	4.18688	6.00534
3.00007	1.93552	4.01853	4.61295	2.77787	4.15875	4.77194	4.94103	5.0429	6.00852	3.13795	5.21242
2.11992	5.45441	4.79547	4.65251	2.69272	4.88296	2.48593	3.23381	5.76163	4.10055	2.47452	4.29943
3.69881	2.77368	3.51392	4.50248	2.37317	7.98814	2.60994	2.68553	4.49774	3.80245	3.77169	6.26614
2.82192	1.79814	4.82204	4.76615	4.10854	3.71461	7.42184	4.30216	5.6658	5.09094	2.63382	6.119
5.12663	2.25814	4.66149	3.26708	3.61288	5.67984	5.60731	2.8438	3.5655	5.93644	3.83453	4.86779
7.04563	2.17921	5.35147	2.43069	4.5392	3.95813	4.86167	7.9479	1.53308	2.79779	6.21095	3.56259
10.2574	4.88183	3.83086	2.75429	5.67831	2.79493	4.01039	6.741	2.90669	5.00019	4.51161	4.26056
2.4147	3.4521	3.0411	7.50186	5.4944	3.23889	3.84701	5.96056	4.29475	4.10659	7.30166	3.97
2.4571	2.79688	11.2213	5.57203	4.15068	4.04406	5.98145	3.79388	3.20265	7.9528	3.94648	5.99273
4.02224	3.74125	4.12634	4.12241	4.09988	7.22186	3.69398	5.64647	3.61072	4.48021	5.02271	3.19068
4.06514	3.2877	13.7313	4.08829	3.92259	4.20488	5.82622	3.09286	5.58262	3.08761	3.69286	2.43082
6.48392	1.76645	13.5478	4.93671	4.19233	7.43202	3.86791	4.94362	6.43396	5.55142	2.61624	3.54229
4.75052	6.59673	4.24819	3.65313	3.81731	6.47836	5.68433	5.74632	5.98864	3.59615	4.64542	2.30737
6.11419	3.25462	3.89839	3.08619	3.65422	3.36415	4.9467	5.18092	4.72417	4.50584	10.1864	4.60403
2.56641	4.15702	7.64878	3.12196	7.55284	1.61139	5.66663	6.80683	1.97093	4.34275	4.32969	3.14982
1.87864	3.97733	5.66375	3.09679	4.58111	3.01789	3.43341	6.81284	5.60161	1.47212	3.22825	5.1543
5.43974	2.41078	4.46047	2.83671	7.4012	5.01957	8.88029	6.06047	2.8166	3.69569	7.60805	5.91304
4.25851	4.73756	5.09153	6.01305	5.934	4.32406	3.16848	5.29402	5.36977	1.65796	7.01185	3.94286
4.58369	2.09875	2.99369	4.25444	5.55768	5.40526	6.80675	7.25254	5.79885	5.72236	6.35735	4.64367
2.84895	5.06166	2.08957	7.64017	3.56262	6.00835	5.88188	3.07768	2.3075	7.20766	2.67148	7.07456
3.41553	3.47234	5.81146	3.38911	3.65684	7.42118	5.74948	2.53839	3.30669	5.62371	4.19238	6.60581
5.18742	3.47913	2.00578	1.99715	2.51883	6.84585	4.57818	4.38801	5.17826	6.65552	5.18078	4.13981
6.82499	6.87961	4.62374	5.06959	3.03746	3.35019	5.91054	3.7599	3.5361	3.00287	5.45094	7.49929
7.72701	5.47353	6.73344	4.54128	6.08036	8.56172	7.31228	2.05052	6.05555	4.22149	7.06486	3.02635
3.07279	4.59591	6.74599	5.12918	1.75084	5.27739	4.93333	4.24237	4.69234	3.84749	5.60402	4.99333
4.69885	6.29693	5.4163	9.10675	6.04254	7.06693	6.13198	7.48486	3.37497	8.00677	8.34703	2.39766
3.31431	11.698	4.00118	4.88645	5.19212	3.94935	5.758	6.58368	6.58501	5.09265	6.56776	1.35877
7.02971	8.81781	2.66485	4.32154	4.63769	5.01725	3.62717	2.22016	3.91584	3.61484	6.44288	3.28677
6.23952	1.93749	7.16393	4.51296	4.19315	11.0731	6.56245	5.79654	4.71342	3.06722	3.67671	5.5403
5.35214	3.34648	6.78427	4.03024	3.84157	4.87796	6.36109	5.7787	3.39096	2.92197	7.17504	5.21571
2.65521	4.03501	2.82142	2.1211	6.36954	2.30155	6.15363	5.58539	4.60761	4.49808	7.21037	4.93302
2.67109	3.15135	6.31508	4.19725	2.73059	4.41617	4.98433	10.2371	6.51372	3.41446	5.97794	4.52225
4.86909	5.36171	8.77651	3.1774	3.01714	3.50335	4.43601	8.44648	4.55001	6.60817	7.695	2.81531

