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Investigation of Natural Perchlorate Levels in Antarctic Snow in Relation to the Ozone Hole

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ABSTRACT

Snow samples collected from the West Antarctic Ice Sheet Divide in January 2013 were analyzed for perchlorate concentrations to determine a possible seasonal variation. These samples were dated for the years 2007 through 2012 using annual variations in sulfate concentration as determined by an ion chromatography (IC) method. Perchlorate concentrations in the samples were measured with an IC instrument paired with tandem mass spectrometry detection. The concentration data show that perchlorate reached an annual maximum concentration in the austral autumn seasons, assuming that snowfall was relatively constant throughout the course of each year. Comparison with published total column ozone density at the South Pole shows that the perchlorate annual maximum does not occur in the same seasons when ozone is at a minimum level.

INTRODUCTION

Perchlorate is a common naturally occurring anion found throughout the world. Although little research has been done on the specific sources of perchlorate in nature, research suggests that it forms primarily in the atmosphere. It has been suggested that perchlorate is

formed in the atmosphere when chlorine species are oxidized by ozone[1]. This project has focused on this proposed formation by investigating how perchlorate concentrations relate to stratospheric ozone. Specifically, the project's focus was to determine if a seasonal relationship exists between perchlorate in Antarctic snow and stratospheric ozone over Antarctica, which is known to drop in the austral spring during the appearance of the ozone hole.

The Polar Regions have been a primary location for investigation of trends of naturally occurring chemical species due to the preservation of atmospheric content in snow. Dating of such ice samples can be completed for a depth of snow using knowledge of annual snow accumulation as well as seasonal variations of ions in snow. Sulfate, for example, is known to reach an annual maximum Antarctic concentration during the austral summer months of January to February[4]. Published research articles have utilized polar snow to measure perchlorate in Arctic Snow[1]; however, few have involved Antarctic snow, and none have investigated seasonal variation in perchlorate in relation to ozone. Some preliminary measurements of preserved snow and ice samples had shown perchlorate levels to increase around the late 1970s, when the ozone hole was known to begin forming. These measurements suggested a possible relationship between perchlorate and the ozone hole.

The original hypothesis of this study revolved around the idea that the depletion of stratospheric ozone over the Antarctic continent would allow additional ultra-violet (UV) light to pass through to the troposphere. This increased UV radiation would then lead to increased tropospheric ozone, as well as airborne chlorine radicals as reactants in perchlorate formation. By Le Châtlier's Principle, the addition of ozone and chlorine radicals should cause an increase in perchlorate concentration. To test this hypothesis, we

created a seasonal timeline of perchlorate in Antarctic snow to determine in which season elevated levels occurred.

METHODS

In a 3-meter deep snow pit, 88 snow samples from West Antarctic Ice Sheet Divide (WAIS) (112.085 °W, 79.467 °S) were taken from the surface downward in 3 cm depth increments in January 2013. These snow samples were stored in sealed plastic cups and transported frozen to a laboratory for analysis. The snow was allowed to melt and then directly poured into clean autosampler vials which were then capped and analyzed by two separate sets of instrumentation.

Each sample was automatically injected and analyzed with an Ion Chromatography (IC) apparatus consisting of a Dionex IC600 system with a GP50 Gradient Pump, Dionex IonPac® AS11 3 x 250 mm column with CSRS300 2 mm suppressor, and ED50 Electrochemical Detector to detect sulfate concentrations at the part-per-billion (ppb) level. An eluent of 1.5 mmol NaOH was used at a flow rate of 0.60 mL/min. The area of the sulfate peak in the chromatogram was calibrated with a set of standard solutions to determine the sulfate concentration. The uncertainty of ion concentrations measured with this method is less than 10%.

Additionally, a method including a similar IC instrument system paired with a SCIEX QTRAP electrospray-ionization tandem mass spectrometry detector allowed for detection of perchlorate at concentrations down to the low or sub-part-per-trillion (ppt) level. This method was similar to that outlined by Jiang, et al.[2], with a precision of 10%.

RESULTS

The data of the snow samples are presented in Figure 1 against a plot of ozone density assuming snow depth and time are perfectly proportional. The gap in perchlorate at depth 224 cm corresponds to a sample of insufficient volume to successfully gather a perchlorate concentration.

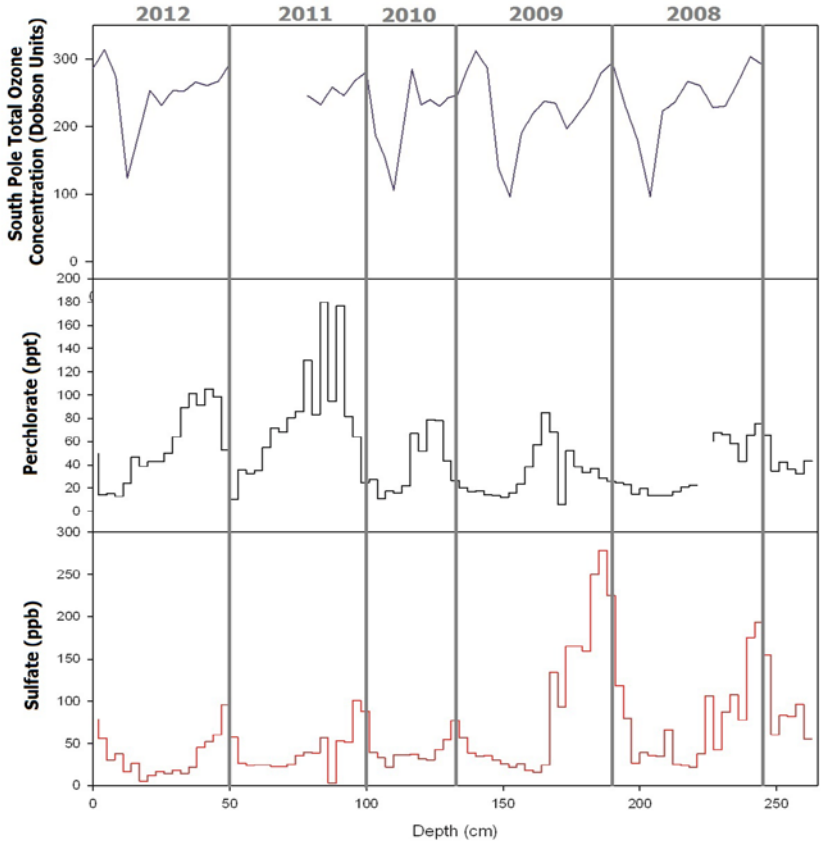

Figure 1: Measured Sulfate and Perchlorate Concentrations in Antarctic Snow Samples, and Published Total Ozone Levels[3]

The sulfate concentrations are used to mark the beginnings of each year as sulfate concentrations in Antarctica peak in the months of January and February[4]. According to the years as shown in Figure 1, it appears that perchlorate concentration reaches an annual maximum in the austral autumn, approximately the months of March and April. The lowest concentrations generally appear in the austral spring, approximately September and October, when the ozone hole is present. This is highlighted by the extreme dips in the total ozone density for this season as measured at the South Pole[3]. The collected data regarding the perchlorate concentrations shows such seasonal variation consistently for all of the years studied.

DISCUSSION

The results contradict our hypothesis in that perchlorate levels are at an annual minimum when the ozone hole is present. However, the consistency of seasonal variation in perchlorate concentration suggests there might still be a relationship between perchlorate levels and the ozone hole, although different than initially hypothesized. It is possible that the atmospheric production of perchlorate occurs primarily in the stratosphere rather than the troposphere. The decreased level of stratospheric ozone during the spring season could be a limiting factor in stratospheric perchlorate production during that season.

Extensive study on the creation of the ozone hole shows that the ozone is depleted during the austral spring because air temperature is low enough to allow formation of polar stratospheric clouds (PSCs) in the stratosphere during a period when daylight is present enough for a UV initiation of ozone depletion[5]. These PSCs act as a necessary factor in chlorine catalyst regeneration in that Cl_2 and HOCl can collect on them and return to chlorine radicals by UV photolyzing[5]. For the rest of the year, these PSCs are not present;

chlorine catalyst regeneration may not occur and thus ozone depletion is limited. It is possible that the formation of perchlorate occurs in the stratosphere alongside this chlorine catalyst regeneration reaction in such a way that they are competing. In one case, PSCs are present and allow for the chlorine catalysts to predominantly reform after ozone depletion. In the other case, PSCs are not present, and thus, perchlorate formation predominates and can be precipitated out of the atmosphere into snow. It has already been suggested that perchlorate acts as a natural sink for chlorine catalysts[1]; the data from this study reinforces this proposition.

In conclusion, the collected data establishes a seasonality in perchlorate concentrations. In comparison to total column ozone density, it appears that the presence of the ozone hole relates to decreased perchlorate concentrations. The data suggests that perchlorate production may be acting as a sink for stratospheric chlorine and thus competing with the chlorine catalytic depletion of ozone. Seasonal trends in Antarctic perchlorate concentration may be attributable to the presence of polar stratospheric clouds during the spring season.

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High Definition 3D Printing – Comparing SLA and FDM Printing Technologies

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ABSTRACT

For consumer level additive manufacturing, there are currently two main methods to 3D print objects: Stereo lithography and Fused Deposition Modeling. Both processes add material, layer by layer, to create objects. Stereo lithography (SLA) uses a UV light source to selectively cure resin while Fused Deposition Modeling (FDM) extrudes semi-liquid plastic in a specific layout to create objects. As with most technologies, both styles of printing have advantages and disadvantages. The biggest advantage of SLA 3D printing is very high resolution. SLA 3D printing can produce objects with more than double the resolution of FDM printers. The mUVe 3D SLA printer and a Makerbot Replicator 2x were used in this paper. The mUVe 3d SLA printer is different from most SLA 3D printers that use a galvanometer or Digital Laser Projector (DLP) as a light source, the mUVe 3D printer uses a UV laser mounted on a Cartesian coordinate gantry system to cure the resin. This research focuses on the difference between the Cartesian coordinate gantry system and the standard galvanometer or DLP approach. Portions of the mUVe 3D printer were redesigned and the function of the 3D printer was tested by comparing print quality after the redesign was implemented. After the design revisions were implemented, the machine produced smoother parts and consistent functionality was improved. The resolution, determined by microscope measurement analysis, of the mUVe 3D printer was found to be significantly better than an FDM printer, however, SLA printers are more difficult for consumers to calibrate and use at home. This better resolution was the result of the much smaller deposition (laser) diameter on the mUVe compared to an FDM printer (nozzle).

INTRODUCTION

3D printing has been changing the manufacturing and prototyping industries since the late 1980's [1], but it wasn't until 2009 that "desktop" 3D printers were readily available to the public [1]. A desktop 3D printer is industry jargon for a smaller, less expensive 3D printer that a typical consumer can buy. It is simple enough to operate that an average consumer can navigate the 3D printer controls without extensive training. Typically, these printers are the Fused Deposition Modeling (FDM) type which use an extruder that melts a plastic filament to build parts. In 2011, Stereo lithography (SLA) desktop 3D printers became available with the B9Creator [2] and the Form 1 [3]. SLA 3D printers use a UV (ultraviolet) curable resin and a UV light source to make solid objects. Typically, one of three different UV light sources is used: a DLP (digital light processing) projector, a laser tuned to a specific wavelength, or LEDs.

As with most differing technologies with the same end goal, there are tradeoffs between SLA and FDM. One of the main advantages of SLA 3D printing is that it has a very high resolution with very thin layers. This allows intricate details to be printed in the objects. However, the liquid resins used for SLA 3D printing can be difficult to work with – especially at the consumer level. The SLA printing process is also slower than the FDM process. The FDM process cannot print with the same resolution as an SLA printer, but the spools of plastic filament for the FDM printer are much easier to use and they are more readily available.

Since 2009, when some of the most restrictive 3D printing patents expired, the 3D printing community (also known as "makers") generated a significant amount of knowledge on the FDM process and shared this knowledge with the world via open source websites and forums. However, very little information is available on SLA printing via lasers mounted on a Cartesian coordinate gantry system, the mUVe 3D printer was chosen for this project. Purchased as a kit, this printer was built (according to specifications) and tested, re-designed with improvements to the original design, re-built, and tested. The mechanics and process of printing with this printer were compared to an FDM printer. In addition, the resolution of parts produced by each printer was compared via the layer height and "nozzle" diameter specifications.

The higher resolution of SLA style 3D printing allows makers to do more with 3D printing. One common application of SLA printing is prototyping medical instruments that require small accurate parts. FDM type 3D printers do not have high enough resolution to print these types of objects at the proper scale. Many other applications exist that would benefit from small, accurate prototypes; however, until an inexpensive and reliable SLA printer is developed, this technology will likely be used only by well-funded projects.

Technology Comparison – SLA vs FDM

Machine Setup

SLA printers require several steps to initiate a print operation and obtain a finished product. First, the reservoir is attached to the frame of the machine. Screws and an L-bracket, shown in Figure 1, are used to clamp the reservoir so that is doesn't lift/shift during printing. Next, the build plate is inserted into its mounting device. After these parts are assembled, the resin is prepared and poured into the reservoir. Proper and careful completion of each step is required for a successful print job. All of these steps require a skilled and trained operator. After each step has been completed, the print job is ready to be initiated.

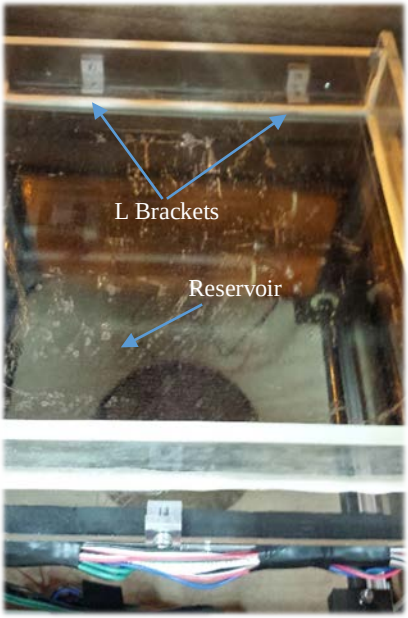

Figure 1: L Brackets and Reservoir

The operator time required to setup a traditional FDM printer is significantly less than an SLA printer and the setup procedures is much simpler, although the overall setup time is similar. The setup for an FDM print operation includes installing a spool of filament and pre-heating the machine to the proper temperatures. Neither process requires significant training or precision.

Although the setup times are roughly equivalent for the two types of printers, the setup difficulty level is much different. For SLA printing, the liquid resin can be very messy as it is difficult to keep the resin from spilling throughout the setup. Therefore, clean up after a job has finished is also time consuming. Cleaning up an SLA print job requires the removal of the entire build plate and removal of the part from the build plate. In addition, the reservoir has to be removed and leftover resin is placed back into storage. If the resin is left in the reservoir too long after the job is completed, it will cure in the reservoir, damaging the reservoir and wasting material. After the part is removed from the build

plate, it is rinsed with isopropyl alcohol to remove excess resin and give the part a nice smooth finish. At this point, the part isn't fully cured, it must still be placed in a UV chamber or in direct sunlight to finish the curing process. The cleanup process for an FDM print job only consists of removing the part from the build plate. The FDM printer is then ready for another project.

Printing Time

The ability to create objects/prototypes quickly was one of the main reasons 3D printing was created. The time and money required for a 3D printer to create a prototype is significantly less than the time and money needed to manufacture the same prototype using traditional manufacturing processes. Although printing parts on the mUVe SLA printer is still much faster than traditional manufacturing, it is much slower than an FDM printer. Printing the same part will generally take the mUVe printer anywhere from 2 to 4 times longer than a desktop FDM printer. This is due to the laser diameter being much smaller than the nozzle diameter of FDM printers and the vertical layers being much smaller on the mUVe printer. Although the SLA printer takes longer to print, it can make objects that have a greater resolution than an FDM printer. The "SDSU Campanile" referenced in Table 1 is shown in Figure 2 (Campanile is 25mm tall). This object was printed using the mUVe 3D printer. Printing would not be feasible (at this scale) on an FDM printer because of the level of resolution required. Table 1 shows a comparison of printing times for the mUVe printer and a standard desktop FDM printer. (Note – Print times provided are theoretical time calculations generated by the printing software used to operate the printer, Repetier-Host [5]. While these times are usually not exact, they do provide approximate printing times.)

Table 1: SLA and FDM[1] Printing Time Comparison

Part	mUVe Time (min)	FDM Time (min)
SDSU Coin	129m	27m
Campanile	167m	45m
Tensile Test Sample	260	115m

Figure 2: SDSU Campanile Printed on mUVe 3D

Resolution

There are several methods used across the industry to compare 3D printer resolution. The simplest and most common method uses the nozzle (or laser) diameter and the minimum vertical layer height. By this standard, the mUVe performs much better than the FDM printer. The mUVe 3D printer can support a laser diameter as small as 0.1mm and a vertical layer height of 0.1mm. The standard nozzle diameter for a desktop FDM printers is 0.4mm and a vertical layer height of 0.2mm.

mUVe 3D Printer Design

mUVe 3D Printer Mechanical Systems

The SLA printer used in this research study (the mUVe 3D printer) was equipped with a 50mW 405nm laser mounted to a Cartesian coordinate gantry system that allows the laser

to slide along an X and Y axis to cure the resin. For this particular style of SLA printer, the build plate only moves up in the Z axis, allowing the objects to be built layer by layer.

The mUVe 3D printer kit was assembled as specified by the manufacturer. Figure 3 shows the fully assembled printer and the location where the resin is poured into the reservoir. In this figure, the build plate is shown above the reservoir. To initiate the print job, the build plate is lowered into the reservoir so that the first layer of the print is cured onto the build plate. The X and Y axis are controlled by four stepper motors, two on each axis – this is two more motors than required for most FDM style printers.

To build the first layer of the object, the build plate is lowered to a distance of one layer thickness away from the reservoir bottom and then raised by increments of one layer thickness after each layer is completed. After each layer is cured by the laser, the resin is attached to both the reservoir bottom and the build plate (or the previous layer of the part). If the build plate is raised equally and simultaneously on both sides of the build plate, there is a high probability that the printed object will either become separated from the build plate and stay attached to the reservoir, or the object will fragment into several pieces with some pieces sticking to the reservoir and some pieces sticking to the build plate. Thus, the build plate is "peeled" away from the reservoir one side at a time, with the hope that the printed layer only adheres to the previous layer instead of the reservoir. In a "peel move", the left side of the build plate is slowly raised to peel the left side of the object from the reservoir, followed by the right side of the build plate raising to complete the peel. Both sides of the build plate are then slowly lowered to a distance of one layer thickness above the previous z-axis position.

Due to the peel move, the Z-axis of the machine needs to be able to tilt/flex to allow the build plate to be peeled away from the reservoir. As specified by the manufacturer, the Z-axis setup shown on the machine in Figure 3 was unstable and did not slide smoothly up and down. In addition, when the peel move was being executed the entire Z-axis frame bent to accomplish this move. As a result, a new Z-axis was designed and built as shown in Figure 4. This new design allows the cross member connecting the two threaded rods to rotate freely at the connection point. The frame of the Z-axis is rigid, but the peel move can still be executed because the build plate rotates. Each SLA printing company tackles this issue of peeling the parts in a slightly different manner. Some use the Z-axis tilt method,

while others tilt the reservoir instead of the build plate, and some rotate the reservoir to shear the bond between the part and the reservoir. While the Z-axis for the mUVe 3D printer was being redesigned, the mUVe company was also developing a similar redesign of the Z-axis.

In addition to the peel movement, a non-stick coating is applied to the reservoir to help prevent the parts from sticking to the reservoir. The non-stick coating process is also a tedious and difficult process for SLA printing preparation. This task may be too difficult for the average consumer.

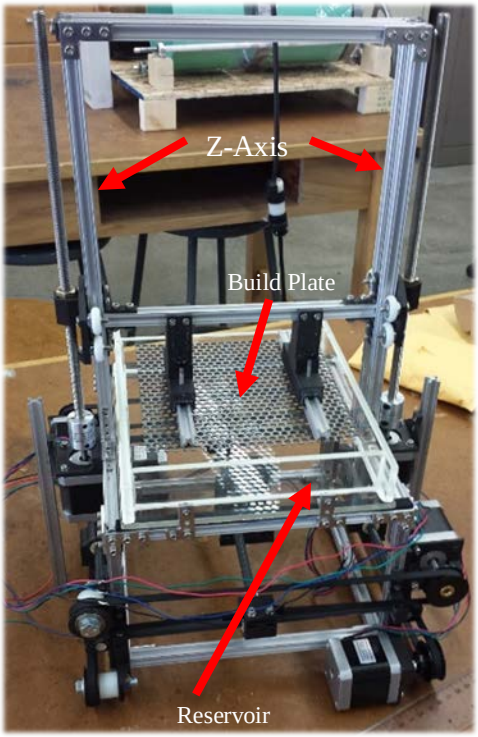

Figure 3: mUVe 3D SLA Printer Build Process

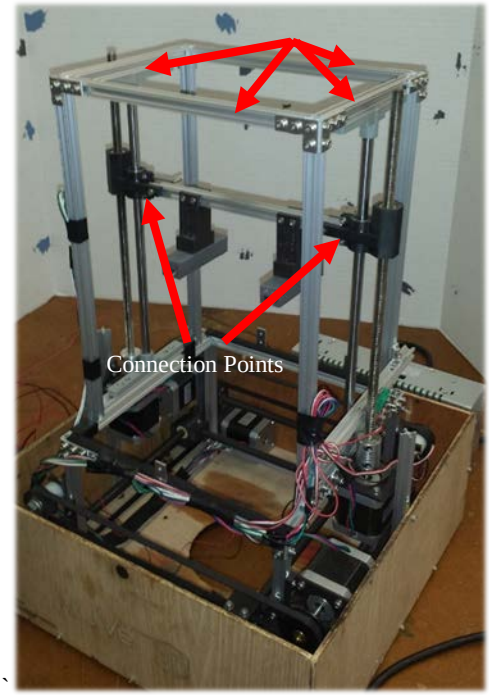

Figure 4: New Z-Axis Design

mUVe 3D Printer Electronics

An Arduino Mega control board with a RAMPS 1.4 shield were used to control the printer along with a modified open-source “Marlin” code, all of which is commonly used within the 3D printing community. A RAMPS 1.4 shield is a separate printed circuit board that attaches to an Arduino Mega microcontroller which controls stepper motors as well as extruders and fans [4]. The mUVe 3D printer is custom enough that several modifications to the Marlin code were required to account for these differences, including an increased number of motors to drive the X and Y axis, the removal of all temperature sensing and limitations based on temperatures, and the reconfiguration of end stop locations to ensure the machine knew the correct location of the laser and would not crash and damage itself.

Several options available within the RAMPS 1.4 Arduino shield to adjust the micro stepping of the motors. For most 3D printers, 1/16 micro stepping is used, however, the mUVe 3D printer uses 1/16 micro stepping only for the x- and y-axis, while a¼ micro stepping is used for the z-axis to provide the Z-motors more torque to peel the object from the reservoir.

The electronics to control and power the 50mW laser are a significant deviation from the standard FDM printer electronics. The laser is powered by an external laser driver and a transistor to control voltage. The RAMPS 1.4 shield is configured to use FDM extruders, so the addition of laser drivers is a significant change from normal wiring and operation. All of the electronics are designed to move the laser along the gantry system according to G code commands (similar to FDM 3D printers and traditional CNC machining).

In the mUVe system, the software package “Repetier-Host” [5] was used to control the mUVe printer along with sending G code commands to the printer. To generate the proper G code commands, the software package Slic3r [6] was used to generate a G-code, which “slices” the object to be printed into layers which are then printed layer by layer. This combination of Slic3r and Repetier-Host is very common in the additive manufacturing community and is considered standard among the open sourced desktop maker community.

Laser Calibration Process

After the printer was assembled, all electronics were connected properly and the correct software was loaded to the Arduino, the machine still required adjustment and calibration. Adjusting the laser is a two-step process, including focusing the laser and adjusting the diaphragm to filter out any diffraction. The laser and diaphragm assembly are shown in Figure 5.

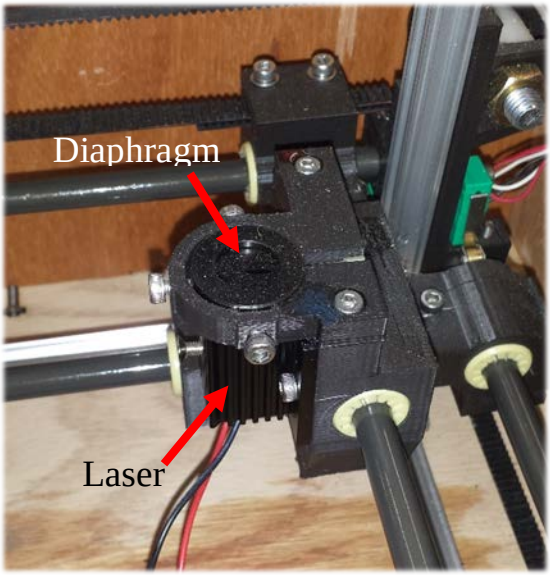

Figure 5: Laser and Diaphragm

The laser was focused by placing a sheet of paper in the reservoir and turning the focus adjustment screw on the laser until there was a point beam shown on the paper. Next, the diaphragm was installed and adjusted until the point on the paper was as small and bright as possible.

One of the most important and difficult calibration steps is determining the diameter of the laser beam. In a traditional FDM 3D printer, the nozzle is a fixed diameter hole drilled through a brass cone, and thus the diameter of each line is known exactly. In the case of the mUVe 3D printer, the “nozzle” diameter is the diameter of the laser. For this laser and diaphragm, the diameter should nominally be 0.1 mm, but can vary by adjusting the diaphragm opening. Because of the small size of the laser point and the manual hand adjustment required to adjust the diaphragm, it is very difficult to get the laser diameter to the exact nominal measurement. It is also very difficult to measure the exact size of the laser diameter using traditional measuring techniques (i.e. a caliper or ruler). With this in mind, the laser diameter was measured by creating a hollow square in which there was a

known number of laser passes based on the assumed laser diameter. For example, if the laser diameter was assumed 0.1mm, the hollow square was made with a wall thickness of 1 mm which would mean 10 passes of the laser. Figure 6 shows a theoretical laser path for the hollow square with 1mm thick walls. This theoretical rendering is provided by the Repetier-Host software to allow the user to understand how the part will be manufactured before the process starts.

