

Brigham Young University BYU ScholarsArchive

Theses and Dissertations

2019-06-01

# Frequency Response and Recovery of Muscles and Effects of Wrapping the Lower Leg on Surface Velocity Measurements

Cameron David Smallwood Brigham Young University

Follow this and additional works at: https://scholarsarchive.byu.edu/etd

#### BYU ScholarsArchive Citation

Smallwood, Cameron David, "Frequency Response and Recovery of Muscles and Effects of Wrapping the Lower Leg on Surface Velocity Measurements" (2019). *Theses and Dissertations*. 7503. https://scholarsarchive.byu.edu/etd/7503

This Thesis is brought to you for free and open access by BYU ScholarsArchive. It has been accepted for inclusion in Theses and Dissertations by an authorized administrator of BYU ScholarsArchive. For more information, please contact scholarsarchive@byu.edu, ellen\_amatangelo@byu.edu.

Frequency Response and Recovery of Muscles

and Effects of Wrapping the Lower Leg

on Surface Velocity Measurements

Cameron David Smallwood

A thesis submitted to the faculty of Brigham Young University in partial fulfillment of the requirements for the degree of

Master of Science

Jonathan D. Blotter, Chair J. Brent Feland Kent L. Gee

Department of Mechanical Engineering

Brigham Young University

Copyright © 2019 Cameron David Smallwood All Rights Reserved

#### ABSTRACT

## Frequency Response and Recovery of Muscles and Effects of Wrapping the Lower Leg on Surface Velocity Measurements

# Cameron David Smallwood Department of Mechanical Engineering, BYU Master of Science

This thesis is comprised of two studies. The objective of the first study was to find the frequency response and stiffness of the biceps brachii muscle group during recovery from exercise induced damage and to determine whether these data could be used to track muscle recovery by correlating changes in the frequency response with changes in muscle stiffness. Stiffness moduli were collected using Shear Wave Elastography (SWE) which were then applied to a proportional first mode frequency analysis. Data were collected for the muscle stiffness and frequency response for fifteen subjects ( $25.6 \pm 4.5$ ). By comparing the proportion of the square root of the SWE results, the variation in stiffness showed a less than 2 Hz change in first mode resonance for the control group. Frequency response results for the control group agreed with the modified SWE results and the proportion analysis. SWE results for the damage protocol group showed an average increase of 4 Hz. Frequency response results for the damage protocol group were sorted into three categories: three subjects had a change in frequency of peaks of at least 4 Hz in the positive direction; four subjects had an increase in amplitude, but no change in frequency of peaks; three subjects showed mixed responses like fewer resonance peaks, variable amplitudes, changes in peak bandwidth. This research allowed for the documentation of the in-vivo frequency response of the biceps brachii muscle. We believe that the frequency response of a muscle group may be used in the future to evaluate recovery from exercise induced damage. Lessons learned were also recorded for helping future studies in their efforts using an SLDV with human body testing.

The second study focused on finding the effects on the surface velocity of tissue above and below a region of the lower leg wrapped in an elastic band when excited by an external source. Ten male subjects between the ages of 18-25 were seated in a chair with one foot placed on a vibrating platform. Two excitation frequencies were separately applied while three points along the leg were measured. A repeatability analysis, using results without the leg wrap, showed a 6.5%, 2.5%, and 10.5% variance in the x-, y-, and z-directions respectively, applying a 20 Hz frequency. With a 40 Hz frequency, the variations were 24%, 23.8%, and 28.4% respectively. A change in displacement of +38% and +10% occurred above the knee in the x-direction with 40 Hz and in the y-direction with 20 Hz, respectively. A change in displacement of -20% occurred below the knee in the x-direction with 20 Hz. A change in displacement of -24% occurred below the wrap location in the y-direction with 40Hz. With a confidence interval of 93%, surface velocity of the tissue located above the wrap increased, while the surface velocity of the tissue below the wrap decreased.

Keywords: Frequency Response, Shear-wave Elastography, skeletal muscle, exercise-induced muscle damage, recovery, elastic modulus, stiffness

#### ACKNOWLEDGMENTS

I would like to give my deepest gratitude to my wife and children for supporting me in my efforts to complete this work. Their sacrifices made it possible for me to be successful. I would also like to thank my advisors, Dr Jonathan Blotter, Dr Brent Feland, and Dr Kent Gee, for their guidance and support in aiding me in the efforts of the performed research to find meaningful data that would contribute to their fields. I also want to thank my research partner, Garrett Jones, for his efforts to make our research possible as well.

	F TABLES	V
LIST O	F FIGURES	vi
Chapte	1 Introduction	1
1.1	Introduction: Frequency Response and Recovery of Muscles	1
1.2	Introduction: Effects of Leg Wrap on Acceleration	3
Chapte	Muscle Damage and Frequency Response of the Biceps Brachii	5
2.1	Experimental Setup and Pilot Studies	5
2.2	Measurement Location Validation	11
2.3	Skin Absorptivity and Reflection	13
2.4	Data Collection	15
2.5	Control Group Results	16
2.6	Damage Protocol Results	20
2.7	Discussion	31
2.8	Conclusion	33
2.9	Lessons Learned	35
Chapte	<b>Data Collection, Discussion, &amp; Results, Leg Wrap Study</b>	37
3.1	Overview	37
3.1 3.2	Overview	37 38
3.1 3.2 3.3	Overview	37 38 38
3.1 3.2 3.3 3.4	Overview	37 38 38 40
3.1 3.2 3.3 3.4	Overview	37 38 38 40 41
3.1 3.2 3.3 3.4	Overview	37 38 38 40 41 48
3.1 3.2 3.3 3.4	OverviewEquipmentMethodsMethods3.4.1Repeatability3.4.2Leg Wrap vs No Wrap3.4.3Direction of Change	<ul> <li>37</li> <li>38</li> <li>38</li> <li>40</li> <li>41</li> <li>48</li> <li>67</li> </ul>
<ul> <li>3.1</li> <li>3.2</li> <li>3.3</li> <li>3.4</li> </ul>	OverviewEquipmentMethodsMethods3.4.1Repeatability3.4.2Leg Wrap vs No Wrap3.4.3Direction of ChangeConclusion	<ol> <li>37</li> <li>38</li> <li>40</li> <li>41</li> <li>48</li> <li>67</li> <li>68</li> </ol>
3.1 3.2 3.3 3.4 3.5 <b>REFER</b>	OverviewEquipmentMethodsResults3.4.1Repeatability3.4.2Leg Wrap vs No Wrap3.4.3Direction of ChangeConclusion	<ul> <li>37</li> <li>38</li> <li>38</li> <li>40</li> <li>41</li> <li>48</li> <li>67</li> <li>68</li> <li>69</li> </ul>
3.1 3.2 3.3 3.4 3.5 <b>REFER</b> Append	Overview   Equipment   Methods   Methods   Results   3.4.1   Repeatability   3.4.2   Leg Wrap vs No Wrap   3.4.3   Direction of Change   Conclusion   ENCES  ix A Frequency Response of Biceps Brachii, Pilot Arm Position Evaluation Results	<ul> <li>37</li> <li>38</li> <li>38</li> <li>40</li> <li>41</li> <li>48</li> <li>67</li> <li>68</li> <li>69</li> <li>72</li> </ul>
3.1 3.2 3.3 3.4 3.5 <b>REFER</b> Append	Overview       Equipment         Equipment       Methods         Methods       Results         3.4.1       Repeatability         3.4.2       Leg Wrap vs No Wrap         3.4.3       Direction of Change         Conclusion       Conclusion         ENCES       Image: State of Biceps Brachii, Pilot Arm Position Evaluation Results         ix A       Frequency Response of Biceps Brachii         ix B       Frequency Response of Biceps Brachii	<ul> <li>37</li> <li>38</li> <li>38</li> <li>40</li> <li>41</li> <li>48</li> <li>67</li> <li>68</li> <li>69</li> <li>72</li> <li>79</li> </ul>
3.1 3.2 3.3 3.4 3.5 <b>REFER</b> Append B 1	Overview       Equipment         Equipment       Methods         Methods       Results         3.4.1       Repeatability         3.4.2       Leg Wrap vs No Wrap         3.4.3       Direction of Change         Conclusion       Conclusion         ix A       Frequency Response of Biceps Brachii, Pilot Arm Position Evaluation Results         ix B       Frequency Response of Biceps Brachii         Control Group Results       Control Group Results	<ul> <li>37</li> <li>38</li> <li>38</li> <li>40</li> <li>41</li> <li>48</li> <li>67</li> <li>68</li> <li>69</li> <li>72</li> <li>79</li> <li>79</li> </ul>

# LIST OF TABLES

2.1	Data Collection Schedule	15
2.2	Calculated Probability Analysis, Control Group	17
2.3	SWE Results, Control Group	17
2.4	Simply Supported Beam Model Results, Control Group	18
2.5	Elastography Results, Damage Protocol Group	21
2.6	Calculated Probability Analysis, Damage Group	22
2.7	Beam Model Average Results, Damage Protocol Group	22
3.1	Average Percent Variation, Repeatability Study	48
3.2	Paired T-test, Two-Tailed P-value Results	66

# LIST OF FIGURES

2.1	Velocity surface plot of the lower leg when relaxed.	6
2.2	Velocity surface plot of the lower leg when flexed	7
2.3	Frequency response comparison for the upper arm for the relaxed and flexed states	8
2.4	Vertical arm orientation for pilot study, with marked measurement locations	9
2.5	Extended arm orientation for pilot study and approximate point locations for all arm	
	evaluations	10
2.6	Bent arm orientation used for this study.	10
2.7	Pilot evaluation of measurement point 1 and subject 1 in the bent arm orientation	11
2.8	Pilot evaluation of measurement point 2 for subject 1 in the bent arm orientation.	12
2.9	Pilot evaluation of measurement point 3 for subject 1 in the bent arm orientation.	13
2.10	Ultrasound configuration used for gathering shear-wave elastography data from study	
	participants.	16
2.11	Ultrasound wand placement on subjects' arms	16
2.12	Average frequency response function at point 2 for the control group	19
2.13	Average frequency response of the damage protocol group for all measurement days.	20
2.14	Frequency response of subject 2 at point 2, for all measurement days	23
2.15	Frequency response function for subject 5 of damage protocol group at point 2	24
2.16	Pre-damage protocol frequency response for subject 10 at point 2	25
2.17	Post-damage protocol frequency response for subject 10 at point 2	25
2.18	Frequency response of subject 3 at point 2 for all measurement days.	26
2.19	Frequency response function for subject 6 of damage protocol group at point 2	27
2.20	Frequency response of subject 9 at point 2 for all measurement days.	28
2.21	Frequency response of subject 4 at point 2 for all measurement days.	29
2.22	Frequency response of subject 1 at point 2 for all measurement days	30
2.23	Frequency response of subject 8 at point 2 for all measurement days.	31
3.1	Direction of axes with respect to the body position of the subjects and measurement	
	point locations on the side of the leg, and reference point location at the top of the knee.	38
3.2	Modifications made to vibration platform to control location and movement of foot	
	during testing	39
3.3	Pilot configuration for testing surface response of the lower leg	40
3.4	20 Hz average displacement results for all scans with no leg wrap for each subject and	
	their standard deviation in the y-direction	42
3.5	40 Hz average displacement results for all scans with no leg wrap of each subject and	
	their standard deviation in the y-direction	43
3.6	20 Hz average displacement results for all scans with no leg wrap of each subject and	
	their standard deviation in the x-direction	44
3.7	40 Hz average displacement results for all scans with no leg wrap of each subject and	
	their standard deviation in the x-direction	45
3.8	20 Hz average displacement results for all scans with no leg wrap of each subject and	
	their standard deviation in the z-direction	46
3.9	40 Hz average displacement results for all scans with no leg wrap of each subject and	
	their standard deviation for point 1 in the z-direction	47

3.10	Average displacements at point 1 for the wrapped and no wrap results with a 20 Hz	40
	excitation frequency in the y-direction.	49
3.11	Average displacements at point 3 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the y-direction.	50
3.12	Average displacements at point 2 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the y-direction.	51
3.13	Average displacements at point 1 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the y-direction.	52
3.14	Average displacements at point 2 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the y-direction.	53
3.15	Average displacements at point 3 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the y-direction.	54
3.16	Average displacements at point 1 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the x-direction.	55
3.17	Average displacements at point 2 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the x-direction.	56
3.18	Average displacements at point 3 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the x-direction.	57
3.19	Average displacements at point 1 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the x-direction.	58
3.20	Average displacements at point 2 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the x-direction.	59
3.21	Average displacements at point 3 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the x-direction.	60
3.22	Average displacements at point 1 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the z-direction.	61
3.23	Average displacements at point 2 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the z-direction.	62
3.24	Average displacements at point 3 for the wrapped and no wrap results with a 20 Hz	
	excitation frequency in the z-direction.	63
3.25	Average displacements at point 1 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the z-direction.	64
3.26	Average displacements at point 2 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the z-direction.	65
3.27	Average displacements at point 3 for the wrapped and no wrap results with a 40 Hz	
	excitation frequency in the z-direction.	66
A.1	Pilot evaluation of measurement point 1, in the extended arm position for subject 1	72
A.2	Pilot evaluation of measurement point 2 in the extended arm position for subject 1	73
A.3	Pilot evaluation of measurement point 3 in the extended arm position for subject 1	74
A.4	Pilot evaluation of measurement point 1 in the extended arm position for subject 2	75
A.5	Pilot evaluation of measurement point 2 in the bent arm position for subject 2	76
A.6	Pilot evaluation of measurement point 2 in the extended arm position for subject 2	76
A.7	Pilot evaluation of measurement point and arm position for subject 2 point 3	77
A.8	Pilot evaluation of measurement point and arm position for subject 2 point 3	78

<b>B</b> .1	Average frequency response function over all subjects that participated in the control	
	group for measurement point 2	79
<b>B</b> .2	Average frequency response function over all subjects that participated in the control	
	group for measurement point 3	80
<b>B</b> .3	Frequency response of subject 7 at point 2 for all measurement days	81

#### CHAPTER 1. INTRODUCTION

This thesis consists of two focused efforts. The first was to determine if the frequency response could be used to measure the existence of damage, and recovery of a muscle group that had been damaged due to exercise. The second was to determine if wrapping the leg to mount an accelerometer, as is common practice, impacts the dynamic response of the lower leg. These two efforts are introduced in this chapter.