3.46339	6.48862	4.5355	5.09178	7.9271	2.68088	3.87595	4.39311	3.60918	5.62588	5.67862	2.35443
3.43831	4.52754	3.05156	1.28756	6.32994	6.31606	6.99789	6.19521	3.77796	3.0277	2.45067	3.81482
2.89428	4.22135	3.10369	3.64626	2.94891	6.65156	4.07196	6.15398	7.79887	3.27505	3.14763	2.0833
2.35381	2.06032	2.65231	1.63467	4.44596	2.26483	3.85031	7.09865	2.9882	4.02034	3.87367	4.39059
7.5369	5.22186	6.75199	2.64236	7.01065	1.4375	3.77536	8.47743	3.49929	5.45773	2.11336	5.03244
4.62195	7.20255	5.88765	5.59399	5.45889	2.88017	3.92395	9.91678	4.73789	4.01583	7.34558	2.32503
3.3943	7.79935	4.46661	4.69237	2.96942	2.02765	4.66764	5.78392	5.58802	4.41888	6.56475	6.34442
6.7168	4.86135	6.21314	5.17668	5.17168	4.22641	7.6785	3.60638	4.16155	4.80082	6.68879	6.94773
5.00811	4.53806	6.97567	2.29293	2.77228	4.61392	5.28414	3.78142	3.53264	4.63792	4.43045	1.9026
6.83941	6.55306	3.40954	5.28365	5.69797	6.99937	4.69511	6.08234	7.75263	7.04282	2.57899	3.65273
4.84177	5.91198	8.29571	4.13373	6.73513	4.12591	9.7842	8.06942	5.71383	5.22061	4.76735	2.87633
5.1027	4.90871	7.0179	4.48322	7.85112	4.61119	3.88526	3.38211	2.98162	4.06273	3.21186	4.56564
6.23851	10.2088	4.54009	7.82119	7.19626	3.15852	2.97201	3.39463	2.80012	2.817	5.47648	2.93671
6.05515	3.33843	5.50048	7.18052	5.50414	4.74144	1.91874	5.09922	5.97478	3.80721	5.24601	5.05932
6.63365	3.84809	3.2512	5.00085	5.69181	2.48113	2.76848	3.48952	3.87445	7.31067	5.60599	5.3042
4.48851	3.0467	6.12593	9.24678	3.95448	3.38204	3.02379	5.41831	4.88359	7.31	6.46646	6.73856
2.57687	10.0418	6.26219	5.25766	3.17879	3.34546	3.3659	3.00338	3.11185	4.70838	2.18638	9.13798
3.31612	4.53341	3.10117	4.64718	4.497	4.81318	7.20272	3.78626	8.17454	3.72749	3.12425	3.54796
4.64649	1.95184	4.42636	2.57297	4.4568	5.59398	3.35825	2.53816	2.72118	4.06115	5.67491	7.76728
2.07274	2.3101	1.5432	2.27103	3.0397	4.1604	6.0696	6.7163	2.80892	4.28944	4.66531	8.18674
12.0721	6.72074	5.02189	4.0346	3.13296	3.69181	3.91483	5.16371	6.03589	6.661	3.55928	6.48291
3.15285	6.12626	7.9852	5.19429	5.8299	3.75663	4.57492	8.23369	5.001	4.26388	5.09681	4.75522
1.70648	3.83835	5.2353	7.6722	5.0939	6.1288	3.36382	8.17802	4.21462	2.33899	5.28399	7.75621
2.28707	4.07021	4.13917	5.65207	6.3794	4.23458	2.84549	4.18549	6.61111	4.11124	5.52055	5.96994
4.37683	7.52209	5.89842	5.78874	4.41559	2.81128	6.77082	7.50912	7.68386	1.88679	4.79464	7.72928
5.45311	6.26286	5.61648	3.23511	3.9709	4.72447	7.94184	3.90558	5.4734	2.94714	2.82516	4.55911
4.45601	3.95056	3.68985	3.45921	4.61222	6.0976	5.72857	7.61333	5.09961	4.27641	3.1108	6.27466
4.63332	5.86492	4.49437	4.17151	6.48935	4.0451	6.45237	4.15117	3.10783	4.00373	2.05057	6.95978
3.17942	3.66826	3.50962	2.41125	2.56921	2.90404	8.16267	5.97779	6.03926	4.35797	1.60303	7.57633
3.57093	4.57047	1.81334	1.27846	5.93069	6.74382	3.72204	5.24708	7.20795	3.18122	7.99041	2.68412
3.06698	5.90992	4.29877	2.09961	5.6955	4.46831	4.79549	3.65264	6.24576	4.13236	2.55422	4.02481
2.2993	3.78419	3.41033	4.45166	4.71103	4.81588	5.2757	3.39247	3.41693	7.81311	1.99811	2.60048
4.70183	7.02344	7.10909	4.48976	2.07788	2.50935	5.87265	3.56807	1.90465	5.25632	3.91995	3.373
7.9198	7.55695	12.8872	3.7221	2.89917	4.44471	3.30478	6.04731	4.76803	3.5982	7.56398	2.41343
4.73558	3.71363	4.57997	2.91514	3.68868	2.62299	3.03279	5.78033	4.50161	2.78308	5.81279	5.84262
2.84782	3.97156	4.22671	4.06752	3.29489	3.6022	5.71952	1.93462	6.13126	3.59969	5.36685	9.62027
5.04667	5.33545	2.97333	2.42254	9.29757	1.72393	4.79299	4.32513	3.96514	3.71642	8.24704	2.78678
3.12328	4.83225	2.55725	3.97991	6.65692	2.98971	2.85886	1.77674	3.8044	4.38038	5.36743	1.21162
4.39582	2.4101	3.39373	2.92821	4.09295	8.93147	6.65406	4.93811	6.00254	6.00285	4.02995	4.32552