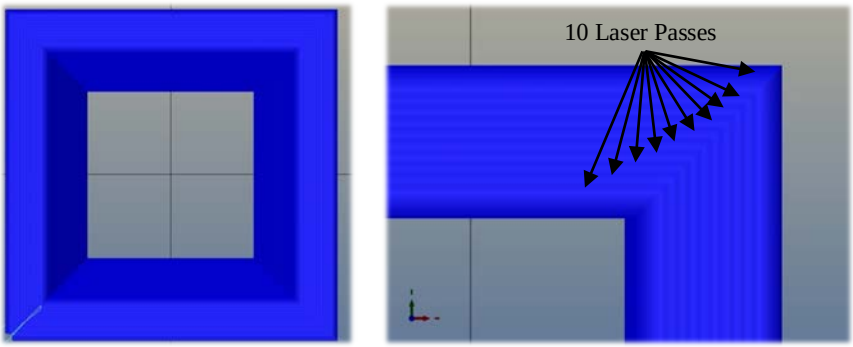

Figure 6: Calibration Cube Theoretical Rendering

This is still too small to accurately measure using traditional measurement techniques, so the calibration cube was analyzed under a microscope and the individual passes were counted and line thicknesses were measured. If the lines created by the laser had gaps in between them, the laser was smaller than assumed. If the lines overlapped, the laser was larger than assumed. Using this technique, and several iterations, the laser diameter was measured and proper adjustments were made in the software to account for the proper laser diameter. This is not a procedure that the average consumer could be reasonably expected to recreate without significant investment in microscopy equipment.

RESULTS

Laser Diameter

The laser diameter was measured using the process previously discussed. The laser diameter can be changed at any time by adjusting the diaphragm connected to the laser, so this process is necessary any time the laser diaphragm is adjusted, the printer is moved or significantly bumped. Before the calibration process, the laser diameter was assumed to be 0.1mm. A hollow cube was made with 1mm wall thickness (10 passes of the laser) as shown in Figure 7. The picture on the left shows the entire hollow cube at 20x magnified. At 20x, the object looks fine, but we know from previous attempts at printing complex objects that the laser diameter is not adjusted properly. At 150x, it is difficult to tell where one laser pass begins and one ends, suggesting overlapping laser paths. This indicates that the laser diameter is greater than 0.1mm.

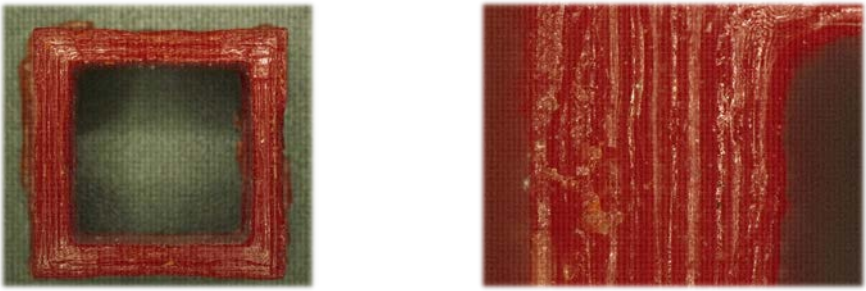

Figure 7: 0.1 mm Laser Diameter (20x – left, 150x – right)

Next, the laser diameter was adjusted in the Repetier-Host software to 0.2mm. The same hollow square was made – except with a wall thickness of 2mm (10 passes of the laser). Figure 8 shows the result of this print with the updated settings. As shown in the figure, the laser diameter was less than 0.2mm because there are clearly gaps in between passes. The microscope was then used to measure the thickness of a single pass. The passes were measured to be an average of 0.14mm.

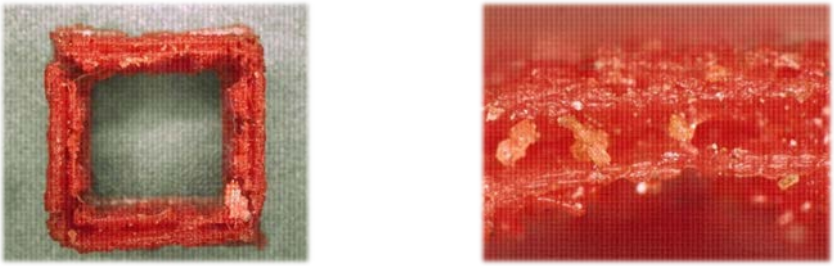

Figure 8: 0.2mm Laser Diameter (20x – left, 200x – right)

With the new laser diameter setting of 0.14mm, another hollow cube was made with a wall thickness of 1.4mm (10 passes). A laser diameter of 0.14mm is a somewhat odd diameter to use, however, it is easier to adjust the laser diameter size in the software to match the actual laser diameter rather than adjusting the diameter of the laser and starting the process over again. Figure 9 shows that with this setting, the hollow cube had very smooth layer lines and nice connection between toolpaths, further indicating that this is the correct laser diameter. The laser paths aren't perfect because of the variations in the laser travel due to the gantry system moving the laser.

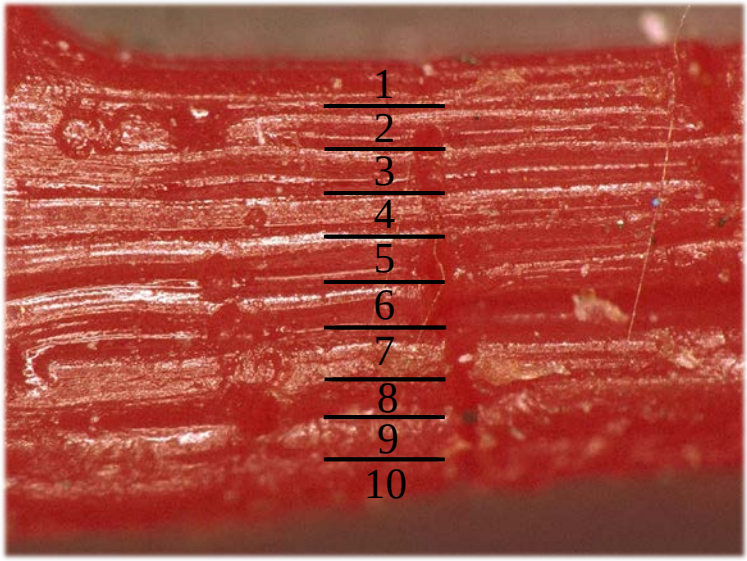

Figure 9: Correct Laser Diameter: 0.14mm (200x)

Z-Axis Design

The Z-axis design specified by the manufacture was not very stable and did not operate smoothly. As seen in Figure 3, the two Z-axis support members were mounted very close together, leaving the system very unstable and inconsistent. Furthermore, the rollers that were used to slide the Z-axis did not perfectly align with the channels they were designated to slide through, decreasing the consistency because sometimes the rollers would catch in their grooves causing the Z-axis to not be perfectly level. For these reasons, the Z-axis was redesigned. With the new design implemented, parts were repeatedly made with identical results showing the consistency of the new design. In fact, no parts were successfully printed using the original Z-axis design because the travel of the axis was not smooth enough to allow for a single layer to build properly.

DISCUSSION

Many ideas and projects require parts with complex geometry and small features that a standard FDM 3D printer is not capable of printing. An inexpensive, reliable, high definition SLA 3D printer will allow projects to be completed faster and cheaper through rapid and simple prototyping. Although successful prints were produced with this machine, there are opportunities to make this machine more reliable and produce higher quality.

Another option to cure the resin is to use a DLP projector as a UV light source instead of the laser on a gantry system that is currently used. This upgrade would increase the quality and reliability of this 3D printer and ease of use for the common user. The DLP projector would remove the gantry system that is responsible for many of the inconsistencies and problems with the current system. The laser diameter measurement process would also be avoided; another cause of significant set up time and frustration.

Another benefit of the DLP upgrade is the speed of a DLP system. Because a DLP projector produces an entire layer at a time, the print time is significantly reduced. In DLP projection systems, the speed is reduced to print times similar to FDM printers.

CONCLUSION

FDM and SLA 3D printers operate on the same principles of creating objects by adding layers of material, also known as additive manufacturing. As with many competing technologies, it is difficult to compare and determine which type is “better”. Both types of printers have strengths and weaknesses. An SLA printer can produce products with a much higher resolution than an FDM printer, but does so at the expense of speed. In addition, the process of using an SLA printer is more complicated than an FDM printer because the resin can be difficult to work with and overall the setup and clean up times are longer.

There are many applications to support both types of printers. In many general 3D printing applications, an FDM printer is appropriate. The setup and cleanup is simple and the printing process will be quick. However, when extra detail is needed, FDM printers will

not be adequate and an SLA printer will be required. Therefore, both pieces of equipment are essential for modern day engineering fabrication and prototyping laboratories.

ACKNOWLEDGEMENTS

A special thanks to my advisor, Dr. Todd Letcher, for initiating this research and all the advice and help throughout the research project. Also, thank you to Mr. Joel McCue for the tour of Falcon Plastics in Brookings and for the insight to the Form 1 printer used at their facility. Last, thank you to the Mechanical Engineering Department and the METLAB for use of testing equipment.

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Applications of Ultrasonic Non-Destructive Testing in 3D Printing

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ABSTRACT

To determine the capabilities of ultrasonic non-destructive testing methods in 3D printing, test specimens were designed to evaluate advantages and limitations in using ultrasoundC-scanning to determine 3D printing effectiveness. A Matlab program was written to process the data obtained from the UTwin software and ultrasound tank, as well as produce 3D models of the parts being tested. Thickness, types of features, and various printing methods were evaluated to determine how these characteristics vary in effectiveness of C-scan imaging. Results from the UTwin software C-scans were compared to the Matlab program's imaging and three-dimensional modeling. After determining the capabilities of ultrasonic non-destructive testing methods in 3D printing, applications where the technology could be applied to further the additive manufacturing industry are discussed, and the economic potential of the technology in the industry was identified.

Keywords: Non-Destructive Testing, 3D Printing, C-scan, capabilities, applications

INTRODUCTION

The emergence of 3D printing as a viable, effective method of manufacturing in the past few decades [1] has led to great strides in the technologies and methods used in additive manufacturing process. As excitement increased around this relatively new manufacturing method, the technology and sophistication of these machines also increased rapidly [2]. More organizations are turning to additive manufacturing processes to aid in their current

operations, as well as develop new and innovative ways to use the technology. One of these organizations is NASA, which on November 24, 2014 became the first organization to 3D print a part in space at the International Space Station [3]. With additive manufacturing processes opening new doors in the manufacturing industry, some of the focus has now moved from expanding the capabilities of 3D printing, to testing and improving the methods for inspecting 3D printed components. Non-destructive testing methods of completed 3D printed parts is one of these new focus areas as researchers search for effective ways to verify the quality and integrity of 3D printed parts.

Using non-destructive testing (NDT) methods to verify quality and integrity of 3D-printed parts is obviously desirable because checking the part for failures and deficiencies while not altering or damaging the part is essential. Testing during the printing process may also offer material and time savings because as a failure is detected, the printer could be automatically shut down before additional parts are manufactured that don't pass quality standards. Because of the wide variety of capabilities and applications of additive manufacturing, an NDT method which allows for detection of both surface and interior features of a part is valuable. The ultrasonic non-destructive testing methods used in this study fulfill both of these criteria.

Ultrasonic non-destructive testing methods use sound waves to both measure and detect features or failures on or within a specimen. This is done by using a high frequency pulsating wave signal which is sent and received by a transducer. The waves sent from the transducer diffuse through a couplant, often water or a custom couplant product, and then through a test specimen. The waves then reflect off discontinuities in the material, which can be a surface, flaw, feature, or defect. As the transducer receives the wave signal after reflection, the time difference between the sent and received waves can be correlated to a depth or distance from the transducer at which the reflection took place. Figure 1 provides a simplified graphic of wave reflection within a part and the receiving transducer signal.

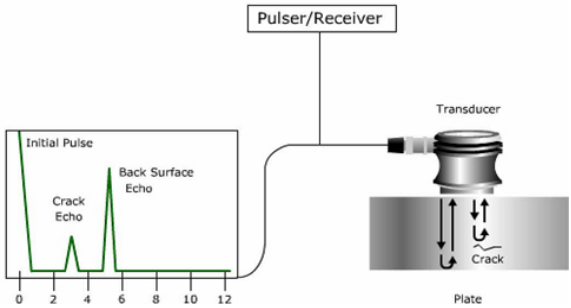

Figure 1: Wave Reflection and Receiver Signal [4]

By using this testing method over a surface, a C-scan image (a 2D image in which colors represent height or depth) can be determined by analyzing the waveform signal, which describes the features of a specimen along a plane of the part or on a surface.

The goal of this study is to test whether ultrasound testing is a viable option for non-destructive inspection of plastic 3D printer parts. Ultrasound techniques are usually very effective methods for non-destructive testing of bulk consistent materials, however, 3D printed parts are not normal bulk materials. Odd surfaces and air gaps exist between layers and may cause significant signal disruptions for ultrasound waves. Test specimens were designed to determine the effectiveness of ultrasonic NDT methods in evaluating different features of the 3D printed parts created. The capabilities of ultrasonic non-destructive testing in 3D printing were then evaluated, as well as a description given of the methods developed to test these capabilities and evaluate the signal data received from the transducer. Finally, the applications of this NDT method in the additive manufacturing industry are discussed.

METHODS

Before an evaluation of the capabilities of ultrasonic NDT methods could be completed, effective test specimens needed were designed to evaluate testing capabilities. When these test specimens were designed, specific characteristics were designed into the specimen to evaluate both the capabilities of ultrasonic NDT as well as any possible limiting factors. The two characteristics focused on during the evaluation process were thickness of the test specimen and internal or backside surface features.

To determine if the thickness of a test specimen related to testing effectiveness, the specimens were designed with varying thicknesses and the same distinct features on the backside surface. By taking a C-scan of the test specimens and comparing the features detected on the backside surface of the specimen, a determination of whether thickness affects ultrasonic NDT capabilities could be justified. A sample test specimen used to determine the effects of thickness on ultrasonic C-scan capabilities is shown in Figure 2. Also considered for evaluation were the effects of feature geometry on a test specimen. The test specimen shown in Figure 2 attempts to vary types of surface features in size and geometry to better understand the capabilities of ultrasonic C-scan capabilities.

Figure 2: Backside Feature of Test Specimen

After the specimen shown in Figure 2 was designed and 3D printed, a specimen devoted to studying the effects of variations in size and geometry was created. Figure 3 displays this specimen, which evaluates the accuracy and precision of the C-scan testing. Surface features were designed as small as 1 mm.

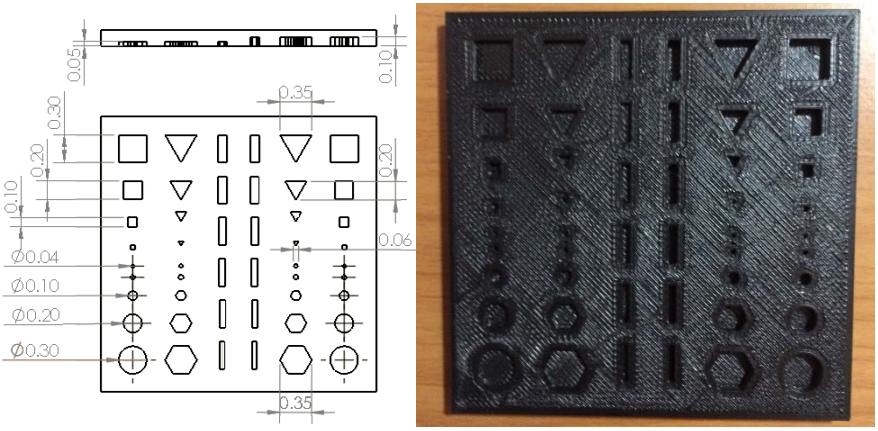

Figure 3: Precision Evaluation Specimen

One of the focuses of the research was the detection of internal features of an object. Using ultrasound C-scan technology, features on the backside surface of a solid object can be detected [4]. However, it is unclear how effective this technology will be when detecting internal features of 3D printed specimens due to the layers of plastic and the different tool paths used to create the layers resulting in disturbances in the ultrasound waves. Furthermore, it was desired to determine if secondary internal features and backside surfaces could be detected along with these types of primary features. The specimen shown in Figure 4 was designed to determine the capabilities of ultrasound NDT in detecting internal features. The boxes shown in dotted lines are located within the specimen; therefore, from the outside, this specimen looks like a rectangular block of plastic.

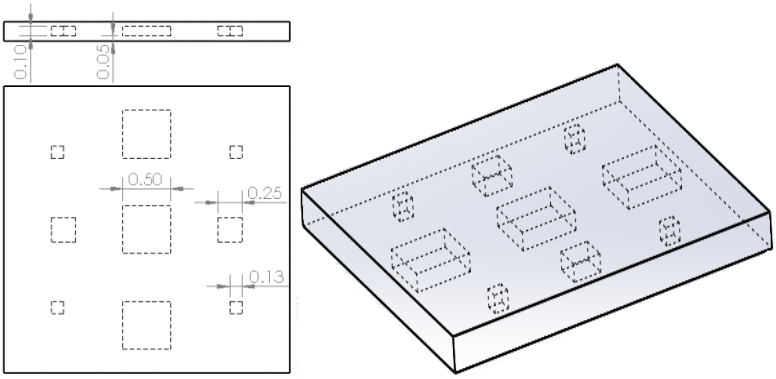

Figure 4: Internal Features Specimen

The three specimens discussed above possess characteristics which aid in the determination of the effectivenss of ultrasound C-scanning. All specimens were printed in Acrylonitrile Butadiene Styrene (ABS) plastic – one of the two most common 3D printing materials. After successful specimens were printed at 100% infill, alternative specimens at varying infills, between 20% and 40%, were tested to determine if infill and hollowness would be a limiting factor in ultrasound NDT of 3D printed specimens.

After the design process and printing of the models was completed, the UTwin software was utilized with the ultrasound tank to evaluate the specimens using a C-scan test. Before running a test, calibration and positioning of both the 10 MHz transducer and specimen to be scanned was completed. The UTwin software's C-scan settings were configured to a resolution of 0.5 mm and a C-scan speed of 10 mm/sec. Figure 5 shows the setup used for scanning a specimen.

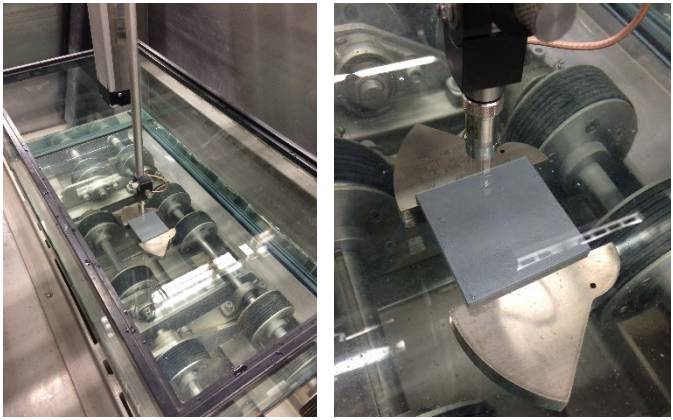

Figure 5: Ultrasound Tank Setup for C-scan (left), Close up of Specimen Setup (right)

When scanning a specimen, a waveform output of the C-scan was produced by the UTwin software. A data file was also generated which contained the waveform depiction of each pixel of the C-scan which was further analyzed in Matlab and used to produce both 2D and 3D representations of the specimens scanned.

RESULTS

The Matlab code was originally written to provide a verification that data logged by a C-scan test could be utilized to produce models, surface images, etc., that went beyond that of the current software used with the equipment. Though the code written did provide this, the most valuable asset it offered related to the time it took to administer a single C-scan test using the UTwin software compared to using the developed Matlab code.

The UTwin software uses “gate” position (“gates” are time period specified to look for signal changes), width, and amplitude to analyze the waveform data received from the transducer. UTwin offers the ability to set up a maximum of four gates which then output four different C-scan results. Although the ability to attempt four different scans at once is valuable, the draw back to the UTwin software is the time it takes for a useful, accurate scan to be obtained. For a high resolution scan of the samples, the average time to scan each sample was 1.5 hours. Furthermore, if a scan is unsuccessful due to gate

misplacement, another scan of equal time must be produced to conduct another analysis. In the early stages of the research process, this was identified as a weakness and was then targeted as a secondary goal of the project.

The Matlab program offers three distinct advantages over UTwin. First, only a single ultrasound scan with proper calibration needs to be run with the raw data exported, which allows the data to be analyzed away from the equipment. Because all of the original data is exported, there is never a need to run another scan due to gate placement failure. The Matlab program is able to run a C-scan test on previously exported data and produce a C-scan image in an average of 15.32 seconds, which saves a significant amount of time rather than run another 1.5 hour scan using only the UTwin software.

The two other major advantages of the program dealt with expanding the capabilities of the UTwin software. Writing a custom Matlab program to analyze the data means there are no limitations on the number of gates used to yield a successful C-scan image. The Matlab program could be expanded to as many gates as necessary, providing the ability to determine multiple surfaces during a test and compile those surfaces into a three-dimensional model.

The third advantage of the Matlab program over the UTwin software is the ability to produce three-dimensional models as well as easily analyze specimen thicknesses. Using the program's ability to analyze multiple surfaces, three-dimensional models could easily be compiled by knowing the depth or thickness at which the surface features occurred. The results and capabilities of the program are discussed below.

C-scan Results and Comparisons

To determine if ultrasonic NDT is a viable method of testing 3D printed parts, the specimen shown in Figure 2 was first tested to detect its backside surface features. This test was first run using the UTwin software. The initial test was meant to determine if ultrasonic NDT could be used to pick up backside surface features of a 3D printed part. A portion of the results from UTwin are shown in Figure 6.

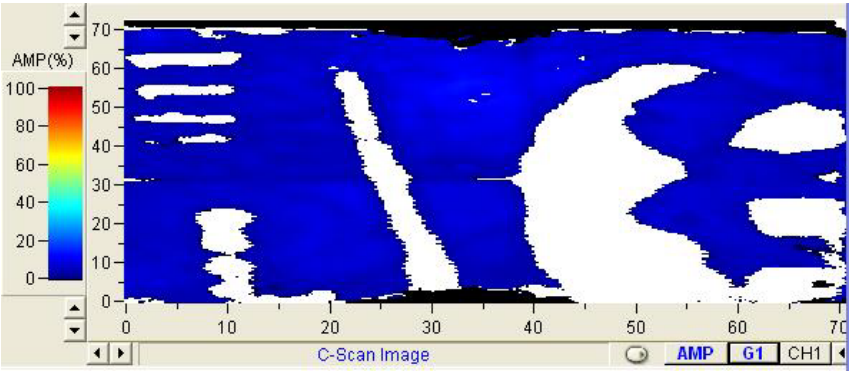

Figure 6: UTwin C-scan Initial Test Results

From Figure 6, it was concluded that the C-scan was able to locate backside surface features designed into the part such as the long diagonal line, the multiple slots, and the randomly shaped round feature. This provided a basis to move forward in developing the Matlab program to further analyze the ultrasound data as well as determine the capability of ultrasonic testing to be used in the additive manufacturing industry.

Next, the Matlab program developed was used to analyze the same data and yielded the 3D model shown in Figure 7 of the specimen pictured in Figure 2 (similar to what is shown in Figure 6 using the UTwin software).

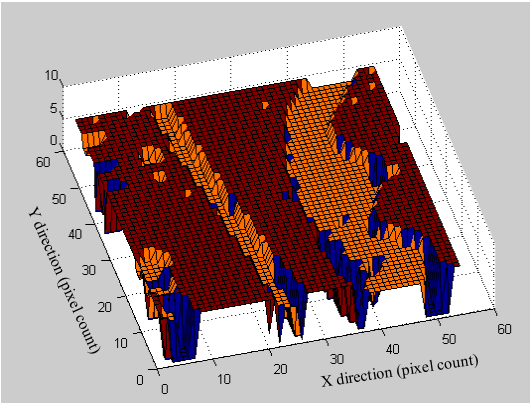

Figure 7: Initial thickness test (in µs) for backside surface geometry detection

The Matlab program was written so each surface depth corresponded to a specific color. The backside (the direction of the specimen facing the direction of the UT sensor) is shown in red, while the surface features designed into the specimen are shown in orange. The blue features of the specimen correspond to data that should be neglected or were not captured by the two gates set during the test. From the average gate position used to detect the backside surface of the specimen, the thickness of the specimen was determined to be 6.7 mm. This compared to the actual specimen thickness of 7 mm, yielding a 4.29% error (thickness determined using nominal wave propagation speeds through non-3D printed ABS plastic sheets [5]).

To evaluate the capabilities of ultrasonic NDT to detect small details on a 3D printed part, the precision evaluation specimen was tested with its features facing up. Although it has been proven that ultrasonic testing is able to pick up these types of features on the top surface of a specimen, 3D printed parts may vary due to the difference in how they are produced. Figure 8 displays the image yielded from the C-scan results produced in the Matlab program.

