THE EFFECTS OF CORN ZEIN PROTEIN COUPLING AGENT ON MECHANICAL PROPERTIES OF FLAX FIBER REINFORCED COMPOSITES

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Title

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The Supervisory Committee certifies that this ***disquisition*** complies with North Dakota State University's regulations and meets the accepted standards for the degree of

MASTER OF SCIENCE

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ABSTRACT

In the field of renewable materials, natural fiber composites demonstrate the capacity to be a viable structural material. When normalized by density, flax fiber mechanical properties are competitive with E-glass fibers. However, the hydrophilic nature of flax fibers reduces the interfacial bond strength with polymer thermosets, limiting composite mechanical properties. Corn zein protein was selected as a natural bio-based coupling agent because of its combination of hydrophobic and hydrophilic properties. Zein was deposited on the surface of flax, which was then processed into unidirectional composite. The mechanical properties of zein treated samples where measured and compared against commonly utilized synthetic treatments sodium hydroxide and silane which incorporate harsh chemicals. Fourier transform infrared spectroscopy, chemical analysis, and scanning electron microscopy were also used to determine analyze zein treatments. Results demonstrate the environmentally friendly zein treatment successfully increased tensile strength 8%, flexural strength 17%, and shear strength 30% compared to untreated samples.

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DEDICATION

To my loving wife Samantha, and loving Parents

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ADF	Acid detergent fiber
ADL	Acid detergent lignin
ANOVA	Analysis of variance
CGM	Corn gluten meal
DDGS	Dried distillers grains with solubles
E	Epoxy
FTIR	Fourier transform infrared spectroscopy
NaOH	Sodium hydroxide
NDF	Neutral detergent fiber
NDSU	North Dakota State University
NZ	Sodium hydroxide plus 2.5% zein
SEM	Scanning electron microscopy
UNT	Untreated
VE	Vinyl ester
Z1	1% zein
Z2.5	2.5% zein
Z5	5% zein

LIST OF APPENDIX TABLES

CHAPTER 1. INTRODUCTION

This section introduces flax fiber composites by describing the benefits and disadvantages of the material. The properties of flax fibers as well as treatment methods will be briefly introduced. Finally, a short section on why zein protein was chosen as a coupling agent is presented.

The use of flax fiber polymer matrix composites in engineering applications has increased in recent years. Industry is now geared toward finding renewable and sustainable materials. Research in the field of natural fiber composites has reached the point where natural fibers can be a viable option to replace glass fibers with comparable properties. Currently, the majority of flax fiber composites are used in the production of interior door panels in the automotive industry (1). It is expected the use of natural fiber composites will continue to increase as research efforts strive to improve the performance of these composites (2). The current limitation of natural fiber composite properties can be attributed to their poor adhesion to polymer matrices (3, 4). The poor interfacial adhesion limits the mechanical properties of natural fiber composites by causing the composite to fail by fiber pullout or interfacial shear as opposed to fiber fracture. The majority of research work in the field of natural fiber composites investigates improvement of the interfacial bond between fiber and matrix with surface treatments or coupling agents (3).

Currently, chemical treatments and coupling agents for natural fibers involve the use of harsh chemicals. It is attractive to develop a treatment which is non-toxic and derived directly from a renewable natural source. Corn zein protein has been identified having the properties needed to be a potential natural coupling agent. Zein protein is a molecule with a mix of hydrophobic and hydrophilic properties (5). This mix of hydrophobic and hydrophilic groups on the molecule suggests zein protein to be a candidate to increase bond strength between hydrophilic natural fibers and hydrophobic polymer thermosets. The purpose of this paper is to investigate the effect of zein protein from corn as a coupling agent on the mechanical properties between flax fiber and thermoset polymer matrices.

1.1. Flax fiber advantages

Flax fiber composites have many advantages over conventional mineral fiber composites. The advantages include: low density, high specific properties, increased toughness compared to glass fiber, increased sound absorption, improved biodegradability, environmentally clean, and rapidly renewable (1, 3, 6). Natural fiber composites also offer the possibility of being burned for energy recovery (3, 6). It has also been estimated that flax fibers consume 80% less energy to produce than the production of glass fibers (7). Flax fiber composites have also been shown to possess an advantage in cyclic loading compared to glass fibers. One researcher concluded flax fiber composites do not experience an irreversible strain until the composite is near failure. At failure, flax fiber composites exhibit an irreversible strain of 0.02%. In contrast, glass fiber reinforced composites exhibit a large degree of irreversible strain which increases each cycle linearly up to 0.13%. It has also been shown for cyclic loading, flax fibers experience a much lower drop in tensile modulus (up to about 5% drop) compared to glass fibers which can experience a 37% decrease in modulus during cyclic loading to failure (8). This research demonstrates the potential advantages for flax fibers in cyclic loading compared to glass fibers (8). The reduction in fatigue damage in flax fiber composites is due to toughness of flax fiber compared to glass.

1.2. Flax fiber disadvantages

Flax fiber composites also have several disadvantages. The largest disadvantage of flax fiber compared to glass fiber is poor interfacial adhesion (8, 9). The reason for poor interfacial adhesion can be attributed to the tendency for flax to absorb water. Flax fibers are hydrophilic and therefore exhibit high moisture absorption (8, 9). The hydrophilic nature of flax fibers tends to agglomerate the fibers as opposed to a desired even dispersion of fibers throughout the polymer matrix (10). The hydrophilic nature also results in insufficient wetting of the fibers (10). It has been shown the hydrophilic nature is less of a detriment when the matrix is chosen to be a polar polymer such as unsaturated polyester or epoxy resin (10). Natural fibers are also susceptible to moisture absorption in wet or damp environments which can cause dimensional swelling and a decrease in interfacial strength (10). The final disadvantage of flax fibers is their geometrical and mechanical property variability (8, 9). Unlike glass fibers, flax fiber have a finite length. This prevents flax fibers from being woven as continuous rovings and instead have to be spun into a yarn. Their properties are also highly dependent on the growing season, harvesting, and processing method (7, 11).

1.3. Flax fiber physical properties

The variability of fiber properties is attributed to the environmental factors during the growing and harvesting seasons. Since the flax plant grows to a limited size, flax fibers have a limited length as opposed to synthetic fibers which can be produced in a continuous process. This prevents the use of continuous natural fibers in composites. Natural fibers can be formed into a continuous twine by mechanically interlocking fibers together through twisting. However, because of the twisted nature of twine, it is difficult to achieve the same level of fiber volume fraction as a synthetic fiber roving. Flax fiber specific properties are comparable to glass fibers. A comparison of glass and flax fiber is shown in Table 1.

Table 1: Properties of Flax and E-Glass Fibers (8)

Fiber Type	Fiber length [mm]	Fiber diameter [μm]	Tensile modulus [GPa]	Tensile strength [MPa]	Strain to break [%]	Density [g/cm^3]	Specific Modulus [GPa/(g/cm^3)]	Specific Strength [MPa/(g/cm^3)]
E-glass	continuous	8-14	76	1400-3500	1.8-3.2	2.56	29.69	547-1367
Flax	20-70	10-30	40-100	600-1100	1.5-2.4	1.5	26.7-66.7	400-733

As shown, the tensile strength of flax fiber is significantly lower than that of glass fiber. Tensile strengths of the fiber measure mean values of 850 MPa and 2450 MPa for flax and E-glass fibers respectively. However, when the specific properties are measured, flax becomes competitive with glass fibers with mean specific strength values of 566 and 957 MPa/(g/cm^3) for flax and E-glass respectively. For many applications, such as car interior panels, the stiffness of the material is more important than the strength. The modulus of flax and E-glass fibers are 40-100 GPa and 76 GPa respectively. For specific modulus, flax is able to outperform E-glass fibers with values of 26.7-66.7 GPa/(g/cm^3) and 29.69 GPa/(g/cm^3), respectively. Based on these numbers, it would be theoretically feasible to produce a flax composite panel with a specific stiffness nearly twice that of a comparable E-glass composite panel. Based on the advantages of flax fiber, it should be expected flax fiber will continue replacing E-glass fibers in structural composite applications as improvements continue to be made.

As previously stated, in the field of natural fiber composites development, there is an emphasis on investigating methods to improve interfacial bonding and mechanical properties. This study investigates the use of zein protein as a coupling agent for natural fiber reinforced composites. Currently, there are limited journal articles which investigate this promising natural candidate for improving interfacial bonding strength.