2.59682	7.01881	3.72997	6.53748	4.47307	6.84769	7.35532	8.44904	4.94304	4.24446	6.51842	2.03618
4.15447	6.96811	5.35018	1.98432	2.01293	4.1661	3.77048	4.78588	3.22382	3.03677	3.26741	4.4132
5.10564	4.46694	5.07382	5.02042	5.36264	5.58969	3.28816	5.46471	4.71549	3.40473	3.57833	2.3283
3.27957	2.76658	3.57946	4.46639	3.2251	7.31183	4.55628	3.19244	9.00157	3.91919	4.99249	4.87479
9.9372	3.91831	3.7059	4.65735	4.15884	3.79101	6.45468	4.49994	6.52003	6.15253	5.17082	4.19382
2.9677	5.44759	5.7007	6.7055	3.23359	5.73414	5.50264	4.90393	2.96477	4.60814	2.00213	1.77586
3.40396	6.54114	5.8618	3.60939	2.43207	3.68608	2.77907	4.07706	5.29446	5.4943	4.91249	2.98061
2.34373	1.9154	5.02795	1.95465	2.50929	5.13876	3.59867	1.82174	3.62767	3.21315	3.19324	5.41431
6.41456	3.60003	6.41404	4.85085	5.04073	5.7843	2.49303	2.41227	3.46415	3.74409	4.31	4.37675
5.12598	2.98501	5.63614	1.90068	9.02602	4.78616	3.56955	2.11862	1.77725	5.46269	3.86353	6.16855
4.20726	4.31604	5.99258	7.41236	6.99267	6.30194	4.69689	8.45186	5.88355	4.56874	5.24466	3.5804
4.76114	5.75427	3.74314	7.17649	5.97995	2.30717	3.81064	2.5197	2.57646	6.59751	5.41436	4.20305
3.45439	6.5316	3.42431	4.89979	4.89083	4.60027	4.57634	3.47602	4.02718	6.28307	2.83069	1.96674
3.33931	5.72895	2.70042	5.34668	4.80022	4.79581	3.68573	5.846	1.89625	6.4395	5.71224	3.36249
4.89333	6.01627	5.54684	2.57323	5.01784	3.20766	4.1446	5.05019	7.13921	4.32667	9.7131	2.52202
5.29237	2.9587	4.74362	4.45195	4.52365	5.32305	6.46748	4.24643	6.51101	5.69608	6.77962	3.71372
6.57294	4.24094	5.21053	5.00176	2.15789	5.82496	2.12401	6.01282	3.18317	7.66663	4.243	3.68005
2.72225	3.50321	3.76823	2.66028	2.23191	4.51265	5.64584	8.42765	4.5303	3.34626	4.70369	7.1257
8.8137	2.02417	6.03245	2.39371	2.83619	5.26854	2.78802	3.04662	5.67565	3.85848	3.2571	3.98917
5.95198	6.6505	1.60285	4.49045	3.64536	4.29432	4.21963	4.30177	5.49169	2.80827	3.2402	3.65784
4.4642	2.64732	5.11799	2.77957	3.61131	7.00586	2.50109	3.20802	7.57383	4.01721	3.29431	4.10742
5.4879	6.77171	6.13431	5.40999	3.59442	5.72612	7.79442	1.64488	4.40303	7.16993	3.73254	3.90856
1.82615	4.93908	3.88449	5.89312	3.52779	5.18365	6.26217	2.55312	5.57904	4.66202	5.99619	4.52876
3.46687	4.64935	2.98718	6.43394	3.64168	6.67828	6.03493	3.88473	3.02524	3.20985	6.99602	4.81286
4.31329	2.51635	5.24868	2.18059	5.10228	1.81808	2.27674	4.71106	4.55527	1.22745	5.02113	5.5369
3.06914	2.51231	7.75555	6.15189	8.01919	5.86975	4.41559	5.95846	4.50413	6.78987	4.94402	2.88556