#### 1.1 Introduction: Frequency Response and Recovery of Muscles

The ability to measure quantifiable in-vivo properties of muscle groups has historically been limited to taking biological samples and using magnetic resonance imaging [1, 2]. However, in the last twenty years, advances have been made in measurement techniques and technology, which has allowed for the development and validation of measuring properties of biological material using ultrasound techniques. To develop this, shear wave elastography (SWE) stiffness measurements were first correlated with electromyography muscle activation measurements [3]. Recent studies that used SWE have included studying changes in stiffness of muscle groups during long-term stresses [4], and during delayed onset muscle soreness (DOMS) [5].

SWE uses a high energy ultrasonic pulse to induce a travelling shear wave through the target tissue. This travelling shear wave introduces deformations proportional to the stiffness of the material. By utilizing material properties, and assuming small deformations, the ability to measure properties of tissues in-situ is possible, and has been validated through various studies [6].

Electromyography is a widely accepted and used method of gathering data on muscle activity, and has been used as a primary benchmark for emerging measurement technologies. When SWE was introduced as a method of measuring muscle activation indirectly, it was evaluated against electromyography [3]. After being shown to be comparable to electromyography, methods of measuring properties of a muscle group, such as elasticity, were developed [7]. The primary use of electromyography is to determine muscle activation by measuring the voltage potential generated by a muscle group. However, in 1972, electromyography was used to find the frequency response of a muscle group in-vivo in a cat [8]. In 1979, electromyography was also used to find the frequency response of a muscle group in a human subject in vivo for the frequency range of 1 to 15 Hz [9–11].

This study will focus on using SWE stiffness measurements of the biceps brachii during a 1-week recovery period to create a baseline with which to compare the frequency response of a muscle group. As of yet, a direct comparison between SWE stiffness measurements and the frequency response of a muscle group has not been reported.

Laser Doppler vibrometry (LDV) has been used for various biomedical applications to collect non-intrusive measurements of surface velocities from which accelerations and displacements can be computed [12]. LDV has been used to measure blood flow in superficial arteries as well [13]. In 2013, scanning laser Doppler vibrometer (SLDV) results were compared to mechano-myography, being correlated with force output and muscle activation values [14]. Similarly, in 2013, SLDV results were compared against electromyography results in the measurement of muscle activation [15]. The use of an SLDV has thus far been limited to measuring mechanical frequency response of muscle groups due to natural movement and muscle activity. An effort to measure frequency response during muscle recovery with an SLDV has not been reported.

The human body provides several indirect markers that indicate when muscle damage occurs, including plasma levels of muscle enzymes, muscle strength performance measurements through maximum voluntary isometric contraction (MVIC), local swelling and inflammation markers, and (more recently) magnetic resonance imaging [1]. Recent efforts to identify exerciseinduced damage in muscles was by metabolically profiling urine to identify metabolites that are expelled by the body after damage has occurred [16]. An inexpensive quantitative non-invasive method for measuring muscle damage has not been developed. Tracking recovery in a muscle group is handled in much the same way as damage is, requiring either expensive imaging equipment, invasive biopsies, or collecting body fluid. An indirect non-invasive quantitative method for measuring recovery was one objective of this project.

Results from an SLDV have been correlated with the dynamic frequency response collected through mechano-myography, and separately correlated with the electrical frequency response col-

lected through electromyography. However, the mechanical frequency response of muscle groups has not been documented above 15 Hz. This study documented the measured mechanical frequency response of a muscle group due to an external excitation for the frequency range, 1 Hz to 100 Hz.

Previous studies have successfully correlated both SWE and SLDV measurements with electromyography, in measuring muscle activation during muscle loading [3, 15]. However, the correlation between SLDV and SWE results has not yet been reported. The proposed research attempted to perform this correlation across changes caused by exercise damage in a muscle group. The proposed work accomplished the following objectives:

- Design and build a test fixture to aid in collecting consistent frequency response data using an SLDV system.
- 2. Collect SWE data for a muscle group before, and then at various periods after a damage protocol.
- 3. Collect frequency response data for a muscle group using the SLDV system before, immediately after, and then at various periods after a damage protocol.
- 4. Analyze the data from objectives 2 and 3 to determine any correlation.
- 5. Compare results of the previous four objectives from a control group with a group that underwent a damage protocol.

The muscle group that was chosen for evaluation under this investigation is the Biceps Brachii of the right arm. This muscle group was chosen for the ease of causing exercise-induced damage, as well as for the fact that this muscle group is accessible for the measurements taken in this study.

#### **1.2 Introduction: Effects of Leg Wrap on Acceleration**

The accelerometer has generally been accepted as an inexpensive and reliable method to capture motion and activity data in human subjects. Several commonly used accelerometers used in clinical studies were evaluated and presented by Godfrey et al. [17]. The list of commonly used

accelerometers used for clinical studies was then updated by Pedisic et al. [18]. Research has been performed using accelerometers to capture data on human reactions to whole body vibrations to understand more clearly the reaction of the body at various frequencies and postural positions [19]. These findings have aided in understanding and creating models for use in implementing safety parameters in commercial atmospheres. One such research article explored the transmission of energy to the head of a seated individual, a serious concern for employees using heavy equipment [20]. Recent studies have explored the nature of how these acceleration data were analyzed, and how the analyses affected the outcomes and understanding of the results [21]. Throughout these studies, data were collected by using wearable accelerometers, as opposed to accelerometers attached to the body with adhesives. Wearable accelerometers are commonly attached to the body by way of wrapping the body part with an elastic band with an accelerometer either attached to the band, or wrapped inside the band and around the limb under investigation [17, 18]. Research performed for this body of work evaluated whether modifying the lower leg by using an external wrap appreciably affects the vibration response.

## CHAPTER 2. MUSCLE DAMAGE AND FREQUENCY RESPONSE OF THE BICEPS BRACHII

#### 2.1 Experimental Setup and Pilot Studies

For this study, an SLDV was used to measure the surface velocity of the biceps muscle to find the dynamic frequency response over the course of a 1-week recovery period. A GE Logiq S8 with a 9L head was used to collect shear wave elastography (SWE) data. These data were collected to try and correlate the stiffness of the muscle with the frequency response collected with a scanning laser doppler vibrometer (SLDV) during the recovery period. Data collected through SWE returns the stiffness modulus of the muscle group, in kiloPascals (kPa). All of these values were converted to an elastic modulus for use in the analysis.

Other data that were collected for this study included: maximum voluntary isometric contraction (MVIC), used to determine the resistance required for the damage protocol; and bio-metric data including height, weight, and gender.

Subjects that volunteered for the study were required to be 18 - 40 years of age, and to have not performed resistance exercise using their upper body during the past three months. Subjects were instructed not to change their routine during the data collection process.

Prior to testing and data collection, an explanation of the objective and purpose of the study was given to the subjects. They were informed of any potential risks that were involved with the study prior to participation. All subjects that participated signed an Institutional Review Board (IRB) approved consent form. Approval for this study was granted by the IRB at Brigham Young University.

Prior to beginning this project, it was unknown whether SLDV surface velocity measurements would indicate a change in the stiffness of a muscle group. Several evaluations were performed on different muscle groups to determine whether stiffness could be detected by a surface measurement of the skin. The first validation was evaluated by measuring the surface response of the lower leg. A subject placed their foot on a test stand attached to a shaker. The heel of the foot was placed against a small bracket to prevent lateral movement of the foot. The side of the foot was marked on the platform surface to aid in preventing movement of the foot during data collection. The foot was then excited using a pseudo-random signal. Figure 2.1 shows the surface velocity plot of the lower leg while the subject was seated with the muscles of the leg relaxed. Figure 2.2 shows the same evaluation, but the subject flexed the gastrocnemius muscle group. The differences in these plots of magnitude and distribution indicate that changes in sub-cutaneous stiffness can be measured at the surface. The vibration is concentrated in the lower portion of the leg when the muscle group is flexed, while the surface velocity plot of the relaxed leg shows the energy is more distributed. These observations showed that a change in tension of the substrate may be identified by measuring changes in response at the skin's surface.



Figure 2.1: Velocity surface plot of the lower leg when relaxed.



Figure 2.2: Velocity surface plot of the lower leg when flexed.

A follow-up analysis was also performed on the upper arm. Velocities were found to be lower for the relaxed arm, when compared to the flexed arm.

Figure 2.3 shows the frequency response of the upper arm when the arm is relaxed, and when it is flexed. The relaxed arm results show three major peaks located near 5 Hz, 7 Hz, and 10 Hz. When the muscle group is flexed, the frequency response changes. Figure 2.3 shows that the bandwidth of all the identified peaks narrowed, and a distinct peak was also identified near 13 Hz. Based on the evaluations performed in this pilot study, stiffness changes in subcutaneous tissues can be identified by a surface velocity measurement.



Figure 2.3: Frequency response comparison for the upper arm for the relaxed and flexed states

Other points of consideration for this project were: the arm position that was chosen during data collection, and the reflectivity of the subjects' skin.

Three different arm positions were evaluated for repeatability and for the subject's body position and the potential difficulty to hold still for several minutes during data collection. This was required because the amount of time required to complete a single scan varied during the pilot activities and determining how much resolution would be necessary to capture the frequency response of the muscle group accurately. Figure 2.4 shows the first arm position that was evaluated for this project. Figure 2.4 also shows the location of measurement points used for all data acquisition activities for this project. Points on the arm were identified and located as follows: Point 2 is located near the center of the biceps muscle. Point 1 is located approximately 2 inches towards the elbow, making sure to be located on the main mass of the biceps muscle. Point 3 is located approximately 2 inches toward the shoulder joint, making sure to be located on the biceps muscle.



Figure 2.4: Vertical arm orientation for pilot study, with marked measurement locations.

The vertical arm orientation that was initially investigated was not ideal for two reasons: first, it was difficult for the subject to hold still enough to take reliable measurements. Second, it was very difficult for the subjects to control how much load bearing was on the arm. This caused residual tension in the arm that was challenging to regulate. Part of why this setup was difficult for subjects to be consistent, was that it was difficult to regulate the location of the platform and the chair to allow for subjects to rest in a seated position with their upper arm in a vertical position on the platform. The ergonomics of this setup is different for everyone, making this arrangement not feasible for controlling body position and repeatability.

Figure 2.5 shows an alternate arm position evaluated as a potential orientation for data acquisition, and Figure 2.6 shows another arm orientation evaluated. These alternative orientations eliminated the issues that were seen with the vertical arm position, and limited the required adjustments to changing the vertical position of the chair, ensuring that the upper arm was at a parallel with the floor. The arm orientation that was used for this study, as seen in Figure 2.6, was chosen because the muscle under investigation was at its most neutral position, and therefore would be less likely to be affected by involuntary muscle activation during testing. A test stand was constructed to allow for free translation in the direction facing the SLDV. The chair on which the subjects sat was adjustable for height, allowing the subject to sit with arm raised laterally at a 90° angle relative to the body.



Figure 2.5: Extended arm orientation for pilot study and approximate point locations for all arm evaluations



Figure 2.6: Bent arm orientation used for this study.

#### 2.2 Measurement Location Validation

Figure 2.5 shows the approximate locations of the measurement points along the axis of the biceps brachii muscle group as described previously. These locations were measured for all pilot and final data collection activities.

A pilot study was conducted to determine which, of the planned points to be measured, accommodated for the most repeatable data. Three subjects were assessed in the two arm configurations shown in Figures 2.5 and 2.6. The scan points in each configuration were measured three times. Subjects were instructed not to move between scans, and each scan was taken immediately after the previous scan. This procedure allowed for an evaluation on how much variation can be expected over a very short amount of time.

Figure 2.7 shows the results of the repeatability study for subject 1 and point 1 in the bent arm orientation. Results indicated varying amplitude in the velocity, but showed peaks near 26 Hz, 34 Hz, and 83 Hz for all three scans. Below 20 Hz, a greater degree of variation between the three scans was observed (Extended arm orientation results can be found in Appendix A).