The use of zein surface treatments offers the potential to increase mechanical properties to comparable levels of traditional treatments and coupling agents. Zein protein treatment uses only nontoxic chemicals as opposed to traditional treatments which use strong bases, or toxic chemicals. Zein offers the increase in benefits while being environmentally safe. This goal aligns itself with one of the main purposes of natural fibers- being environmentally sustainable.

Zein protein is extracted from low cost industrial corn byproducts. By developing a zein treatment which uses a solution from these byproducts, it offers the potential to be a low cost alternative to traditional treatment methods while adding value to the growing and harvesting of corn.

CHAPTER 2. BACKGROUND/LITERATURE REVIEW

This chapters provides relevant information about flax fiber structure, harvesting, processing, and treatments. Background on zein protein is also discussed including structure, and why it is of interest to investigate as a coupling agent for natural fibers.

2.1. Structure of flax fiber

The mechanical properties of flax fibers are highly variable. This is due to the variance in growing, harvesting and processing conditions. The spread in mechanical properties is also due to the non-uniform geometry of flax fiber. As explained, flax fibers have an irregular cross sectional shape with a hollow lumen of variable size in the center. The odd shaped geometry creates a challenge to precisely calculate the stress in the fiber.

In a flax stem, elementary fibers are joined in groups of 10-50 fibers (8). These fibers are bound together by pectin (3, 12). For use in composites, it is necessary to split the groups into individual fibers to increase surface area for bonding to the matrix. It has been shown that flax fibers do not have a circular shape as mineral or synthetic fibers, but have a polygonal shape with typically 5-7 sides (13). Figure 1 demonstrates a cross section bundle of polygonal shaped flax fibers.

Fibers within a flax plant have been shown to be thicker near the base of the plants stem and thinner near the top (13). It has been reported fiber diameter can range between 5-76 μm and the length of the fiber range between be 4-77 mm (13). Flax fibers themselves can be thought of as a natural composite. They are composed of four main elements: cellulose, hemicellulose, lignin, and pectin (13, 14).

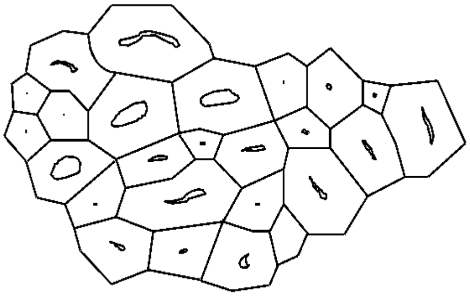

Figure 1: Section of a bundle of fibers (13).

Cellulose is regarded as the most abundant naturally occurring branched linear polymers. The cellulose polymer forms a stiff rod like structure. The strength and stiffness of the fiber is attributed to the cellulose which makes up a majority of the fibers content. It is reported that fully crystalline cellulose nanofibrils can achieve a tensile strength of approximately 10 GPa annd theoretical modulus between 110-220 GPa. (15, 16). Other sources report the cellulose structure of flax fiber is approximately 44% crystalline (17). The higher the crystallinity of the cellulose, the higher the properties of a natural fiber. A cellulose polymer unit cell is shown in Figure 2.

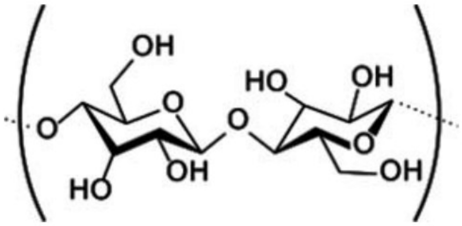

Figure 2: Cellulose polymer unit cell (18).

The composition of flax fibers is shown in Table 2 (13). The structure of the fiber is complex and made up of concentric layers of crystalline cellulose fibrils bonded in an amorphous matrix of lignin and hemicelluloses (13, 19). There are two main concentric phases within the flax fiber, the primary wall and the secondary wall. This structure is shown in Figure 3. In the outermost primary wall the cellulose fibrils are randomly oriented. The secondary wall accounts for the majority of the fiber cross section. In this phase, the cellulose fibrils are aligned at a constant tilt angle with respect to the axis of the fiber (13). The tilt angle of the cellulose micro fibrils in flax fiber has been recorded to be approximately 11° within the primary walls (13). As previously stated the center of the fiber is hollow and is referred to as the lumen (13). The density of flax fibers have been reported to be between 1.44-1.53 g/cm^3 (18, 20).

Table 2: Composition of Flax Fiber (13)

Cellulose (%)	hemi-cellulose (%)	pectin (%)	Lignin (%)	Water soluble (%)	Wax (%)	Water (%)
68.0	14.0	1.8	2.3	3.9	2.0	9.0

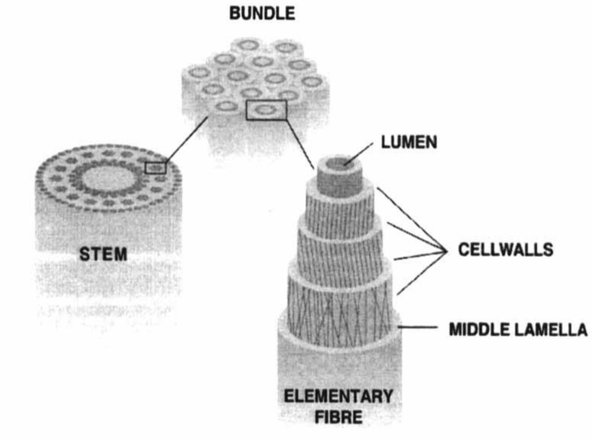

Figure 3: Flax plant and fiber structure (21).

A working knowledge of the structure of flax fiber is necessary to understand their use in polymer matrix composites (13). It has been shown that fiber strength increases and modulus decreases as moisture content increases (22). An increase in moisture content also has a significant increase on the elongation to failure for flax fibers (23). It has been reported the change in mechanical properties due to moisture content can be attributed to hydrophilic nature of hemicellulose within flax fiber (23). The hemicellulose within the fiber draws moisture into the fiber structure and reduces the cohesion of the elements within the fiber increasing the ductility while decreasing the stiffness (23).

As with all materials, defects within the flax fiber structure are detrimental to fiber strength (13). A common defect of flax fibers are nodes. Nodes appear as thickened regions around the flax fiber (13). The sudden change in fiber diameter causes a stress concentration at locations near the nodes. As a result, fibers typically fail at a location of a node (13). Figure 4 depicts a node on a flax fiber.

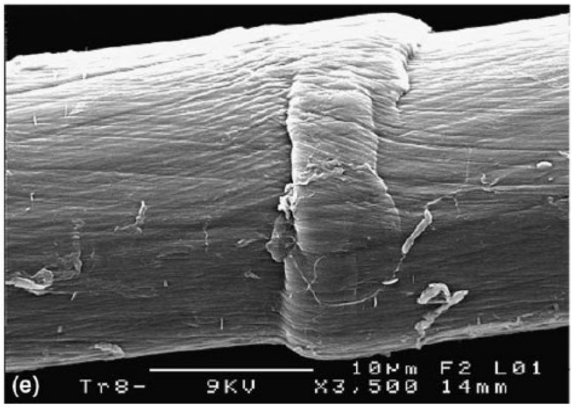

Figure 4: Flax fiber node (12).

2.2. Growing, harvesting, and processing

The cultivation and use of flax (*Linum usitatissimum L.)* has been dated back to ancient Egyptian civilizations (1). Currently, most of the high quality long fiber flax linens are produced in Europe, while short fiber varieties are grown in the U.S. and Canada (1). Throughout history, flax fibers have been used to create textiles and paper products (1, 14). Flax plants create two categories of valuable products, fiber and seed. The seed from the plant is used to create linseed oil and meal (1). The fiber within the flax plant is generated between the outer and inner regions of the stem (7). The fiber region of the stem containing fibers appears as a concentric ring when viewed as a cross section (7). The non-fiber region, called shive, accounts for a majority of the plant mass. Approximately 20-30% of the plant mass is useable fiber (1, 7). A typical yield of flax per acre is reported to be between 7000-8000 lbs (1). Thus, the average useable fiber yield per acre is 1400-2400 lbs (1). The average yield of flax seed per acre for linseed varieties is approximately 1300 lbs., nearly 20% of the total weight (1). In order to produce useable fiber, the shive material must be removed. If shive is not removed it remains attached to the fiber which is detrimental to the spinning process (7). If shive is present in the spinning process of flax fiber, the result is coarser twine with decreased mechanical properties (7). Similarly, shive is also detrimental to composite properties if it is not removed. If shive is attached to the plant fiber when processed into a composite it can result in lower fiber volume fraction and lower properties overall. In order to separate the flax fiber from the shive a series of processes must be accomplished (7).