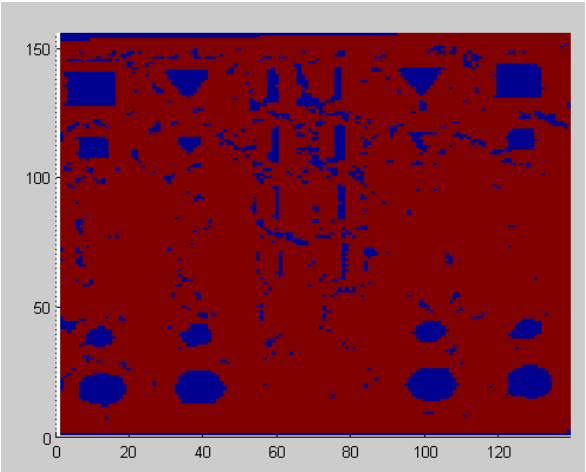

Figure 8: C-scan Image of Precision Evaluation Specimen by pixel

The C-scan was able to detect all of the shape features with a size greater than or equal to 5 mm and parts of features less than 5mm, as shown in Figure 8. The inability of the ultrasound C-scan to pick up the smaller features of the part was unexpected, but is most

likely attributed to the uneven, layered surface of the specimen due to the additive layering process of 3D printing. Both depths tested in this specimen yielded similar results, indicating feature depth on the top surface of a part is not a limiting factor in testing capabilities. Noise picked up during testing also caused some error within the imaging of the specimen as shown in Figure 8. These inaccuracies in the imaging due to noise could possibly be filtered out using a function within the Matlab code, and should be considered in future testing.

The internal features specimen (shown in Figure 4) was then tested to evaluate the capabilities of the ultrasound tank in detecting features enclosed within a specimen. Because of the ability to measure the outside features of 3D printed parts rather easily, this feature was identified as one of the more valuable assets of ultrasound NDT. Figure 9 displays the results yielded from the UTwin software when the initial C-scan was run.

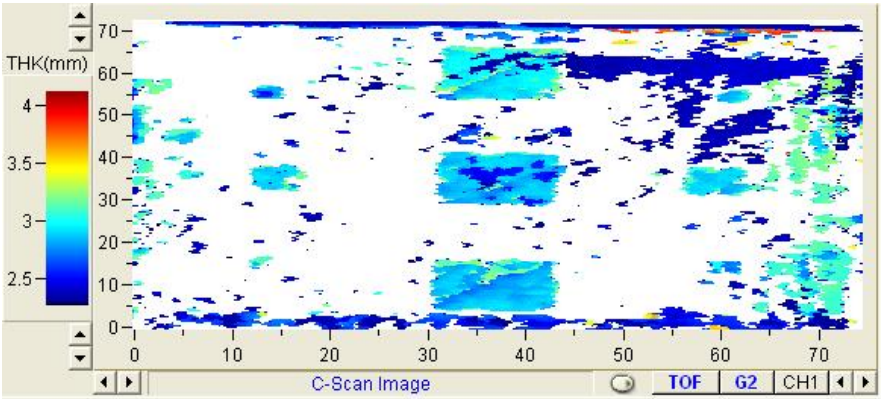

Figure 9: UTwin C-scan Image

From Figure 9, internal features of the specimen were detected (as determined by the UTwin software). The aqua color in the figure definitively represents the feature designed into the specimen. Although this initial scan was successful in finding the hidden features, a relatively large amount of noise also distorted the results. The raw data of Figure 9 were imported into the Matlab program to be further analyzed. Figure 10 displays the C-scan image results from the Matlab program.

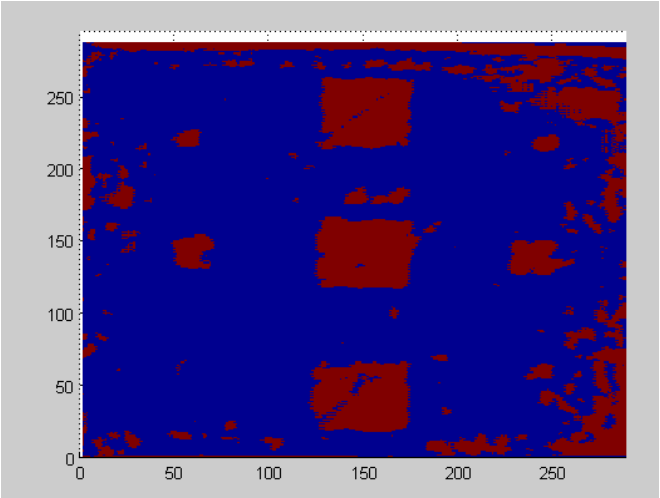

Figure 10: Internal Features Detected by Matlab Program

For this specimen, the image yielded from the Matlab program indicated a thickness of 1.31 mm before the internal features occur. This compares very well to the design thickness of 1.27mm and yields a 3.15% error. This error is again most likely attributed to the difference in the published wave speed through uniform bulk plastic compared to the plastic layers which make up a 3D printed part. The data was further analyzed to detect the secondary surface of the hollow internal features, but these secondary surfaces were not detected. However, from the C-scan results of both the UTwin software and the Matlab program, it was determined that an internal feature as small as 10.9 mm^2 can be detected on a surface located 1.27 mm away from the external surface of a part.

After the specimens printed as solid objects (100% infill) were tested, the initial test specimen was printed with 30% infill to determine if this type of print would pose a problem in producing an effective C-scan image. Figure 11 shows a cross section of a specimen printed with 30% infill.

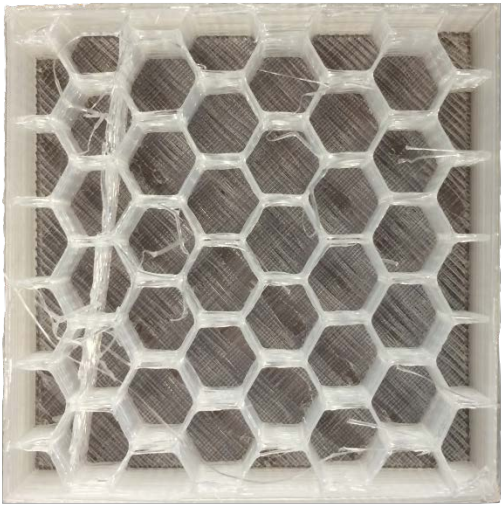

Figure 11: 30% Infill Specimen Cross Section

A C-scan image of the 30% infill specimen produced from the UTwin software is shown in Figure 12. Following the results of the internal features specimen, these results are expected.

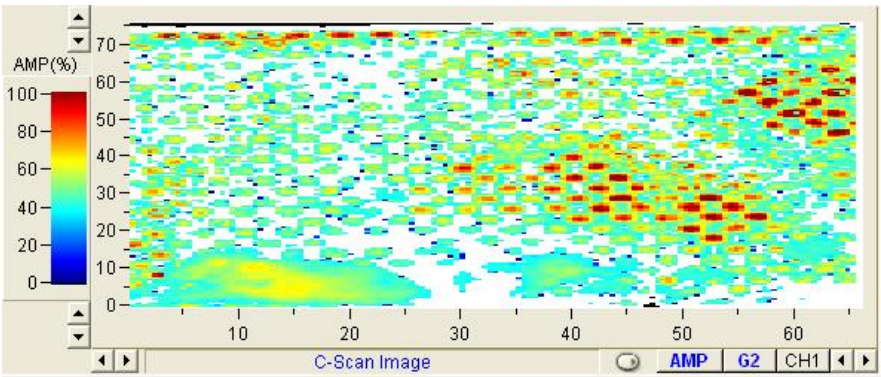

Figure 12: Initial Specimen C-scan Image with 30% Infill

From Figure 12, the honeycomb pattern, which the 3D printers use to fill a specimen partially as support, can be seen very clearly. This type of result was expected due to the air within the honeycomb structures possessing a very low density, which reduces/eliminates

the sound wave propagation back to the transducer. Although this type of scan was not successful in detecting the backside surface features of the specimen, it should be considered that the infill was detected by the ultrasound tank fairly effectively. This could be valuable to manufacturers using infill within their 3D printed parts where structural integrity is essential.

DISCUSSION

Through the testing of multiple types of specimens, the capabilities and limitations of ultrasonic NDT methods in the 3D printing industry were determined, and different types of applications of this technology should be considered. The C-scans obtained of the initial test specimen and the internal features specimen identified that ultrasonic testing is able to detect backside surface and internal features of 3D printed parts. This type of application could be very useful to manufacturers who are unable to measure tolerances and part features within an assembly or printed part, but desire to verify the accuracy and success of their print. The major limitation of this testing method is that the testing would need to be completed after the part is manufactured because a high density fluid must be present between the part and transducer.

Two major limitations of ultrasonic NDT of 3D printed specimens were determined from the findings discussed above. Because wave propagation and reflection are related to the density of the material being tested, the reflection of the waves on the second and third layers of a part decreased rapidly, thus posing problems for this type of testing as thickness of a part increases. This, along with the fact that additive manufacturing uses layers to build a part, makes deciphering the waveforms being received by the transducer difficult. This difficulty poses an even greater burden as thickness is increased, and as the layers propagate a greater amount of waves and reduce the return amplitude.

Another limitation determined during the research was the occurrence observed with the infill specimens. Because air is within the specimens when infill is used, wave propagation to layers beyond the air located within the specimen is nearly impossible as air’s density is so small relative to water and the many types of materials tested.

Through the testing of these specimens, two relevant applications for NDT methods were identified. First, through the use of ultrasonic NDT methods after a part is printed, the accuracy, integrity, and quality of the internal features of a part can be tested. Although ultrasonic NDT methods may not be practical in real-time 3D printer monitoring due to the need of a transfer fluid/couplant, a laser or other type of NDT method could be used in conjunction with a program such as Matlab to verify successful progress of a print job by scanning the outside features of an ongoing print. A program would then cross-reference the expected shape and features of the part being printed at the given time or step and determine if the print is proceeding successfully or not. This type of program could save both material and money during large prints, which require a significant amount of time and material.

Although the applications of ultrasonic NDT methods in the 3D printing industry appear limited, the Matlab program developed during this project to better analyze the ultrasound data collected is still a valuable tool that can be used in other material analysis projects completely unrelated to 3D printing. Further testing and analysis should be done to explore more types of applications for the Matlab program and to determine how well the program handles a type of sample/material that is better suited for ultrasonic NDT methods.

ACKNOWLEDGEMENTS

I would like to thank Dr. Jikai Du for his introduction into NDT methods and the uses and theory of ultrasonic testing applied to C-scan imaging. I would also like to thank the SDSU METLAB and its staff for providing the equipment used during testing. Last, I would like to thank my advisor for this project, Dr. Todd Letcher for all the hard work he puts forth solely for his students and the betterment of their education.

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The Legal Minimum Drinking Age: What Good Does it Do?

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ABSTRACT

Underage drinking is a huge problem in the United States. Rational choice and deterrence theories suggest that the best way to deal with the problem is to create and enforce strict laws against underage drinking. However, are these truly effective mechanisms in this regard? This paper explores these issues by analyzing underage alcohol consumption patterns in three sets of states classified as “lax”, “strict” and “typical” in terms of their underage drinking laws using data from the 2012 National Survey of Drug Use and Health (NSDUH). T-tests did not show significant differences between underage alcohol use and binge drinking in the six randomly sampled typical states, strict states and the national averages. T-test suggested that alcohol use in the lax states was somewhat lower than the national average. Logistic regression results did not show a significant difference in the rates of underage alcohol use and binge drinking across the three different categories. They do, however, show a slight decrease in underage drinking over time, holding state type constant. The findings suggest that perhaps tougher laws, in and of themselves, do little to address this problem of underage alcohol consumption.

Keywords: underage drinking, alcohol legislation, deviant behavior

INTRODUCTION

Consumption of alcohol by minors has an estimated societal cost of $61.9 billion per year (Lipperman-Kreda et al, 2010). Alcohol consumption by underage Americans is implicated

in some 3,500 deaths, 1,200 cases of fetal alcohol syndrome, and triggered 57,000 admissions into alcohol treatment programs (Bonnie and O'Connell, 2004). Then there are the unwanted pregnancies, the suicides, the kids who use or abuse alcohol and get kicked out of school (Bonnie and O'Connell, 2004). Consequently, we try and fix the problem the way we know best – legislation. One hypothesized way to reduce alcohol use is by restricting access (Erickson, 2014). Exploring the efficacy of current alcohol legislation is essential to developing a comprehensive plan to combat the social costs of underage alcohol consumption.

Background

A discussion of underage drinking is best prefaced with a historical understanding of the evolution of the minimum drinking age standard. During much of our nation's history, alcohol was predominately free of laws and regulations until the zeal of a growing temperance movement climaxed with the passage of the 18th amendment in 1919 and the beginning of the Prohibition era in the United States (Wolfson and Hourigan, 1997). The Volstead Act of 1920 provided legislation for enforcement of the newly adopted 18th amendment and with the amendment's repeal in 1933, birthed a wealth of alcohol related regulations (Wolfson and Hourigan, 1997). A legal minimum drinking age of 21 years was established across the nation and individual states classified underage possession and/or consumption as a statutory offense (Wolfson and Hourigan, 1997).

In 1971, the 26th amendment was ratified, spurring 31 states to drop their minimum legal drinking age in response to the lowering of the voting age from 21 to 18 (Wolfson and Hourigan, 1997). After seven years with a reduced minimum drinking age, Michigan was the first in a slow trend to restore the 21 years of age requirement. By 1984, states without a legal minimum drinking age of at least 21 were effectively forced by the federal government and faced losing significant funding for highways if the age requirement was not adopted (Office of Inspector General, 4). By 1988, the legal minimum age of 21 was collectively applied to the U.S with each state ratifying its own variation of legislation (Wolfson and Hourigan, 1997). Although the federal government ostensibly mandated states to legislate a minimum age of 21, the implementation of such a standard was beyond federal purview.

Legislated exceptions to the “21” rule and inconsistent enforcement of the law lead to a variety of outcomes when discussing the mandatory minimum drinking age. Even beyond the differing legislation among the states coupled with a lack of uniformity in enforcing the laws, Lipperman-Kreda et al. (2010) asserted, “Research indicates that there is considerable variability in alcohol policy and enforcement at the community level” (249). Currently 45 states permit underage consumption under prescribed circumstances ranging from religious purposes to educational purposes. For example, the Catholic Church endorses a sip of wine as part of the Eucharistic sacrament with the first communion taking place once the child has achieved the age of reason at 7years-old. It is also seen with culinary students, to allow underage consumption as long as the underage person is in view of a family member who is over the age of 21 (45 states, 2014). The discrepancies within policies are in conflict with the ideals of many who see criminalization of underage drinking as an effective way to combat youth-related problems that are exasperated by alcohol consumption.

Sociological Perspectives on Underage Drinking

Several theories attempt to explain underage drinking from a sociological standpoint. Classical deterrence and rational choice theorists would assert that deviance, such as underage consumption of alcohol, takes place when a person’s hedonistic calculus finds the benefits of the deviant behavior are greater than the costs. While deterrence application is the foundation of criminalizing behaviors, Weil asserted that criminalizing substance use is misguided (Mosher and Akins, 2014). Weil concluded the drive of humans to alter consciousness is a universal quality that is often labeled as hedonistic, but that the appropriate course of action is to supply people with a true and unbiased picture of costs (such as health risks) and benefits (actual positive effects or sensations often associated with the substance use) so that the decision is rationally informed (cited in Mosher and Akins 2014). Critics of rational choice theories pointed to instances of deviance where the costs significantly outweigh the benefits and cases where emotion produces irrational choices.

Applying social bond theory to deviant behavior such as underage consumption of alcohol presumes that the more ties one has to the community in the form of attachment, commitment, involvement and belief, the less likely he or she is to engage in illegal

possession or consumption of alcohol (Mosher and Akins, 2014). The difficulty lies in applying the deviance label to status offenses, such as underage drinking, which places the behavior of drinking alcohol in the deviant category. This is based upon age since drinking alcohol is approved of within the culture of the United States.

Differential association theory purported that deviant behavior is learned and is the result of associating with others who engage in similar behaviors (Mosher and Akins, 2014). Criminologists would identify peer groups among those to hold the greatest influence in advocating pro-deviant behaviors; therefore, drinking with peers while underage would encourage the deviant act of underage drinking, creating unique subcultural norms. Critiques of this viewpoint suggested that while differential association is implicated in the acquisition of deviant norms, it does not explain the continuation of the deviant behavior after undergoing serious negative sanctions, which is often the case in repeated criminal activities (Mosher and Akins, 2014).

Social learning theory, as developed by Ronald Akers and Robert Burgess (1966), explained the phenomenon as an interplay between reinforcement and punishment with social, psychological, physiological rewards and the influence of projected messages upon the learning of behavior. This outlook implicates the forces of operant conditioning as a cultural learning technique found within individual societal contexts. While social learning theories may make sense, the complexity of rewarding and punishing values at the individual and structural levels makes operationalizing this for research purposes cumbersome; consequently, there has been little empirical support generated.

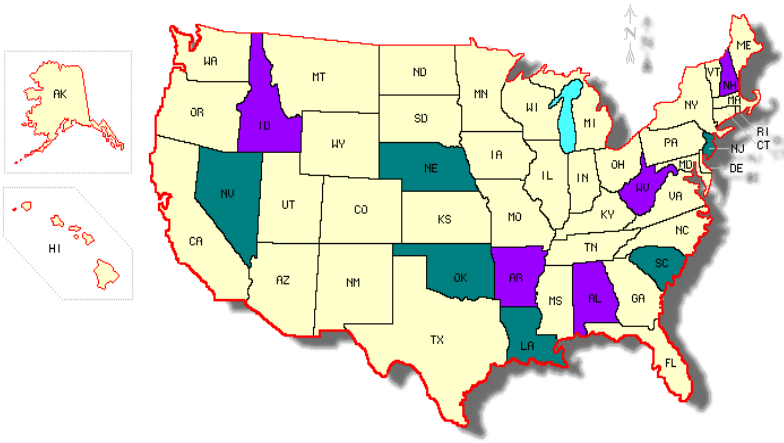
Within the context of these theories, there is found two ways to decrease underage drinking rates. Rational Choice Theory, which maintains that people act in their own self-interest and the rational self will balance the cost of an action against its possible rewards leading to a decrease in underage drinking rates with increased criminality or negative sanctions (Mosher and Akins, 2014). Alternatively, Social Bonding Theory holds that deviant actions are reduced when the number and strength of social bonds is augmented, predicting an inverse relationship between the rates of underage drinking and the level of positive societal attachment (Mosher and Akins, 2014). The objective of this study is to explore one aspect of this debate: the extent to which enhanced enforcement, in the form of stricter legislation, affects the likelihood of underage alcohol consumption. This research has

important policy implications, given the powerful influence of deterrence theory in the US Criminal Justice System.

DATA AND METHODS

Data

Using self-reported survey data collected by the Substance Abuse and Mental Health Services Administration, this study compared drinking rates in different states according in their approved exclusions for application of the legal minimum drinking age to identify legislative efficacy upon selected outcomes. To this end, it analyzed self-report data from the National Survey on Drug Use and Heath (NSDUH) for 2003, 2009, 2010, 2011, 2012, and 2013 (https://nsduhweb.rti.org/respweb/homepage.cfm), which were the only years that could be accessed through NSDUH. NSDUH employs a random probability sampling method and sampling rates are preset for state and age group. This study uses data from the 12 to 20year-old samples. This survey was administered by the Center for Behavioral Health Statistics and Quality for the Substance Abuse and Mental Health Services Administration (SAMSHA). Data for individual states were grouped into three categories based upon their policies regarding enforcement of the minimum legal drinking age of 21: strict states, lax states, and typical/moderate states (See Figure 1).



Key: Purple=Strict States Green=Lax States Yellow=Moderate/Typical States

Figure 1: State-level enforcement of underage drinking laws.

Strict States are the only five states to provide no exemption to the twenty-one minimum legal drinking age. The law does not allow for underage drinking under any circumstances in these states. These states are Alabama, Arkansas, Idaho, New Hampshire, and West Virginia (n=5). *Lax States* provide the most exceptions to the minimum legal drinking age and permit underage drinking as long as the alcohol is consumed on private property where liquor is not sold. This is the only restriction upon underage consumption in these six states. Parental consent is not necessary. These states are Louisiana, Nebraska, Nevada, New Jersey, Oklahoma, and South Carolina (n=6). *Typical or Moderate States* include the majority of the underage drinking policies in the United States with 39 of the 50 states falling into this category. These states set certain exemptions to the 21 minimum drinking age, variable by state, which include religious ceremonies, drinking under the supervision of parent or guardian, and educational purposes such as culinary students. These states fall in the middle of the Strict States and the Lax States in regards to the number of exemptions. Six typical states were selected as follows. The typical states were listed in alphabetical

order and assigned a number between one and thirty-nine. Six numbers were randomly selected using the integer generator at Random.org. The states corresponding to these numbers were included in the analysis. These typical states were Alaska, California, Georgia, Minnesota, New York, and Texas.

This study uses two dependent variables, underage alcohol use in the previous month and underage binge drinking in the previous month. The NSDUH data conceptualizes binge drinking as having consumed five or more drinks in one setting. Both variables are coded 0 for “no” and 1 for “yes”. The states were coded into a variable (state type), which was coded 0 for typical states, 1 for lax states and 2 for strict states. Year was coded in two different ways. First as an ordinal variable coded 0 for 2003, 1 for 2009, 2 for 2010, 3 for 2011, 4 for 2012 and 5 for 2013. Year was also coded as continuous variable. Ultimately, the latter version was employed for simplicity’s sake, as regression models yielded similar results with both versions of the year variable.

Methods

Various analyses were employed in this study. First, current trends in underage alcohol use and binge drinking by state enforcement type were compared to the national average using t-tests. A t-test is used to determine whether there is a significant difference between the combined averages of the typical, lax, and strict states compared to the national average. Two-tailed tests were employed to gauge whether the states under study fell significantly below (left tailed) or above the national average (right tailed). Time series charts were created to discern any changes in underage alcohol use or binge drinking in the typical, lax, and strict states over time. Logistic regression was employed to determine whether there is a significant difference in the odds of underage alcohol use and binge drinking in the lax states versus typical states and the strict states versus typical states. Logistic regression was employed because two dependent variables, underage drinking and binge drinking are dichotomous.

ANALYSIS RESULTS

Current Trends in Alcohol Use and Binge Drinking

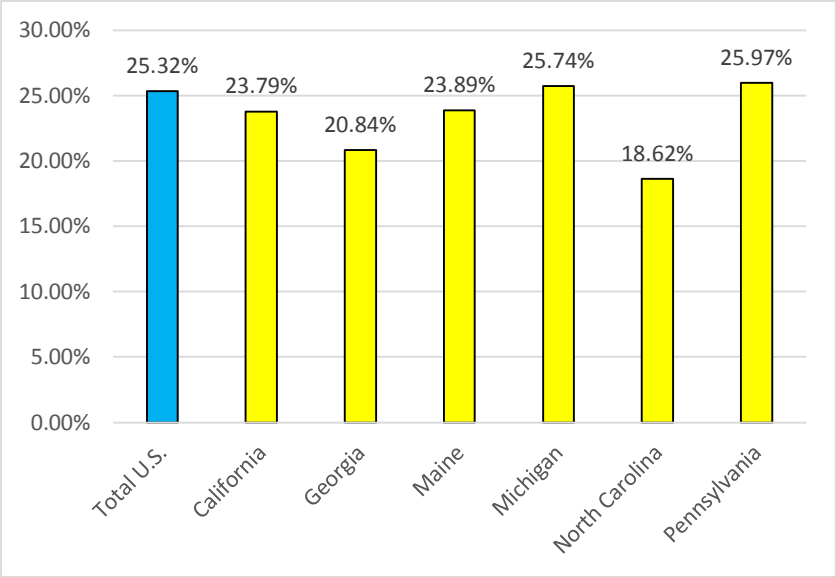


Figure 2: National average versus selected typical state averages of underage alcohol use in the past month, 2013 NSDUH Data.

Figure 2 compares underage alcohol use in the last month for the six randomly selected typical states with the US average for 2013. Most of the selected states are below the national average with the exception of Michigan and Pennsylvania. A t-test shows the combined average of the selected typical states were significantly less than the US average (p=.000).

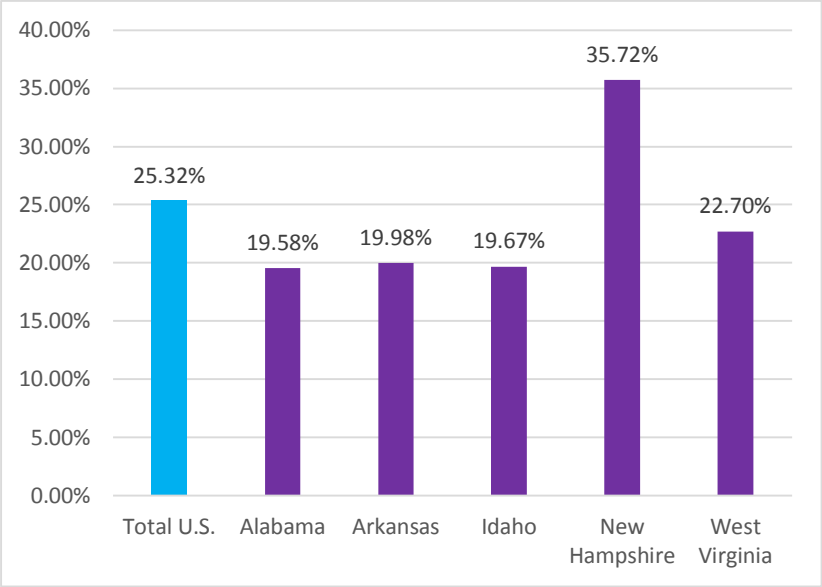


Figure 3: National average versus strict state averages of underage alcohol use in the past month, 2013 NSDUH Data.

Figure 3 compares underage alcohol use in the last month for the strict states with the US average for 2013. Most states were below the national average, except for New Hampshire, which was much higher. A t-test did not indicate a significant difference between the US average and the combined average of the strict states ($p>.05$).