Figure 2.7: Pilot evaluation of measurement point 1 and subject 1 in the bent arm orientation.

Figure 2.8 shows results from point 2 in the bent arm orientation. Results showed some variation in amplitude below 15 Hz, however, a peak near 4 Hz was identified on two of the three scans. Above about 15 Hz, the three scans were nearly identical in both peak frequency locations, and amplitude. Additionally, peaks were identified near 25 Hz, 49 Hz, and 70 Hz.



Figure 2.8: Pilot evaluation of measurement point 2 for subject 1 in the bent arm orientation.

Figure 2.9 shows results for subject 1 at point 3 in the bent arm orientation. Results showed variation in amplitude, but consistency in peak frequency above 20 Hz. Below 20 Hz, variations in peak frequencies and amplitudes were again identified. Peaks were identified near 24 Hz and 84 Hz, the 84 Hz peak frequency being the highest resonance identified for the pilot testing, and similar to a resonance peak for the point 1 results shown previously. Some variation between all three points in peak frequencies was noted for these analyses. One explanation for some of the variation was that the volume of tissue being measured at each point was different from point to point. Interactions between tendons and other muscle groups with the muscle group being evaluated may have also caused change in the frequency response between points.



Figure 2.9: Pilot evaluation of measurement point 3 for subject 1 in the bent arm orientation.

Based on the results from all the pilot subjects, it was found that points 1 and 2 were similar in their consistency from one scan to the next. It was decided that a focus on point 2 would provide the simplest measurement situation because it was farthest from connective tissues and other muscle groups that may affect data clarity. Results for the other pilot subjects and points can be found in Appendix A. It was also decided to use the bent arm configuration during testing because we were concerned for subjects' ability to hold still during data collection especially after muscle damage was induced. By keeping the arm in a bent position, we assumed that it would be easier for the subject to keep the arm relaxed during testing.

#### 2.3 Skin Absorptivity and Reflection

A challenge we encountered during pilot activities and data collection efforts was the quality of the signal received by the SLDV. Reflectivity of light off the skin is a function of both the absorptivity of the skin, and the size of the particle that is causing scattering. Absorptivity of the dermal layer is dependent on both the wavelength of the light, and in large part, the hemoglobin and melanin content in the skin. Hemoglobin is a protein made up of several polypeptide chains. These proteins are bonded to a heme (Hb A) and are responsible for absorption of the majority of light in blood [22]. According to Lister et al., Hb A appears to have three zones of excitation due to light: a blue light region, a green light region, and a red light region.

Scattering is highly dependent on size of particles and can be categorized into two separate mathematical models: Rayleigh scattering, and Mie scattering [23]. Rayleigh scattering is highly dependent on wavelength and describes the nature of scattering off smaller particle sizes. Mie scattering is attributed to scattering against larger particles and is not wavelength-dependent.

Generally, it is believed that between 4 and 7% of visible light is reflected from the skin surface, independent of skin color or wavelength [22]. Little information regarding absorption and reflection behavior of skin is understood above a wavelength of 750 nm. However, as the SLDV is a narrow-wavelength dependent measuring device whose wavelength is 1,550 nm, it is imperative to maximize the reflective nature of the surface being measured.

Several commercially available skin coatings were used to enhance scattering of the laser beam to improve the signal received by the SLDV. The first product that was used was Albedo 100<sup>TM</sup>, which offers a pet-safe spray to use on animals. The Aerosol spray uses a transparent glue as a base and reflective microspheres as the interactive material. While this material worked well in improving the signal received by the SLDV, it was found that the aerosol did not provide enough product for a large number of applications. The aerosol can would run out of spray relatively quickly. The second product evaluated for this project was SafetySkin®, a direct rub-on skin application product. This product is wax based and contains micro-reflectors as well. For ease of application, number of applications, and quality of scattering, the SafetySkin®product was used for this project. It was found that when these products were used, the quality of reflected laser light that returned to the SLDV system was improved in some cases as much as 50% as observed by the laser light return indicators supplied on the laser head units and the software.

#### 2.4 Data Collection

Data were collected from fifteen human subjects. Five subjects were randomly assigned to a control group, while ten subjects were designated to the experimental group. The control group participated in the prescribed data measurements and did not undergo a damage protocol.

The control group was tested by collecting SWE and evaluating the biceps muscle's frequency response on three consecutive days. Members of the control group were instructed not to deviate from their regular daily activities during testing.

The experimental group participated in a 7-day test schedule as outlined in Table 2.1. On the day labeled PRE, measurements were taken prior to the damage protocol being performed on each subject to establish a baseline. Following pre-testing, the experimental group underwent a muscle damage protocol. The damage protocol consisted of determining the maximum voluntary isometric contraction (MVIC) for the right arm, then using 50% of the maximum value to perform 10 sets of 10 repetitions of a dumbbell curl. A rest period of 1 minute was used between each set. If the subject struggled for more than two sets to complete the required ten repetitions per set, the weight was decreased by 5 pounds, and the damage protocol was resumed. Measurements of SWE, frequency response, and VAS were then assessed at 24 hours, 48 hours, and 1 week after the damage protocol was performed.

Day Number	MVIC	Elastography	Frequency Response	VAS
PRE	X	X	X	X
1		X	X	X
2		X	X	X
6		X	X	X

Table 2.1: Measurements taken for each day of participation as designated by the labels listed in column 1. Biometric data included recording the maximum isometric force for each subject, gender, and age data.

SWE measurements were collected by having subjects lie flat on a table, with their arm extended at their side as shown in Figure 2.10. The data were taken in the distal 1/3 of the muscle group. Figure 2.11 shows the manner that the ultrasound head was placed on on the subjects' arms

so that the probe was aligned parallel with the longitudinal axis of the biceps brachii. Marks were made on the arm using a permanent marker to outline the exact placement of the probe to assure that follow-up measurements were made in the same location. Data calculated from a region of interest on the ultrasound image were expressed in kilopascals, representing tissue shear stiffness.



Figure 2.10: Ultrasound configuration used for gathering shear-wave elastography data from study participants.



Figure 2.11: Ultrasound wand placement on subjects' arms

## 2.5 Control Group Results

SWE measurements for control group subjects were evaluated using a paired t-test probability analysis to determine whether the variation between the values were due to chance, or were caused by damage in the muscle group. The paired t-test analysis provides a probability value that communicates the confidence interval in which results are considered statistically significant and considered valid. For this study, data were considered statistically significant if the p-value was less than 0.07, indicating a confidence interval of 93% that results are statistically significant. Table 2.2 shows the results of the paired t-test analysis. The p-values were greater than 0.07, indicating that the variation observed between the different days is not statistically significant and is likely due to random chance. Day 6 data was collected for only a small number of control group participants due to scheduling problems. It was decided to omit day 6 data from the control group analysis. A table of stiffness results for the control group can be found in Table 2.3.

Table 2.2: Calculated probability analysis ofSWE results for control group.

	DAY 2	DAY 3
<b>P-Value</b>	0.359311	0.995788

Table 2.3: Measured muscle stiffness, in kilopascals, for the control group.

SUBJECT	DAY 1	DAY 2	DAY 3
1	34.06	37.2	36.56
2	44.24	38.82	42.24
3	48.24	48.23	50.31
4	48.14	40.84	49.23
5	44.6	38.35	40.84

As described previously, results from SWE analysis are expressed as an average shear stiffness of the muscle group. An evaluation of the relationship between the shear modulus and the elastic modulus indicate that the relationship between the two is linear, and in the case of muscle tissue, the relationship is

$$3\mu = E \tag{2.1}$$

where  $\mu$  is the stiffness modulus and E is the elastic modulus. This relationship is true, when the poisson's ratio of muscle tissue is rounded to 0.5. This is generally acceptable, as the average poisson's ratio was found to be about 0.47 [6] for muscle tissue. Based on this analysis, the SWE results were used to evaluate the expected change in first mode resonance of a material based on the first mode resonance model of a beam.

Using the measured muscle stiffness values found in Table 2.3, it was desired to understand how these results would affect the frequency response. In a beam, the first mode resonance frequency is proportional to the square root of the Young's Modulus of the material [24]. By understanding the relative shift in resonance, it is possible to determine what effects may be expected from the muscle group after damage occurs. Values for Young's modulus in the proportional analysis were substituted for the stiffness values from SWE measurements from each subject.

The application of control group results to the proportional analysis described previously are shown in Table 2.4. This evaluation shows a variation of 0 to 1.5 Hz between days for all subjects. These results informed the opinion that if the same analysis yields greater than a 2 Hz change, then damage is present in the muscle group.

	Day 2 $\Delta$	Day $3\Delta$	Std Dev
Subject 1	0.71	0.57	0.375
Subject 2	-1.13	-0.41	0.574
Subject 3	-0.002	0.39	0.230
Subject 4	-1.48	0.21	0.918
Subject 5	-1.31	-0.78	0.658

Table 2.4: First mode frequency deviations, in Hz, of an elastic proportion analysis with stiffness results of the control group.

Figure 2.12 shows the averaged frequency response across all subjects who participated in the control group for measurement point 2. The in-vivo frequency response of the biceps brachii

muscle group has not been previously reported, and is a contribution that is provided by this research. Frequency response results show that resonance peaks varied little between the three consecutive measurement days. Based on the average results for the control group, the peak near 11 Hz on day 1 is related to the peak near 12 Hz on day 2, and 13 Hz on day 3. Similarly, a peak was identified near 8 Hz on day 1, near 9 Hz on day 2, and near 10 Hz on day 3. These results suggest an average variation of 2-3 Hz between days. The average frequency response at point 2 for the control group provided a first look into the frequency response of the biceps brachii for this research. It was found that there were three primary resonance peaks that continued to be present for all of the subjects tested. The first resonance that was identified that was present in all of the subjects was a resonance peak between 10 and 20 Hz. The second was one that was present between 20 and 40 Hz, while the final resonance peak that was identified multiple times was a peak between 60 and 80 Hz. These resonance peaks are found in this average frequency response plot as well.



Figure 2.12: Average frequency response function at point 2 for the control group

#### 2.6 Damage Protocol Results

An analysis of the results gathered from the damage protocol group follows. The analysis was initially performed using average results from the group in its entirety, however, it was found that the average values for the frequency response analysis showed little to no changes in resonance frequency. While this was not ideal in determining what effects may be identified for muscle damage, it did provide another opportunity to find the average frequency response of the biceps brachii muscle group.

Figure 2.13 shows the average frequency response plot for the duration of the data collection cycle. An analysis of the frequency response shows that there are four resonance peaks between 0 Hz and 100 Hz. These peaks are located near 17 Hz, near 30Hz,near 35 Hz, and near 66 Hz. While these values vary from subject to subject, trends showed that generally speaking, the first resonance frequency was located between 10 and 20 Hz, and the second resonance frequency was located between 20 and 40 Hz. Multiple smaller resonances were also present above those, with a higher resonance peak visible between 60 and 80 Hz.



Figure 2.13: Average frequency response of the damage protocol group for all measurement days.

Stiffness results for the damage protocol group are shown in Table 2.5. The average standard deviation calculated between all subjects in the damage protocol group was 14.23 kPa. The average standard deviation of the stiffness results for the control group was 2.79 kPa. Because of the large disparity between measurement results at the points, this was the first indication that muscle damage occurred.

SUBJECT	PRE	<b>DAY 1</b>	<b>DAY 2</b>	DAY 6
1	73.85	114.35	118.85	75.22
2	44.69	73.81	50.66	52.63
3	41.21	81.28	53.48	63.12
4	33.65	55.45	56.81	37.94
5	50.19	79.18	80.78	79.27
6	61.04	72.75	78.55	67.37
7	51.03	94.38	98.64	61.00
8	67.43	89.11	92.06	73.10
9	67.75	75.78	88.34	69.71
10	71.67	83.35	93.9	85.01

Table 2.5: Raw data of SWE results for all subjects in damage protocol group, in kPa.

Table 2.6 shows the calculated probability evaluation performed on stiffness results of the damage protocol group. The p-values for day 1 and day 2 were well below the 0.05 threshold for the calculated probability. This showed that the variation between the stiffness of the muscle group was not due to random chance, but rather was likely due to the damage present in the muscle group. The day 6 p-value was slightly higher than the 0.05 threshold. This may indicate that the damaged muscle group had begun to recover because of the likelihood of variation being due to chance.

	Day1	Day2	Day6
<b>P-Value</b>	0.000345	0.003214	0.083876

Table 2.6: Calculated probability analysis for SWE results of damage protocol group.

Stiffness results for the damage protocol group were compiled into a set of average values. The beam model proportional analysis was applied to the average values in order to determine whether the average change in stiffness measured for the subjects would yield an appreciable difference in the first mode frequency. Based on the results found in Table 2.7, the average frequency shift for the first mode frequency was greater than 4 Hz. From the control group evaluations, it was determined that variation between days that causes greater than a 2-3 Hz shift was requisite for identifying muscle damage.