The first process to separate the fiber from the shive is called retting (1, 7, 14). Retting is a process where bacteria break down the pectin which bonds the fiber to the plant. Retting allows for the fiber to be easily removed from the plant structure. The most common method of retting is dew retting (1, 14, 24). When dew retting is preformed, the plant is cut about 5cm above the ground and left lying in the field. The moisture from dew allows natural microbes to break down the pectin and lignin which binds the fibers together within the plant (1, 14). This dew retting processes can take as long as a month to complete (1). Other types of retting methods include water and enzyme retting. Water retting involves submerging flax stalks underwater. This can be done in natural reservoirs or man-made tanks. Water retting has been shown to provide consistently high properties (11). However, this method is no longer popular because it produces a large quantity of polluted waste water (11). The water retting method also requires an additional drying step (11). The third method of retting involves spraying the stalk with enzymes to break down shive and pectin in as few as 24 hours (11). The enzyme retting method is currently not used in mass production of flax fiber. The major drawback of enzyme retting is the high cost of enzymes. The chosen retting process factors have a large influence on the resulting mechanical properties of the fibers (7).

After the retting process, it is necessary for further mechanical treatments to remove shive material (7). Some mechanical processes to remove shive include: scutching, hackling, carding, sharking, and roller calenders (7, 14). These processes remove shive by bending, pounding, or combing the flax stalks. Figure 5 depicts the rolling process to remove shive. After mechanical processing the fiber is graded based on several properties: weight, strength, fineness, length, luster, straightness, and etc. (14).

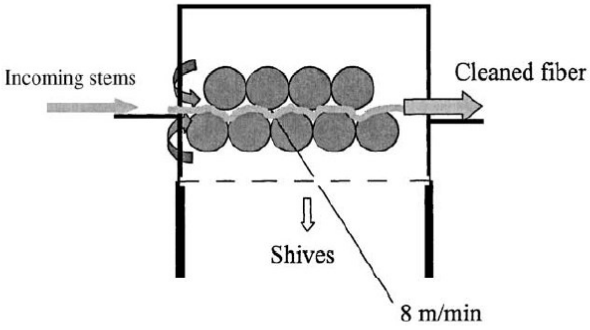

Figure 5: Shive removal by rolling process (7).

2.3. Surface treatments on flax fibers

This section investigates two common surface treatments that are routinely used to treat natural fibers for composite applications. Surface treatments have a large influence on the mechanical properties of natural fiber composites. Surface treatments modify the fiber surface chemically or mechanically to enhance bond strength at the fiber matrix interface.

2.3.1. Alkaline treatment

The most commonly used treatment to change the structure of the fiber is the alkaline treatment. Alkaline treatments use the chemical sodium hydroxide (NaOH) to modify fiber chemical properties (6). This treatment changes the fiber properties by changing the natural flax cellulose which is arranged in a cellulose-I lattice to a different polymorphous form cellulose-II (25). This transformation takes place as a result of cellulose swelling during the reaction (6). In solution, NaOH dissociates into a Na^+ and OH^- ions. The Na^+ bonds to cellulose OH-groups which imbeds the large Na^+ ion into the spaces in the lattice structure which as a result causes the cellulose to swell. When the fiber is washed with water, the Na^+ ions are removed. However, the larger space in the lattice structure remains. This structure is referred to as cellulose-II which is more thermodynamically stable than cellulose-I (6).

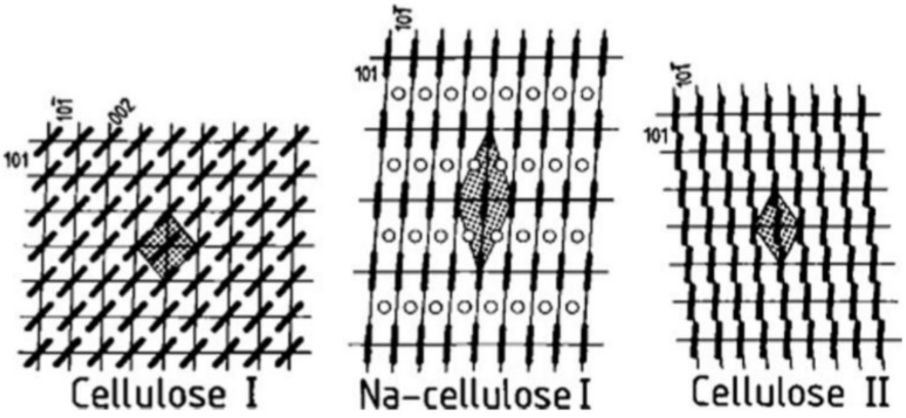

Figure 6: Cellulose I to cellulose II transformation by NaOH treatment (6).

Alkaline treatments also improve properties by removing hemicellulose and pectin from the fiber (14). By removing the hemicellulose and pectin, the fiber bundles are separated into elementary bundles. The separation of elementary fibers and an increase in fiber surface roughness increases the area for the fibers to bond to the polymer matrix (6). It has been shown for concentrations of NaOH at 6% and above, the fiber is reduced to elementary fibers (26). It has also been shown at appropriate concentrations the treatment requires 10 minutes to complete with little change in properties with longer treatment times (6). If the treatment continues for too long of a period a decrease in fiber strength can result from fiber degradation (6, 27). Alkaline treatments have the disadvantages of safety due to use of strong bases and disposal of chemicals after treatment (28).

2.3.2. Silane treatment

Literature reports there are two main mechanisms which increase interfacial bonding. The first type works by changing the structure of the fiber to form stronger bonds with the matrix as discussed in the alkaline treatment section. The second category of treatment is the use of a coupling agent. A coupling agent is a chemical bonded to the surface of the fiber to create a chemical bridge between the fiber and matrix (10). Coupling agent molecules have one group which can achieve a strong bond with the fiber and another group to form a strong bond to the matrix. A proper coupling agent can be used to bond two materials that otherwise would not form strong bonds with one another (10). Coupling agents currently used for natural fiber polymer composites are maleated polyethylene, maleated polypropylene, and most commonly aminopropyltriethoxysilane (silane) (10). The anhydride group found in these compounds is able to form an ester bond with the hydroxyl groups on the surface of natural fibers (10). The chemical bonding of silane and flax fiber is shown in Figure 7. Silanes are an effective type of coupling agent used for both inorganic and natural fibers that have active hydroxyl groups (10). Silane coupling agent surface treatments can be performed by spraying or immersing in silane solution. The disadvantages of silane treatments include: high cost of the chemicals, nonrenewable, toxic, and disposal of chemicals (10).

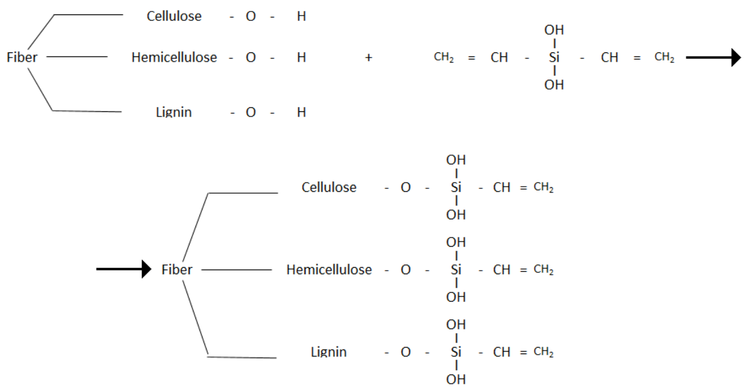

Figure 7: Reaction between fiber and silane (29).

2.4. Zein protein

Zein protein accounts for between 45-50% of the protein found in corn. It has been reported 6-12% of a dry corn kernel is protein by weight. 75% of this protein is contained in the endosperm of the corn kernel (30).

In industry, zein is typically used as a coating because of its desirable properties as a film: tough, glossy, hydrophobic, flexible, and microbial resistant (30). Zein films are used for pill capsules, adhesives, and binders (30).

One journal article reported using zein to treat a nonwoven fabric in a polypropylene matrix (31). The effect of the zein treatment was reported to be a 14% increase in tensile strength, 100% increase in flexural strength, and 40% decrease in impact strength (31). The article supports zein to be an effective coupling agent for increasing interfacial bond strength and thus, increases mechanical properties.