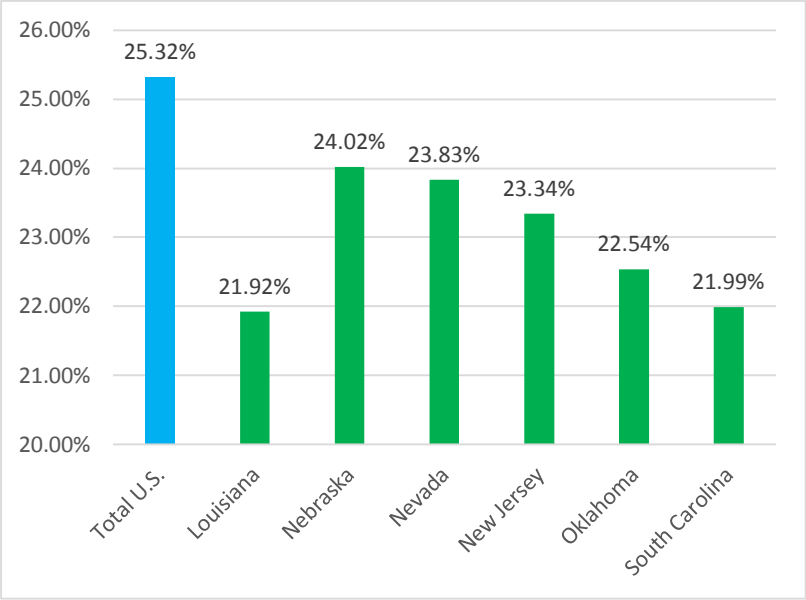


Figure 4: National average versus lax state averages of underage alcohol use in the past month, 2013 NSDUH Data.

Figure 4 compares underage alcohol use in the last month for the lax states with the US average for 2013. Underage alcohol use was lower than the national average in all of the lax states. A t-test shows that the combined average of the lax states was significantly lower than national average.

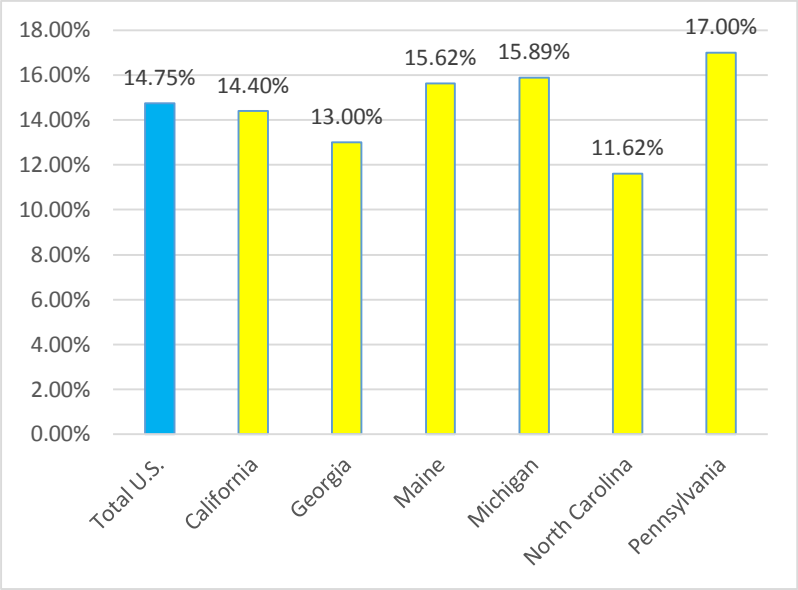


Figure 5: National average versus selected typical state averages of underage binge drinking in the past month, 2013 NSDUH Data.

Figure 5 compares the binge drinking averages of the 6 randomly selected typical states with the national average for 2013. Most of the typical states are close to the national average with the exception of North Carolina, which is about 3 percent higher and Pennsylvania which is 2.5 percent higher. A two-tailed t-test comparing the US average with the combined average of the typical states was not significant ($p>.05$). This suggests that there is no significant difference in binge drinking rates between the selected typical states and the US average.

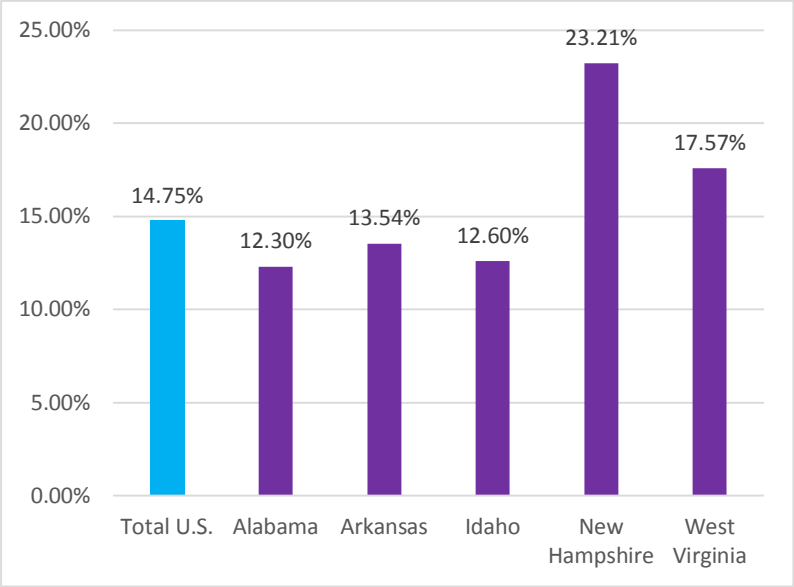


Figure 6: National average versus strict state averages of underage alcohol use in the past month, 2013 NSDUH Data.

Figure 6 compares the binge drinking averages of the 5 strict states with the national average for 2013. Three strict states are slightly lower than the national average. West Virginia is about 3 percent higher and New Hampshire is nearly 10 percent higher. A t-test comparing the combined average of the strict states with US strict states average to be higher than the national average, although the results were only borderline significant (p=.06).

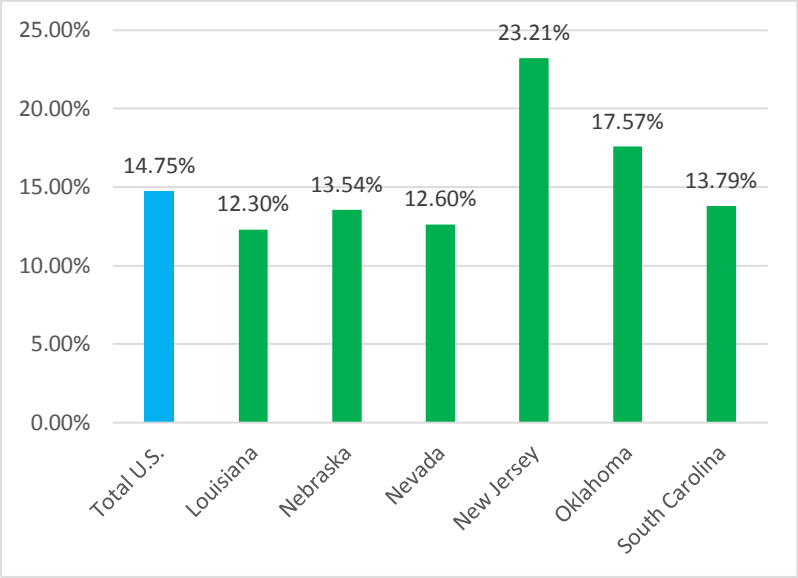


Figure 7. National average versus lax state averages of underage binge drinking in the past month, 2013 NSDUH Data.

Figure 7 compares the binge drinking averages of the 6 lax states with the national average for 2013. Three lax states are slightly lower than the national average. Four states, Louisiana, Nebraska, Oklahoma and South Carolina fall beneath the national average. Oklahoma is about 3 percent higher and New Jersey is almost ten percent higher. A t-test comparing the combined average of the lax states with US national average did not indicate a significant difference.

Temporal Trends in Underage Alcohol Use and Binge Drinking

Next, temporal trends of underage drinking were analyzed between 2003 and 2013 by state type, using time series charts. Please note some years are missing because NSDUH would not allow access to this data. Nonetheless some patterns still can be discerned.

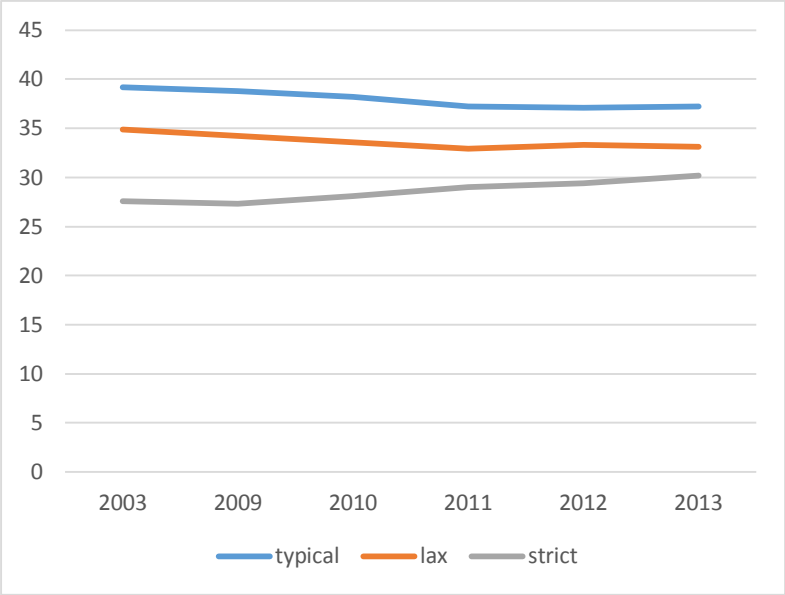


Figure 8: Underage alcohol use by state type in past month, 2003-2013 NSDUH Data.

Figure 8 shows the changes in percentages of 12-20 year olds who indicated having consumed alcohol in the previous month from 2003-2013. Alcohol use initially was higher in the typical states and much lower in strict states; however, the data indicate a convergence of rates over time. This is due to decreased alcohol use in the typical and strict states and increased use in the strict states.

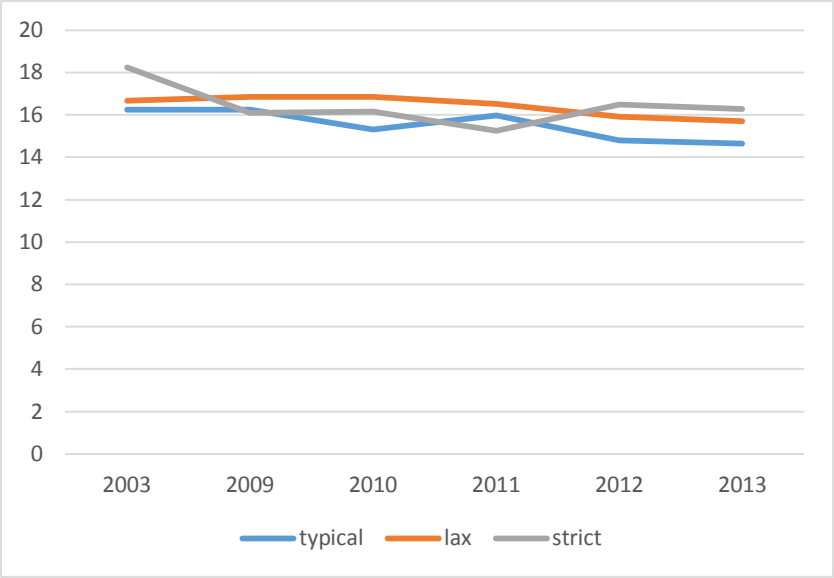


Figure 9: Underage binge drinking by state type in past month, 2003-2013 NSDUH Data.

Figure 9 shows the changes in percentages of 12-20 year olds who indicated binge drinking behavior from 2003-2013. Binge drinking initially was higher in the strict states and a bit lower in typical and lax states. Binge drinking declined in the strict states from 2009-2011, but appears to be rebounding. Binge drinking appears to be declining slightly over time in the typical and lax states.

Logistic Regression Analysis Results

Table 1shows the odds ratios from the logistic regression of underage alcohol consumption one month prior to the survey on the independent variables. The results do not indicate that underage drinking is more likely to occur in lax and strict states versus typical states controlling for year, as the p value is greater than .05. The data indicated, however, that the likelihood of underage drinking has declined over time. With each passing year, 12 to 20-year-olds were only.898 times as likely to have consumed alcohol in the previous month then in the previous year, controlling for state type.

Table 1: Odds ratios from logistic regression of underage alcohol use in previous month on state-level deterrence and year (p-values in parentheses).

Variable	Results
Lax State	1.05 (.383)
Strict State	1.06 (.459)
Year	.898 (.000)
N	5,937

Table 2 shows the odds ratios from the logistic regression of underage binge drinking on the independent variables. The results do not indicate a greater likelihood of binge drinking in either the lax and strict states versus typical states, controlling for year as the p value is greater than .05. The data indicate, however, that the likelihood of underage drinking has declined over time, controlling for state-type. With each passing year, 12 to 20-year-olds were only .901 times as likely to have binged drink in the previous month than in the previous year.

Table 2: Odds ratios from logistic regression of underage binge drinking on state-level deterrence and year (p-values in parentheses).

Variable	Results
Lax State	1.01 (.096)
Strict State	1.03 (.775)
Year	.901 (.000)
N	5,926

DISCUSSION

The analysis of self-reported data supports a conclusion that differences in alcohol legislation do not necessarily indicate a change in rates of underage consumption. Classical deterrence and rational choice theories would predict fewer underage drinkers when harsher sanctions are imposed which is not supported by the findings of this research (Mosher & Akins, 2014). Results of a study by Lipperman-Kreda, et al (2010). suggest that personal drinking beliefs may mediate the relationships of community norms about adolescents' alcohol use.. Local enforcement of underage drinking laws may allow for a community norm that supports underage drinking with levels of enforcement that do not coincide with the current legislation, which may be a limitation of this project. Social learning theory could explain the lack of significant variation among states with lax, moderate, and strict policies regarding legal minimum drinking age. Social learning theory would support a lack of recognized deviance in the act that supersedes its actual criminality. If present, this variable would vary greatly among jurisdictions. This view is supported by an Australian study in which high risk youth who lacked protective factors such as community

attachment, school opportunities for prosocial involvement, family attachment, and social skill (Stockwell et al., 2004). A cross-sectional analysis of youth in the study found that 67.1% of binge drinkers fell within the average risk category with only 4.3% categorized at high risk (Stockwell et al., 2004). Further research is needed to identify causal factors in underage alcohol use, but our findings show no significant differences between states that have legislation that allows for underage drinking on private property with no parental permission required versus those who provide absolutely no exception to the minimum drinking age and rates of underage consumption or bingeing. The results of this research therefore challenge the efficacy of minimum legal drinking age legislation as a solution to the societal harms created by the act. Without further inquiry, one must address the notion that if criminalizing underage drinking may not be the most effective control of underage alcohol consumption.

LIMITATIONS

The results of this study are preliminary and limited due to the fact that NSDUH restricted my access to the data. NSDUH only allows access to the complete data once a year to a limited number of researchers based on competitive research proposals. I plan to submit a proposal when the window opens again later this year, so I can conduct more sophisticated analyses with control variables such as gender and race and ethnicity.

ACKNOWLEDGEMENTS

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Four-Digit Hand: Anatomy of Ray II Amputation

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Department: Biology- Anatomy

ABSTRACT

The South Dakota State University anatomy program received a male cadaver, age eighty-four, in January 2014. Upon arrival, the cadaver was observed for any signs of surgery, skin ulcerations, or abnormality. The inspection revealed that his right hand only had four digits. In order to observe such a unique extremity, a dissection was conducted to understand the anatomy of the four-digit hand. This dissection would search for standard anatomical landmarks as well as abnormalities that could have been in relation to the absent finger. In this case study, both amputation and a possible birth defect were taken into account and researched. There were no signs of a birth defect, such as malformed fingers on either of the hands. The dissection showed many hallmark signs of amputation.

Keywords: hand, deviations, amputation, dissection, case study

INTRODUCTION

It is a way of investigating the world, a tool of dexterity, a source of force, an extension of leverage, and a way of expressing emotion (Smith, 2014). The hand is a vital part of the human body, allowing us to conduct very strong maneuvers to very fine touch (Smith, 2014). Not only are the hands important for day-to-day physical tasks, the hands have a significant role in different emotional aspects, such as body image (Beasley, 2003). Since hands are so individualized and personable, many people hold self-worth to the appearance and function of their hands, just as someone would value his or her face (Beasley, 2003). This understanding is very important in the subject of hand surgery, including amputation, because surgeons and physicians must be receptive to the emotional stress a disabled hand may cause the patient.

Grip is a very common role performed by the hand; one can grab a book off the shelf, hold the handle of a heavy bag of groceries, unscrew a cap off a bottle, and even pull a needle through fabric (Edwards et al., 2003). Even the index finger, also known as the second phalange, has many of its own types of grasps (Boscheinen- Morrin et al., 1992). In one study, each finger of the hand was evaluated to see how much it contributed to the function of the hand; the thumb (first phalange) supplied forty percent, while the second and third phalange each contributed twenty percent, and the fourth and fifth phalange supplied each ten percent of functionality (Boscheinen- Morrin et al., 1992).

Besides grip and functionality, the fingers also have an incredible ability to feel much more perceptively than most other parts of the body (Smith, 2014). According to homunculus drawings, the motor and sensory areas of the brain have a large percentage dedicated to the hand in proportion to other areas of the body (Martini et al., 2009). Not all areas of the hand are innervated equally; the finger pads contain many more neurovascular bundles than other aspects of the hand, especially in comparison to the dorsal surface. If there is damage or removal to a digit, the hand loses a certain amount of ability to operate normally; hand surgery must be done with care in order to limit the amount of sensory and motor functionality lost (Lister, 1984). This is the science behind surgery and amputation of any portion of the hand (Murdoch and Wilson, 1998).

There are many reasons for altered hand structure including: birth defects, injuries, and surgery. Birth defects can cause many issues with the ability of the hand to perform its many tasks, all depending on the type and severity of the deformity (Flatt, 1994). Although birth defects are not extremely common (seven percent of live births), the amount of anomalies that occur on the upper limb include one in every 626 of the congenital anomaly births (Flatt, 1994).

Injuries of the hand are very common; almost every task involve the hands. Depending on the severity of the injury, reconstructive surgery and microsurgery may be performed. If the injury cannot be fixed by reconstructive surgery, or the surgery did not work properly, amputation is another intervention that would be selected to provide the most functionality of the injured hand (Murdoch and Wilson, 1998). Certain amputations allow the hand to have more range of motion or better grip from the removal of a dysfunctional stump (Murdoch and Wilson, 1998).

The South Dakota State University anatomy lab received an 84 year-old (at death) male cadaver from the University Of South Dakota Sanford School Of Medicine medical anatomy laboratories. It appeared that his second digit was missing on his right hand, leaving only four digits. This was an extraordinary opportunity for me to pursue in-depth dissection in order to explore how the gentleman came to have a digit missing and how the hand adapted to the absent digit.

METHODS

This cadaver body was dissected for the South Dakota State University anatomy class, and done so in the manner of prosection for teaching purposes. The right arm had been dissected down to muscle; few superficial veins that were kept on the forearm included the basilic and cephalic. The nerves that were kept on the forearm included the median, ulnar, and radial. The skin on the hand was intact until the exploratory dissection began. The hand was amputated in order to allow more mobility of the extremity during the dissection. I completed the hand dissection and it was done separately from the full body prosection done for the anatomy class.

RESULTS

The dissection will be described using standard terminology: supination of the extremity so the palm of the hand is exposed upwards with the digits extended; medial refers to being closer to the midline of the body with lateral being the opposing term; the medial aspect of the hand in anatomical position refers to the pinky finger and the relating palmar side; distal is a term used to describe something that is further from the core of the body than something else. The fingers are distal to the palm of the hand, whereas the palm of the hand is proximal to the fingers. Anterior and ventral both mean the front of the body, while posterior and dorsal relate to the back of the body. The fingers are referred to as phalanges, and a single finger is a phalange. The phalanges are named laterally to medially from one to five. The first digit is known as the thumb, and the fifth digit is known as the pinky. The bones that make up the phalange are the metacarpal and the proximal, middle and distal phalanx bones (Martini et al., 2009). The dissection findings will be compared to the standard set by Netter in The Atlas of the Human Body, 2006.

Skin

The skin of the hand was very thin and fragile which could be contributed to the age of the body with no signs of trauma or scars from an amputation. Initially, only the epidermis and dermis were removed, noting that a straight line was visible while holding the skin to the light. This line traveled between the third digit and the first digit and began on dorsal side of the hand at the area of the second metacarpal head, moving towards the palmar side, with a lateral angle and is consistent with the Ray II amputation. The skin on the posterior aspect of the hand was much thinner with an estimated three times more cutaneous nerves and neurovascular bundles attached to the skin on the anterior or palmar surface of the hand compared to the posterior surface.

Dorsal Superficial Veins and Nerves

Around the most distal aspect of the second metacarpal showed a venous network of interconnecting veins that did not appear around any of the other knuckles of the hand. This venous network created an arch with many branches off of it, running on the medial aspect

of the thumb, arching towards the head of the first metacarpal, and then moving medially towards the third metacarpal. This network formed an arch around the distal portion of the head of the second metacarpal. The typical route of the cephalic vein travels on the lateral aspect of the dorsal side of the wrist, then branch into two digital veins; one of these veins would travel to the medial side of the thumb while the other one would continue to the second digit, branching again to both sides of the second digit (Netter, 2006). This differentiation appeared to be a sign of possible amputation, as the venous system seemed to have adapted to some sort of trauma to the area.

The superficial nerves of the posterior aspect of the hand were also observed. The superficial branch of the radial nerve was terminated at the area of the second metacarpal head. The typical path of this nerve starts at the dorsal lateral aspect of the wrist, and branches to the first phalange as well as the second phalange; the branch that runs to the second phalange is the digital branch demonstrated on the cadaveric hand (Netter, 2006). The end of the nerve was thicker than the more proximal end, possibly indicating a neuroma. The dorsal branch of the ulnar nerve seemed to follow the pathway described in the Atlas (Netter, 2006).

Dorsal Tendons and Correlating muscles

The next layer on the back of the hand was exposed, which demonstrated the extensor tendons. The branch of extensor digitorum that belonged to the third digit was not linear; the tendon curved laterally toward the first digit. This was a clear indicator that the symmetry of the hand was shifted, as the third digit extensor digitorum tendon travels down the midline (Netter, 2006).

The extensor digitorum branch was supposed to terminate on the distal portion of the second digit (Netter, 2006). Instead it was tethered to the other tendon in close proximity, which was the extensor indicis. This tendon was supposed to travel to the second digit, but was interwoven to the deep side of the extensor digitorum without possibility of separation. The fibers of both tendons were woven into each other, making a mesh of tendon that did not dominantly belong to one tendon or the other. These tendons together fanned out and attached to the head of the second metacarpal, the extensor digitorum to the third digit, and to the second dorsal interosseous tendon. Just proximal to the fanning of the tendons, the

tendon appeared atrophied in diameter, darkened in color, and slightly translucent which did not seem to be caused by the preservation of the body.

The fan of the tendons was fibrous and expansive with no obvious insertion. From observing how the tendon was attached, the extensor digitorum and extensor indicis to the second digit may have acted on extending the third metacarpal. Although no muscle measurements were done, the extensor indicis appeared to be an average size; if the indicis muscle was not being used, the muscle belly would be extremely atrophied.

Ventral Structures

On the ventral portion of the hand, the palmar fascia was easy to find and follow, except in the area where the second digit was missing where it was almost impossible to find traces of the fascia. After the removal of the remaining fascia, all of the underlying structures were exposed. There was very little adipose or connective tissue in the distal area between the first and third metacarpal, leaving the arteries and nerves very superficial.

Ventral Superficial Arteries

The palmar arch was followed from the ulnar artery on the medial portion of the hand. Instead of forming an arch, the artery formed a tortuous pattern. The ulnar artery was followed until the flexor digitorum tendon of the fourth digit, a few centimeters distal to the origin of the thenar muscles. The artery then turned a ninety degree angle, heading laterally towards the thumb. At the angle, an artery branched off of the palmar arch and headed distally. This section only continued for about a centimeter then stopped and created a forty-five degree angle continuing medially for about a centimeter. At the angle, a branch went straight and distally to supply blood to the lateral aspect of the fifth digit and the medial portion of the fourth digit. Continuing medially after the second angle, the artery created a second forty-five degree angle towards the fifth digit. It then continued distally, formed a full loop on itself, and then traveled to the medial side of the fifth digit. This is demonstrated in Figure 1.

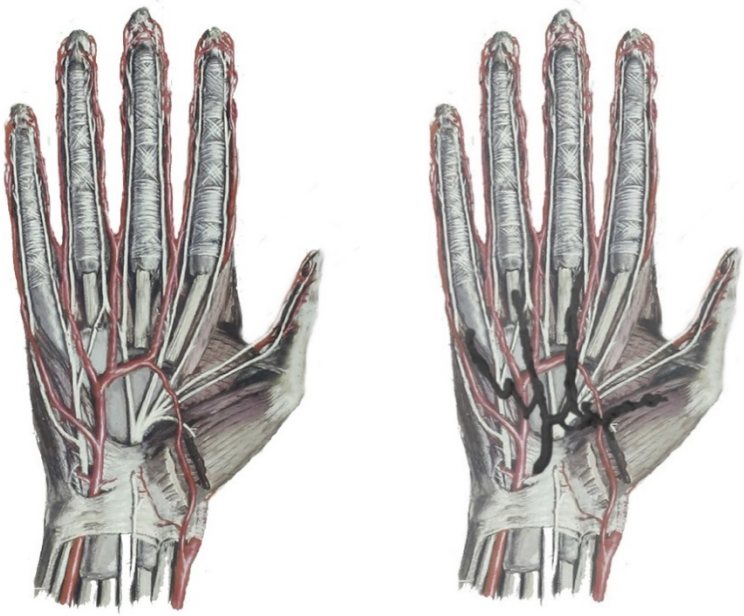

Figure 1: The palmar arch according to *The Atlas of Human Anatomy* (left) compared to vasculature of observed hand, demonstrated with black lines (right) (Netter, 2006).