Table 2.7: First mode frequency shift based on pre-damage results for an elastic modulus proportion beam analysis for the damage protocol group.

Subject	Day 1	Day 2	Dov 6
Identifier	Day I	Day 2	Day 0
1	5.655651	6.216889	0.213702
2	5.134118	1.164946	1.534123
3	6.992058	2.406514	4.108196
4	4.432221	4.676684	0.966062
5	4.885272	5.126206	4.898889
6	1.929926	2.828113	1.064179
7	6.925715	7.509716	1.795716
8	3.308065	3.725483	0.911099
9	1.277006	3.145562	0.318389
10	1.787872	3.29771	2.031525
Average	4.23279	4.009782	1.784188

Figure 2.14 shows the frequency response at point 2 for subject 2 for all measured responses. The pre-damage frequency response curve shows peaks near 14 Hz, 33 Hz, and 47 Hz. A smaller peak was also found near 67 Hz. After the damage protocol, and for the following consecutive measurements, a frequency peak was identified near 36 Hz, with a change in amplitude. The amplitude of the peak near 14 Hz decreased by almost the same factor as the increase in amplitude at the peak near 36 Hz. A peak was also identified near 44 Hz, with a slight decrease in amplitude from the peak near 47 Hz on the Pre-damage response. A change in frequency for the resonance peak from 33 Hz to 36 Hz correlates well with the proportional analysis performed on the SWE results, where the expected change in frequency was aout 4 Hz.



Figure 2.14: Frequency response of subject 2 at point 2, for all measurement days

A similar response as what was observed for subject 2 was also observed for subject 5. The amplitude of the peak near 15 Hz for all scans decreased from pre-damage levels, but the peak frequency did not change between days. Prior to the damage protocol, a peak was identified near 34 Hz. After the damage protocol, a resonance peak was located near 38 Hz with a wider bandwidth than what was observed from the pre-damage peak near 34 Hz. This change in resonance

peak location agrees well with the proportional analysis and SWE results, showing a change in resonance frequency of 4 Hz. It is interesting to note, however, that these changes in frequency are not occurring on the lowest resonance peak, but an intermediate peak, located between 20 and 40 Hz. Figure 2.15 shows results collected for subject 5.



Figure 2.15: Frequency response function for subject 5 of damage protocol group at point 2

Subject 10 experienced a similar trend as subjects 2 and 5, but at a lower frequency. Figure 2.16 shows the pre-damage frequency response for subject 10. The results for the pre-damage frequency response showed a substantially lower amplitude for the entirety of the plot when compared to the post-damage results. Figure 2.17 shows the post-damage frequency response for subject 10. Results for subject 10 show that pre-damage frequency response amplitudes are several magnitudes smaller than post-damage frequency response amplitudes. Nevertheless, resonance peak frequencies were collected from the pre-damage response plot for subject 10. A resonance peak was identified near 7 Hz. Post-damage frequency response results show a resonance peak near 12 Hz. The frequency response shift in these peaks correlate well with the muscle stiffness analysis,

again because the expected change in frequency was 4 Hz, and the change was identified at a lower resonance peak than the other subjects placed in this category.



Figure 2.16: Pre-damage protocol frequency response for subject 10 at point 2



Figure 2.17: Post-damage protocol frequency response for subject 10 at point 2

Although some subjects indicated a change in peak frequencies, others saw a change in amplitude while peak frequencies remained unchanged. An example of this can be seen in Figure 2.18, where the frequency of the peaks for subject 3 didn't change, but the amplitude of the peaks changed. Resonance peaks were identified near 17 Hz and 30 Hz for all results, but the amplitude initially decreased after the first post-damage measurement. Amplitudes continued to vary for the duration of the data collection process for the lower frequency peak. The amplitude consistently increased for the higher frequency peak near 30 Hz for all scans of subject 3 at point 2.



Figure 2.18: Frequency response of subject 3 at point 2 for all measurement days.

Figure 2.19 shows frequency response results for subject 6 at point 2. In the pre-damage results, three primary resonance peaks were identified: near 14 Hz, 26 Hz, and 41 Hz. In the post-damage results, three peaks were also identified for each of the scans. For all three post-damage results, a peak was identified near 14 Hz, however, the amplitude varied from scan to scan. This was the same case for the peak identified near 24 Hz for all post-damage results as well. The amplitude varied, but the peak frequency did not. The third peak identified in the post-damage results was different for each scan. For Day 1, the peak was near 32 Hz, Day 2 results showed the

peak near 33 Hz, and Day 6 results indicated a peak near 34 Hz. These results suggested that for lower frequency peaks, damage may be identified through a change in amplitude of the resonance peaks.



Figure 2.19: Frequency response function for subject 6 of damage protocol group at point 2

Figure 2.20 shows the frequency response results for subject 9 at point 2. Resonance peaks were identified and located near 15 Hz for all scans, and smaller peaks near 22 Hz, 29 Hz, and 35 Hz. For the peak near 15 Hz, the amplitude varied between all of the scans, with the highest amplitude on the Day 1 post-damage results. The amplitude for this peak then decreased for each subsequent scan result. Variation in peak location for the other identified peaks varied for the Day 1 and Day 2 results. Day 6 results indicated the same pre-damage peak frequencies, with elevated amplitudes. These results indicated that for higher frequency peaks, some variation in both frequency and amplitude may occur due to damage. For the low frequency peak, the amplitude increased after the damage protocol, and indicated a slow decrease from the maximum amplitude over the course of the recovery period.


Figure 2.20: Frequency response of subject 9 at point 2 for all measurement days.

Frequency responses for subject 1, subject 4, and subject 8 showed mixed results, as demonstrated by results for subject 4 at point 2 in Figure 2.21. In the pre-damage frequency response, three peaks were identified: the first peak was identified near 12 Hz, the second near 25 Hz, and the third peak near 40 Hz. The post-damage frequency responses showed peaks near 15 Hz, 23 Hz, and 35 Hz. These results indicated both a potential increase in resonance frequency at the low frequency peak, and a decrease in resonance frequency at the high frequency peak. These results also showed the amplitude of the lower frequency peaks increased, then gradually decreased by the time Day 6 data collection had occurred. The higher frequency peak had a decrease in amplitude from the pre-damage results. Subject 4 was categorized as difficult to sort, because the frequency peaks. Variation in the frequency response above 50 Hz also was indicated by the post-damage results. Day 6 results showed an overall decrease in amplitude for the frequency response below 50 Hz, when comparing against Day 1 and Day 2 post-damage results.



Figure 2.21: Frequency response of subject 4 at point 2 for all measurement days.

The frequency response of subject 1 at point 2 is shown in Figure 2.22. Before the damage protocol, it appeared that there were two clear frequency peaks, one near 10 Hz, one near 16 Hz. Several smaller peaks were also indicated between 30 and 45 Hz. After the damage protocol, it appeared that the frequency response was consolidated into two peaks, one near 17 Hz, and a wider peak near 32 Hz. This indicated that the first resonance of the muscle group was somehow eliminated from the frequency response of the muscle group altogether, or that perhaps the first resonance peak had its energy shifted enough to have combined with the second resonance peak. For Day 2 post-damage results, it appeared that the higher frequency peak had continued to shift, as a small resonance peak was visible near 37 Hz, but the amplitude was significantly decreased. The resonance peaks may have been beginning to return to pre-damage locations and amplitudes by day 6, as indicated by the three resonance peaks identified: one near 16 Hz, one near 25 Hz, and one near 35 Hz. When comparing the change in amplitude, it appears that the amplitude of the peaks on day 6 were also decreasing from day 2 and day 3 levels as well.



Figure 2.22: Frequency response of subject 1 at point 2 for all measurement days

Figure 2.23 shows the frequency response comparison for subject 8. Pre-damage results indicated three resonance peaks; one near 14 Hz, one near 26 Hz, and one near 48 Hz. Two smaller peaks were identified near33 Hz and 72 Hz as well. Day 1 results indicated three peaks: one near 16 Hz, one near 28 Hz, and one near 36 Hz, with a smaller peak around 48 Hz. These values suggest that the two lower frequency peaks had an increase of about 2 Hz, the intermediate peak had an increase of about 3 Hz, and the high frequency peak showed little to no change in frequency. Day 2 results showed three peaks located near 15 Hz, 25 Hz, and 49 Hz. Day 6 results showed peaks near 10 Hz, 20 Hz, 26 Hz, and 40 Hz, with smaller intermediate peaks between the higher amplitude peaks. The amplitude of the frequency response was also elevated above 60 Hz, when comparing the amplitude of the other days. The frequency responses for subject 8 showed significant variation of the amplitude for the resonance peaks on each day. The amplitude was smallest for the low frequency resonance peaks on day 6. Variation in amplitude occurred between all scans for subject 8.



Figure 2.23: Frequency response of subject 8 at point 2 for all measurement days.

### 2.7 Discussion

The data collection efforts conducted for this research provided for an opportunity to record resonance frequencies of the biceps brachii muscle over a large frequency range. Little information is available in the literature regarding in-vivo passive resonance frequencies of skeletal muscles. The data provided in this thesis shows that there are three major resonance frequencies for the biceps brachii muscle group between 10 and 40 Hz, the largest being between 10 and 20 Hz.

Pilot activities aided in the decision to focus analysis on point 2 for this project. It was theorized that points 1 and 3 were located near enough to the elbow and shoulder joints respectively, that connective tissue, instead of purely bicep brachii muscle was being measured. This may have caused some of the variation in the repeatability of the measurements that were observed during pilot evaluations.

Pilot activities also aided in determining the configuration that subjects would be positioned for data collection activities. It was decided that a configuration that allowed for the most neutral position of the body would be most advantageous because it would be less difficult for subjects to hold a neutral position for a longer period of time, avoiding as much as possible, natural movements and involuntary muscle activation. However, looking back on data collection, and configuration, a future study should repeat the measurements taken in this study with the subjects in the extended arm position. It is hypothesized that by placing the arm in the extended position, the change in stiffness of the muscle group would be emphasized in the frequency response results.

Table 2.4 shows the converted stiffness results for the control group, applied to the proportional analysis based on the beam model to find the relative shift in frequency expected from changes in stiffness of the muscle group. Based on this evaluation, a change in the first mode frequency of 2 Hz or greater may indicate potential damage in the muscle group. A paired t-test evaluation also showed that the variability observed in the stiffness results between days was most likely due to random variation for the control group. These results support the theory that control group measurements were consistent with acceptable variation, and normal activities were maintained by subjects during data collection. Data collected from the control group indicated that a shift of greater than 2 Hz in a first mode resonance peak may identify muscle damage.

Frequency response results for the control group showed strong correlation with stiffness results from the same subjects. Resonance peaks identified in the average frequency response plots varied 2 to 3 Hz between scans.

A similar analysis was performed using the stiffness results from the damage protocol group. Results of this evaluation are shown in Table 2.7. An analysis using the proportional model used with control group results and converted stiffness results from the damage protocol group showed an average change in first mode resonance of 4 Hz, which is greater than the resonance peak threshold as outlined by the control group analysis.

The analysis of the frequency responses of each of the subjects that participated in the damage protocol group necessitated that there were three categories created to sort the observed changes in frequency response that were documented throughout the recovery period.

Three subjects (2, 5, and 10) showed a shift in a resonance peak of at least 4 Hz. These responses agreed well with the stiffness analysis results. It was interesting to note that the shifts in frequency didn't always occur at the first peak. An explanation for this effect was not found.

32

Four subjects (3, 6, 7, and 9) showed an increase in amplitude of acceleration, but not a shift in frequency response. Subject 3 had a change in amplitude, but the reaction was mixed. At the higher frequency peak, the amplitude increased, and by day 6, had begun decreasing.

Three subjects (1, 4, and 8) elicited mixed responses when comparing pre-damage frequency responses with the post-damage frequency responses. The pre-damage frequency response for subject 1 showed two distinct peaks, but after the damage protocol, the energy from the two peaks may have combined at the higher frequency peak. The frequency response of subject 4 showed an opposing response, with a high frequency peak decreasing in amplitude, and low frequency responses increasing in amplitude. Results from subject 8 had two distinct peaks indicated in the pre-damage measurements. Post damage results showed two peaks, but the frequency of the resonance peaks had increased, and the amplitude of the low frequency peak increased. Day 3 results showed three peaks as well, and the amplitude of the peaks had returned to pre-damage levels. Day 6 results showed an overall decrease in amplitude. Peaks for Day 6 results were more complex in nature when compared to other results.

Based on these observations, it was determined that results for the frequency response of subjects that experienced muscle damage were not consistent. Some subjects saw an increase in frequency of modal peaks, while others saw a decrease in frequency.

For the purposes if this study, it was found that the frequency response of a muscle group did change when the muscle group was damaged. However, because there was not a clear pattern that the changes followed, more work must be done in the future to determine whether this may be a way to identify and track muscle damage and recovery.

#### 2.8 Conclusion

The objective of this project was to determine whether the frequency response of a muscle group could be used to track changes attributed to muscle recovery after a damage protocol. A natural consequence of this objective was that the resonance frequencies of the biceps brachii muscle group was recorded over a wide range of frequencies. It was found that three primary resonance frequencies between 10 and 40 Hz were common.