2.4.1. How zein is extracted

The two main corn processing methods which result in protein rich products are wet milling and dry milling (30). Wet milling is intended to extract oil and starch while dry milling is the processes used to produce ethanol (30). The byproducts from wet milling and dry milling are corn gluten meal (CGM) and distiller dried grains with solubles (DDGS), respectively. CGM and DDGS both possess relatively high concentrations of zein protein with 65% and 27% by weight, respectively (30). Pure zein is traditionally extracted from CGM because of its high protein percentage. The method of extraction involves dissolving the zein out of CGM with a 50-90% ethanol/water mixture (30). The remaining solid components of CGM are filtered out of the solution. Then, the zein is extracted from the ethanol/water mixture by evaporation or precipitation (30).

2.4.2. Zein structure

The α-zein molecule occurs in two molecular masses 19 KDa and 22 KDa (32). These two size classes are referred to as Z19 and Z22, respectively. It is reported the purpose for the zein protein in corn is to store carbon and nitrogen for the developing seed (33). α-zein molecules are composed of 20 amino acids. Analysis of the constituent amino acids reveals a majority of the amino acids are hydrophobic leading to the hydrophobic properties of zein (5). The constituent amino acids group to form 9 or 10 repeated helical structures. Glutamine sections within the helical structure act as sites for hydrogen bonding to occur between adjacent helical groups. The same glutamine sections are also the polar amino acid groups of the zein protein. Figure 8 shows the structure of an individual helical structure. The boxed groups are the polar groups which form hydrogen bonds with adjacent helices to form the entire structure of the zein protein. The entire protein structure is shown in Figure 9.

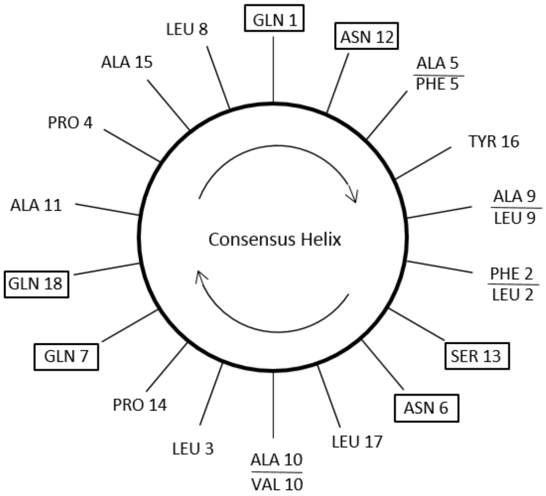

Figure 8: Helical wheel model zein helix (5).

As shown, each helix has three polar groups available for hydrogen bonding. Two of the polar groups are used to form bonds with adjacent helices and the remaining group is available to form hydrogen bonds with neighboring zein proteins. The ends of the helices bond through glutamine rich turn loops. There exists two end terminals on the protein. One end terminal is characterized as the NH_2 terminal and the other the COOH terminal (5). The overall protein has a cylindrical structure with a length to width ratio of 2:1 (5, 32). Figure 10 depicts a model for the intermolecular packing of zein proteins. As shown, the proteins are bonded by a combination of hydrogen bonds and van der Waals interactions. This stacking structure allows zein to form an effective membrane which is why it is commonly used as a coating.

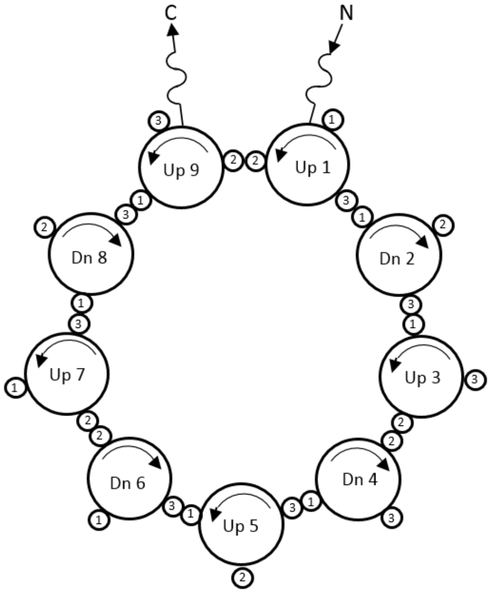

Figure 9: Nine helical protein structure of zein (5).

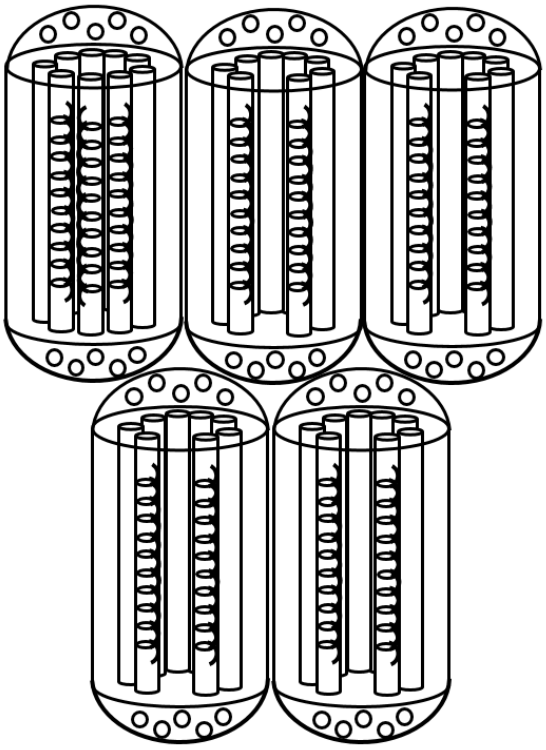

Figure 10: Proposed model for zein molecular stacking (5).

2.4.3. Zein as a natural fiber coupling agent

The mix of polar and nonpolar regions on the zein molecule lend zein to be used as a coupling agent. Zein can be deposited on the surface of natural fibers by immersing the fibers in a zein solution. After being deposited it is expected there will be an interaction between the polar groups of the zein and fiber surface forming hydrogen bonds. The hydrophobic edges of the zein protein will be exposed to provide increased bonding interactions with the polymer thermoset. Thus, the addition of zein protein will enhance interfacial bonding.

CHAPTER 3. OBJECTIVE FOR THE RESEARCH

The objective for this research is to improve mechanical properties of flax fiber composites to advance the technology and use in industry. This research seeks to advance this technology by investigating a potential bio-based coupling agent, corn zein protein. In order to investigate using zein as a coupling agent, it is important to investigate the concentration of zein solution which results in the highest increase of mechanical properties. Part of this study will also investigate if zein treatment benefits are sensitive to matrix type. Therefore, this study will investigate zein treated fiber in two common thermoset matrices- epoxy and vinyl ester. Since, the cost of pure-food-grade zein is high, there is motivation to develop a method to zein treat fiber using a solution made from low cost industrial byproducts: corn gluten meal (CGM) and/or dried distiller grains with solubles (DDGS). For this study, the zein treatments are performed with food grade zein to isolate the effect of the protein on mechanical properties. The industrial corn byproducts offer potential to use zein at a thousandth the cost of the food grade.

To fully understand the mechanisms which influence the treatment effectiveness, the interactions at the microscopic level will be assessed by analyzing the fracture surface by scanning electron microscope (SEM). SEM is helpful to gather qualitative data to support the results of the mechanical tests. Finally, it is important to investigate the effect of the zein protein on interfacial bonding from a molecular perspective by chemical analysis and Fourier transform infrared spectroscopy (FTIR). These six objectives are listed below:

1. Optimize method for zein treatment of flax fibers
2. Characterize and compare mechanical properties for zein treated fiber against common treatments for flax fiber composites
3. Compare effect of zein treatment in epoxy vs. vinyl ester thermosets

4. Develop a method to zein treat fiber from a low cost industrial product zein source (CGM, DDGS)
5. Analyze failure surface for evidence of increased interfacial bonding
6. Investigate chemical interaction of zein protein by chemical analysis and FTIR

CHAPTER 4. EXPERIMENTAL PROCEDURES

4.1. Materials

Unidirectional biotex flax fabric (275 g/m^2) was obtained from Composites Evolution in Chesterfield UK. The epoxy resin used in this study was Araldite 8601/Aradur 8602 epoxy resin system from Freeman Mfg & Supply Co located in Avon, OH. The vinyl ester resin used was Hydropel® R037-YDF-40 vinyl ester resin system from AOC resins. The hardener used to cure the vinyl ester was a 2-butanone peroxide (Luperox® DDM-9) solution from Sigma-Aldrich Co. The food-grade zein protein was purchased from monomer-polymer. Sodium hydroxide (NaOH) and 95% ethanol, were obtained from the Chemistry Department at North Dakota State University (NDSU). Amino ethyl amino propyl tri (methoxy silane) - coupling agent was purchased from Dow Corning. Corn gluten meal (CGM) was obtained from the Department of Animal Sciences at NDSU. Finally, Dried distillers grains with soluables (DDGS) was sourced from Tharaldson Ethanol.