From the stem of the ulnar artery that took a 90 degree angle, the artery continued straight for about a centimeter, and then divided into two branches, one diving into the thenar muscles, and one creating a 45 degree angle towards the third digit. This pattern of 45 degree angles continued until the artery became too minute to follow. There were also two branches off of the palmar arch that traveled into the thumb, which were very irregular in shape.

The digital arteries that branched off the palmar arch continued distally towards the fingers, doubled back, made loops, and became small towards the distal phalange. The artery between the fourth and the fifth digit dove into the digital nerve, splitting it into two and then rejoining again on the far side of the artery. The arteries running towards the medial side of the third digit had a very interesting weaving relationship with the digital nerves. The artery traveled with the nerve, so much that it was impossible to separate the two structures without compromising the integrity of each. This artery also made four tight

loops while traveling toward the digit. The artery on the lateral side of the third digit was too small to be followed more than halfway of the metacarpal. There was only one arterial branch that separated from the palmar arch and traveled proximally instead of distally. This artery branched between the third and fourth metacarpal and dove deep into the space between the third metacarpal tendons and the thenar (thumb) muscles.

The ulnar artery appeared to have what appeared to be a stent placed in it from the flexor retinaculum to the palmar arch. It was assumed to be a stent because there was very distinct circular bands that were visible with the naked eye. Under a dissecting microscope, the circular bands were shown to be the muscular layer of the artery layered between the fibrous layer and the tunica interna. Since the muscular layer was visible by the naked eye, this could indicate stenosis of the artery. When the dissection of the ulnar artery was performed under the dissecting microscope, the smooth muscle layer was cut, and the circular bands fell apart into sharp fragments, which would be consistent of stenosis in the arteries. These circular bands were observed on the ulnar arteries on both the right and the left hand.

Ventral Nerves

When the carpal tunnel was exposed, the median nerve was not visible. A cut of the carpal tunnel on a normal hand would be done in line with the third digit, and the median should sit most superficially once the carpal tunnel was opened. After searching, the median nerve was found more laterally and deep, in close proximity to the origins of the thenar muscles. This was consistent with adaption, as the symmetry of the hand had changed to compensate for the absent second digit.

There was a very interesting relationship between the arteries and the nerves as previously indicated in the information about the ventral superficial arteries.

Instead of one branch, there were two digital branches from the median nerve that traveled to the space between the fourth and third metacarpals with the destination of the medial aspect of the third digit. It was impossible to separate the digital nerve branches from the digital artery without causing damage to the structures. The digital branch of the median nerve that traveled to the lateral aspect of the same third digit was surrounded in scar tissue. This made it very difficult to follow the nerve distally. On this same side of the third digit,

there were two masses at the end of the two separate nerve branches. These neuromas were also embedded in scar tissue, making it hard to provide accuracy of their size. The approximations of their dimensions were 6 mm long by 4 mm wide, and the second one measured at 5 mm wide and 6 mm long. These are shown in Figure 2.

The median nerve was followed to the palmar then digital branches that would innervate the absent second digit. The end of this branch terminated around the space of the thenar muscles where a large neuroma was found, an indicative sign of amputation. The neuroma measured 8 mm long and 8 mm wide.

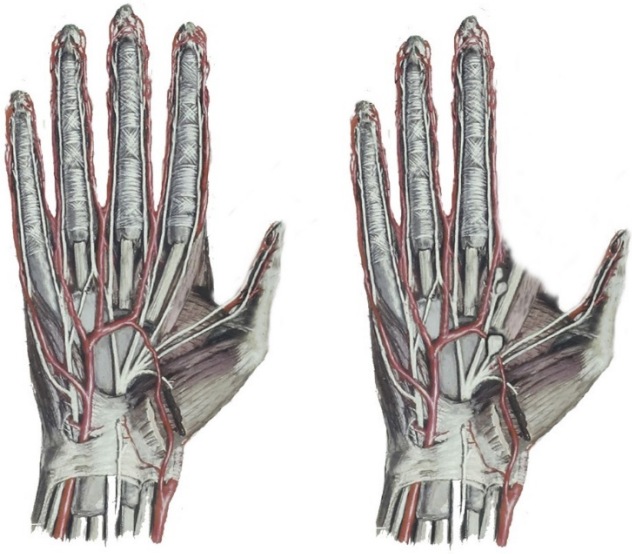

Figure 2: Three neuromas are shown on the right image with the atlas comparison on the left. Two neuromas are on the end of the digital branch of the median nerve that would travel up the medial aspect of the second digit. The third neuroma developed from the digital branch of the median nerve that would travel on the lateral aspect of the second digit (Netter, 2006).

Ventral Tendons and Musculature

Just as the tendons of the back of the hand demonstrated asymmetry, the flexor digitorum tendons for the third digit were curved laterally, with the tendons for the fourth digit appearing to be at the center of the hand. The palmaris brevis was very large, stretching to the thenar musculature, while the palmaris longus was absent.

Flexor digitorum superficialis and profundus were found for the second digit. The two flexors for the second digit each had different insertions at which they created a fanlike structure that spanned across the fibrous scar tissue between the third and first digit. The flexor digitorum profundus inserted more proximal than the flexor digitorum superficialis. Both of their proximal to distal paths together seemed to dive under adductor pollicis and continue to the distal aspect of the hand to the tendinous mass that terminated before the boundary of the head of the third metacarpal. These tendons dove more laterally under the thenar muscles than observed in the atlas (Netter, 2006). They were also very difficult to follow distally as they were embedded in scar tissue just proximal to the insertion of the tendons. Their appearance changed from glossy tendon to atrophied, darkened, and semi-translucent tissue, similar to the extensor tendons discussed earlier. The tendon fibers attached to the fibrous tissue on the lateral aspect of the third metacarpal proximal to the head of the bone and also expanded to join with the second dorsal interosseous muscle and first palmar interosseous. This area was also where the third digital branch of the median nerve on the lateral side was embedded as well.

The first lumbrical muscle was located originating on the flexor digitorum profundus tendon for the second digit at the base of the opponens pollicis brevis. The original insertion point of this muscle was absent, so the muscle stayed continuous along the tendon.

Tendons from the flexor digitorum profundus and superficialis that traveled to the fourth and fifth digit were found in this layer as well. The sheath enclosing the flexor digitorum superficialis tendons was fibrous and almost impossible to separate from the tendons without causing damage. The tendons within the sheath were brittle. The tendon traveling toward the fourth digit divided in the same sheath, and then would weakly reconnect distally. They appeared damaged because of fraying and a rough surface of the tendons. These findings were not thought to be a result of preservation.

In relation to the overall size of the hand and the other muscles of the hand, it appeared that the thumb muscles, the adductor pollicis, the flexor pollicis brevis, the abductor pollicis brevis, and the abductor pollicis transversus appeared hypertrophied although no measurements were done. The transverse head of the adductor pollicis appeared to have the most distal portion of its origination on the sheath of the flexor digitorum superficialis.

Dorsal Deep Musculature

Once removing the tendons of the palmar and dorsal sides of the hand, the deep muscles were visible. The first dorsal interosseous muscle, which was supposed to insert on the lateral side of the second proximal phalanx, inserted on the distal tip of the shortened second metacarpal where the shortened muscle belly was still attached.

First palmar interosseous muscle belly was shorter as well, reaching the whole length of the second metacarpal then attaching to the fibrous area on the lateral portion of the third metacarpal. Another origination point on the second metacarpal bone was a portion of the oblique head of the adductor pollicis.

Deep Structures

Just as the superficial palmar arterial arch was tortuous, the deep palmar arterial arch followed a similar pattern with less distinct angles.

When dissecting the musculature from the second metacarpal bone, the muscles were easiest to remove from the medial aspect of the bone. All other areas of the bone had well attached muscle fibers. The measurements from the second metacarpal were taken once it was removed, and consist of 40 cm long, 2.5 cm wide, and 5 cm deep. The bone was shaped like a triangle, with a wide proximal base, moving to a distal tip. There was a flat edge angled at a downward toward the medial aspect of the metacarpal. The bone appeared to be about half the length of the other metacarpal bones. At the end of the bone, there was a large tuberosity on the palmar side which could have been a bone scar.

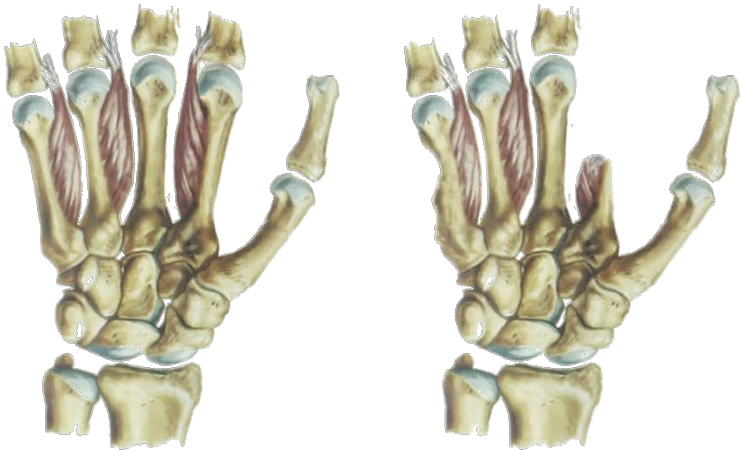

Figure 3: The right image shows the shortened metacarpal bone as well as the fracture of the fifth metacarpal bone (Netter, 2003).

The musculature was removed from the other metacarpals and it was found that the fifth metacarpal had a healed fracture with poor alignment of the bone. The proximal portion of the metacarpal was in alignment with the other metacarpals. At the fused fracture, the bone jutted anteriorly, and then continued in a straight line few centimeters anteriorly than the rest of the metacarpals which is demonstrated in Figure 3.

The carpal bones were all present. The trapezoid bone was dark with a reddish, dark brown color and cracked. The capitate also had a darkened area; it was located in the center of the dorsal aspect of the bone. It had a central, dark area moving outward into a lighter-colored bone, then more outward into the normal bone color. At the articulation of the radius and the scaphoid, both bones had a darkened area which was aligned when the hand was straight with the forearm. This may have been a result of a fall that happened before death.

DISCUSSION

Overall, the entire dissection indicated signs of amputation in the early life. Since there were signs of adaptation as well as no evidence of scars or stitches, it can be concluded that the amputation was performed at a young age. Tendons are left to retract after an amputation, so distinguishing between adaption and surgical placement is difficult (Beasley, 2003).

When dealing with amputation of the digits, maintaining length is very crucial unless the boundary of the proximal interphalangeal joint is met (Murdoch and Wilson, 1998). If the amputation must be done below the interphalangeal joint, the stump is deemed a burden to the hand and a Ray Amputation is performed (Beasley, 2003). If the second digit is severely injured proximally to this boundary, the pinch function between the first digit and the second digit is dysfunctional (Murdoch and Wilson, 1998). The remaining stump would impair the pinching ability between first digit and the adaptive third digit (Murdoch and Wilson, 1998).

A Ray Amputation is the process of removing the entire digit down to the lower aspect of the metacarpal (Beasley, 2003). It is normally performed when a previous amputation of a finger at a more proximal joint is deemed dysfunctional to the hand (Murdoch and Wilson, 1998). When performed on the second digit, this amputation is called Ray II (Atkins and Meier, 1989). The main reason for this type of amputation is to restore some sort of pinch and grip function; by removing the body of the metacarpal, the distance between the first and third digit is shortened, increasing the likelihood of the third digit to act as the second digit (Beasley, 2003). This amputation is performed by making an incision on the dorsal aspect of the hand, following the groove between the second and third metacarpal. (Wilhelmi, 2013). From this, the skin is purposefully left long so that the flaps are not pulled tight; if skin flaps are taut over this area, web contractures are possible (Green et al., 1999). The incision lines are shown in Figure 4. The second metacarpal is cleaved at an angle; the bone should not be cut proximally enough to interfere with the insertion of the flexor carpi radialis (Wilhelmi, 2013); A tenotomy (process of dividing a tendon) is performed with the tendons cut shorter than the cut length of the metacarpal (Wilhelmi, 2013); (Green et al., 1999). Arteries and veins are cauterized or ligated, and nerves are cut

proximally in order to try to prevent neuromas (Wilhelmi, 2013). By doing this type of amputation, the third digit is then capable to act as the second digit, with the same pinch grip that is performed with the first and second digit (Boscheinen- Morrin, 1992). The amount of leverage and strength exhibited by the hand does decreases after this type of amputation because the width of the palm is almost decreased by one fourth of the size (Lister, 1984). This makes actions as simple as swinging a hammer difficult (Lister, 1984). As for emotional adaption to this surgery, the end result is fairly cosmetically pleasing (Beasley, 2003). While first observing the cadaver's hand, it was not obvious that his finger was absent.

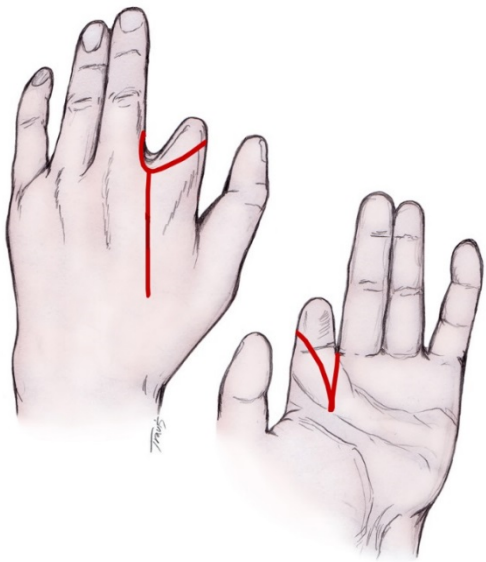

Figure 4: The red line indicates the incision for Ray II Amputation (Medscape, 2013).

The venous system on the dorsal aspect of the hand formed an arch with many branches around the remaining knuckle of the second digit. This was not present around any of the other knuckles and could have developed because of the ligation performed during surgery.

The flexor and extensor tendons that were to act on the second digit were found to have formed tendinous fanlike structures with tough scar tissue surrounding it. Studies have shown that tendons can produce collagen after being transected (Tubiana, 1981). In the

book *The Hand*, this process of tendon growing was described as “Newly synthesized connective tissue adjacent to loose areolar tissue appears to be induced to form disorganized, loose fibrillar and fibril patterns that favor longitudinal slipping of subunits and lengthening of connective tissue strands” (Tubiana, 1981). This explanation of tendon growth after a cut is consistent with the findings of tendinous masses during dissection as shown in Figure 5.

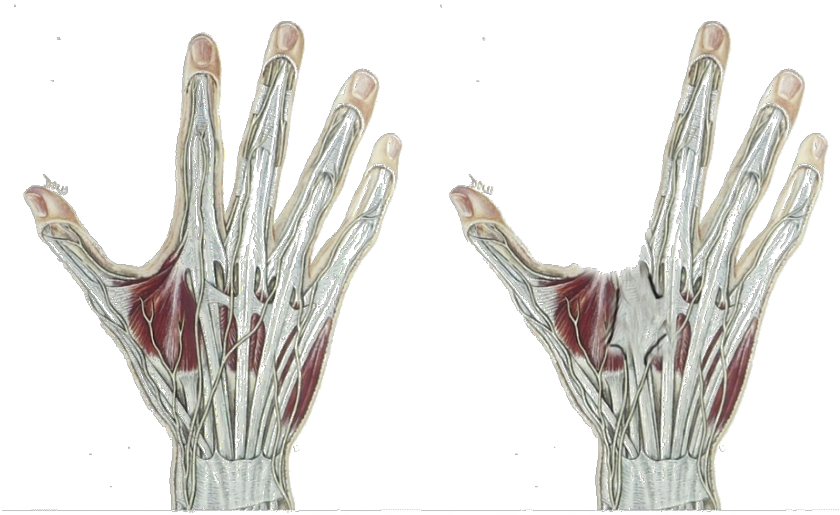

Figure 5: The tedinous mass found on the back of the hand is demonstrated on the right side, with an atlas comparison on the left (Beasley, 2003).

Both of the digital branches of the median nerve that traveled to both the medial and lateral aspect of the second digit were cut during the amputation. When dissected, three neuromas were found. The process of forming a neuroma consists of a nerve that begins to “extrude a new axoplasm, and in the absence of distal Schwann tubes into which to regenerate, the nerve endings curl up and become encapsulated in a fibrous mass called a neuroma.” (Beasley, 2003). Two of the neuromas are consistent with the medial and lateral digital branches running to the second digit. The third neuroma is the terminal end of the division off of the lateral branch.

There are many different ways to amputate the bone in the Ray II amputation; in the most recent explanation of the surgery, the metacarpal bone was described as “transected” meaning to cut at an angle (Green et al., 1999). In older texts, the bone was described as having to be kept cylindrical with most cuts being completely longitudinal (Green et al., 1999). There was no definitive description of how the bone was cut. Since bone constantly remodels, there is a possibility that the bone changed its shape after amputation to accommodate for the new situation.

The fifth metacarpal of this hand had a rejoined facture known as a transverse mid-shaft metacarpal fracture (Beasley, 2003). The cause of injury is normally due to a direct force to that area (Beasley, 2003). It is unknown if this injury was done to the second digit at the same time as the fifth metacarpal.

CONCLUSION

Learning about the human body allows health care providers make better decisions that affect many people. Constantly, procedures are being evaluated and changed to promote the best outcomes possible. Surgery and amputation performed on portions of the hand have been performed and evaluated for centuries (Vasconcelos, 1945). The importance of establishing certain procedures that could be shared to other physicians about the hand increased during the time of World War II (Vasconcelos, 1945). By examining the hand with four digits, more knowledge was gained about the Ray II Amputation and the effects on the hand.

This dissection exposed many different anatomical variations from the altas (Netter, 2006). Although it is not possible to say all of the abnormalities found were due to the Ray II Amputation, it can be speculated that the hand did adapt after the surgery. The tendinious fans that were found can be attributed to the growth after the amputation, but the tortious arteries may have a different cause. Finding these abnormalities was the goal of this dissection, in order to understand how an amputation can affect the hand.

Only assumptions can be made as to the situations that led up to this gentleman having his second digit amputated and his fifth metacarpal broken. The dissection exposed the important anatomical structures of the hand with four digits. The range of capabilities that

the hand could perform is information that is not able to be confirmed from dissection or research.

LIMITATIONS

The limitations that occurred during this research project included the lack of information about the donor, including a medical record, health history, and personal information such as occupation. There were also stipulations regarding the photographic documentation of the dissection, which forced more verbal description of the findings. The University Of South Dakota Sanford School Of Medicine emphasized the importance of confidentiality for the donor. The skill required to dissect the delicate and intricate structures of the hand was also a barrier as only sixty hours of dissection training was achieved before the initiation of this project.

ACKNOWLEDGEMENTS

This research was funded by the Joseph F. Nelson Undergraduate Mentorship program, and I extend my greatest gratitude for helping make this project possible. I would like to thank Dr. Scott Pedersen for helping and encouraging me throughout this project. Another thank you is extended to Mr. Ethan Snow for seeing potential in my abilities and having confidence in me to complete such an amazing project. Finally, I would like to sincerely thank every person, especially this gentleman with a unique hand, who gave back to the anatomy program by the amazing gift of donating their bodies to science.

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Optimization and Structural Characterization of Dimethyl Trisulfide (DMTS) Oxidation Product

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ABSTRACT

Cyanide poisoning is a public concern, and there are many shortfalls in current cyanide treatments. Dimethyl trisulfide (DMTS) is a cyanide antidote candidate that overcomes these shortfalls. Currently, there are limited published reports related to the analysis of DMTS. Therefore, an analytical method to detect and analyze DMTS from a biological matrix is vital for it to become available as a therapeutic agent against cyanide poisoning. The motivation of this project is to develop an HPLC-MS/MS method for analysis of DMTS and its degradation products; however, DMTS is difficult to ionize, a requirement for MS analysis, due to its nonpolar nature. In this study, DMTS was oxidized to a more polar compound that should enable its MS-MS analysis. The oxidation reaction was optimized to maximize product yield and, therefore, improve the accuracy of the analytical technique. The optimized oxidation reaction increased the yield of oxidized DMTS by 17.4% and decreased the amount of un-oxidized DMTS by 88.5%. In addition, initial characterization of the reaction product was preformed, using GC-MS. The preliminary results indicated the DMTS was fully oxidized.

INTRODUCTION

Cyanide is a potent metabolic poison. Upon exposure, cyanide enters mitochondria and binds to cytochrome c stopping production of ATP, effectively killing the cell[1]. Exposure to

cyanide can occur due to consumption of food containing cyanide, such as cassava, almonds, and apricot seeds. Other sources of exposure include anthropogenic sources: mining industry, smoking cigarettes, and smoke inhalation from fires. In addition, exposure can result from malicious use of cyanide, such as a terrorist weapon. Due to the numerous sources of exposure, there is a need for an efficient cyanide antidote.

Several types of cyanide antidotes have been developed. One class of cyanide antidotes is sulfur donors. According to the Centers for Disease Control and Prevention (CDC) guidelines from October 2014, sodium thiosulfate is the primary sulfur donor based antidote for cyanide poisoning[2]. Sodium thiosulfate is utilized by the enzyme rhodanese to donate sulfhydryl groups that bind with cyanide to form the less toxic thiocyanate, preventing the binding to cytochrome c[1].

Dimethyl trisulfide (DMTS) is a potential novel cyanide antidote that acts as a sulfur donor. DMTS does not require rhodansese to form thiocyanate in the body. DMTS is an ideal candidate for intramuscular injection, creating an antidote that could be readily administered in a mass exposure scenario[3].

In order for DMTS to be available as an FDA approved cyanide antidote, a method must be developed to analyze DMTS and DMTS degradation products in blood. A High Performance Liquid Chromatography Tandem Mass Spectrometry (HPLC-MS/MS) method is highly desirable for measurement of both DMTS and DMTS degradation products. Due to the non-polar nature of DMTS, it does not ionize in electrospray ionization and cannot be detected in Electrospray Ionization Mass Spectrometry (ESI-MS). Oxidizing the DMTS molecule to a polar molecule could allow for detection in an ESI-MS instrument.

METHODS

Optimization of Oxidation Reaction

DMTS solution (19.8 mM) was prepared by diluting a 396 mM stock solution with HPLC grade methanol (MeOH). The initial oxidation method was as follows: 1350 µL of deionized H_2O, 200 µL of DMTS sample, 400 µL of 30% H_2O_2, and 50 µL of 1 mM nitric acid were added, along with a stir bar, to the reaction vial. The reaction vials were placed in

a heat block with a thermometer on a hot plate. The power of the hot plate was adjusted until the temperature of the thermometer on the heat block stabilized at 30 °C. The reaction proceeded for 1 hour.

Oxidation parameters, such as temperature, reaction time, and amount of nitric acid were systematically changed to maximize the product (oxidized DMTS) yield. Triplicates of samples were used for analysis.

After oxidation, the oxidized DMTS was extracted by adding 600 µL of ethyl acetate to reaction vials, mixing, and then settling. The organic layer was separated and removed from each vial. The organic layer was washed twice with 500 µL of H_2O, then separated and filtered through a Teflon filter and analyzed using an Agilent Technologies 1200 Series HPLC-UV-Vis instrument. Gradient elution was used with 30% MeOH and 70% H_2O for 4 minutes, linearly increased to 100% MeOH over 5 minutes, held constant for 2 minutes, and linearly decreased to 30% MeOH over 3 minutes at a flow rate of 0.35 mL/minute. A multiwavelength UV detector at wavelength 280 nm was used to measure the absorbance. The column was a 2.1 x 150 mm ZORBAX Eclipse XDB-C-18 reverse phase column, with a 160 Å pore size.

Isolation of Oxidation Product by Column Chromatography

Before structural characterization of oxidized DMTS, a normal phase gravity column (7 cm in length, 1.5 cm diameter) containing 60Å silica beads (230-400 mesh particle size) was used to separate the oxidized DMTS from the other components of the reaction vial. The mobile phase was ethyl acetate.

Gas chromatography-mass spectrometry (GC-MS)

An Agilent Technologies 5975B Inert XL EI/CI GC/MSD system with an Agilent Technologies 7683B Series injector and a gas chromatography column with a length of 30 m and internal diameter of 0.250 mm was used to analyze the isolated oxidation product. The GC-MS instrument had an inlet temperature of 150 °C and gas flow rate of 58 mL/min. The oven temperature was controlled starting from 50 °C for 1 minute with a ramp of 120 °C/min for 2 minutes, and held at 290 °C for 1 minute.

Data Analysis

The percent change in peak area for the oxidation product and DMTS was calculated using Equation 1. The standard deviation of the percent change was calculated using Equations 2 and 3.

$$\%\,change\ in\ Peak\ area = \frac{Average\ Peak\ area_{final} - Average\ peak\ area_{initial}}{Average\ peak\ area_{initial}} \times 100\% \quad (1)$$

$$Std.dev\ of\ the\ percent\ change\ (s_f) = \sqrt{s_{initial}^2 + s_{final}^2} \quad (2)$$

$$Percent\ Std.dev\ of\ the\ percent\ change\ (s_f) = \frac{s_f}{Average\ peak\ area_{initial}} \times 100\% \quad (3)$$

Equations 1-3: The equations above show calculations performed during data analysis.

RESULTS

Through systematic variation of the initial oxidation reaction parameters, the effect of each parameter on oxidized DMTS formation was determined. The effects of changing parameters can be seen in Table 1. The only parameter that significantly increased the oxidized DMTS yield was an increase in the reaction time. When the temperature was increased from 30 °C to 45 °C, the peak area of oxidized DMTS decreased slightly (by 6%). When the temperature was increased to 60 °C, the peak area of oxidized DMTS decreased by 67%.

Table 1: The percent change of peak area and standard deviation of DMTS and oxidized DMTS with changing reaction parameters compared to initial reaction conditions.