SWE measurements were taken and the results used as a baseline for determining the state of the muscle group. It was determined that a shift in the first mode frequency of more than 2 Hz

was required to identify damage in a muscle group, based on an analysis performed on the stiffness results. Stiffness results for the damage protocol group indicated that an average shift of 4 Hz had occurred based on the same stiffness analysis. Based on evaluations of SWE data from both the control and damage protocol groups, it was determined that damage had occurred in the muscle group for the damage protocol participants.

Frequency response results for the control group showed little variability between days for individual subjects. It was found that minor changes in frequency response from day to day was expected for the control group participants. Frequency response results for the damage protocol group showed mixed results, leading to three different potential outcomes that were identified: Three subjects whose post-damage frequency responses showed a positive correlation with results from the stiffness analysis; Four subjects whose post-damage frequency responses showed an increase in amplitude, but no shift in resonance peaks; three subjects whose post-damage frequency responses were affected by damage in a muscle group, but more testing and a variation of arm position should be explored to determine the relationship between stiffness and frequency response of a muscle group.

To emphasize the stiffness changes in the muscle group during recovery, it is proposed that the same experimental methods used in this project be repeated with the straight arm orientation in the future. SWE measurements were taken with the arm in a straight position because when the arm is in a bent position, it was found that the results in pilot studies showed the arm to have isotropic properties. With the arm in a straightened position, the results were more homogeneous, and provided a better data collection method. By taking data with the SLDV in the same manner, we believe that the frequency response results will be more repeatable and consistent, and will be able to gather better data regarding the effects of muscle damage on frequency response. This analysis was performed with only 10 subjects in the damage protocol group. Because there is no general data to compare SWE data against as a check, more subjects should be tested to determine what a general SWE response should be for the Biceps Brachii muscle group. It is expected that with the speed at which SWE and elastography research in general is being conducted, that perhaps a better understanding of general responses for muscle groups should be. More research into how connective tissue affects the frequency response of a muscle group should also be conducted.

### 2.9 Lessons Learned

A major portion of this project was spend learning best practices in collecting frequency response data for a muscle group. There were several takeaways that improved efforts in collecting repeatable data and the quality of the signal from the laser vibrometer.

We found that body position and keeping the measurement surface as static as possible was a considerable challenge to overcome. There were multiple attempts to determine the most comfortable and repeatable position for subjects to take during testing. It was found that a seated position, when possible, was advantageous, because it prevented natural movements from the legs and torso to affect measurements taken at the arm. An evaluation of repeatability for a subject lying down is advisable in the future to further prevent natural movements in the body from affecting the area of interest during data collection.

The arm position that the subjects were instructed to use for the data collection efforts had their elbow bent at 90 degrees, with their forearm resting on the test fixture, while the arm position used for the SWE measurements had subjects keep their elbow straight, and at their side. In retrospect, it would be beneficial to repeat the experiment, taking SLDV measurements with the subject's elbow straight. It is hypothesised that this change in arm configuration will improve clarity of the frequency response after the damage protocol by placing more emphasis on the change in stiffness of the muscle group, as well as minimizing effects from swelling and increased fluid levels in the tissue after the damage protocol.

The test fixture used for data collection on the biceps brachii muscle group was also an important factor in improving data collection efforts. The arm had to be supported to avoid subjects from having to use any muscle power to hold their arm. Any muscle activation was deemed to have potential in affecting the results of the study. The platform that was built also had to avoid direct contact with the muscle group under investigation. Any outside influence from the test fixture would have affected the frequency response of the muscle group.

Previously, a description of different surface coatings was presented. The surface coating was necessary to improve reflection of the laser to boost the return signal from the skin's surface to the laser head. We found that natural skin reflectivity was insufficient for what was considered good for collecting data with the SLDV system. We found that the SafetySkin®rub-on product was superior to other products, especially aerosolozed products for several reasons: liquids tended

to dry out too quickly, limiting the shelf life of the product; Aerosols were required to be applied in a separate room from the SLDV system to prevent contamination of the lenses. We also found that the Albedo 100<sup>TM</sup> product leaked after the first use, thereby rendering it useless after the first application. The SafetySkin®product tended to have the best shelf life because it is not a liquid. The product was easy to apply, and could be applied while the subject was seated for data collection. The SafetySkin®product proved to be the most reliable for improving the reflectivity of the skin surface.

## CHAPTER 3. DATA COLLECTION, DISCUSSION, & RESULTS, LEG WRAP STUDY

### 3.1 Overview

While it is general practice when gathering clinical data to use a wearable accelerometer, typically attached to the body by way of using a strap system, the objective of this study was to determine the repeatability of the test setup, and whether there was an appreciable change in the response of the system due to the wrap that was applied to the lower leg.

Three points, located nominally, were measured. Point 1 was located nearest the knee at the bony marker for the head of the fibula. Point 2 was located approximately at the center of the largest circumference of the gastrocnemius muscle group. Point 3 was located approximately the same distance from point 2 as point 1 is located from point 2, as shown in Figure 3.1. A reference point was located at the top of the knee to aid subjects in holding the leg position throughout testing, labeled "R" in Figure 3.1. A red laser source was suspended above the knee, and was aimed at the point at the top of the knee. Subjects were instructed to keep the red laser light aligned with the reference point throughout testing. Two excitation frequencies were selected for analysis for their prevalence of use in whole body vibration studies [25]: 20 Hz, and 40 Hz. Each subject was measured 6 times for each applied frequency: three times with the leg not wrapped, and three times with the leg wrapped at the largest circumference of the lower leg. Each scan for the different configurations were taken immediately after one another. Several other test setups were attempted to try and solve the issue of keeping the leg in the correct location and position throughout testing. by using the reference laser at the top of the knee, and the brackets at the feet, no undue hardware was present to introduce errant vibrations at any other location along the length of the leg. other setups that were attempted used different bracket and fixture configurations, but were found to be unreliable in preventing poor data from being collected.



Figure 3.1: Direction of axes with respect to the body position of the subjects and measurement point locations on the side of the leg, and reference point location at the top of the knee.

# 3.2 Equipment

A 3-dimensional SLDV was used to measure the surface response of the leg for this study. The focus of this analysis was to determine how the displacement along the axis of the leg, the y-axis, was affected by the change in the system created by wrapping the leg in self-adhesive tape. The x-axis was categorized as the positive direction moving in the forward direction of the body. The positive z-direction moves laterally from the body as shown in Figure 3.1.

# 3.3 Methods

Data were collected for ten male subjects ages 18-25 for this study. A vibration plate was modified with several brackets to create a fixture where the subject placed their right foot on the platform and secured the front part of the foot with a hook and loop strap as shown in Figure 3.2.

The heel was placed against a bracket to prevent any protraction or retraction of the foot. The subject was seated in an adjustable chair to bring the knee to a  $90^{\circ}$  angle.



Figure 3.2: Modifications made to vibration platform to control location and movement of foot during testing

The point marked on the top of the knee was done using a permanent marker, and a red laser pointer was aligned with that point to aid in holding the leg position by the subjects. The subjects were instructed to keep the laser and the point on the leg aligned for the duration of the data collection period. Figure 3.3 shows the seated body position used for all subjects that participated in this pilot study.



Figure 3.3: Pilot configuration for testing surface response of the lower leg

Each subject was tested at two different frequencies and in two different states: 20 Hz, and 40 Hz; no tape, and tape. A self-adhesive elastic tape (3M Coban) was used to wrap the leg during the tape scans. The tape was applied at the largest circumference of the lower leg. To be as consistent as possible with the tension during application, the tape was stretched to its elastic limit as it was applied to the leg. A total of 3 scans per configuration were performed for each subject to find the average response at each scan point. Four analyses were identified for this study:

- 1. Repeatability was evaluated for each configuration
- 2. The average at each point was used to compare results between the no-tape configuration and the tape configuration
- 3. A paired t-test analysis was performed to determine if wrapping the leg affects the displacement and if that effect is statistically significant
- 4. The direction in which the effects occurred (positive or negative shift) was also analyzed

# 3.4 Results

Two different evaluations were performed on the data collected for this study: the first was a study on the repeatability of the test setup and fixture. Multiple fixture designs were evaluated in the past, with poor results. The setup for this evaluation needed to be verified to ensure that the data would be repeatable. The second analysis performed was that on comparing the displacement results for when the leg was wrapped versus when the leg was not wrapped. A statistical analysis was also performed on this data, to find whether the results showed statistically significant changes from one configuration to the other.

### 3.4.1 Repeatability

An evaluation of repeatability for all axes of the lower leg was necessary to prove that the test fixture allowed for enough control in all three planes of motion. For the purposes of this project, particular attention was given to both point 1 and point 3. To determine repeatability of the setup that was used for this pilot study, each scan at the two test frequencies (20 Hz and 40 Hz) were compared for percent variation by finding the percent variation for each subject across the three scans taken at each point, then finding the average of those percentages over all 10 subjects. This resulted in an overall average percent variation for each point under investigation. The analysis of repeatability did not include the scans that were made on the subjects in the taped configuration.

Figure 3.4 shows the average displacement in the y-direction results for each subject at points 1, 2, and 3, and their standard error. 7 of 10 subjects had increasing displacements from point 1 to point 3, while 2 subjects had similar displacement values for points 2 and 3. One subject had the opposite results, when compared to the other subjects. Point 1 had the greatest displacement, while point 3 had the smallest displacement. This reflects that the excitation source is closest to point 3, and as the vibrations travel through the leg, some of the energy will be absorbed by the tissue. Overall, the standard deviation for the points showed very little variation. The standard deviation for point 1 in the y-direction was calculated to be 2.14%, and standard error bars in the plot show minimal variation between the three scans taken for each subject. An analysis of the percent variation for point 3 was calculated to be 2.39%, indicating a slightly better repeatability as compared to point 1. The percent variation analysis of point 2 showed a percent variation of 1.84%, indicating that point 2 was the most repeatable of all three measurement locations. The repeatability analysis for the y-direction showed that the overall percent variation for all three points with a 20 Hz excitation frequency was 2.47%, and was the most repeatable of the three axes

under investigation. This suggested that the test setup was sufficiently repeatable for the y-direction and the 20 Hz excitation frequency.



Figure 3.4: 20 Hz average displacement results for all scans with no leg wrap for each subject and their standard deviation in the y-direction

The same analysis that was done for the 20 Hz results was carried out for the 40 Hz results. It was found that there was more variation in the results at the higher excitation frequency, as demonstrated by the larger range indicated by the error bars. Figure 3.5 shows the average displacement results for the 40 Hz excitation frequency. It was noted that the displacements of each of the points were flipped, when comparing to the trends identified with the 20 Hz results. Point 1 showed the greatest average displacement, and point 3 showed the smallest average displacement for the 40 Hz results. This may indicate that with the higher excitation frequency, involuntary muscle spasms were activated, causing greater movement at point 1, which was located approximately on the head of the fibular bone. An activation of the muscle group would have caused more movement of the bony structures underneath. For the variation analysis, the average percent variation between the scans for all of the subjects was calculated at 24.06% for point 1. When evaluating the results from point 3, variation between points was greater than at point 1 for the same excitation frequency. This was made apparent when calculating the average percent variation, which was 32.02% for point 3.



Figure 3.5: 40 Hz average displacement results for all scans with no leg wrap of each subject and their standard deviation in the y-direction

The results of the repeatability analysis for the 40 Hz excitation frequency were substantially higher than the repeatability results for the 20 Hz excitation frequency, though the only thing that changed between the two evaluations was the excitation frequency. This would suggest that the change in applied excitation frequency caused the system, meaning the leg, to experience an increase in instability in the y-direction.

Figure 3.6 shows the results from the same repeatability exercise performed for the ydirection evaluation for the 20 Hz excitation frequency in the x-direction. Y-direction displacements for the 20 Hz excitation frequency showed that a majority of subjects showed the highest displacements at point 3. For the x-direction, exactly half of the subjects showed the greatest amount of displacement at point 1, and half showed the greatest displacement at point 3. 8 of the 10 subjects showed the lowest displacements at point 2. These results indicate a split in responses for the x-direction. The calculated average percent variation from the mean for all subjects for point 1 was calculated at 9.02%. The results for point 3 found that the average percent deviation was 4.87%, more than half as much variance being present in point 3. Point 2 results were similar in the x-direction showing an average variation of 5.51%. The average variation in the x-direction is larger than in the y-direction, indicating that more movement was apparent in this direction.



Figure 3.6: 20 Hz average displacement results for all scans with no leg wrap of each subject and their standard deviation in the x-direction

Like what was observed for the y-direction evaluation, results for the 40 Hz excitation frequency indicated an increased amount of error between the three scans for all subjects. Standard deviation values were larger than results for the 20 Hz excitation frequency. Figure 3.7 shows the results at point 1 in the x-direction. The average percent variation at point 1 for the 40 Hz excitation frequency was calculated to be 28.85%. Point 3 in the x-direction showed similar results as what was seen at point 1. The average percent variation for point 3 in the x direction was calculated to

be 12.16%. Point 2 in the x-direction was most repeatable, when comparing the average variation. Point 2 had an average variation of 12.16%.