4.2. Material processing

Flax fiber composites were produced by submersing the fiber in a boiling bath of solution for a period of two hours. The formulation of the solution varied depending on the treatment type. Once the treatment was finished, the fiber was oven dried. Next, the fiber was processed into a panel by a hand-layup compression molding method. After curing, these panels were cut into test sample specimens using a wet tile saw. The details of each step are described below.

4.3. Surface treatments

To achieve the objectives of the research, eight treatments where used to produce the fiber to be processed into panels. The procedures for each treatment are outlined in the following sections.

4.3.1. Alkaline treatment

The alkaline treatment was performed by dissolving NaOH into 2.5L 95% ethanol/ distilled water mixture at a ratio of 10g/L. The solution is brought to boil, and then 200g of flax fabric was submerged into the solution. The fiber remained in the boiling solution for two hours. Once the fiber had been in solution for two hours, the fiber was rinsed in a stream of tap water until the waste water did not appear brown/yellow in color. The fibers washing step was completed once the waste water remained clear after passing through the fiber fabric layers. The fiber was then dried at 80°C for 24 hours.

4.3.2. Silane treatment

The silane treatment requires the fibers first be treated using the Alkaline treatment method. The silane treatment was prepared by mixing 2% by weight organo-silane into 80% ethanol/water mixture which was made by diluting 95% ethanol/distilled water with tap water. Then, acetic acid was added to achieve a solution pH of 5. This mixture was mechanically stirred for two hours. Then, 200g of flax fabric was submerged into the solution. The ratio of fiber mass to solutions mass was 1:10. Once the fiber had been in solution for two hours, the fiber was removed and excess solution was removed with paper towel. The fiber was then dried at 80°C for 24 hours.

4.3.3. 1%, 2.5%, and 5% zein treatments

Zein treatments were performed by dissolving food-grade zein protein into 2.5L 80% ethanol/water mixture which was made by diluting 95% ethanol/distilled water with tap water. The 80% ethanol/water mixture was chosen to maximize the solubility of the zein protein. The solution ratios used were 1%, 2.5%, 5% zein mass to solution mass. Throughout this study 1%, 2.5%, and 5% zein treated fibers are referred to as Z1, Z2.5, and Z5, respectively. To perform these treatments, the prepared zein/ethanol/water solution is brought to boil, and then 200g of flax fabric was submerged into the solution. The fiber remained in the boiling solution for two hours. Once the fiber had been in solution for two hours, the fiber was removed and excess solution was removed with paper towel. The fiber was then dried at 80°C for 24 hours.

4.3.4. NaOH + 2.5% zein treatment

The NaOH + 2.5% zein treatment (NZ) was completed by first performing the alkaline treatment method. The fiber was dried for 24 hours. Then, the 2.5% zein treatment was preformed followed by drying the fiber again at 80°C for 24 hours.

4.3.5. CGM, DDGS treatments

CGM and DDGS treatments were performed by dissolving these products into 2.5L 80% ethanol/water mixture at ratios of 7.5%wt, and 18.5%wt, respectively. These ratios were chosen to achieve a solution of 2.5% zein protein by mass. CGM and DDGS were boiled and mechanically stirred for two hours to dissolve the protein. Then, the mixture was filtered to remove the solid particles. The filtered solution was brought to boil, then 200g of flax fabric was submerged into the solution for two hours. Once the fiber had been in solution for two hours, the fiber was removed and excess solution was removed with paper towel. The fiber was then dried at 80°C for 24 hours.

4.4. Composite processing

Composite panels were produced using a hand-layup compression molding process. Twelve unidirectional layers of biotex flax fabric were used to produce each panel. The dimensions of the fiber layers were 120mm wide by 200mm in length. The twelve fiber layers were placed on an aluminum plate at 0° orientation and 150g of resin was slowly poured onto the fiber until it was absorbed. Then, a caul plate of dimensions 200 mm by 92 mm was placed on the fiber and was sealed under a layer of plastic film. The aluminum plate was moved to a press where 3 metric tons of force was applied to the fiber. For the panel size, the applied force results in a pressure of 1.6 MPa. The panel was left in the press, under pressure for 24 hours to allow the resin to cure. A schematic of this process is shown in Figure 11. The test matrix of the panels produced is shown in Table 3. One panel was produced for each fiber type and matrix combination. Three panels were made for untreated (Unt) epoxy (E) to assess the variability due to processing. It should be noted in the results, vinyl ester is abbreviated (VE).

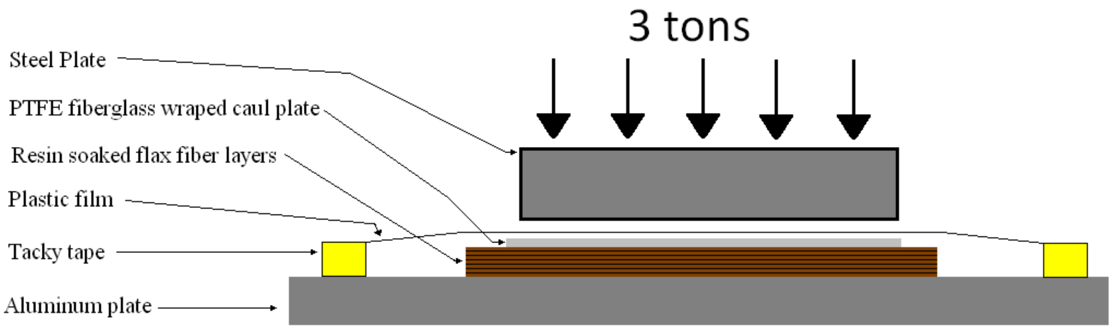

Figure 11: Composite panel processing schematic.

Table 3: Test Matrix and Number of Panels Processed

	untreated	NaOH	Silane	1% zein concentration	2.5% zein concentration	5% zein concentration	NaOH + 2.5% zein concentration	CGM	DDGS
Epoxy	**3**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**
Vinyl ester	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**

4.5. Chemical analysis

Chemical analysis was performed on all nine fiber types by the nutrition lab of the Department of Animal and Range Scenes at NDSU. Data collected from the chemical analysis included: dry matter content, ash, crude protein, nitrogen, neutral detergent fiber %, acid detergent fiber %, acid detergent lignin %, calcium, phosphorus, and crude fat.

4.6. Density testing

Density testing on flax fiber composite was performed using a Mettler Toledo 33360 density determination kit. The density of the composite is calculated:

$$\rho = \frac{w_o}{w_o - w}\rho_o \tag{4.1}$$

Where, w_o is the weight of the composite sample, w is the weight measured when submerged in the fluid, and ρ_0 is the density of the fluid. The density of the composite was used to calculate fiber volume fraction. Fiber volume fraction is calculated by:

$$V_f = \frac{w_f}{w_c}\frac{\rho_c}{\rho_f} \tag{4.2}$$

Where, w_f is the weight of the fiber, w_c is the weight of the composite, ρ_c is the density of the composite, and ρ_f is the density of the fiber. This method of calculating fiber volume fraction is the most accurate because it does not require a value for the density of the matrix material. In practice the density of the matrix will vary because of voids which unavoidably form within the panel. The void content of the panel can be calculated by comparing the experimental fiber volume fraction with the theoretical fiber volume fraction. The theoretical fiber volume fraction is calculated by:

$$V_f = \frac{\rho_f - \rho_r}{\rho_c - \rho_r} \tag{4.3}$$

Where, ρ_f is the density of the fiber, ρ_r is the density of the resin, and ρ_c is the density of the composite. Calculating volume fraction based on known densities of the constituents of the composite and the experimentally measured composite density results in a fiber volume fraction assuming no voids. The void volume fraction of the test panels can be calculated by finding the difference between the theoretical and experimental fiber volume fraction values:

$$V_v = V_{f_{th}} - V_{f_e} = \frac{\rho_f - \rho_r}{\rho_c - \rho_r} - \frac{w_f}{w_c}\frac{\rho_c}{\rho_f} \tag{4.4}$$

It is important to note for this study, the fiber volume fraction calculation is based on measurements of fiber mass and composite mass only within the section of the panel which was compressed under the caul plate. The caul plate section of an as processed panel is shown outlined in Figure 12.