Changed Parameter	**% Change in Oxidized DMTS**	**% Change in DMTS**
Time: 1 hr to 3 hr	17.4 ± 7.1%	-88.5 ± 3.2%
Temp: 30 °C to 45 °C	-6.6 ± 4.5%	-77.9 ± 3.7%
Temp: 30 °C to 60 °C	-66.8 ± 29.1%	-97.9 ± 4.0%
Nitric Acid Volume: 50 µL to 25 µL	-39.3 ± 5.7%	60.7 ± 24.3%
Nitric Acid Volume: 50 µL to 100 µL	-17.7 ± 28.8%	-39.0 ± 33.6%

The optimized oxidation conditions are as follows: 1350 µL of deionized H_2O, 200 µL of DMTS sample, 400 µL of 30% H_2O_2, and 50 µL of 1 mM nitric acid reacted for 3 hours at 30 °C. Optimized oxidation reaction conditions led to an increase of 17.4 ± 7.1% of oxidized DMTS. The optimized reaction conditions decreased DMTS by 88.5 ± 3.2 %. Figure 1 illustrates a comparison between optimized reaction conditions and initial reaction conditions.

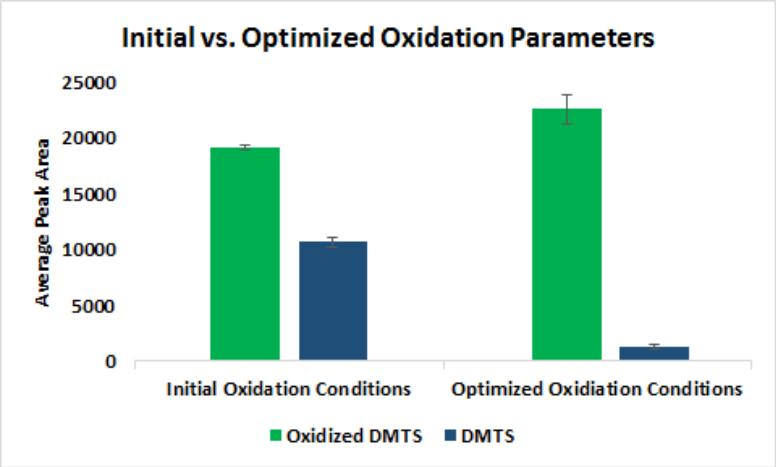


Figure 1: Initial vs. Optimized Oxidization Parameters effect on DMTS and Oxidized DMTS.

The mass spectrum of the oxidation product peak in GC-MS contained ions of mass/charge (m/z) 126, 147, 158, 175, 189, 207, and 221, see Figure 2.

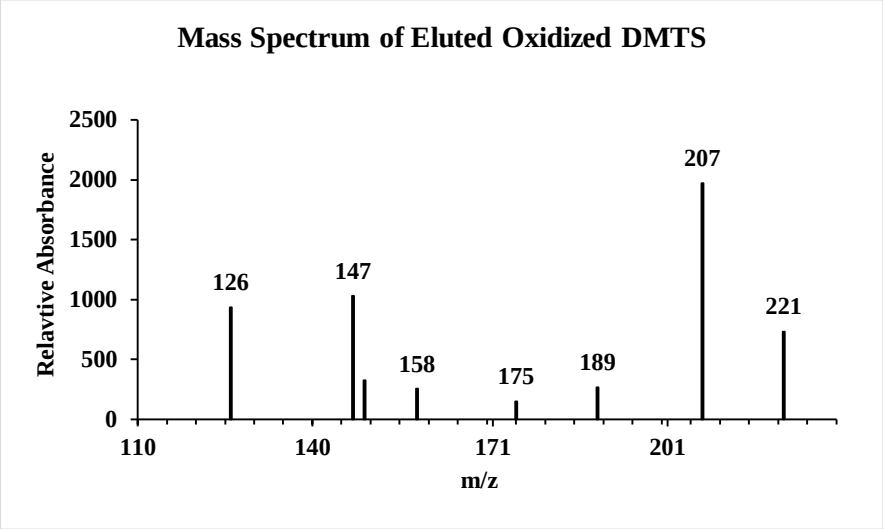


Figure 2: Mass Spectrum of the GC peak at 3.465 min.

DISCUSSION

The loss of product with increasing temperature is likely attributed to the volatility of DMTS and oxidized DMTS, as well as thermal degradation of these compounds. It was also noticed that increasing the reaction time increased the yield of oxidized DMTS; this is likely due to the fact that reaction equilibrium is not reached within 1 hour and extending the reaction to 3 hours allows the reaction to approach equilibrium.

The MS fragmentation pattern of oxidized DMTS suggests that the structure in Figure 3 is likely that structure. The m/z ions of 147, 158, 175, 189, 207, and 221 likely correspond to fragments after the original molecule lost methyl groups, hydrogen atoms, sulfur atoms, and oxygen atoms. For example, the m/z ion of 207 corresponds to a fragment with the loss of a methyl group, and the ion at 175 likely corresponds to a fragment resulting from the loss of a methyl group and two oxygen atoms. The probable structures for the fragment ions can be seen in Table 2. The proposed structure for oxidized DMTS can explain the data obtained through mass spectrometry. However, the proposed molecule of the oxidized DMTS cannot explain all of the data in the mass spectrum in Figure 2. For example, the 147 m/z ion does not fit the proposed structure. Further study of the oxidation product is needed to confirm the proposed structure.

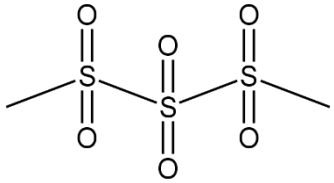
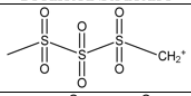
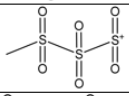
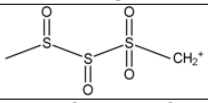
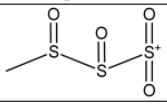
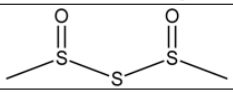


Figure 3: Predicted oxidized DMTS structure,

Table 2: Fragment mass/charge (m/z) ratio from the mass spectrum and predicted structural assignments.

m/z	Predicted Structure
221	
207	
189	
175	
158	

CONCLUSIONS

Increasing the time of the oxidation reaction allowed for an increase of 17.4% for oxidized DMTS. A preliminary structure of oxidized DMTS was determined. Future work will be needed to test these optimized oxidization parameters on lower concentrations of DMTS. Further characterization of oxidized DMTS will also be conducted using LC-MS/MS and ultimately an LC-MS/MS method will be developed. The complete development of this method provides a necessary tool in the approval of DMTS as a cyanide antidote.

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The Relationship of High School Size, Gender and First-Year Retention Rates at South Dakota State University

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ABSTRACT

This study examines selected demographic characteristics of six South Dakota State University 2008 – 2013 cohorts of first-time, full-time freshmen who graduated from South Dakota public high schools. The purpose of our study is to explore the relationship between high school size, gender and student retention at South Dakota State University.

Information that was both gathered and analyzed about these students includes their high school size and gender. Our examination of this data uses descriptive statistics to identify characteristics of students who were retained after their first year at SDSU. This study identifies two findings of interest: first, students graduating from high schools with populations of 200-399 students were retained at the highest level; second, gender matters for retention as females were more likely to return than males.

Key words: high school size, gender, postsecondary education, postsecondary retention, higher education

INTRODUCTION

University retention rates are currently garnering considerable attention. According to a 2008 study by ACT, only 40.3% of college students complete their degree (ACT, 2008). A review of the literature shows an increased emphasis coming from the public and the government on the efficiencies and efficacy of the higher education system to graduate students. Selingo (2015) explains, “After years of focusing on increasing access to higher education, federal and state higher-education policymakers recently have begun to shine a spotlight much more on the results of those efforts” (p. 8). Federal mandates and proposed legislation require that universities significantly improve retention and graduation rates. In a report published in the Chronicle of Higher Education (Selingo, 2015), higher educational systems identify the need for decision-making to be driven by big data as a powerful tool; they even consider modeling university systematic and programmatic efforts after leading corporate entities, much like Netflix and Amazon promote videos and books specific to an individual consumer’s interests. This approach is known as predictive analytics. The data retrieved can be used to “steer students to more appropriate courses, and majors,” thereby reducing resources expended in university systems (Selingo, 2015, p. 27).

In studying retention at universities, educational leaders explore K-12 school effectiveness. A commissioned report for the National Symposium on Postsecondary Student Success emphasizes the importance of a student’s academic preparation in high school: “The quality of the academic experience and intensity of the high school curriculum affects almost *every dimension* of success in postsecondary education” (Kuh, Kinzie, Buckey, Bridges, & Hayek, 2006, p.19). Further, Hayali (2013) concludes that academic preparation at the high school level is “one of the most – if not the single most – important predictors” of enrollment and retention in college, even more so than socioeconomic factors (p. 40-41). Thus, the quality and depth of curriculum provided at the high school level becomes a significant factor in future academic success for many students.

State departments of education also work to validate the effectiveness of their schools by collecting information on college graduation rates. Considerable research exists on the relationship between high school size and variable factors, such as academic achievement (Fetler, 1989; Walberg & Walberg, 1994) and dropout rates (Felter, 1989; Pittman &

Haughwout, 1987). However, the results from research on high school size and academic gains are inconsistent. For example, a study of a nationwide population of approximately 9,000 students found that students from moderate-sized high schools (enrollments between 600 and 900) had, on average, higher academic achievement advances in reading and mathematics than their student peers from small or large high schools (Lee & Smith, 1997). However, a study of a national representative population of almost 14,000 secondary students found little to no relationship between secondary academic achievement and high school size (Lindsay, 1984). Looking specifically at dropout rates, a 1989 study examining all public high schools in California found higher dropout rates correlated with higher school enrollments (Felter, 1989). A national study by Pittman and Haughwout (1987) found similar results. Out of a representative sample of 744 high schools across the nation, higher dropout rates were associated with larger school size. This study estimated that for every additional 400 students, the dropout rate increased by one percent (Pittman & Haughwout, 1987).

Many universities have studied and continue to study student retention by examining characteristics of students who were retained and those who were not retained. According to a Winona State University study in 2011, the “most significant predictor” of a student’s post-secondary retention is his or her high school class size; the larger the size of a student’s high school, the more likely the student will be retained (Yu, Lin, Chen, & Kaufman, 2011, p. 26). When examining gender, Yu et al. (2011) found that females are less likely to be retained.

Colleges employ student affairs and admissions staff to track student retention as a way to measure the effectiveness of university programs and recruiting efforts. In *Impact 2018*, a strategic vision for South Dakota State University, the Division of Student Affairs notes a retention rate target for first-time, full-time students of 80% (Division of Student Affairs, SDSU, 2013). *Impact 2018* more narrowly indicates a fall-to-spring academic semester target retention rate of 94%. In the Fall 2013 Bachelor’s Degree cohort, the university reported a retention rate of 77% (South Dakota State University, 2013). It is of utmost importance to more closely examine the factors that impact retention, because “retention can affect every aspect of higher education” (Stillman, 2009, p. 2).

This descriptive research study looks at retention by examining selected demographics of freshmen who were retained after their first year at South Dakota State University. We identified two research questions: (1) What is the relationship of high school size and first-year retention at South Dakota State University? (2) What is the relationship of gender and retention at South Dakota State University?

Higher education administrators who explore retention research will be able to use the results of this study to develop programmatic efforts to increase retention rates.

METHODS

This study uses descriptive statistics to examine specific characteristics of students who were retained after their first year at South Dakota State University (Weirsma, 2000). Specifically, we looked at two variables, the high school size and the gender of freshmen who were retained and of freshmen who did not return after their first year of college. We wanted to see if there was a pattern in student retention based on these two variables.

After completing the Institutional Review Board requirement, we requested and received data from the Office of Administrative Information Services for six first-time, full-time, Bachelor's Degree seeking fall cohorts from the years 2008 to 2013, which includes 6,714 student data observations. The data set includes only students graduating from public school districts in the state of South Dakota. Due to the lack of available data consistent with the South Dakota Department of Education reports, we decided to exclude freshmen from private, parochial and tribal schools.

Using Excel spreadsheets we tabulated the data to determine the total number of students retained and not retained. We also calculated the total number of males and females in the population.

To identify any pattern with the variable of high school size, we categorized the freshmen in our data into five high schools groups, according to school sizes. We determined the size of the high schools according to South Dakota Department of Education School District Profiles (SD DOE, 2014). To select the ranges and grouping of the schools, we compared the school populations to the groups defined by the South Dakota High School Activities

Association (SDHSAA, n.d.). In addition, the range in numbers appeared to have naturally occurring divisions in the Excel spreadsheet.

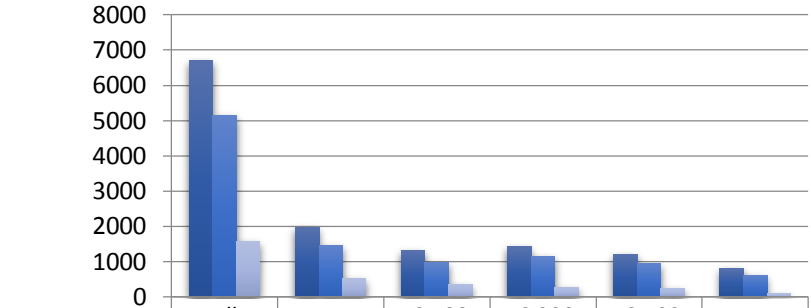
- Group 1 High Schools: > 899 students
- Group 2 High Schools: 400 to 899 students
- Group 3 High Schools: 200 to 399 students
- Group 4 High Schools: 100 to 199 students
- Group 5 High Schools: < 100 students

RESULTS

Our findings yield interesting results. In examining selected demographic characteristics of six South Dakota State University 2008 – 2013 cohorts of first-time, full-time freshmen who graduated from South Dakota public high schools, we discovered several patterns in regard to high school size, gender, and student retention at South Dakota State University. We found that out of 6,714 total freshmen in the six cohorts studied, 1,975 freshmen graduated from high schools of less than 900 students; 1,308 freshmen graduated from high schools with between 400 and 899 students; 1,412 freshmen graduated from high schools with between 200 and 399 students; 1,197 freshmen graduated from high schools with between 100 and 199 students; and 822 freshmen graduated from high schools of less than 100 students. Of the 6,174 students studied, 76.6% of the freshmen returned for their sophomore year; 23.4% of the freshmen did not return. In total, 5,145 students were retained and 1,569 were not retained.

Table 1 illustrates these numbers and percentages. It also shows the numbers of students retained and not retained in each of the high school size categories.

Table 1: School Size Comparison of Retention Rates



	All Students	HS>900	HS 400-899	HS 200-399	HS 100-199	HS<100
Total	6714	1975	1308	1412	1197	822
Retained	5145	1462	959	1140	955	629
Not Retained	1569	513	349	272	242	93

We also disaggregated the data according to gender. With an equal distribution of 3,357 males and 3,357 females, males have a retention rate 3.7% lower than their female counterparts. Males have a retention rate of 74.8% with 2,511 being retained and 846 not retained. Females have a retention rate of 78.6% with 2,634 being retained and 723 not retained. Table 2 illustrates these numbers and percentages.

Table 2: Total Number of Students Retained with Gender Breakdown

CATEGORY	DESCRIPTION	NUMBERS	PERCENTAGE
All Students	Total	6714	
	Retained	5145	76.6%
	Not Retained	1569	23.4%
	Males	3357	
	Males Retained	2511	74.8%
	Males Not Retained	846	25.2%
	Females	3357	
	Females Retained	2634	78.5%
	Females Not Retained	723	21.5%

For the 1,975 total students from high schools with populations above 899, a retention rate of 74% is reported with 1,462 students being retained and 513 not retained. Males have a retention rate of 71.4% with 720 retained and 289 not retained. Females have a retention rate of 76.8% with 742 retained and 224 not retained. Table 3 illustrates these numbers and percentages.

Table 3: Number of Students Retained According to >899 School Size with Gender Breakdowns

CATEGORY	DESCRIPTION	NUMBERS	PERCENTAGE
Group 0 Schools	Total	1975	
HS >899	Retained	1462	74.0%
	Not Retained	513	26.0%
	Males	1009	
	Males Retained	720	71.4%
	Males Not Retained	289	28.6%
	Females	966	
	Females Retained	742	76.8%
	Females Not Retained	224	23.2%

For the 1,308 total students from high schools with populations of 400 to 899, a retention rate of 73.3% is reported with 959 retained and 349 not retained. Males have a retention rate of 73.4% with 485 retained and 176 not retained. Females have a retention rate of 73.3% with 474 retained and 173 not retained. Students from high schools with populations of 400 to 899 have the lowest retention rate at 73.3% being retained and 26.7% not retained. Table 4 below illustrates these numbers and percentages.

Table 4: Number of Students Retained According to 400-899 School Size with Gender Breakdowns

CATEGORY	DESCRIPTION	NUMBERSS	PERCENTAGE
Group 1 Schools	Total	1308	
HS 400 to 899	Retained	959	73.3%
	Not Retained	349	26.7%
	Males	661	
	Males Retained	485	73.4%
	Males Not Retained	176	26.6%
	Females	647	
	Females Retained	474	73.3%
	Females Not Retained	173	26.7%

For the 1,412 total students from high schools with populations of 200 to 399, a retention rate of 80.7% is reported with 1,140 retained and 272 not retained. Males have a retention rate of 79.2% with 549 retained and 144 not retained. Females have a retention rate of 82.2% with 591 retained and 128 not retained. Students from high schools with populations of 200 to 399 have the highest retention rate of all categories at 80.7% being retained and 19.3% not retained. Table 5 below illustrates these numbers and percentages.

Table 5: Number of Students Retained According to 200-399 School Size with Gender Breakdowns

CATEGORY	DESCRIPTION	NUMBERSS	PERCENTAGE
Group 2 Schools	Total	1412	
200 to 399	Retained	1140	80.7%
	Not Retained	272	19.3%
	Males	693	
	Males Retained	549	79.2%
	Males Not Retained	144	20.8%
	Females	719	
	Females Retained	591	82.2%
	Females Not Retained	128	17.8%

For the 1,197 total students from high schools with populations of 100 to 199, a retention rate of 79.8% is reported with 955 retained and 242 not retained. Males have a retention rate of 78.7% with 470 retained and 127 not retained. Females have a retention rate of 80.8% with 485 retained and 115 not retained. Table 6 below illustrates these results.

Table 6: Number of Student Retained According to 100-199 School Size with Gender Breakdowns

CATEGORY	DESCRIPTION	NUMBERS	PERCENTAGE
Group 3 Schools	Total	1197	
100 to 199	Retained	955	79.8%
	Not Retained	242	20.2%
	Males	597	
	Males Retained	470	78.7%
	Males Not Retained	127	21.3%
	Females	600	
	Females Retained	485	80.8%
	Females Not Retained	115	19.2%

For the 822 total students from high schools with populations less than 100, a retention rate of 76.5% is reported with 629 retained and 193 not retained. Males have a retention rate of 72.3% with 287 retained and 110 not retained. Females have a retention rate of 80.5% with 342 retained and 83 not retained. Interestingly, students from high schools with populations less than 100 have the highest retention percentage difference between males (72.3) and females (80.5). Table 7 below illustrates these numbers and percentages.

Table 7: Number of Students Retained According to <100 School Size with Gender Breakdowns

CATEGORY	DESCRIPTION	NUMBERS	PERCENTAGE
Group 4 Schools	Total	822	
<100	Retained	629	76.5%
	Not Retained	193	23.5%
	Males	397	
	Males Retained	287	72.3%
	Males Not Retained	110	27.7%
	Females	425	
	Females Retained	342	80.5%
	Females Not Retained	83	19.5%

DISCUSSION AND CONCLUSION

In this study, we examined the relationship of high school size, gender, and student retention of six 2008-2013 cohorts of first-time, full-time freshmen at South Dakota State University. Of the 6,174 students studied, 76.6% of the freshmen returned for their sophomore year; 23.4% of the freshmen did not return. In total, 5,145 students were retained and 1,569 were not retained. Students from high schools with populations of 200 to 399 have the highest retention rate of all categories at 80.7% being retained and 19.3% not retained.

In regard to gender, with an equal distribution of 3,357 males and 3,357 females, males have a retention rate 3.7% lower than their female counterparts. Males have a retention rate of 74.8% with 2,511 being retained and 846 not retained. Females have a retention rate of 78.6% with 2634 being retained and 723 not retained.

Within this section, we will first discuss the relationship of high school size and retention and briefly compare our findings with other studies. We then consider possible explanations and considerations to explain the variances. Next we will discuss the relationship of gender and retention and briefly compare our findings to other studies. We then consider possible explanations for the variances.

HIGH SCHOOL SIZE AND RETENTION

Our findings show that the highest retention rate for university freshmen in South Dakota comes from students who graduated from high schools whose size ranges from 200 to 399. In this size category, a retention rate of 80.7% is reported with 1,140 retained and 272 not retained. Students graduating from high schools with enrollments of 400 to 899 had the lowest retention rate of all the sizes, which was a rate of only 73.3% retained and 26.7% not retained. This result does not support our review of the literature, which reveals a wide range of findings related to the ideal high school size Considerable research identifies high schools with enrollments of 500 and above as ideal for student achievement (Conant, 1967; Lee & Smith, 1997; Yu et al., 2011). A renowned 1976 study by Conant claims that high schools with populations lower than 750 are unable to deliver an inclusive educational program. Further, a study by Lee and Smith (1997) characterizes high schools with populations ranging from 600 to 900 students as most effective in helping student achieve academic success.

Our results contradict these studies in light of university retention rates because students graduating from high schools with populations between 200 to 399 were retained at the highest rates at South Dakota State University. This size is smaller than the ideal numbers cited by others (Conant, 1967; Lee & Smith, 1997; Yu et al., 2011). With this contradiction in mind, we seek to further explore the specific theoretical and conceptual occurrences that differentiate high schools with populations from 200 to 399 students from their peer institutions. The next section examines three possible areas that may directly or indirectly influence postsecondary retention: 1) Curriculum Quality, 1) Academic Achievement, and 2) Student Engagement.

CURRICULUM QUALITY

Curriculum quality affects retention because the quality and rigor of a student's academic experience in high school affects success in the postsecondary environment (Kuh, Kinzie, Buckey, Bridges, & Hayek, 2006). Some experts argue that larger schools can offer a more diverse, comprehensive curriculum than smaller schools. However, Cotton's extensive review of over 69 documents that identify a relationship between school size and student success concludes that the research does not show a reliable and justifiable relationship

between that of curriculum quality and school size (Cotton, 1996). Slate (2010) argued that although larger schools are able to offer more diversity in their curriculum, this fact does not necessarily transfer into higher curriculum quality. According to Howley (1994), as cited by Slate (2010), "The value of offering a wide range of specialized courses might be overstated, and that small school with a strong required core curriculum could produce student achievement at high levels" (p. 5).

STUDENT ACHIEVEMENT

Thus, students who are most prepared coming out of high school have a higher chance of succeeding in the post-secondary environment "regardless of who they are, how much money they have, or where they go" (Kuh et al., 2006, p. 19). Multiple studies have found a positive correlation between student achievement and school size (Howley, Smith, & Bickel, 2000; Bingler et al., 2002). Further, Cotton (1996) contends, "the states with the largest schools and school districts have the worst student achievement, affective, and social outcomes" (p. 13).

According to Howley et al. (2000), as cited in Darling-Harmond (2006), "recent literature relating district size to school performance rests almost entirely on an indirect relationship in which socioeconomic status and size work jointly to influence school performance" (p. 30). Fowler and Walberg's (1991) comprehensive study, controlling for factors like socioeconomic status and school expenditures, found that smaller secondary schools in New Jersey "produced higher achievement and higher passing rates on several state tests" (as cited in Darling-Hammond, Milliken, & Ross, 2006, p. 9).

STUDENT ENGAGEMENT

Astin (1997), as cited by Noel-Levitz (2008), states that the "keys to success or graduation are involvement and connection. Involvement refers to both formal academic as well as co-curricular activities" (p. 7). A study by Wehlage and Smith (1992), as cited in Weiss et al. (2010), found that smaller high schools are more likely than larger ones to promote conditions that support and foster student engagement. Similarly, a 2003 study by the National Research Council states that small-school settings foster higher student engagement experience, which, in turn, can improve achievement academically, reduce disaffection and dropout rates. The benefits of student engagement are diverse: higher

grades (Finn & Rock, 1997), lower dropouts rates before completing degrees (Crosnoe et al., 2002), fewer disciplinary issues (Gutman & Midgley, 2000), higher scores on standardized tests (Roeser et al., 1996).

The “expectancy theory” provides a possible explanation by suggesting students are “predisposed” to seek out certain kinds of activities during college (Kuh et al., 2006, p. 14). Perhaps students who are familiar with being engaged within their secondary environment are more apt to pursue similar activities (such as extra-curricular involvement, connection with faculty and advising) on the post-secondary level, thereby improving their overall success and retention. We suggest that expectancy theory plays a role in student engagement in the postsecondary environment and recommend exploring the expectancy theory and student engagement as an area for future study.

Although we found extensive research related to high school size showing an intensive interest in this factor from 1967 to 1992, there appears to be a lack of recent study in this area. Our findings demonstrate that education officials might benefit from a renewed focus on the high school pipeline moving students into the university system. We conclude that a fresh examination of high school size, as well as other relevant demographic factors, would contribute to the current discussion on university retention rates. In addition, researchers should further examine student persistence and retention through the lens of high school size, focusing on curriculum quality, academic achievement, and student engagement. Is the high school’s quality of its curriculum an intervening variable related to size? How does student persistence relate to school size and academic achievement? Is student engagement an intervening variable related to school size? Further research should also explore the relationship of these factors in student retention as possible confounding factors (Weirsma, 2000).