Figure 3.7: 40 Hz average displacement results for all scans with no leg wrap of each subject and their standard deviation in the x-direction

Repeatability was evaluated to determine how wrapping the leg affected the displacement in the z-direction. Like what was seen with the x-direction results, it was found that half of the subjects showed the greatest displacement at point 1, and half of the subjects showed the greatest displacement at point 3. Because of this split, it was not determined which point was most affected by the excitation frequency. It was found for point 1 at the 20 Hz excitation frequency, that the deviation between points was less repeatable when compared to either of the two other directions. The average percent variation for point 1 at 20 Hz was calculated to be 6.1%. Figure 3.8 shows the results of each subject, and their standard deviations for all points. Point 3 with the 20 Hz excitation frequency showed similar results to point 1. It was observed that the average percent variation for the group was calculated to be 9.62% for point 3. Average variation results for point 2 were calculated to be 15.88%, indicating that in the z-direction, point 2 was the least repeatable point with the 20 Hz excitation frequency.



Figure 3.8: 20 Hz average displacement results for all scans with no leg wrap of each subject and their standard deviation in the z-direction

Results for points 1, 2, and 3 with the 40 Hz excitation frequency are similar to what we have shown previously. 5 of the 10 subjects had the greatest displacement at point 3, while the other 5 had the greatest displacement at point 1. The average percent variation was calculated to be 33.24% for point 1. The average percent variation for point 3 was calculated to be 28.68%. The average variation for point 2 was calculated to be 28.31%. Figure 3.9 shows the plotted results for these points. The error bars shown in figure 3.9 are wider than in all of the other directions. The percent standard deviation for point 1 was 35.09%, while for point 2 the standard deviation was 28.60%, and for point 3 was calculated to be 49.99%.



Figure 3.9: 40 Hz average displacement results for all scans with no leg wrap of each subject and their standard deviation for point 1 in the z-direction

The standard deviation was highest for point 3 in the z-direction out of all of the other points. The next highest standard deviation calculated was point 2 in the x-direction, at 40.78%. It was found that both the average percent variation and the standard deviation was highest with the 40 Hz excitation frequency.

A summary of all the average percent variations and standard deviations are found in Table 3.1. Based on the evaluations done for the repeatability study, it was found that the results for the y-direction and the 20 Hz excitation frequency were most repeatable, indicating that controls that were set up to control against unwanted movement were adequate for this direction. The most variable direction was found to be the z-direction. This was expected, considering the degrees of freedom available in both the ankle, and in the hip. Controls for these degrees of freedom were difficult to establish, and were left to the individual subjects to control through the instruction to keep the laser point aligned with the point on the top of the knee. Further discussion on the results

as well as why repeatability at the 40 Hz excitation frequency may be less reliable will be addressed in the discussion section of this paper.

	% Variation	X-Dir	ST Dev	Y-Dir	ST Dev	Z-Dir	ST Dev
20 Hz Excitation	Point 1	9.02	9.16	3.20	2.14	6.13	2.63
	Point 2	5.51	4.81	1.84	1.42	15.88	19.22
	Point 3	4.87	3.70	2.39	2.11	9.63	5.00
40 Hz Excitation	Point 1	28.85	24.73	24.06	30.36	33.24	35.09
	Point 2	28.76	40.78	9.40	11.00	28.31	28.60
	Point 3	12.16	10.27	32.02	30.29	28.68	49.99

Table 3.1: Percent variance and standard deviation of both excitation frequencies.

## 3.4.2 Leg Wrap vs No Wrap

A comparison of average displacement results in the y-direction for each subject were evaluated for differences between conditions of when the leg was not wrapped versus when the leg was wrapped. The points above the leg wrap, point 1, and the point below the leg wrap, point 3, were a particular focus to determine the effects on the surrounding tissue. With the repeatability analysis previously, it was found that the point that experiences the maximum displacement is dependent upon the applied excitation frequency. With the 40 Hz excitation frequency applied, point 1 showed the largest amount of displacement in the y-direction. With the 20 Hz excitation frequency applied, it was found that point 3 experienced the largest amount of displacement. A paired t-test was then performed on the data to determine if the results were statistically significant. A result was considered statistically significant if the calculated p-value was less than 0.07, indicating that the results are considered significant with a 93% confidence interval.

Figure 3.10 shows the average displacement at point 1 for all the subjects for both test configurations at an excitation frequency of 20 Hz. For most subjects, a positive shift in displacement occurred after the leg was wrapped, with 8 of the 10 subjects showing an increase in displacement. The subjects who did not see an increase in displacement did not experience an appreciable change in displacement between the two configurations. The average percent increase in displacement for all subjects at 20 Hz excitation frequency was 10%, though some subjects saw an increase of 20% between the two configurations.



Figure 3.10: Average displacements at point 1 for the wrapped and no wrap results with a 20 Hz excitation frequency in the y-direction.

A paired t-test analysis was performed to determine whether the changes observed due to the wrap on the leg were statistically significant. Using 9 degrees-of-freedom and a two-tail probability analysis, the t-test results indicated a p-value of 0.023 for results at point 1 in the y-direction and 20 Hz excitation frequency. Previously, it was stated that results with a p-value of 0.07 or less was considered statistically significant. The calculated p-value for this point indicates that the changes were due to the wrap on the leg, and not due to random variation.

Figure 3.11 shows the average displacement at point 3 for the 20 Hz excitation frequency under the same evaluation done for point 1. It was once again observed that there were changes to the response for all subjects. However, the nature of the responses at point 3 were mixed. 4 of 10 subjects experienced an increase in displacement, while 6 of 10 subjects experienced a decrease in displacement at this point. While nearly half of the subjects did see an increase in displacement,

the increases shown were small magnitude changes. The average percent change in displacement was calculated to be -4%, meaning there was an overall decrease in the average displacement of this point for all subjects. A paired t-test was performed on the results for this point as well. A two-tailed analysis showed that the p-value was 0.64, much larger than the 0.07 threshold that was established. These results indicated that the variation observed was most likely due to random chance.



Figure 3.11: Average displacements at point 3 for the wrapped and no wrap results with a 20 Hz excitation frequency in the y-direction.

Results for point 2 were also evaluated for the 20 Hz excitation frequency. The average response for point 2 showed that half of the subjects had a small decrease in displacement, and the other half showed a slight increase in displacement. Two subjects did not see any appreciable difference in displacement between the two configurations. Because the change in displacement for the majority of subjects was small in both directions, the potential for the results for this point of being statistically significant are low. An overall average percent change in displacement was calculated to be 4% in the positive direction. Based on the observations of the plot, this result was

reasonable, indicating that there were no significant outliers in the data. The paired t-test showed that the results for point 2 were not statistically significant. The two-tailed p-value was calculated to be 0.54, significantly higher than the 0.07 threshold. Figure 3.12 shows the results for point 2. These results point to random chance being the cause of the variation observed.



Figure 3.12: Average displacements at point 2 for the wrapped and no wrap results with a 20 Hz excitation frequency in the y-direction.

Previously, the analysis for repeatability showed that there were larger variations in results when the 40 Hz excitation frequency was applied. It was expected that results for the leg wrap comparison would also show large variations in the two configurations. Figure 3.13 shows the results for point 1 in the y-direction with the 40 Hz excitation frequency. 7 of the 10 subjects had a decrease in displacement at point 1 when the wrap was applied. The overall average percent change was found to be -14%. This was likely due to the large changes in displacement for three of the subjects. 2 of the 10 subjects showed little or no change in displacement between the two configurations. While the -14% change in displacement might indicate a substantial change in displacement at point 1, the two tailed t-test resulted in a p-value of 0.26 for this point, indicating

that the results were not statistically significant. These results indicate that the variation observed was due to random chance.



Figure 3.13: Average displacements at point 1 for the wrapped and no wrap results with a 40 Hz excitation frequency in the y-direction.

Figure 3.14 shows the results for point 2 in the y-direction with the 40 Hz excitation frequency. 7 of the 10 subjects showed a decrease in displacement when the leg was wrapped. all but three of the 7 subjects showed significant decreases in displacements. Results for this point showed an average overall percent change in displacement of -24%. Because there was a majority of subjects who experienced a decrease in displacement, it was expected that the paired t-test analysis would indicate that these results were statistically significant. The percent change for this point was considerable, and the two-tailed t-test resulted in a p-value of 0.07, meeting the threshold designated for this analysis. These results indicate that the variation observed, at 93% confidence, was due to the wrap being applied to the leg. These results suggest that the wrap has a damping effect on the tissues located below the leg wrap.



Figure 3.14: Average displacements at point 2 for the wrapped and no wrap results with a 40 Hz excitation frequency in the y-direction.

Figure 3.15 shows the results for point 3 in the y-direction with the 40 Hz excitation frequency. Results showed that half of the subjects experienced a decrease in displacement at point 3, and half experienced an increase in displacement. 5 subjects experienced little to no change in displacement due to the wrap being applied. Based on these observations, it is likely that the results for this point excitation frequency are not statistically significant. No conclusions can be drawn from these data to determine the likely effect that the leg wrap causes on the tissue located near point 3. The average overall percent change in displacement was found to be 12% in the positive direction. These results show that a majority of subjects did experience an increase in displacement at point 3. The two-tailed t-test analysis was performed on the data for this point and the p-value for point 3 in the y-direction was found to be 0.69, far greater than the 0.07 threshold established. These results indicate that the results for this point were not statistically significant, and therefore were likely due to random chance.



Figure 3.15: Average displacements at point 3 for the wrapped and no wrap results with a 40 Hz excitation frequency in the y-direction.

An analysis was performed for all points in the x-direction in the same manner that was performed for the points in the y-direction. An evaluation of the change in displacement was performed, as well as an analysis of the average change in displacement was performed. A paired t-test was then used to explore statistical significance of the results, and determining whether the changes in displacement are likely caused by wrapping the leg, or whether they are likely due to random chance.

Figure 3.16 shows the results for point 1 in the x-direction for both the wrapped and the nonwrapped configurations with the 20 Hz excitation frequency. 4 of the 10 subjects that were tested showed an increase in displacement, while 6 of 10 showed a decrease. the majority of changes were small in magnitude. At least two of the subjects saw almost no change in displacement when comparing the two configuration results. The overall average shift in displacement was calculated and resulted in a 6% increase in displacement. Because the average shift in displacement was small, and the number of subjects that experienced a decrease versus an increase in displacement were nearly the same, it was predicted that the data for this point were not statistically significant. The paired t-test evaluation showed that the two-tailed p-value was 0.26, much larger than the 0.07 threshold established earlier. These results point to random chance being the source of variation for this point.



Figure 3.16: Average displacements at point 1 for the wrapped and no wrap results with a 20 Hz excitation frequency in the x-direction.

Figure 3.17 shows the results for point 2 in the x-direction for both the wrapped and the non-wrapped configurations with the 20 Hz excitation frequency. It was found that 6 of the 10 subjects saw an increase in displacement, while 4 of the 10 saw a decrease in displacement. Four subjects saw smaller changes in displacement, the majority of whom indicated an increase. The calculated average change in displacement for point 2 was 7% in the negative direction, indicating an overall smaller displacement in the x-direction. There were, however, several subjects that showed significant differences between the non-wrapped displacement and the wrapped displacement. The largest shift due to wrapping the leg that was observed was from subject 8, with a decreased response 63% less than the non-wrapped displacement. The results of the paired t-test

showed that the two-tailed p-value was 0.15, only slightly larger than the 0.07 threshold applied. These results indicate that the variation was likely due to random chance, and likely not due to the wrap being applied to the leg.



Figure 3.17: Average displacements at point 2 for the wrapped and no wrap results with a 20 Hz excitation frequency in the x-direction.

Figure 3.18 shows the results for point 3 in the x-direction for both the wrapped and nonwrapped configurations with the 20 Hz excitation frequency. The results for this point showed a general trend among all of the subjects but 1, of a decrease in displacement due to the wrap at the center of the leg. Three of the subjects did not see a significant change in displacement between the two configurations. The average shift in displacement was calculated and resulted in a 20% decrease overall in the x-direction displacement at point 3. The decrease in displacement was nearly universal. A paired t-test was performed, and the two-tailed p-value that was calculated was 0.07. These results indicate that at 93% confidence, the variation was due to the wrap being applied to the leg. The results for point 3 in the x-direction suggest that tissues located below the wrapped portion of the leg experiences a decrease in displacement due to the wrap being applied to the leg.



Figure 3.18: Average displacements at point 3 for the wrapped and no wrap results with a 20 Hz excitation frequency in the x-direction.

Figure 3.19 shows the results for point 1 in the x-direction with the 40 Hz excitation frequency. For most of the subjects at point 1 with the 40 Hz excitation frequency, the change in displacement was in the positive direction, with 7 of the 10 subjects showing an increase in displacement at point 1. 5 of the subjects showed small changes in displacement The average change in displacement was calculated and showed that for all subjects, the change in displacement was 42% in the positive direction. Though the average change in displacement was large, it was influenced by significant displacement changes that some subjects saw. The largest change that was recorded showed a positive change in displacement up to 115% higher with the leg wrapped than with the leg not wrapped. The change in displacement observed at point 1 for the 40 Hz excitation frequency in the x-direction returned a two-tailed p-value of 0.03, suggesting that the changes observed were likely due to the application of the wrap to the leg, with a 97% confidence interval.