The area of the caul pate was calculated to be 74% of the area of fiber. Since the fiber has a constant areal mass, the mass of the fiber can be assumed to be 74% of the total fiber mass that was processed into the panel. The final composite panel mass is then measured once the sections beyond the caul plate were removed. If these steps were not taken, the fiber volume fraction would have been calculated based on the mass of the total fiber placed into the panel, and the as processed mass of the composite panel. This calculation would not be as accurate as the method used. The area beyond the caul plate was much thicker with the same number of fiber layers.

Therefore, it can be assumed the fiber volume fraction outside the caul plate region had a much lower value. The error induced would have had a large effect on the final normalized results which could have erroneously skewed the findings of this study.

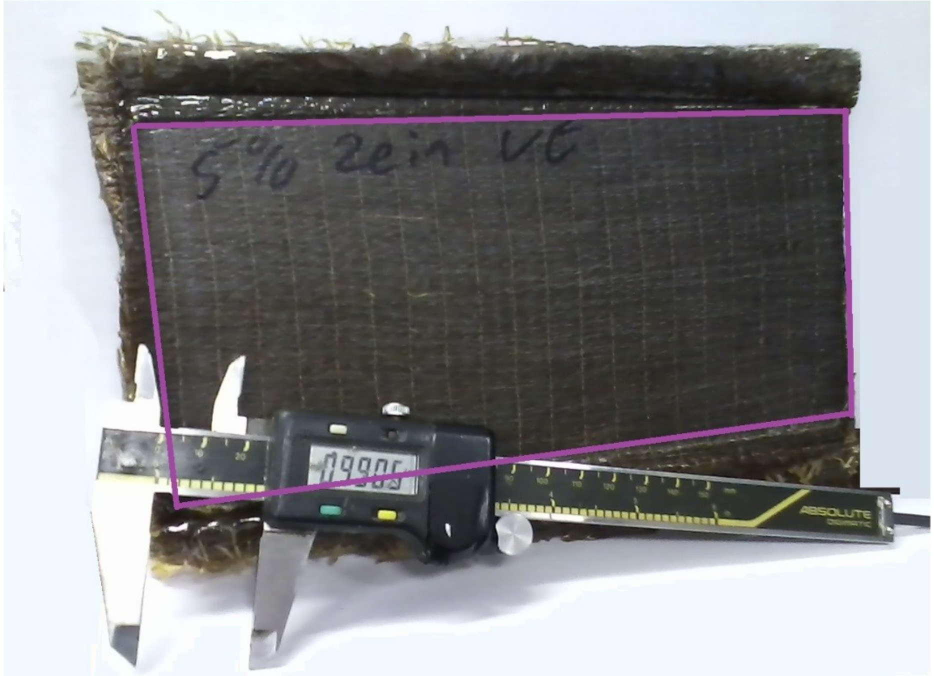

Figure 12: Caul plate section of processed panel.

The fiber volume fraction is important because mechanical properties has been shown theoretically and empirically to correlate with fiber volume fraction. It is impossible to maintain a constant value of for fiber volume fraction for all composites in the test matrix because of variables in the processing such as: treatment type, press pressure, fiber packing in layup, etc. Therefore, mechanical properties are normalized to an average fiber volume fraction between all test panels. The mechanical properties are normalized by:

$$V_n = V_e \left(\frac{V_{f_{target}}}{V_f}\right) \tag{4.5}$$

Where, V_e is the experimental mechanical property, $V_{f_{target}}$ is the target fiber volume fraction, and V_f is the experimental fiber volume fraction. Normalizing by fiber volume fraction was chosen because it does not change the units of the results measured from the experimental testing. For example, if it were chosen to normalize by density a tensile strength measurement when normalized would result in units of MPa/(g/cm^3). While it would be an acceptable method to normalize results, it would be difficult to compare properties to other known materials. For this research, the density of each test panel was measured using tested short beam shear specimens because of their convenient size which worked well with the available testing equipment.

It is important to note, for this study it was desired to keep the fiber volume correction factor to a minimum. A correction factor greater than 15% may skew the data and reduce confidence in the results of the study. Knowing this, all steps were taken during processing to ensure the most consistent manufacturing methodology was achieved.

4.7. Fourier transform infrared spectroscopy and scanning electron microscopy

FTIR spectra were collected for all nine fiber types in the test matrix using a Nicolet 6700 FTIR spectrometer. The equipment was setup to read thirty two scans for each sample between a frequency range of 650 to 4000 cm^{-1}.

SEM captured images of the fracture surface of tensile specimens for samples Unt VE, and Z5 VE. SEM images were also captured for fiber surfaces of untreated, and 5% zein treated fabrics. Samples were prepared for SEM by being sputter coated with gold-palladium (Model SCD 030, Balzers, Liechtenstein.). The SEM images were captured at the Electron Microscopy Center, NDSU using a JEOL JSM-6490LV scanning electron microscope (JEOL, Peabody, Massachusetts, USA) at an accelerating voltage of 15 kV.

4.8. Mechanical testing

This section explains the tests used to evaluate mechanical properties of the composite. These tests were critical to achieve the objectives of the research. The three tests chosen to evaluate mechanical properties of the flax fiber composites were: tensile, flexural, and short beam shear. The three tests were chosen because of their simplicity and real world applications. The tensile test examines the tensile strength, modulus, and strain to failure. These properties are important for direct comparison between capabilities between flax fiber composite and other materials. Flexure test is important because it gauges the ability of the composite to handle a mix of tensile and compressive stresses within the material and also shear within the composite. Natural fiber composites are commonly used in paneling or flooring applications which most closely resembles a flexural loading. Short beam shear testing is important because it is the clearest indicator of fiber matrix interfacial bond strength. This test measures the shear stress at which there is interfacial delamitation. An increase in short beam shear shows in increase interfacial bond strength. The object of this research is to increase interfacial bond strength, which makes the results of the short beam shear test highly significant.

4.8.1. Tensile testing

Tensile testing was performed according to ASTM standard D3039. A minimum of 5 specimens were used to test each panel. The standard does not specify a specific geometry required for test samples. In this study, the tensile specimens were cut on an Allied tile saw model# 70-30010 to a width of 2.5 times the thickness, and a length of 200 mm. The thickness of the samples were the thickness of the panel which varied between 5.1 mm and 4.0 mm. Specimens were conditioned in a Boekel desiccator model# 134041 for a minimum of 24 hours prior to testing. Specimens were tested using an Instron model 5567 load frame. The test speed used was 2 mm/min. This speed was chosen to achieve failure within the specified one to ten minutes. Strain was captured with an MTS extensometer model# 632.25B-20 until stress reached 80 MPa, at which the extensometer was removed. Stress was calculated by:

$$\sigma_i = \frac{P_i}{A} \tag{4.6}$$

Where, P_i is the load, and A is the initial cross sectional area. Strain was captured directly from the extensometer. Modulus was found by determining the slope of the initial linear region of the stress-strain curve. The test was run until the test specimen failed.

4.8.2. Flexural testing

Three point flexure testing was performed according to ASTM standard D790. A minimum of 5 specimens were used to test each panel. The standard specifies a specific geometry required for test samples. In this study, the flexural test samples were cut on an Allied tile saw model# 70-30010 to a width of 2.5 times the thickness. The length of the specimens was 19 times the thickness. Specimens were conditioned in a Boekel desiccator model# 134041for a minimum of 24 hours prior to testing. Specimens weretested on an Instron model 5567 load frame. The test speed used was:

$$R = \frac{0.01\, L^2}{6d} \tag{4.7}$$

Where, L is the span of the test fixture, and d is the thickness of the test specimen. Flexural stress was calculated by:

$$\sigma_f = \frac{3pL}{2bd^2} \tag{4.8}$$

Where, p is the load on the sample, L is the span of the test fixture, b is the width of the sample, and d is the thickness of the sample. Flexural strain was calculated by:

$$\epsilon_f = \frac{6Dd}{L^2} \tag{4.9}$$

Where, D is the deflection at the center of the beam, d is the thickness of the panel, and L was the span of the test fixture. Modulus was found by determining the slope of the initial linear region of the flexural stress-flexural strain curve. The test fixtures were set where the span was 16 times the average thickness of the test samples for a given panel type. This fixture was placed so the center loading fixture was positioned in the middle of the two end fixtures. The test fixture supporting the sample at the ends had a diameters of 3.18 mm and the test fixture applying load at the center was 6.40 mm in diameter. The test is stopped when either, there was a 40% drop in the load or flexural strain exceeded 5%.