GENDER AND RETENTION

Our study contradicts some existing research in regard to the relationship of gender and retention. Alarcon and Edwards (2013) report “females were 1.59% more likely to leave than males” (p. 135). Today, however, the demographics of college populations have changed and most campuses have larger populations of females than males. Our study finds females were 1.049% more likely to be retained in comparison to their male counterparts.

A study published by the National Center for Education Statistics found that females enrolled in a post-secondary institution had completed their program at a rate of 52% in comparison with their male counterparts at a rate of 46% (Ross, Lauver, Le, Davis, Langley, Carlstrom, 2004). The most influential background variables that influence postsecondary retention, as cited in Clark (2015, p. 87), are "high school grades, the rigor of the high school curriculum, and class rank (Conger & Long, 2010; Buchmann, 2009; Ewert, 2012). Clark (2015, p. 87) continues: "Generally speaking, girls have higher grades and rank, and are more likely to take rigorous courses, particularly in math and science" (Buchmann, 2009; DiPrete & Buchmann, 2013; Peter & Horn, 2005; Reynolds & Burge, 2008; Sax, L.J. 2008).

The findings of our study have implications for higher education administrators who study, oversee, and implement retention practices and programs. Educational leaders at the high school level might also consider how factors such as student engagement and persistence could improve preparation of high school students for post-secondary success.

LIMITATIONS

As with any study, limitations exist. First, this study sought to discover how high school size and gender affected retention specifically at South Dakota State University. Our data reflect whether or not freshmen enrolled in a second year at SDSU. We do not have data to indicate if the students who were not retained had transferred to another institution or if they dropped out of postsecondary education. Secondly, we examine retention only through high school size and gender. Our study does not include relational factors, such as socioeconomic status, parental educational attainment levels, extracurricular engagement, and so forth.

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Unveiling White Privilege in South Dakota

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ABSTRACT

Past research has shown white privilege, or the tendency of a certain group of people to receive increased opportunities and access to the benefits of society, to be commonly experienced by people identifying themselves as "white" (McIntosh**,** 1989). In this study, we explored the relationship between the race of participants and their reported experience of white privilege. The present study examined if this concept is applicable and holds true in current society among Midwestern undergraduate students and the general public in a Midwestern state. A total of 329 participants (46 undergraduate students and 283 from the general public) in a nonrandom sample answered a modified version of Peggy McIntosh's White Privilege Survey (1988) to measure their experience of white privilege. The results revealed that whites experienced the highest degree of white privilege among the general public's responses, the undergraduate students' responses, and my own personal responses. However, the general public reported a lesser perception of white privilege (M=1.8601) overall compared to the undergraduate classes (fall: M=1.6850, spring: M=1.7461) and my own responses (M=1.1975). The results supported my hypothesis that white privilege is a factor in the general public in the state and in the fall undergraduate class of which I was a student. I also predicted that females would report less white privilege experience compared to males, which was confirmed. Identifying the occurrence of white privilege serves an important purpose of highlighting its presence and potential negative effects to those who are unaware of its influence.

INTRODUCTION

According to the US Census Bureau (2014), 85.9% of South Dakotans define themselves racially as "white," or Caucasian. This statistic indicates that a large portion of the Midwest may experience what is known as "white privilege." McIntosh (1989) captures the essence

of this concept by comparing it to an "invisible weightless knapsack [full] of special provisions, maps, passports, code books, visas, clothes, tools, and blank checks" (p.10). In other words, white privilege is the current system in society that allows whites to have more access to benefits, advantages, and opportunities solely because of the color of their skin. These advantages may include anything from readily being approved for a loan, being judged less likely to shoplift, or being less likely to suffer from hate crimes. For instance, Rankin and Reason (2005) discovered that college students of racial and gender minority groups experienced a higher rate of harassment on college campuses than their white and male counterparts. This finding displays an active manifestation of discrimination that stems from unfavorable judgments and stereotypes. Kottak and Kozaitis (2012) claim "unequal access to strategic resources, such as employment and education, disproportionately affects peoples of color" (p. 129). However, this advantage often goes unnoticed by the group reaping the rewards.

White privilege is commonly overlooked and the dominant group is generally not taught how their privileged status affords them benefits in society (McIntosh, 1988). For instance, Peggy McIntosh (1998) maintains that white people are not actively aware of their advantages. Instead, whites are socialized and conditioned not to be conscious of this privilege, which is more often than not blatantly obvious to members of the minority groups. As McIntosh (1998) notes, it is "dizzying" to suddenly be taught of this imbalance and "hard to learn you are being de-centered when you were not aware of being central to begin with" (p. 7). Meanwhile, this factor also makes us prone to be unaware of the privileges given to the dominant group concerning character judgments in society.

It is not uncommon to unconsciously judge minority groups as less favorable, reliable, or honest in everyday life. This tendency is often due to engrained stereotypes that allow people to believe that it is just the way minority groups are "supposed to act" (Kottack & Kozaitis, 2012, p. 80). As McIntosh (1998) notices, "The bad behavior of a person of color, when it occurs or is thought to occur, is unfairly projected onto his or her entire group, whether it is the stereotype of inscrutable shiftiness attributed to Asian Americans, or the gangsterism attributed to Latino men…" (p. 4). However, we do not see a mass generalization applied to whites by the general public or media. Therefore, I wanted to

investigate the presence of white privilege in character judgments using Peggy McIntosh's White Privilege Survey (1988).

METHOD

Participants

There were 329 total participants in the current study. Specifically, the data consists of the responses of 46 students enrolled in an undergraduate sociology course at a Midwestern university, as well as 283 participants from the general public, who were associates of the students. There were 190 females and 139 males. For the purposes of the study, it is important to note that of the total participants for all groups, approximately 75% labeled their race as white, 8% as Native American, 3% as African American, 3% as Asian, and 10% as two or more races. The sample was from a predominantly Midwestern population.

Design and Materials

The study used a correlational design to establish the relationship between the dependent variable of white privilege and the independent variable of race. In addition, gender differences in relation to reports of white privilege are also briefly examined. The survey used in this study is an abridged version of Peggy McIntosh's White Privilege Survey (1988). The current survey consisted of 10 questions instead of the original 46 questions in order to use a more concise method of investigating the participants' experience of white privilege. The hypothesis predicted that participants who identified as being a member of the white race would correlate with a greater level of white privilege, as would being male. In addition, it was predicted that there would be more perception of white privilege among the undergraduate groups than among the general public. This is due to the fact that these undergraduate participants were enrolled in a course that studied race and ethnic relations. Therefore, these participants may be more cognizant of white privilege and have a heightened awareness of these advantages. Thus, they would be more likely to be aware and report its occurrence.

Procedure

Students who were enrolled in a spring and fall semester undergraduate sociology course each completed the survey. Following the individual completion of the survey, each student recruited 10 associates from the general public to take part in the survey. The recruited participants consisted of fellow undergraduate students, as well as other associates of the students from across the state. The responses from the associate participants were compiled into a data set to represent the general public. In addition, all responses collected from each researcher were compiled in a database for further analysis. All participants verbally consented to taking part in the study and the data was recorded anonymously to ensure confidentiality.

For the purpose of this study, I focused my analysis on three particular questions: (2) "I can go shopping alone most of the time, pretty assured that I won't be followed or harassed by store staff." (5) "I can swear, or dress in second hand clothes, or not answer letters, without having people attribute these choices to the bad morals, the poverty, or the illiteracy of my race." (7) "I can do well in a challenging situation without being called a credit to my race" (questions 2, 5 and 7 of the abridged version of the survey). Participants rated the degree to which they agreed with the statement on a four-point scale, with 1 meaning strongly agree, 2 agree, 3 disagree, and 4 strongly disagree. A lower score indicated a greater presence of white privilege experienced by that participant. The three questions chosen for an in-depth analysis specifically concern the degree to which the public judges a person's character to be favorable, reliable, and honest. As stated, I hypothesized that participants who identified as being a member of the white race would correlate with a greater level of white privilege. Therefore, I specifically predicted that white people in both the general public and undergraduate class would feel that their character is judged more favorably by those around them and would consequently be closer to "strongly agree" on the rating spectrum on all three of these questions. I also predicted that my responses and those of the fall 2014 class would show that I would be more likely to strongly agree than the averages of other races in the general public. Once again, this is because I would be more aware of the influence of white privilege and I would be more likely to notice and report its occurrence.

RESULTS AND DISCUSSION

General Public

As a whole, the general public displayed lower levels of white privilege compared to the undergraduates, but still widely experienced white privilege (*M*=1.8601). An average score of 1.8601 suggests that, on average, the general public agreed that they could function in everyday life without generally experiencing these setbacks, which indicates the influence of white privilege. However, this mean is greater than the mean for the rest of the sample, as it is approaching "disagree" on the Likert rating scale. For the three particular statements in question, the general public reported that they felt they could shop alone without being harassed (*M*=1.6784), could swear, dress in clothes, or not answer correspondence without feeling that these choices are attributed to the bad morals of their race (*M*=1.9753), and could do well in a difficult situation without just being called a credit to their race (*M*=1.7138).

For the purpose of this investigation, race is an important factor in the data analysis. The general public consisted of 73.48% identifying as white, 8.60% Native American, 3.58% African American, 3.23% Asian, and 11.11% as two or more races. Table 1 displays the differences in racial makeup of the samples. Clearly, the general public had a greater racial diversity in the sample than the undergraduate class. Therefore, the contrasting racial demographics exemplify the concept of white privilege, as demonstrated by the differences in results between the general public and rest of the sample discussed below.

Table 1: Percentage Race Composition.

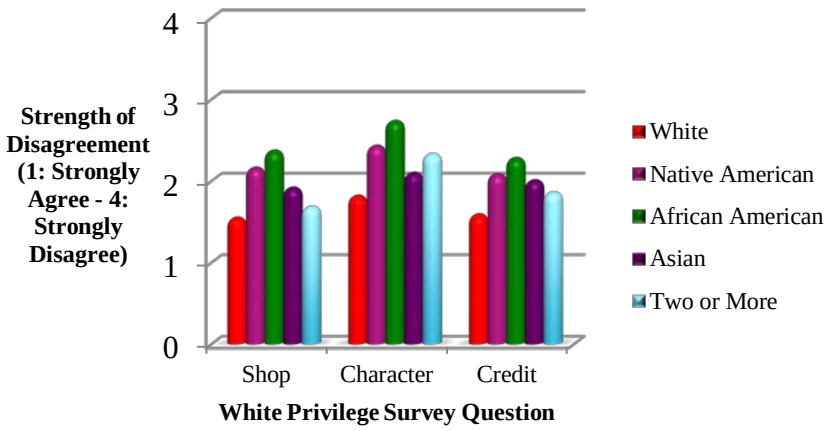
Race	General Public	Fall 2014 Class	Spring 2013 Class
White	73.48	89.34	87.00
Native American	8.60	5.26	4.35
African American	3.58	0	4.35
Asian	3.23	5.26	4.35
Two or more	11.11	0	13.04

**Note.* Based on the data of 279 participants in the general public, 19 participants in the fall 2014 class, and 26 participants in the spring 2013 class.

Fall Class Versus the General Public

First, it is worth noting that for the three questions overall (combined spring 2013, General Public, fall 2014), whites leaned the most towards strongly agree on average, while African Americans were the farthest from strongly agreeing on the continuum (Figure 1). This demonstrates that white people, on average, are more likely to strongly agree that they will not be followed while shopping, will not have people negatively judge their character by their appearance or behavior in relation to their race, and won't surprise people with their achievements compared to stereotypes of others of their race. Meanwhile, African Americans are the most likely to think they will be followed in a store, have others negatively judge their character by their appearance or behavior and apply these factors to their race, and excessively surprise people with their accomplishments.

Figure 1: Mean Race Scores for Question 2, 5, and 7: All Groups

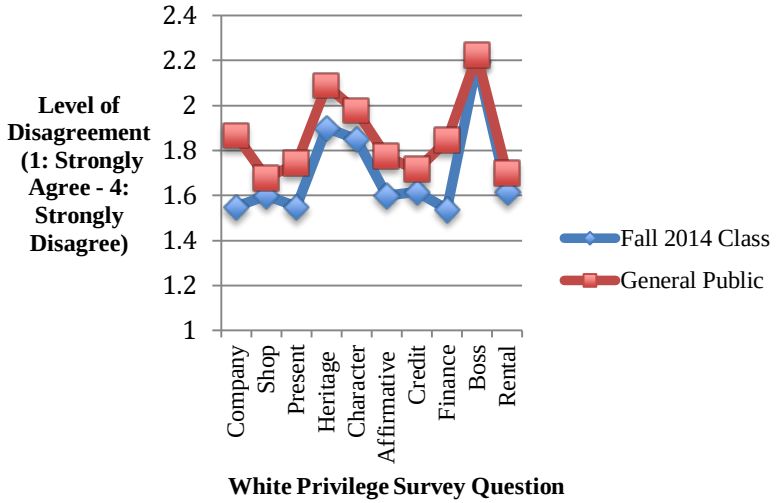


There are noticeable similarities between the general public and our class's responses. For instance, whites in the general public reported an average of 1.5659 for the shop statement, 1.8098 for the character statement, and 1.6000 for the statement concerning credit judgment. This indicates that whites are most likely to agree that they can shop without

being followed, will not be judged for their appearance and behavior in terms of their race, or will not be called a credit to their race for their accomplishments in the general public. This also coincided with the responses of whites in the fall 2014 class for the shop and character questions. Native Americans had one of the highest averages for each question between the general public and the fall 2014 class, meaning they are more likely than whites to think that they will be followed in a shop, may be judged for their appearance and behavior in terms of their race, and may be called a credit to their race for their accomplishments. For example, they scored 2.4583 in the general public and 2.000 for the fall 2014 class for the character question, versus 1.8098 and 1.7059 for whites.

However, there are noticeable differences between the general public and the class of 2014. There was a disparity particularly between the statement on being seen as a credit to their race between the general public and the fall 2014 class. For the general public, whites reported an average of 1.6000, while the fall 2014 class showed an average of 1.4706. This trend held true for all three questions, demonstrating that white undergraduates were more likely to agree to these statements that whites in the general public. Native Americans were also slightly more likely to agree to these statements in the fall 2014 class versus the general public . As a whole for all 10 questions of the White Privilege Survey (1988), the general public was more likely to feel that their race was used by society as a means of preemptively and unfairly judging them, as the general public had a higher overall mean than the fall class (M=1.8601, M=1.6850). However, the overall trend was similar to the responses of the fall 2014 class (Figure 2). As noted in Table 1, the general public had much greater racial diversity. Therefore, it makes sense that a greater number of those participants would be less likely to experience the unfair advantages allotted from white privilege, as more identified themselves as a racial minority in the general public. In addition, the undergraduates were enrolled in a course that studied race and ethnic relations and, therefore, would be more sensitive and aware of white privilege and would be more likely to report its occurrence, as discussed below.

Figure 2: Mean Scores for Question 1-10: Fall 2014 Class and General Public



The fact that the undergraduate classes are a “Race and Ethnic Relations” course and focuses on the relationship between these factors in our society may explain some of this variance. Mainly, this may raise awareness for white privilege among both the fall class and the spring class as well. Therefore, they may be more likely to strongly agree with these statements because we have furthered students’ education on this often unconscious and overlooked privilege. Students who consider themselves of the white race may be more aware of the benefits they have in society and therefore would be more likely to agree with the statements of the White Privilege Survey (1988). This inference is supported by a 2007 study conducted by Case, which discovered that courses in psychology designed to raise student awareness about racism showed an increased awareness of white privilege and an increase in white guilt after the end of the course. Thus, it is not unreasonable to believe that our course has a similar effect on the responses of the fall class, who would, therefore, be more sensitive and aware of white privilege and would be more likely to report its occurrence, as discussed below.

My Responses Versus the General Public

The interactions which form racial advantage and disadvantage occur at the individual level. Further, by examining the responses of an individual to these interactions, a closer understanding of other factors (such as gender) may emerge. As the research data was aggregated, I chose to use my own responses as a point for an individual comparison to the general public. In doing so, I acknowledge that my answers do reflect my status as a student in the area of study, but also point to a larger question of how gender intersects with racial interactions. A comparison of means also showed there were similarities between my own responses and that of the general public. I was also likely to be on the agreeing side of the continuum as I answered mostly 1 to the questions, which is similar to whites in the general public. However, I did have a lower average on the statements concerning shopping and being a credit to my race compared to other whites in the general public, meaning I was more likely to strongly agree to the statements in question. Although, I had a response of 2 to the statement "I can swear, or dress in secondhand clothes, or not answer letters, without having people attribute these choices to the bad morals, the poverty, or the illiteracy of my race," while whites in the public only had an average of 1.8098. This indicates that I was less likely to strongly agree that I will not have my appearance or actions be negatively attributed to my race.

In addition, I recorded a lower response to all three questions compared to the average responses of the general public for every other race (although my score was equal to Asians for the character question). To demonstrate one particular difference, I recorded a 1 for the shop question, which contrasts with African Americans in the general public who recorded an average of 2.3000 to this question (Figure 3). In other words, African American's are noticeably more inclined to believe that they cannot go shopping without being followed or harassed by store staff. As a whole, I had a mean of 1.1975, as compared to the mean score of the general public of 1.8601 (Figure 4). While it is unique to include my personal responses as the researcher in regards to the survey, it offers an individualized perspective on the social phenomenon in question – one that is often difficult to be aware of in relation to one's own life.

Figure 3: My Scores Versus the General Public

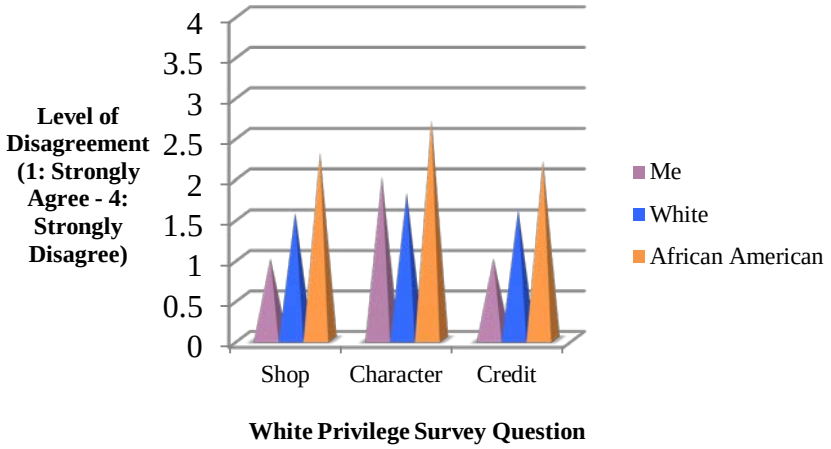
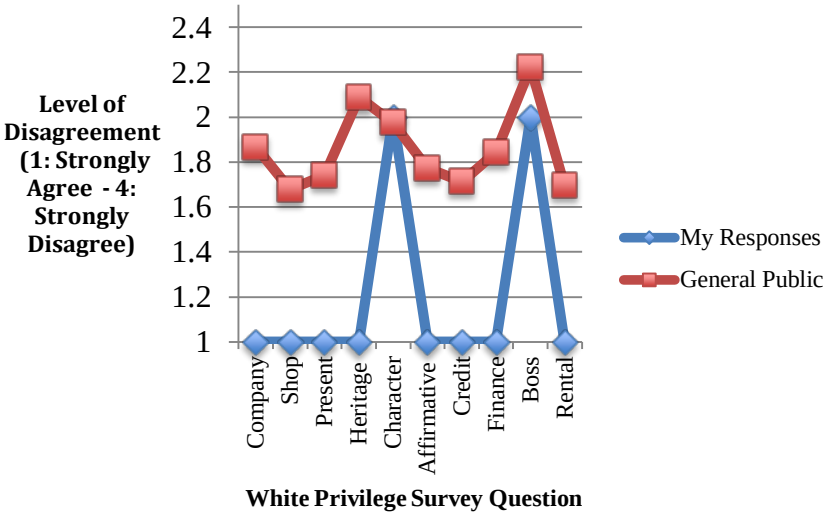


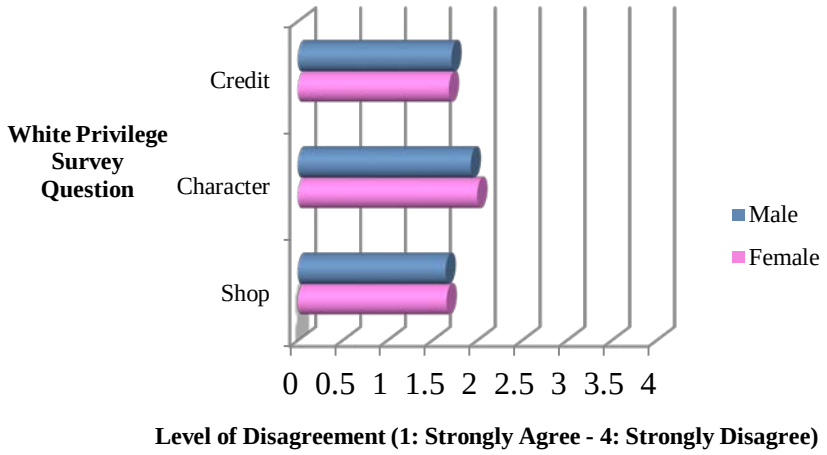
Figure 4: Mean Scores for Question 1-10: My Responses and General Public



Gender Differences

One factor that may contribute to these differences between my scores and the general public responses is gender. Across all the groups, females reported slightly higher scores to the questions concerning shopping and character judgment than males (Figure 5). However, it's interesting that males scored higher on the question concerning credit (1.6842 versus 1.7122). Therefore, females were more likely to think they would be followed in stores or judged by their race for their appearance and behavior, yet they are less likely to think that their achievement in a challenging situation will label them unusual for their race, and therefore, will not be called a credit to their race. Could this reflect the fact that females have more expectations placed on them in the first place? Further investigation is needed to help explain this difference. Due to the fact that I only deviated from the results shown by the analysis in my answer to the shop statement (I was more likely to agree than both males and females, while females overall were less likely to agree than males), I was fairly close to the trends shown with gender differences.

Figure 5: Gender Differences



CONCLUSIONS

Overall, this study displayed that white privilege exists among undergraduate students, as well as the general public. As stated earlier, the results showed that for all three questions whites leaned the most towards strongly agree on average, while African Americans were the farthest from strongly agreeing on the continuum for the combined spring 2013, general public, fall 2014. (Figure 1). This demonstrates that white people, on average, are more likely to have neither their positive nor their negative behaviors seen as racialized, and that any negative judgments would reflect only on the individual, not their race. On the other hand, African Americans are the most likely to think their appearance, achievements and valuation as trustworthy is based not on them individually, but on their race.

My hypothesis stated that the white race is more likely to feel their character judged more favorably by those around them than other racial groups, which is encompassed by the above three questions. According to my analysis in the preceding paragraph, my hypothesis held true for both the general public and the undergraduate classes. My hypothesis that I would be more likely to agree to all three questions than other races in the general public was also confirmed. In addition, the predictions that the general public would report less perception of white privilege than the undergraduate classes, as well as the tendency for females to experience less privilege, was supported, albeit narrowly. Therefore, the results illustrate that white privilege is alive in both society at large and in our individual class.

The research design has limitations that suggest that further study is needed. Firstly, this study only examines 10 questions from the original White Privilege Survey (McIntosh, 1989). Accordingly, this only provides a glimpse into the experience of white privilege among people. Secondly, a study that examines a larger and more diverse sample would be better able to see disparities in everyday experiences influenced by white privilege among differing races. While this current project is an accurate peek into the impacts of white privilege primarily among Midwest individuals, explorations into diverse contemporary populations would provide additional data.

In conclusion we can deduce that the overall findings strongly and clearly indicate white privilege is present in our class and general public. According to the hypothesis of this paper, this privilege also translates into the tendency for white people to be more likely to perceive the public to judge their character to be more favorable, reliable, and honest. This is especially true when comparing whites with African Americans or Native Americans. Accordingly, this illustrates the continuing problem of our society to unconsciously give white people a "knapsack full of special provisions" (McIntosh, 1989, p. 10). While this study concentrates on the often overlooked and unrealized benefits whites are privileged to in society, the demonstration that this discrepancy between races still exists can translate into a more perverse expression of this imbalance. Blatant acts of prejudice and discrimination that still find themselves expressed in society are obvious and outward manifestations of the subtle differences that this white privilege survey unveils. As Kottak and Kozaitis (2012) asserts, "This unequal treatment isn't legal, but it happens anyway" (p. 80). Therefore, the first step to lessening the gap and decreasing these violent and regrettable acts of prejudice is to bring awareness to this imbalance, which is the advantageous purpose of the White Privilege Survey (1988).

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APPENDIX

Abridged White Privilege Survey (McIntosh, 1988)

Please indicate whether you Strongly Agree (1), Agree (2), Disagree (3) or Strongly Disagree (4) with the following statements.

1. I can, if I wish, arrange to be in the company of people of my own race most of the time.
2. I can go shopping alone most of the time, pretty assured that I won't be followed or harassed by store staff.
3. I can turn on the television and see people of my race widely represented.
4. When I am told about our national heritage or "civilization" I am shown that people of my race made it was it is.
5. I can swear, or dress in second hand clothes, or not answer letters, without having people attribute these choices to the bad morals, the poverty, or the illiteracy of my race.
6. I can take a job with an affirmative action employer without having coworkers on the job suspect that I got it because of race.
7. I can do well in a challenging situation without being called a credit to my race.
8. Whether I use a debit card, credit cards or cash, I can count on my skin color not to work against the appearance of my financial reliability.
9. I can be pretty sure that if I ask to talk to "the person in charge," I will be facing a person of my race.
10. I can choose public accommodation or an apartment without fearing that people of my race cannot get in or will be mistreated in the place I have chosen.
11. My gender is: ___Female ___ Male
12. My age is: _____
13. My race is: ____White _____African-American ____Native American ____Asian-American ____Mixed Race
14. My ethnicity is: _____ European _____ Hispanic _____ African _____Asian _____Indigenous ____Other
15. Are you: ____U.S. born? ____ Foreign born?