These results strongly point to the variation in displacement being due to the wrap applied to the leg.



Figure 3.19: Average displacements at point 1 for the wrapped and no wrap results with a 40 Hz excitation frequency in the x-direction.

Figure 3.20 shows the results for point 2 in the x-direction with the 40 Hz excitation frequency. 6 subjects saw an increase in displacement when compared to the non-wrapped leg, with the majority of those indicating relatively small increases in displacement. The average overall change in displacement was calculated and was found to be 35% in the positive direction. The largest change in displacement that was observed was a positive change of 234% in the positive direction from a single subject. This biased the data significantly, when calculating the average change in displacement. Because the responses were nearly split in half, and most of the subjects only experienced a small change in displacement, it is likely that the data collected for this point are not statistically significant. Although the results for this point were mostly in the positive direction, the paired t-test analysis showed a two-tailed p-value of 0.85, much larger than the 0.07 threshold established earlier. These results strongly indicate that the variation was due to random chance, and not influenced by the presence of the wrap on the lower leg.



Figure 3.20: Average displacements at point 2 for the wrapped and no wrap results with a 40 Hz excitation frequency in the x-direction.

Figure 3.21 shows the results for point 3 in the x-direction with the 40 Hz excitation frequency. 4 of 10 subjects experienced an increase in displacement between the two configurations, and two of those subjects experienced minimal increases in displacement. The largest change in displacement was a decrease. The average overall change in displacement was calculated and was found to be 14% smaller than the displacement without the leg wrap. Because the response due to the leg wrap was split nearly in two by the subjects, it is expected that the results for this point and excitation frequency are not statistically significant. The paired t-test analysis showed that the two-tailed p-value for this point at this excitation frequency was 0.31, greater than the 0.07 threshold. The t-test analysis indicates that the variation was likely due to random chance, and not a result of the wrap applied to the leg.



Figure 3.21: Average displacements at point 3 for the wrapped and no wrap results with a 40 Hz excitation frequency in the x-direction.

An evaluation of all the points in the z-direction for both excitation frequencies was performed. A comparison of responses was carried out, and an evaluation of the average percent change in displacement was calculated. A paired t-test analysis was performed on the data to determine whether the results were due to random chance or were indicative of the change in the system due to wrapping the leg.

Figure 3.22 shows the results for point 1 with the 20 Hz excitation frequency in the zdirection. 6 of 10 subjects experienced a decrease in displacement due to the wrap being applied to the leg. A majority of the decreases were moderate in size, meaning that we expected that the average change in displacement be decreasing when the leg wrap is applied. The average overall change in displacement was found to be 7% in the negative direction, which agrees with the observations made earlier. Because only a small majority of the subjects had a decrease, it was expected that the results would not be considered statistically significant. When performing the paired t-test analysis, the two-tailed p-value that was calculated was 0.21, failing to prove the null hypothesis false, indicating that the changes in displacements were likely due to random chance and not due to the wrap applied to the leg.



Figure 3.22: Average displacements at point 1 for the wrapped and no wrap results with a 20 Hz excitation frequency in the z-direction.

Figure 3.23 shows the results for point 2 with the 20 Hz excitation frequency in the zdirection. 6 of 10 subjects saw an increase in displacement, however the largest changes in displacement were held by subjects who had a decrease in displacement. The majority of changes in displacement were only moderate. Again, because only a small majority of subjects followed the same trend, and the changes in displacement were only moderate, it was expected that the average change in displacement would be negative, and small in magnitude. It is also expected that the paired t-test analysis would yield a p-value that suggests that the results for this point are not statistically significant. The average overall change in displacement was calculated to be 75% in the positive direction. Although the average change in displacement was large, most of the change was due to one subject that showed a change of nearly 700%. An average change in displacement was calculated excluding the outlier subject. The results showed an average overall change in displacement of 5% in the negative direction, agreeing correctly with what was anticipated. A paired t-test analysis was performed for the results of point 2 in the z-direction. The two-tailed p-value calculated for these data was found to be 0.37. When the outlier data were excluded, the p-value was 0.26. These results were also anticipated, meaning that the changes in displacement at this point were not statistically significant, and are likely due to random variation.



Figure 3.23: Average displacements at point 2 for the wrapped and no wrap results with a 20 Hz excitation frequency in the z-direction.

Figure 3.24 shows the results for point 3 with the 20 Hz excitation frequency in the zdirection. 6 of 10 subjects saw an increase in displacement at point 3 in the z-direction., while one subject failed to indicate a change in displacement between the two configurations. The largest changes in responses were held by two subjects, one of which had an increase, and the other a decrease in displacement. It is anticipated that the average change in displacement was significant because of the large variations shown by the few subjects that showed them. It is also anticipated that these results are not statistically significant, because a general response was not visible in this analysis. The average overall change in displacement was a 20% decrease in displacement. The paired t-test analysis showed that the two-tailed p-value for this point in the z-direction at the 20 Hz excitation frequency was 0.74, exceeding the 0.07 threshold established earlier. These analyses indicate that the results for this point are not statistically significant, and the changes observed are likely due to random chance.



Figure 3.24: Average displacements at point 3 for the wrapped and no wrap results with a 20 Hz excitation frequency in the z-direction.

Like what was observed with the other directions, and with the repeatability analysis, it was anticipated that the results with the 40 Hz excitation frequency would show more variability in responses, and perhaps larger changes in displacement than with the 20 Hz excitation frequency results. An analysis of the results for the 40 Hz excitation frequency follows.

Figure 3.25 shows the displacement results in the z-direction for point 1 with the 40 Hz excitation frequency. 6 of 10 subjects had a decrease in displacement in the z-direction at this point for the 40 Hz excitation frequency, and like what was observed previously, when only a small majority is identified for the two responses, it is anticipated that the average change in displacement will be small, and the t-test analysis will reveal that the results for this point are not statistically
significant. There were two subjects that had greater than a 100% positive change in displacement, and were considered outliers. When excluding the two subjects, the overall average change in displacement was 12% in the negative direction. The paired t-test showed a two-tailed p-value of 0.90, indicating that the change in displacement for this point and excitation frequency are likely due to random variations.



Figure 3.25: Average displacements at point 1 for the wrapped and no wrap results with a 40 Hz excitation frequency in the z-direction.

Figure 3.26 shows the displacement results in the z-direction for point 2 with the 40 Hz excitation frequency. 6 of 10 subjects saw an increase in displacement for point 2, but the majority of changes in displacement were minor. These results suggest that the p-value will indicate that the changes observed are likely due to random variation. The overall average change in displacement showed a 15% increase in displacement in the z-direction, agreeing with what the majority of subjects showed. The paired t-test analysis showed a two tailed p-value of 0.78, substantially higher than the threshold, 0.07. These analysis support the predicted outcome that the changes observed in the displacement due to the wrap are likely random in nature.



Figure 3.26: Average displacements at point 2 for the wrapped and no wrap results with a 40 Hz excitation frequency in the z-direction.

Figure 3.27 shows the displacement results in the z-direction for point 3 with the 40 Hz excitation frequency. Half of the subjects saw an increase in displacement in the z-direction, already suggesting that the results are not likely to be statistically significant. Three of the subjects saw an increase of greater than 100%, while other subjects did not show a significant change in displacement due to the wrap. When including these displacement values, the average overall change in displacement is calculated to be a 21% increase. When the three largest displacement shifts are excluded as outliers, the overall average displacement shift is 18% decrease in displacement. The two-tailed p-value was found to be 0.96, indicating that the changes observed in the displacement are not statistically significant, and likely due to random variation.

The analysis of the z-direction results indicate that none of the data collected were statistically significant, and showed large variations in the data. A more thorough analysis of the test setup and fixture may be necessary to understand whether they were sufficient to control unwanted movement in the z-direction.



Figure 3.27: Average displacements at point 3 for the wrapped and no wrap results with a 40 Hz excitation frequency in the z-direction.

Table 3.2 shows a summary of t-test results for all evaluations discussed in this chapter. Results that were considered statistically significant to a confidence interval of 93% are italicized.

Table 3.2: Paired t-test results using the two-tailed p-value approach and the average percent change in response. Results were considered statistically significant with a p-value less than 0.07. Statistically significant results are italicized.

		X-Dir	$\% \Delta$	Y-Dir	$\% \Delta$	Z-Dir	$\% \Delta$
20 Hz	Point 1	0.26	-6%	0.02	+10%	0.21	-7%
	Point 2	0.15	-7%	0.55	+4%	0.37	-5%
	Point 3	0.07	-20%	0.64	-4%	0.74	+38%
40 Hz	Point 1	0.03	+38%	0.26	-14%	0.91	-12%
	Point 2	0.85	+35%	0.07	-24%	0.78	+15%
	Point 3	0.31	-14%	0.69	+12%	0.97	-18%

### **3.4.3** Direction of Change

A paired t-test analysis and an evaluation of the shift in displacement for all subjects were performed and recorded. Based on the paired t-test results, the responses recorded showed two data points that had statistically significant results within a 95% confidence: 20 Hz excitation, point 1 in the y-direction; and 40 Hz excitation, point 1 in the x-direction. There were two other points that had statistically significant results within a 93% confidence: 20 Hz excitation, point 3 in the x-direction; and 40 Hz excitation, point 2 in the y-direction. None of the results in the z-direction were considered statistically significant.

The y-direction displacement for the 20 Hz excitation frequency indicated an overall average shift of 10% greater displacement with a leg wrap versus without a leg wrap at point 1. The x-direction displacement for the 40 Hz excitation frequency indicated an overall average shift of 42% greater displacement with a leg wrap versus without a leg wrap at point 1. These results point to an overall increase in displacement of tissue above the wrapped portion of the lower leg when a wrap is applied to the location of largest circumference.

The x-direction displacement for the 20 Hz excitation frequency indicated an overall average decrease of 20% at point 3 with a confidence interval of 93%. Based on the results of this point, a decrease in displacement is expected for tissue below the leg wrap.

During data collection, there was an observation expressed by several of the subjects, that the difficulty in holding still during the scans increased with the excitation frequency. Burke et al. explored the effects on non-contracted muscle when an external vibration is applied. The research described how muscle groups will be activated at the same excitation frequency, or at sub-harmonics of the excitation frequency of an external source [26]. Depending on how long each scan is required to run, this anomaly may have affected the results of these scans, especially for higher excitation frequencies. The effects described by Burke et al. was one possible reason for why some of the data points collected did not result in statistically significant results for the 40 Hz excitation frequency.

### 3.5 Conclusion

The objective of this study was to evaluate both the repeatability of the test setup, and the effects of wrapping the lower leg on the surface velocity using two different excitation frequencies. Repeatability was evaluated based on percent variation between scans and standard deviation values. Large increases in percent variation and standard deviation occurred when the excitation frequency was increased to 40 Hz. Through the analysis of the effects of the leg wrap, an increase in velocity for material located above the wrapped region of the leg can be expected, and the surface velocity may decrease below the wrapped region of the leg. The effects of wrapping the leg are greater for tissue located above the leg wrap, when comparing against the effects below the leg wrap.

A major factor that was identified during data collection was that the ability of the subject to control their leg position decreased as the excitation frequency increased. This was corroborated with a 1976 study conducted by Burke et. al. Control of body position for this study was different from the arm study discussed previously because of two factors: First, this study evaluated results in all three directions; second, because this study evaluated results in all three directions, the setup was required to aid subjects in their ability to hold the leg in the required position without an external aid at the knee. From these observations, it is suggested to ensure that the measurement period for gathering data at higher excitation frequencies can be accomplished, the rate of collection must be higher than the excitation frequency. Ensuring the measurement frequency is greater than the highest excitation frequency may be a viable way to overcome the variation due to this phenomenon.