4.8.3. Short beam shear

Short beam shear testing was performed according to ASTM standard D2344. A minimum of 5 specimens were used to test each panel. The standard specifies a specific geometry required for test samples. In this study, the test samples used had a width of two times the thickness and a length of 6 times the thickness. Specimens were conditioned in a Boekel desiccator model# 134041 for a minimum of 24 hours prior to testing. Specimens were tested on an Instron model 5567 load frame. The test speed used was 1 mm/ min as specified.

Short beam strength was calculated by:

$$F = 0.75 * \frac{p_m}{bh} \tag{4.10}$$

Where, P_m is the maximum load, b is the measured specimen width, and h is the measured specimen thickness. The test fixtures were set to have a span four times the average thickness of the test samples for a given panel type. This fixture was placed so the center loading fixture was positioned in the middle of the two lower fixtures. The test fixture supporting the sample at the ends had diameters of 3.18 mm and the test fixture applying load at the center was 6.40 mm in diameter. The test is stopped when either, there was a drop in the applied load or the head travel exceeded the thickness of the sample. For this test, it is important the sample failed with the proper failure mode. The proper failure mode is determined by the presence of an interlaminar crack or delamination. If a sample showed signs of tensile or compressive failure, the sample test results were not used.

4.9. Statistical methods

Statistical methods are important in the evaluation of data in this thesis. Statistical methods are useful to systematically draw conclusions from the results. Statistics define methods to state if the two sample results are actually different and if a correlation exists, as opposed to just speculation based on observation. The statistical methods used in this research include: the T-test, Least squares linear fit, and the ANOVA test. The statistical tools used are useful to provide solid evidence to support conclusions from the data which may not show clear trends. The methods and mathematics of the statistical operations are explained in the following subsections.

4.9.1. Test statistic and p-value

A test statistic assuming unequal variance is used to determine the probability of two set having a common sample mean. The first step in the test statistic is to determine the t value. The t-value t is:

$$t = \frac{\mu_1 - \mu_2}{\sqrt{\frac{s_x^2}{n} + \frac{s_y^2}{m}}} \tag{4.11}$$

Where, $\mu_1$is the sample mean of the first set of data, $\mu_2$is the sample mean of the second set of data, s_x^2 is the variance of the first set of data, s_y^2 is the variance of the second set of data, n is the number of samples in the first set, and m is the number of samples in the second set. The next step is to calculate the degrees of freedom. The degrees of freedom df is calculated as:

$$df = \frac{\left(\frac{s_x^2}{n} + \frac{s_y^2}{m}\right)^2}{\left(\frac{1}{n-1}\right)\left(\frac{s_x^2}{n}\right) + \left(\frac{1}{m-1}\right)\left(\frac{s_y^2}{m}\right)} \tag{4.12}$$

Where the variables are the same values as in equation (4.11). Since this formula does not result in an integer value, it must be rounded to the nearest integer. The two calculated values are then used to lookup a P-value on a T-table. First, find the row on the table that corresponds to the degrees of freedom. Then look for the columns for which the T-value is in-between. Then following these two columns to the first row, there will be two P values. To approximate the exact P-value for the T-test, an interpolation between the values must be performed. The resulting P-value must be doubled since this is a two sided test. If the final P-value is greater than the predetermined α cutoff value, then there is not enough evidence to reject the null hypothesis. If the P-value is less than α, then the null hypothesis is rejected and is evidence to support the alternate hypothesis.

4.9.2. Least squares linear fit

The least squares linear fit is used to investigate correlations between quantitative variables. Using this method produces a graph for which demonstrates trends on an easy to interpret graph. The least squares linear fit generates a best fit line for data on an x-y scatter plot, for which the x coordinates is the independent variable and the y coordinates is the dependent variable. The best fit line is a linear equation of the form:

$$f(a,b) = a + bx \tag{4.13}$$

Where, a is the y-intercept and b is the slope of the liner model. Values for a and b are found by solving the matrix equation:

$$\begin{bmatrix} a \\ b \end{bmatrix} = \begin{bmatrix} n & \sum_{i=1}^{n} x_i \\ \sum_{i=1}^{n} x_i & \sum_{i=1}^{n} x_i^2 \end{bmatrix}^{-1} \begin{bmatrix} \sum_{i=1}^{n} y_i \\ \sum_{i=1}^{n} x_i \, y_i \end{bmatrix} \tag{4.14}$$

Where, x_i and y_i are the independent and dependent variables value corresponding to the ith values, respectively.

The overall goodness of fit is measured by the correlation coefficient. The correlation coefficient is found by the equation:

$$R^2 = \frac{ss_{xy}^2}{ss_{xx}\, ss_{yy}} \tag{4.15}$$

Where, ss_{xy} is defined as:

$$ss_{xy} = \sum_{i=1}^{n} (x_i - \bar{x})(y_i - \bar{y}) \tag{4.16}$$

ss_{xx} is defined as:

$$ss_{xx} = \sum_{i=1}^{n} (x_i - \bar{x})^2 \tag{4.17}$$

and ss_{yy} is defined as:

$$ss_{yy} = \sum_{i=1}^{n} (y_i - \bar{y})^2 \tag{4.18}$$

An R^2 value from 1.0 to 0.7 is generally regarded as a very strong correlation, from 0.69 to 0.5 is regarded as a good correlation, 0.49 to 0.25 is typically a weak correlation, and below 0.25 is generally not strong enough to conclude a correlation exists.

4.9.3. ANOVA test and pairwise t-test

The analysis of variance (ANOVA) test preforms a function similar to the T-test. The ANOVA test is used to test for a significant difference between the means for three or more sets in a single test. The results of ANOVA test will have a form as shown in Table 4. The most important result of ANOVA is the P-value. The P-value represents the chance of an erroneous rejection of the null hypothesis due to random sampling chance based on the normal distribution. If the P-value is less than α the specified cutoff value (typically 0.05) then there is enough evidence at least two of the sets have a different mean value.

Table 4: Example Results of ANOVA Test

ANOVA						
Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	38.1	2	19.1	13.2	9.36E-04	3.9
Within Groups	17.3	12	1.4			
Total	55.5	14				

The values in the ANOVA table are calculated as following. The SS column represents sum of the squares. $SS_{between}$ is calculated as:

$$SS_{between} = \frac{(\sum X_1)^2}{n_1} + \frac{(\sum X_2)^2}{n_2} + \cdots + \frac{(\sum X_m)^2}{n_m} - \frac{(\sum X)^2}{N} \tag{4.19}$$

SS_{total} is calculated as:

$$SS_{total} = \sum X^2 - \frac{(\sum X)^2}{N} \tag{4.20}$$

SS_{within} is calculated as:

$$SS_{within} = SS_{total} - SS_{between} \tag{4.21}$$

Where, X_1 is the data from set one, X_2 is the data from set two, and so on unit the last data set X_m. Similarly, n_1 is the number of samples in set one, n_2 is the number of samples in set two, and so on unit n_m is the number of samples in the last set. X represents the data from all sets, and N is the total number of data points.

The df column represents degrees of freedom. $df_{between}$ is calculated as:

$$df_{between} = \alpha - 1 \tag{4.22}$$

And df_{within} is calculated as:

$$df_{within} = N - \alpha \tag{4.23}$$

Where, α is the number of data sets, and N is the total number of samples.

The mean square (MS) column is calculated using the same equation for both the between and within rows. MS is calculated as:

$$MS = \frac{SS}{df} \tag{4.24}$$

Where, SS is the sum of the squares for the row, and df is the degrees of freedom in the row.

The F-value is calculated as:

$$F = \frac{MS_{between}}{MS_{within}} \tag{4.25}$$

This F-value is then compared with an F-crit value to determine if as statistical difference exists between any two sets. An F_{crit}-value can be found on an F-table and is a function of $df_{between}$ and df_{within}. If the F-value is larger than the F_{crit} –value then there is at least one statistically difference between two or more sets of data. The exact P-value from the ANOVA test must be found using statistic software packages. It should be noted the ANOVA test can only determine if a statistical difference exists and cannot determine where these statistical differences are found.

To determine where the statistical differences are found, a Pairwise T-test can be performed. A Pairwise T-test is simply preforming a T-test for each combination of sample set. The results is a table which lists the samples types along the first column and the first row and lists the resulting P-value from the T-test comparing the two samples. The number of comparisons preformed in a pairwise t test is:

$$N = \frac{m(m-1)}{2} \tag{4.26}$$

Where, m is the number of sample types in the ANOVA test. For this work, the table is color coded to highlight the results of the test. Values less than α are color coded dark blue to signify a statistical difference exists. Values between 2α and α are color light blue to show there is high probability of a statistical difference but not enough to evidence to suggest their mean values are actually different.