Evaluating Physiological Responses of Ten Alfalfa (*Medicago sativa* subsp. *falcata*) Germplasm to Drought Treatments

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ABSTRACT

Alfalfa is the most widely produced perennial forage legume in North America. However, its use in the semiarid northern Great Plains is limited due to poor stand establishment and persistence under drought condition. The development of drought-tolerant alfalfa cultivars is of great need. Some *Medicago sativa* subsp. *falcata* populations have demonstrated promising drought resistance when compared to *M. sativa*. Morphological and physiological mechanisms play a critical role in drought tolerance by influencing seedling survival, stand establishment and drought recovery. Assessment of variability in physiological responses to drought among germplasm lines to drought is necessary for developing cultivars with improved drought tolerance. A greenhouse study was conducted to evaluate the variations of stomatal conductance, chlorophyll content, and root to shoot (R/S) ratio of eleven alfalfa populations under drought treatments. Eleven entries included one *sativa*-based commercial cultivar, six *falcata*-based populations, and four rangeland naturalized populations. Uniform seedlings from each population were watered to attain drought treatment regime of 100, 50, and 25% of field capacity. The 50 and 25% treatments created mild and severe drought-stress. Two *falcata*-based germplasms originated under annual precipitation of 250mm and 165mm natural environmental conditions demonstrated the drought tolerant associated traits. They showed either the lowest stomatal conductance or the greatest increased leaf chlorophyll content under severe drought among 11 populations. Both exhibited the highest R/S ratio under severe drought. The results

indicated reducing water loss through minimizing transpiration while increasing root water absorption, and the ability of delaying leaf senescence thus retaining photosynthesis are some key traits that may contribute to drought tolerance in these *falcata*-based alfalfa.

Keywords: alfalfa seedling, drought tolerance, stomatal conductance, root to shoot ratio, chlorophyll, *falcata,*

INTRODUCTION

Alfalfa is one of the most important forage legumes in North America and the world. It has been shown that alfalfa has the highest protein production potential among all legumes and grain. Along with its high nutritive value to livestock, legumes like alfalfa provide ecosystem service by fixating atmospheric nitrogen, which benefits other organisms that depend on them. However, lack of soil moisture and frequent drought are the primary limitation to alfalfa establishment and persistence (Misar et al, 2015), and production (Kang et al. 2011). This is especially prevalent in the semi-arid conditions in the Northern Great Plains (NGP) region.

Drought has been defined by the Society of Range Management as "…prolonged dry weather when precipitation is less than 75% of the average amount" (Society for Range Management, 1989). The studies have shown that drought can reduce crop yields by 69% (Boyer, 1982) and caused extensive economic loss for farmers and ranchers. Global climate change is a major driver of more frequent drought. Northern mixed-grass prairie is expected to exhibit greater precipitation variability, which potential result in more frequent and longer-term drought in the future (Kunkel et al., 2013). Development of alfalfa cultivars with improved drought-tolerant at seedling stage is of great need and crucial for successful stand establishment. *Medicago sativa* subsp. *falcata* is commonly referred to as yellow-flowered alfalfa (YFA). Several YFA populations have been observed to have promising drought resistance when compared to *M. sativa.* Assessment of physiological response variability among germplasm lines to drought stress is necessary for developing drought tolerant cultivars.

A major limitation of using YFA in the semi-arid region is due to relatively low seed production and seed shattering (Berdahl et al. ,1986). This has been solved with the finding of the ability to hybridize with purple-flowered alfalfa (PFA) (*M. sativa*) common around the NGP (Oakley and Garver, 1917). Recently, YFA was found naturalized in northwestern South Dakota in the Grand River National Grasslands. This phenomenon indicates that YFA cultivars could be developed for use in harsh environmental conditions (e.g. drought, cold, and grazing) (Boe et al., 2004).

Interest in development of YFA cultivars has existed since it was first introduced to the United States in 1897 from Siberia by Neils E. Hansen (Oakley and Garver, 1917). Oakley and Garver (1917) observed that PFA is predominantly taprooted with a main single large root propelling downward, whereas YFA is characterized by a deep crown with branched and lateral roots sprawling out horizontally. Fibrous roots found in YFA have been shown to be more tolerant of winterkill conditions and tolerate of injury from grazing and drought when compared to PFA (Garver, 1922).

Plants have evolved diverse physiological and morphological mechanisms to adapt to drought-stress conditions (Blum, 1996). One of these mechanisms is altering resource allocation between root and shoot system, such as increase root-to-shoot biomass ratio (R/S) (Harris, 1992). Under drought conditions, plants reduced shoot production to minimize transpiration, but plants often increase the relative proportion of root biomass for exploiting water in soil (Erice et al., 2010). Another mechanism developed in combating drought is through regulating stomatal conductance. Stomatal conductance regulates CO_2 uptake and water loss of plants. Drought tolerant plants can regulate stomatal conductance under drought conditions to limit water loss (Montague et al., 2008). The ability of plants to sustain chlorophyll content under drought conditions is an important adaptation to continue conducting photosynthesis to support growth, particularly root growth. Chlorophyll content deterioration is an early indicator of leaf senescence (Kang et al., 2011). Leaf senescence causes a loss of photosynthetic capacity and ultimately can lead to plant death.

The objective of this study was to determine physiological responses associated with drought tolerance among alfalfa populations under different drought-stress treatments. The hypothesis is that these physiological traits associated with drought tolerance can be used to develop alfalfa with improved seedling survival, stand establishment, and drought recovery in the future.

METHODS

Seed Source

Table 1: Description and marketer/origin of 11 alfalfa populations used to evaluate physiological traits for drought resistance at a greenhouse of South Dakota State University

* indicates the information of PIs from USDA-ARS National Genetic Resources Program

Entry	Description	Marketer/Origin*
PI631677	*M. sativa* subsp. *falcata, NPGS*	Mongolia, Lat. 49°49’32” N, Long. 92°3’48”E Elev. 1,141m
PI631678	*M. sativa* subsp. *falcata, NPGS*	Mongolia, Lat. 49°46’40” N, Long. 91°53’52”E Elev. 1,463m
PI631682	*M. sativa* subsp. *falcata, NPGS*	Mongolia, Lat. 48°10’33” N, Long. 91°45’29”E Elev. 1,232m
PI502441	*M. sativa* subsp. *falcata, NPGS*	Russian Federation, Lat. 46°11’24” N, Long. 43°53’24”E Elev. 55m
PI538984	*M. sativa* subsp. *falcata, NPGS*	Kazakhstan, Lat. 46°11’24” N, Long. 43°53’24”E
NE1010	*M. sativa* subsp. *falcata, NPGS*	Brookings, SD, Lat. 44°18’41” N, Long. 96°47’53”W
SD201	*M. sativa* subsp. *falcata, NPGS*	South Dakota State University experimental for forage and wildlife habitat, Brookings, SD, Lat. 44°18’41” N, Long. 96°47’53”W
SD202 (Coiled)	Predominantly *M. sativa* subsp. *falcata*	South Dakota State University experimental with coil-shaped seed pods collected from a feral population in native rangeland in northwest SD
SD203 (Sickle)	Predominantly *M. sativa* subsp. *falcata*	South Dakota State University experimental with sickle-shaped seed pods collected from a feral population in native rangeland in northwest SD
Wind River (WR)	Predominantly *M. sativa* subsp. *falcata*	Wind River Seed, Manderson, Wyoming, developed by Norman G. Smith, Lodgepole, South Dakota

Persist II	*M. sativa*, Cultivar, Conventional Hay-Type	Millborn Seeds Inc.

Eleven alfalfa populations were evaluated in this study, including 'Persist II', 'SD201', 'SD202', 'SD203', 'Wind River', 'NE1010', and six plant introductions (PIs) (PI631677, PI631678, PI631682, PI502441, and PI538984). The cultivar Persist II is a *M. sativa* type and was purchased from Millborn Seed Company in Brookings, SD. Seeds of SD201, SD202, SD203, and NE1010 were collected from the experimental plots at the Felt Family Research Farm of the South Dakota State University Agricultural Experiment Station. SD201 is a pure *M. falcata* and SD202 and SD203 were derived from naturalized populations in northwestern South Dakota with predominantly *M. falcata* backgrounds. Seeds of Wind River were purchased from Wind River Seed in Manderson , WY with predominantly *M. falcata* base. All PIs were acquired from National Germplasm System and the selection was based on their local distribution latitudes and climate similarity to South Dakota (Table 1).

Experimental Design

The experiment consisted of the eleven populations with seven replicates under three drought intensity regimes (100, 50 & 25% of field capacity) and two drought durations (15 days and 30 days) after imposed to drought regimes initiated. The 50 and 25% of field capacity treatments created mild and severe drought-stress with soil volumetric water content of 12 and 2.2% (Kang et al., 2011). In pre-experiment, field capacity for 100% was determined by watering soil until saturated and letting drain 24 hours while covering the top to prevent evaporation. Then 50 and 25% field capacity was obtained by measuring changes in cone container (Ray Leach "Cone-containers"; Stuewe and Sons, Inc., Tangent, OR) weight daily from 100% of field capacity to 50 and 25% of field capacity without top cover. The amount and frequencies of water added to cone container to maintain relative consistent of 100, 50, and 25% of field capacity were calculated and determined with 10 replicates for each field capacity regime. All experimental units were maintained in greenhouse conditions under 24±3°C, 16-hour light and 8-hour dark photoperiod for the duration of the experiment.

Experimental Procedure and Treatments

For each population, 150 uniform seeds were selected, scarified with sandpaper, and grown in a growth chamber for 7 days at 20°C (Fig. 1A). Uniform seedlings were taken after 7 days and transplanted into 164ml plastic cone containers filled uniformly with ~46g of Sunshine Mix #3 potting soil (Sun Gro Horticulture Canada Ltd. Seba Beach, AB, Canada with two seedlings per container.

Seedlings were first double planted into cone-containers (Fig. 1B) and allowed to grow for 10 days at 100% of field capacity, then thinned to one seedling per cone-container to ensure plants were relatively uniform in size. Seedlings were then grown for 11 more days under 100% of field capacity to reach trifoliate seedling stage with concomitant root development prior to treatment application.

For 100% of field capacity treatments, 21ml of water was given to every other day at the 9 a.m. to noon CT of the day from the beginning of experiment, 50% field capacity was allowed to dry out for 7 days until it just past 50% and watered back to 50% with 11ml of water every other day, 25% field capacity was allowed to dry out for 12 days just past 25% and watered back to 25% with 7ml of water every other day. Water was applied using 10ml pipette mixed with commercial Miracle-Gro® from the Scotts Miracle-Gro Company.

Data Collection

Root and shoot biomass was harvested at completion of each drought duration regime of 15 and 30 days. Soil was removed from belowground tissues by rinsing with water. Root and shoot biomass were separated by cutting at crown position, and dried in an oven at 60°C for 72 hours. Weight of root and shoot biomass was weighed using a 0.0001 gram scale. Stomatal conductance (SC) was measured using a SC-1 Leaf Porometer (Decagon Devices, Inc., Pullman, WA) during 9 a.m. to coon CT on day 15 after drought treatments were imposed with three repetitions per population per treatment (Fig. 1D). Chlorophyll (C) measurements were obtained using a SPAD 502 Plus Chlorophyll Meter (Konica Minolta, Inc., New Jersey) during 9 a.m. to noon CT on day 16 after drought treatments were initiated with 7 repetitions per population per treatment (Fig. 1C).

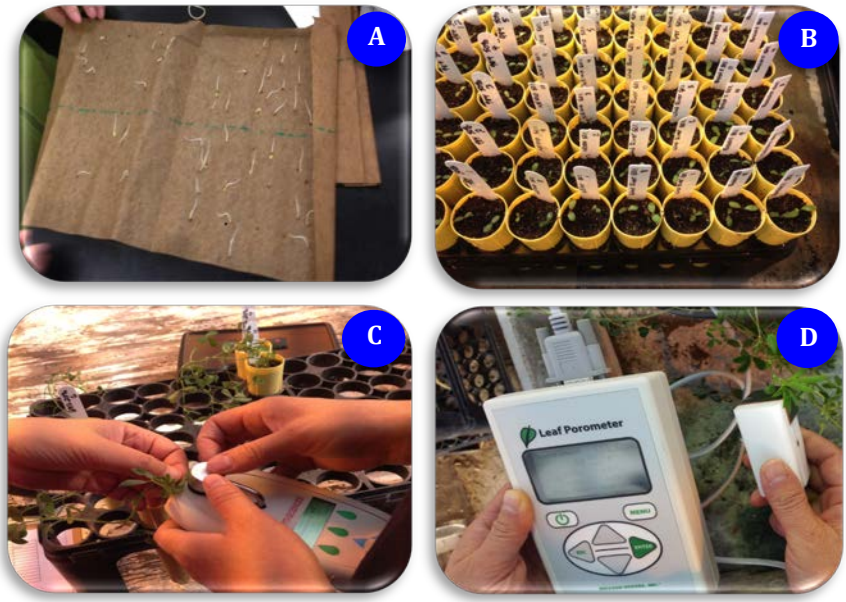

Figure 1: Alfalfa radical growth after 7 days in a growth chamber (A); two emerged alfalfa cotyledon seedlings per container (B); measuring chlorophyll of alfalfa on 16th day after drought treatment initiated (C); and measuring alfalfa stomatal conductance on 15th day after drought treatment initiated (D).

Data Analysis

Root-to-shoot ratio (R/S) was calculated by using the root dry weight divided by the shoot dry. Percent change in chlorophyll content was calculated by finding the difference between the mean chlorophyll content of 100 and 25% of field capacity divided by 100% of field capacity and changing to percent. Differences of physiological traits among populations evaluated with one-way analysis of variance ($p<0.05$) using procedures in SAS statistical analysis software (Institute, 2008) for each drought duration treatment (15 days *vs*. 30 days).

RESULTS

Stomatal Conductance

Stomatal conductance differed among the 11 populations measured at day 15 drought duration treatment at each field capacity (Fig 2). The SC for 25% of field capacity ranged from 11.3 to 36.2 mmol H_2O m^{-2} s^{-1}, 50% of field capacity ranged from 56.5 to 109.3 mmol H_2O m^{-2} s^{-1}, and 100% of field capacity ranged from 71.9 to 135.0 mmol H_2O m^{-2} s^{-1}. The SC of all 11 populations showed the similar response pattern to field capacity treatments, which decreased as field capacity decreased. In general, YFA populations demonstrated lower SC values compared to PFA populations, particularly in 25% of field capacity. PI502441 had the lowest SC and Persist II had the highest SC values (Fig. 2).

Chlorophyll Content

The relative change in chlorophyll content from 100% field capacity to 25% field capacity treatments decreased 25.9% in Persist II and increased 8.7% in PI502441 (Fig. 3). Chlorophyll content increased under severe drought (25% of field capacity) at 15 days drought duration treatment for all PIs (*falcata*-based populations) except PI631682. This indicated that these *falcata*-based populations have the ability of delaying leaf senescence and thus better maintain photosynthesis.

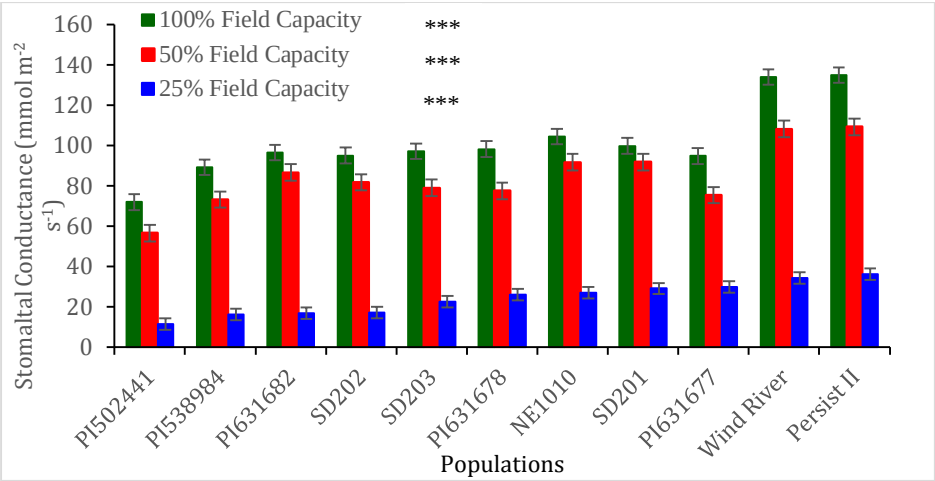

Figure 2: Stomatal conductance for 25, 50, and 100% of field capacity and 15 days drought duration treatment (n = 3) (*** indicates $P < 0.0001$ within each field capacity). Bars indicate standard errors.

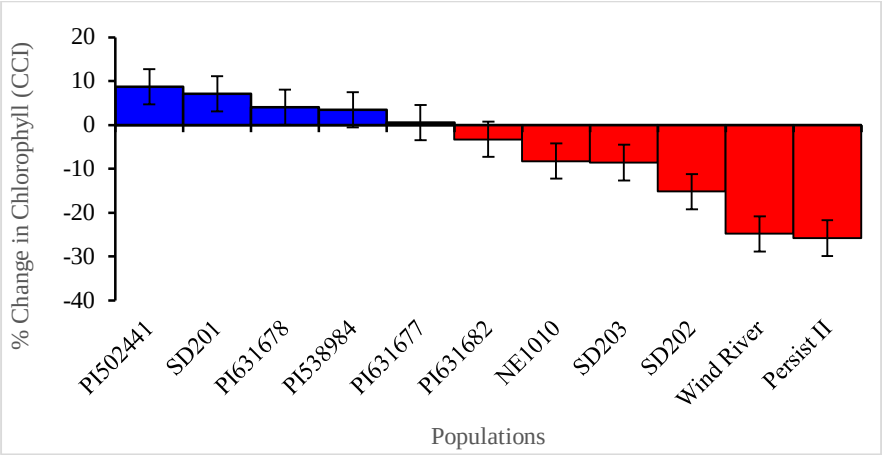

Figure 3: Mean percentage change of chlorophyll content index (based on SPAD readings) from 100% to 25% field capacity (n = 7) at 15 days drought duration treatment ($P < 0.0001$). Bars indicate standard errors.

Root and Shoot Dry Weights

No significance differences in root or shoot dry weight were observed for the 15 days drought duration treatment, whereas for the 30 days drought duration treatment shoot biomass significantly differed among 11 populations for all 3 field capacity treatments. Shoot dry weight for the 25% of field capacity treatment ranged from 0.13 g to 0.39g, for the 50% of field capacity ranged from 0.38 g to 0.66 g, and for 100% of field capacity ranged from 0.84 g to 1.90 g. Shoot biomass decreased as field capacity decreased from well watered to mild to severe drought. Root dry weight for 25% of field capacity ranged from 0.09 g to 0.12 g, no significant difference among 11 populations. For 50% of field capacity, it ranged from 0.18 g to 0.34 g, and 100% of field capacity ranged from 0.09 g to 0.38 g (Fig. 4). Root biomass showed significantly difference among 11 populations under 50% and 100% of field capacities.

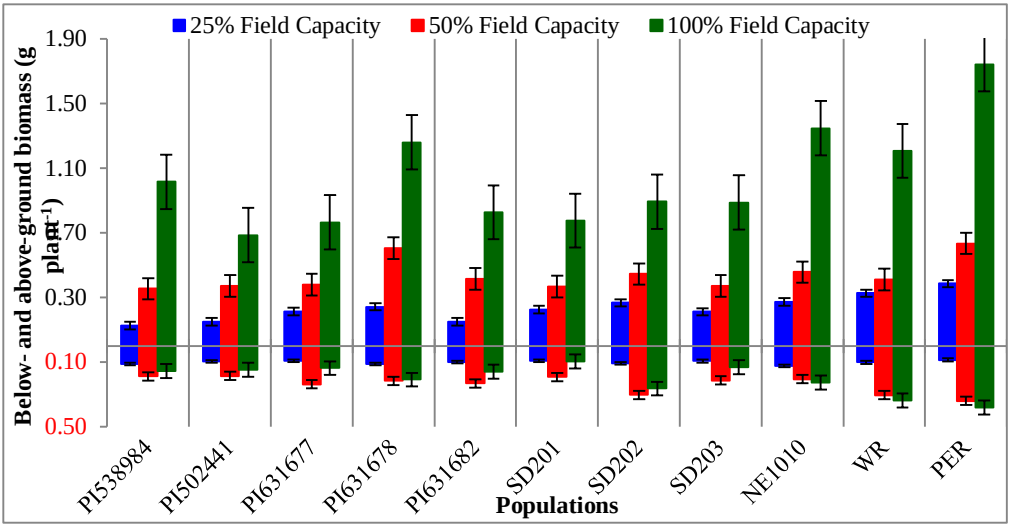

Figure 4: Above and below ground biomass comparison for 25, 50, and 100% field capacity in 30 days drought duration treatment (n = 7). Horizontal line separates aboveground and belowground. Bars indicate standard errors.

The R/S responses were not significantly different in 15 days drought duration treatment for all 3 field capacities. The R/S was significantly different for 30 days under 25% of field capacity, which ranged from 0.23 to 0.97, and PI538984 germplasm had the highest R/S and Persist II had the lowest. The R/S ranked from high to low in order from pure *falcata* lines, predominantly *falcata* lines to *M. sativa* lines, respectively. There was no obvious pattern among 11 populations for 50% field of capacity treatment with R/S ranging from 0.36 to 0.76. For 100% of field capacity treatment, R/S was lower compared to other two drought intensity treatments, but still differed among 11 populations with a range of 0.12 to 0.29 (Fig. 5).

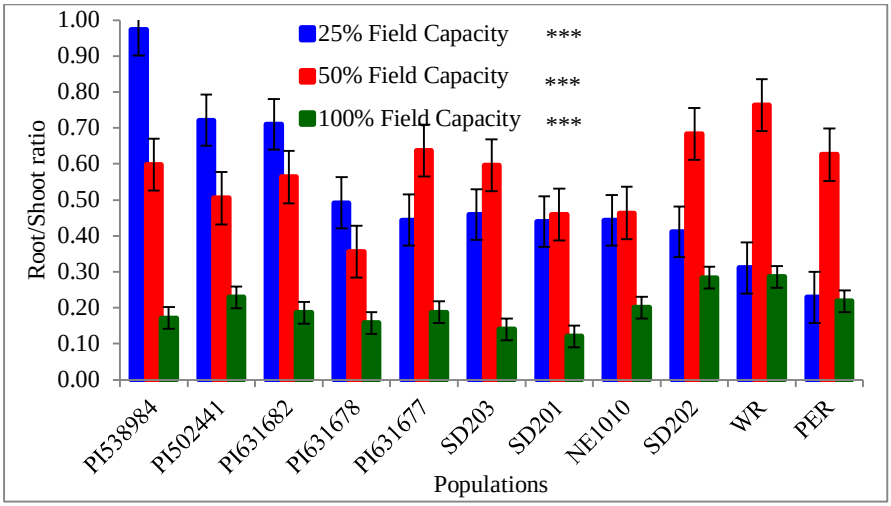

Figure 5**:** Root to shoot ratio for 25, 50, and 100% field capacity and 30 days drought duration treatment (n = 7) (*** indicates $P < 0.0001$ within each field capacity). Bars indicate standard errors.

DISCUSSION

Alfalfa plants responded to drought in a variety of different ways. The main physiological pathways for drought resistance included closing of stomata and reducing of shoot biomass to hinder water loss through transpiration, increasing allocation biomass to root system to facilitate exploiting water absorption, and delaying chlorophyll degradation to maximize resource utilization in needed processes.

Stomatal conductance declined under mild to severe drought and was lower for *falcata*-than *sativa*-based populations. PI502441 had the lowest SC under severe drought (Fig. 2). The results showed *falcata*-based populations are better adapted to regulating transpiration loss under unfavorable soil moisture. Similar results were found by Kang et al. (2011) that showed *falcata*-based populations having a greater SC response to drought. The mechanism for regulating stomatal conductance may be caused by levels of xylem abscissic acid (Tardieu et al., 1992).

Percentage of chlorophyll content change increased under severe drought for all PIs except PI631682. PI502441 had the greatest increase in chlorophyll content, indicating delayed leaf senescence and continued photosynthesis (Fig 3). In contrast, Persist II chlorophyll content decreased more than 25% under severe drought from 100% of field capacity, suggesting chlorophyll degradation, leaf senescence, and reduced photosynthetic capacity. All these physiological processes indicated *falcata*-based populations were more drought tolerant than *sativa*-based populations, which supports Kang, Han et al. (2011). In addition, the chlorophyll content changes under drought conditions showed a correlation with stomatal conductance (Fig 2) (r = -0.5439). With a better control in stomatal conductance the plants can better conserve water to continue photosynthesis.

Root to shoot ratio increased under severe drought for *falcata*-based populations. PI538984 had the greatest R/S, suggesting greater resource allocated to root growth to access water (Fig 5), because water is the most limited factor for their growth under drought condition (Berdahl, et al., 1989). This research found that *falcata*-based alfalfa had dormancy during midsummer drought and slowed down the growth compared to *sativa*-based alfalfa. This leads to better ability to persist through drought conditions to reserve energy and resource. The results of this study showed a slower growth in both above- and belowground biomass under mild to severe drought condition (Fig. 4), which was consistent with Wang et al. (2012).

Falcata-based alfalfa has been found to be tolerant of grazing Misar et al. (2015) and has the ability to develop adventitious shoots from roots (Kannenberg and Xu, 2014). Continued screening to identify germplasm lines will hasten development of better drought, cold, and grazing tolerant cultivars for the semi-arid and arid regions in the world.

LIMITATIONS

This project was conducted in a greenhouse instead of field conditions. Future field-based experiments are needed to better evaluate potential *falcata*-based populations. The implementation of re-watering after drought treatment will allow us to evaluate drought recovery capability among these *falcata*-based populations for resistance and persistence.

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