Table XXVI: Averaged waveforms used in the generation of the sponge water content test plot.

Average Dry	Average 1T	Average 2T	Average Dry (cont.)	Average 1T (cont.)	Average 2T (cont.)
6.377	4.164	5.486	7.187	3.297	3.608
4.616	4.081	4.176	4.094	5.504	5.229
3.522	3.903	5.331	2.984	4.470	3.107
4.967	3.354	4.365	2.041	4.423	4.605
3.597	4.324	3.662	5.259	3.580	5.954
3.795	5.082	5.020	6.435	4.720	5.833
3.396	4.967	4.115	5.582	4.862	4.911
3.320	4.453	4.093	4.620	4.486	4.969
3.822	4.161	4.883	6.403	4.666	5.693
4.665	4.193	4.787	5.038	5.546	4.109
6.530	4.725	4.983	3.700	5.479	5.663
3.997	5.005	4.579	4.844	5.662	3.754
7.036	4.651	3.921	3.196	4.545	5.458
6.750	5.164	5.643	2.554	5.466	5.212
4.833	5.327	5.110	4.103	4.986	4.677
3.413	3.988	4.804	3.882	4.934	4.874
4.976	4.944	4.374	6.207	3.487	3.576
4.246	3.677	4.629	8.055	3.550	4.805
3.236	7.100	4.191	3.736	3.115	4.355
5.281	4.476	4.107	4.089	4.206	3.889
3.116	5.923	6.258	3.577	5.271	4.002
4.930	5.151	4.198	3.790	4.168	3.321
4.224	5.609	3.823	2.911	6.559	5.648
2.494	4.648	5.407	5.762	6.225	5.879
5.524	4.099	3.433	4.768	3.317	3.682
5.583	7.318	4.109	4.854	4.747	4.528
5.490	3.987	4.261	3.604	5.031	5.371
6.940	6.414	6.289	4.094	4.802	5.724
6.862	4.966	6.087	5.951	4.823	4.159
5.268	4.427	3.250	5.337	2.966	4.955
4.538	7.276	4.526	2.966	3.749	2.888
4.720	5.027	4.031	4.955	4.439	3.207
2.993	4.942	4.897	3.507	5.794	3.120
4.555	4.044	6.722	5.907	5.997	6.301
5.772	3.652	6.535	5.558	4.033	3.898
5.372	4.828	4.543	4.952	4.689	4.595
2.956	6.548	4.334	4.592	4.427	4.727