#### REFERENCES

- [1] Foley, J. M., Jayaraman, R. C., Prior, B. M., Pivarnik, J. M., and Meyer, R. A., 1999. "Mr measurements of muscle damage and adaptation after eccentric exercise." *Journal of Applied Physiology*, 87(6), pp. 2311–2318 PMID: 10601183. 1, 2
- [2] Duane A. Morrow, Tammy L. Haut Donahue, G. M. O. K. R. K., 2010. "Transversely isotropic tensile material properties of skeletal muscle tissue." *Journal of the Mechanical Behavior of Biomedical Materials*, 3(1), pp. 124–129. 1
- [3] Gennisson, J. L., Cornu, C., Catheline, S., Fink, M., and Portero, P., 2004. "Human muscle hardness assessment during incremental isometric contraction using transient elastography." *Journal of Biomechanics*, 38, pp. 1543–1550. 1, 3
- [4] Andonian, P., Viallon, M., Le Goff, C., de Bourguignon, C., Tourel, C., Morel, J., Giardini, G., Gergele, L., Millet, G. P., and Croisille, P., 2016. "Shear-wave elastography assessments of quadriceps stiffness changes prior to, during and after prolonged exercise: A longitudinal study during an extreme mountain ultra-marathon." *PLOS ONE*, **11**(8), p. e0161855. 1
- [5] Agten, C. A., Buck, F. M., Dyer, L., Flck, M., Pfirrmann, C. W. A., and Rosskopf, A. B., 2016.
  "Delayed-onset muscle soreness: Temporal assessment with quantitative mri and shear-wave ultrasound elastography." *American Journal of Roentgenology*, 208(2), pp. 402–412.
- [6] Nowicki, A., and Dobruch-Sobczak, K., 2016. "Introduction to ultrasound elastography." *Journal of ultrasonography*, **16**(65), pp. 113–124. 1, 18
- [7] Bercoff, J., Tanter, M., and Fink, M., 2004. "Supersonic shear imaging: a new technique for soft tissue elasticity mapping." *IEEE Transactions on Ultrasonics, Ferroelectrics, and Frequency Control*, **51**(4), pp. 396–409. 1
- [8] Mannard, A., and Stein, R. B., 1972. "Determination of the frequency response of isometric soleus muscle in the cat using random nerve stimulation." *Journal of Physiology*, 229, pp. 275–296. 2
- [9] Zahalak, G. I., and Heyman, S. J., 1979. "A quantitative evaluation of the frequency-response characteristics of active human skeletal muscle in vivo." *Journal of Biomechanical Engineering*, **101**(1), pp. 28–38. 2
- [10] Cannon, S. C., and Zahalak, G. I., 1982. "The mechanical behavior of active human skeletal muscle in small oscillations." *Journal of Biomechanics*, 15(2), pp. 111–121. 2
- [11] Brereton, L. C., and McGill, S. M., 1998. "Frequency response of spine extensors during rapid isometric contractions: effects of muscle length and tension." *Journal of Electromyography and Kinesiology*, 8(4), pp. 227–232. 2

- [12] Tabatabai, H., Oliver, D. E., Rohrbaugh, J. W., and Papadopoulos, C., 2013. "Novel applications of laser doppler vibration measurements to medical imaging." *Sensing and Imaging: An International Journal*, **14**(1), Jun, pp. 13–28. 2
- [13] Scalise, L., and Morbiducci, U., 2008. "Non-contact cardiac monitoring from carotid artery using optical vibrocardiography." *Medical Engineering Physics*, **30**(4), pp. 490–497. 2
- [14] Rohrbaugh, J. W., Sirevaag, E. J., and Richter, E. J., 2013. "Laser doppler vibrometry measurement of the mechanical myogram." *Review of Scientific Instruments*, 84(12), p. 121706.
  2
- [15] Scalise, L., Casaccia, S., Marchionni, P., Ercoli, I., and Tomasini, E., 2013. "Laser doppler myography: A novel non-contact measurement method for muscle activity." *Laser Therapy*, 22(4), pp. 261–268. 2, 3
- [16] Jang, H., Lee, J., Jeon, H., Kim, A., Kim, S., Lee, H., and Kim, K., 2018. "Metabolic profiling of eccentric exercise-induced muscle damage in human urine." *Toxicol Res*, 34(3), pp. 199–210. 2
- [17] Godfrey, A., Conway, R., Meagher, D., and OLaighin, G., 2008. "Direct measurement of human movement by accelerometry." *Medical Engineering & Physics*, **30**(10), pp. 1364– 1386. 3, 4
- [18] eljko Pedii, and Bauman, A., 2014. "Accelerometer-based measures in physical activity surveillance: current practices and issues." *British Journal of Sports Medicine*(49), pp. 219– 223. 4
- [19] Kitazaki, S., and Griffin, M. J., 1997. "Resonance behavior of the seated human body and effects of posture." *Journal of Biomechanics*, **31**(2), pp. 143–149. 4
- [20] Mandapuram, S., Rakeja, S., Marcotte, P., and Boileau, P.-E., 2011. "Analysis of biodynamic responses of seated occupants to uncorrelated fore-aft and vertical whole-body vibration." *Journal of Sound and Vibration*, **330**(16), pp. 4064–4079. 4
- [21] Coyte, J. L., Stirling, D., Du, H., and Ros, M., 2016. "Seated whole-body vibration analysis, technologies, and modeling: A survey." *IEEE Transactions on Systems, Man, and Cybernetics: Systems*, 46(6), pp. 725–739. 4
- [22] Lister, T., Wright, P., and Chappell, P., 2012. "Optical properties of human skin." *Journal of Biomedical Optics*, 9(17). 14
- [23] Ash, C., Dubec, M., Donne, K., and Bashford, T., 2017. "Effect of wavelength and beam width on penetration in light-tissue interaction using computational methods." *Lasers Med Sci*(32), pp. 1909–1918. 14
- [24] Rao, S. S., 2004. Mechanical Vibrations 4th Ed.. Pearson Prentice Hall. 18
- [25] Ritzmann, R., Gollhofer, A., and Kramer, A., 2013. "The influence of vibration type, frequency, body position and additional load on the neuromuscular activity during whole body vibration." *European Journal of Applied Physiology*, **113**(1), Jan, pp. 1–11. 37

[26] Burke, D., Hagbarth, K.-E., Lofstedt, L., and Wallin, B. G., 1976. "The responses of human muscle spindle endings to vibration of non-contracting muscles." *Journal of Physiology*, 261, pp. 673–693. 67

# APPENDIX A. FREQUENCY RESPONSE OF BICEPS BRACHII, PILOT ARM POSI-TION EVALUATION RESULTS

Appendix A shows the frequency response results for subjects 1 and 2 for the arm position evaluation in preparation for the frequency response and muscle damage project. The objective of these evaluations was to determine which position, extended arm or bent arm, provided the most reliable and consistent results for use in the main study. This analysis was necessary in part because a standard mode of measuring resonance frequency of the biceps muscle group has not been reported or established in the literature.

Figure A.1 shows the frequency response of subject 1 at point 1 in the extended arm test configuration. This position was evaluated for potential use in the Frequency response of the Biceps Brachii study. The data suggest that for point 1 in the extended arm position, the frequency response was variable below 20 Hz in both resonance peaks, and amplitude.



Figure A.1: Pilot evaluation of measurement point 1, in the extended arm position for subject 1

Figure A.2 shows the frequency response of subject 1 at point 2 in the extended arm test configuration. Like point 1, point 2 showed good repeatability for the frequency response above 30 Hz, and even indicated a higher frequency resonance around 60 Hz for all three scans. Variability was indicated below 20 Hz in amplitude. Variation in resonance peaks were also indicated below 10 Hz. Resonance peaks for this point were identified near 15 Hz, near 25 Hz near 35 Hz, and near 60 Hz. Comparing these results with the frequency response for subject 1 at point 2 in the bent arm position, a resonance peak was similarly found near 22 Hz, and near 60 Hz. Additional resonances were identified for the bent arm results near 35 Hz, near 50 Hz, and near 70 Hz. The results shown here suggest that by extending the arm, fewer resonance peaks are captured for the muscle group.



Figure A.2: Pilot evaluation of measurement point 2 in the extended arm position for subject 1

Figure A.3 shows the frequency response of subject 1 at point 3 in the extended arm test configuration. Much like what was observed for point 1, repeatability for subject 1 above 30 Hz was good for the extended arm configuration, and showed some variation in amplitude and resonance peaks below 20 Hz. Resonance frequencies were identified near 8 Hz, near 20 Hz near 35 Hz, near 55 Hz, and near 70 Hz. Smaller peaks were shown between 8 Hz and 20 Hz, but were difficult to identify due to the variation between scans. When comparing the variation identified

in the results for point 1 and the variation observed here, the results seem comparable, as far as repeatability is concerned. It was interesting to note the variation in resonance frequencies that were identified from point 1 and point 3. Resonance peaks were similar at near 35 Hz, and 60 Hz, but the lower frequency peaks did not match. A comparison between the extended arm results and the bent arm results for subject 1 at point 3 shows that resonance peaks near 8 Hz, near 22 Hz, and near 60 Hz are close to the resonances identified for the extended arm results. These results don't follow the same pattern identified by the point 2 results, suggesting that there are not fewer resonance peaks in the extended arm position. However, resonances between the two configurations are similar, within roughly 2-3 Hz.



Figure A.3: Pilot evaluation of measurement point 3 in the extended arm position for subject 1

Figure A.4 shows the frequency response for subject 2 at point 1 for the extended arm position. Like results for subject 1 at point 1 in the same configuration, the frequency response for subject 2 was consistent above 18 Hz, while there was variation in resonance peak frequency and amplitude below 18 Hz. Resonance peaks were identified near 25 Hz, and near 37 Hz. When comparing these resonances with subject 1 results in the extended arm position, a similar resonance

for subject 1 is found near 35 Hz, suggesting that a natural resonance of the biceps brachii muscle group is near 35 Hz.



Figure A.4: Pilot evaluation of measurement point 1 in the extended arm position for subject 2

Figure A.5 shows the frequency response for subject 2 at point 2 in the bent arm position. The results in this configuration were similar to results seen at point 1, showing variation in resonance frequencies below 20 Hz, while the results above 20 Hz were very consistent. Resonance peaks were identified near 5 Hz, near 15 Hz, and near 30 Hz for this configuration. Variations in amplitude above 10 Hz between the three scans were apparent, but were only identified below 30 Hz. An evaluation between the bent arm configuration and the extended arm configuration show similar results. Resonance peaks were identified in the extended arm position at near 5 Hz, near 20 Hz, near 45 Hz, and near 65 Hz. These resonance peaks correlate well with the bent arm configuration near the 5 Hz resonance. Consistently, we are seeing a change in resonance between the bent arm configurations are difficult to identify. Figure A.6 shows the results from the extended arm configuration for subject 2 at point 2.



Figure A.5: Pilot evaluation of measurement point 2 in the bent arm position for subject 2



Figure A.6: Pilot evaluation of measurement point 2 in the extended arm position for subject 2

Figure A.7 shows the frequency response for subject 2 at point 3 in the bent arm position. The results for point 3 showed more variation at higher frequencies when compared to results at point 1 and point 2. While amplitudes varied between the three scans, a peak was identified near 22 Hz for all three scans. Results below this frequency varied between the three scans.



Figure A.7: Pilot evaluation of measurement point and arm position for subject 2 point 3

Figure A.8 shows the frequency response for subject 2 at point 3 in the extended arm position. Results for point 3 in this arm position for subject 2 indicated the same trend that was identified at the other points, where above a certain frequency, the results were very consistent, but below that frequency there was some variation between the scans. Resonance peaks were identified near 5 Hz, near 10 Hz, and near 22 Hz for point 3. When comparing resonance peaks in the extended arm position, a single resonance was identified, near 20 Hz. Variation in amplitude was identified for the range between 0 and 40 Hz for all three scans. The analysis between the bent and extended arm configurations identified a related resonance peak near 20 Hz in both configurations, and a similar trend was shown for the frequency response below 10 Hz. It was determined that no significant difference between the two arm configurations was identified.



Figure A.8: Pilot evaluation of measurement point and arm position for subject 2 point 3

Based on the results presented here, both subjects showed consistent trends regardless of arm position, or measurement point. For both arm positions, it was shown that below a certain frequency, between 20 and 30 Hz, the results were mixed, while above the frequency identified, results were very consistent, and showed excellent repeatability. Based on this evaluation, results for the main study will be consistent, regardless of whether subjects use the straight arm position or the bent arm position. While this was the case when muscle damage was not present, this evaluation was not performed while muscle damage was present, and the same conclusion may not be true when muscle damage occurs.

## APPENDIX B. FREQUENCY RESPONSE OF BICEPS BRACHII

Appendix B shows the average frequency response results for the control group for points 1 and 3 and results for subject 7 at point 2 from the damage protocol group. The main body of research for the frequency response study focused on point 2, located at the center of the biceps brachii muscle group.

## **B.1** Control Group Results

Figure B.1 shows the average frequency response for the control group at point 1. Over the three consecutive measurement days, the control group indicated very consistent results for the frequency range under investigation. Point 1 results showed some variation, however, between 15 Hz and 30 Hz.



Figure B.1: Average frequency response function over all subjects that participated in the control group for measurement point 2

Figure B.2 shows the average frequency response for the control group. Results for point 3 did not show good consistency between the different days. Both the frequency of resonance peaks, and amplitudes varied across the frequency range that was evaluated.



Figure B.2: Average frequency response function over all subjects that participated in the control group for measurement point 3

# **B.2 Damage Protocol Group Results**

Figure B.3 shows the frequency response for subject 7 at point 2 of the damage protocol group. Resonance peaks were identified near 18 Hz, 37 Hz, and 48 Hz for the pre-damage frequency response. Day 1 frequency response results show resonance peaks near 17 Hz, 34 Hz, and 48 Hz. Day 2 results show resonance peaks near 19 Hz and 36 Hz. Day 6 results show resonance peaks near 14 Hz, 25 Hz, and 36 Hz. The peak near 48 Hz identified in the pre-damage results had no equivalent peak in the post-damage results after the day 1 data was collected . Amplitudes of the peaks increased after the damage protocol.



Figure B.3: Frequency response of subject 7 at point 2 for all measurement days.