3.657	4.557	5.743	4.712	4.123	5.505
2.116	3.520	4.702	4.051	5.438	5.485
4.872	4.075	5.811	4.818	3.369	5.621
6.228	4.088	6.224	3.311	4.130	5.435
5.653	3.222	5.264	3.483	3.631	4.194
5.417	5.692	4.190	4.248	4.053	4.201
4.602	4.223	3.984	3.515	4.373	4.933
5.082	5.797	6.959	6.105	5.705	4.406
6.114	6.882	6.335	4.906	4.991	4.265
5.470	5.449	3.475	4.690	5.452	3.373
7.523	4.442	3.004	3.315	3.066	3.498
5.340	4.055	4.960	5.473	6.102	5.751
4.033	3.647	4.892	4.823	4.778	4.831
6.140	3.453	5.871			
...	...	...			

Appendix K: Accessories to the Muscle Dehydration Test

Table XXVII: Summary table of the temperatures, weights, and averaged locational data from the muscle dehydration test.

		Sample					
		1	2	3	4	5	6
Set 1	Temp	70.5	70	71.2	70	70.3	70.3
	Weight	52	56	70	70	68	76
	Location 1	12.120	19.061	23.369	25.813	13.661	17.833
	Location 2	14.349	21.389	28.759	14.213	14.194	14.023
	Location 3	19.485	18.992	22.684	14.465	12.621	11.894
	Sample Average	15.318	19.814	24.937	18.163	13.492	14.583
Set 2	Temp	71.8	71.8	71.8	70.9	70.9	70.7
	Weight	50	52	66	66	66	74
	Location 1	20.797	13.847	12.998	13.918	14.730	14.604
	Location 2	18.769	14.677	14.111	14.347	14.987	14.225
	Location 3	15.651	12.708	15.223	13.341	15.241	15.335
	Sample Average	18.406	13.744	14.111	13.869	14.986	14.722
Set 3	Temp	71.7	71.9	71.4	71.3	71.2	71.2
	Weight	44	46	58	60	60	70
	Location 1	16.11	13.75	12.49	14.73	16.67	16.54
	Location 2	16.41	12.76	13.07	13.38	15.24	32.03
	Location 3	14.56	13.56	13.78	12.58	15.51	32.10
	Sample Average	15.6934	13.3564	13.1129	13.5664	15.8037	26.8894
Set 4	Temp	71.4	71.6	71.2	70.5	70.7	69.2
	Weight	38	42	52	50	48	58
	Location 1	4.48	4.35	4.60	4.53	8.37	9.06
	Location 2	4.48	4.76	4.99	4.69	8.28	8.50
	Location 3	4.82	4.61	4.20	4.64	8.75	8.78
	Sample Average	4.59	4.57	4.60	4.62	8.47	8.78

Figure 66: Paired t-test analysis for Set 2 and Set 4.