CHAPTER 5. RESULTS AND DISCUSSIONS

This chapter presents the results obtained by experimentation in this study. The purpose of this section is to determine the impact of various zein treatments in flax fiber composite samples. Mechanical test results are reported first, followed by non-mechanical test results. The non-mechanical test results will be used to help explain the results determined from mechanical testing.

5.1. Density testing results

The density of each test panel was measured using the tested short beam shear samples. The density of the flax fiber is assumed to be equal to 1.44 g/cm^3 for all treated fiber types. The density of the matrix is not an input to determine fiber volume fraction because it is accounted for by the fiber mass, composite mass, and composite density. Density of the samples were found to vary between 1.27 g/cm^3 and 1.35 g/cm^3 with a maximum measurement error of 0.02 g/cm^3. The resulting fiber volume was found to vary between 0.44 and 0.57. It is observed the calculated fiber volume fraction and measured densities vary approximately 27% and 6% from minimum to maximum values, respectively. It is difficult to determine a true error value for the fiber volume fraction. The only portion of error able to be measured is due to the uncertainty from density measurement. However, it is possible to estimate the uncertainty of the measurement by differential analysis.

Using differential analysis the uncertainty in the fiber volume calculation can be estimated based on an assumed level of tolerance in the values used in the formula. The uncertainty due to a single variable in the equation is estimated as:

$$u_i = \frac{\partial f}{\partial x_i}\,\delta x_i \tag{5.1}$$

Where, $\frac{\partial f}{\partial x_i}$ is the partial of the function with respect to the variable of interest, and δx_i is the assumed tolerance in the variable value. Using this equation will result in a series of uncertainties which can be combined using the root-sum-squares method to result in the uncertainty of the calculation. The root-sum-squares method is described as:

$$u_x = \sqrt{u_1^2 + u_2^2 + \cdots u_k^2} \tag{5.2}$$

Where, $u_1,\ u_2,\ \ldots.,u_k$ are elemental errors. The partial derivatives of equation 4.2 are found to be:

$$\frac{\partial V_f}{\partial w_f} = \frac{1}{w_c}\frac{\rho_c}{\rho_f} \tag{5.3}$$

$$\frac{\partial V_f}{\partial w_c} = -\frac{w_f}{w_c^2}\frac{\rho_c}{\rho_f} \tag{5.4}$$

$$\frac{\partial V_f}{\partial \rho_c} = \frac{w_f}{w_c}\frac{1}{\rho_f} \tag{5.5}$$

$$\frac{\partial V_f}{\partial \rho_f} = -\frac{w_f}{w_c}\frac{\rho_c}{\rho_f^2} \tag{5.6}$$

The values for the uncertainty analysis were chosen from sample Unt 1 E. These values are: $w_f = 59.6g, w_c = 99.6g\ , \rho_c = 1.31\frac{g}{cm^3}, and\ \rho_f = 1.438\frac{g}{cm^3}$. The tolerance of the values are: $\delta w_c = 0.05, \delta\rho_c = 0.02, and\ \delta\rho_f = 0.02$. These values are based on the confidence interval of the values. Choosing a value for the tolerance of the fiber mass is not as straight forward. It was assumed the area of the caul plate was within 3% of the calculated 74%.

Therefore the tolerance of the fiber mass was calculated to be: $\frac{wf}{0.74} * 0.03 = \frac{59.6g}{0.74} * 0.03 =$ 2.415g. Thus, the resulting uncertainties due to each terms is:

$$u_{w_f} = \frac{1}{w_c}\frac{\rho_c}{\rho_f}\ \delta w_f = \frac{1}{99.6\ g}\frac{1.31\frac{g}{cm^3}}{1.438\frac{g}{cm^3}}\ 2.415g = 0.0221 \tag{5.7}$$

$$u_{w_c} = -\frac{w_f}{w_c^2}\frac{\rho_c}{\rho_f}\ \delta w_c = -\frac{59.6\ g}{(99.6\ g)^2}\frac{1.31\frac{g}{cm^3}}{1.438\frac{g}{cm^3}}\ 0.05g = -0.000274 \tag{5.8}$$

$$u_{\rho_c} = \frac{w_f}{w_c}\frac{1}{\rho_f}\ \delta\rho_c = \frac{59.6\ g}{99.6\ g}\frac{1}{1.438\frac{g}{cm^3}}\ 0.02g = 0.00832 \tag{5.9}$$

$$u_{\rho_f} = \frac{1}{w_c}\frac{\rho_c}{\rho_f}\ \delta\rho_f = -\frac{59.6\ g}{99.6\ g}\frac{1.31\frac{g}{cm^3}}{(1.438\frac{g}{cm^3})^{\ 2}}\ 0.02g = -0.00760 \tag{5.10}$$

Using the root-sum-squares method the uncertainty of the calculation is found to be:

$$\begin{aligned} u_{Vf} &= \sqrt{u_{w_f}^2 + u_{w_c}^2 + u_{\rho_c}^2 + u_{\rho_f}^2} \\ &= \sqrt{0.0221^2 + (\ -0.000274)^2 + 0.00832^2 + (-0.00760)^2} \\ &= 0.0248 \end{aligned} \tag{5.11}$$

Using equation 4.2 the fiber volume fraction for sample Unt 1 E is calculated to be 0.5475±0.0248. Differential analysis can be performed again on equation 4.5 to determine the resulting uncertainty on the fiber volume fraction normalization factor.

$$\frac{\partial_{cf}}{\partial_{Vf}} = -\frac{V_{f\ target}}{V_f^2} \tag{5.12}$$

$$u_{cf} = \frac{V_{f\ target}}{V_f^2}\ \delta v_f = -\frac{0.5}{0.5475^2}\ 0.0248 = -0.0413 \tag{5.13}$$

This result means it is expected the normalization factor is accurate within approximately 4.13% of the true normalized factor. While it would be preferable to achieve a smaller uncertainty level, this is the best that can be accomplished for the processing methods available. It should be noted on the basis of this analysis mean, values of less than 5% difference should not be deemed as significantly different. Table 5 shows the results of density testing and Figure 13 plots the correction factors used for this study. The error bars in Figure 13 represent the uncertainty interval of the correction factors.

Table 5: Density Testing and Fiber Volume Fraction Results

Sample Type	Composite Density (g/cm^3)	Flax fiber Density (g/cm^3)	Fiber Mass (g)	Caul Plate Area Fiber Mass (g)	Composite Mass (g)	Fiber Volume Fraction	Property Correction Factor
Unt 1 E	1.31±0.01	1.438	80.5	59.6	99.6	0.5475	0.91
Unt 2 E	1.29±0.01	1.438	78.9	58.4	98.0	0.5393	0.93
Unt 3 E	1.31±0.02	1.438	78.8	58.3	94.0	0.5668	0.88
NaOH E	1.28±0.01	1.438	68.6	50.8	101.0	0.4501	1.11
Silane E	1.29±0.01	1.438	72.6	53.7	104.0	0.4653	1.07
Z1 E	1.31±0.01	1.438	77.8	57.6	98.0	0.5369	0.93
Z2.5 E	1.27±0.01	1.438	78.8	58.3	106.0	0.4885	1.02
Z5 E	1.27±0.01	1.438	81.5	60.3	104.0	0.5165	0.97
NZ E	1.29±0.02	1.438	72.4	53.6	104.0	0.4636	1.08
CGM E	1.28±0.01	1.438	84.0	62.2	106.0	0.5247	0.95
DDGS E	1.29±0.01	1.438	82.7	61.2	104.0	0.5320	0.94
Unt VE	1.32±0.02	1.438	79.8	59.1	105.6	0.5157	0.97
NaOH VE	1.34±0.02	1.438	71.9	53.2	102.8	0.4864	1.03
Silane VE	1.32±0.00	1.438	74.1	54.8	114.0	0.4435	1.13
Z1 VE	1.32±0.01	1.438	79.9	59.1	106.0	0.5140	0.97
Z2.5 VE	1.31±0.00	1.438	78.0	57.7	110.0	0.4797	1.04
Z5 VE	1.29±0.01	1.438	84.0	62.2	124.0	0.4507	1.11
NZ VE	1.32±0.01	1.438	72.5	53.7	108.0	0.4572	1.09
CGM VE	1.30±0.01	1.438	81.0	59.9	114.0	0.4771	1.05
DDGS VE	1.32±0.01	1.438	82.4	61.0	116.0	0.4842	1.03

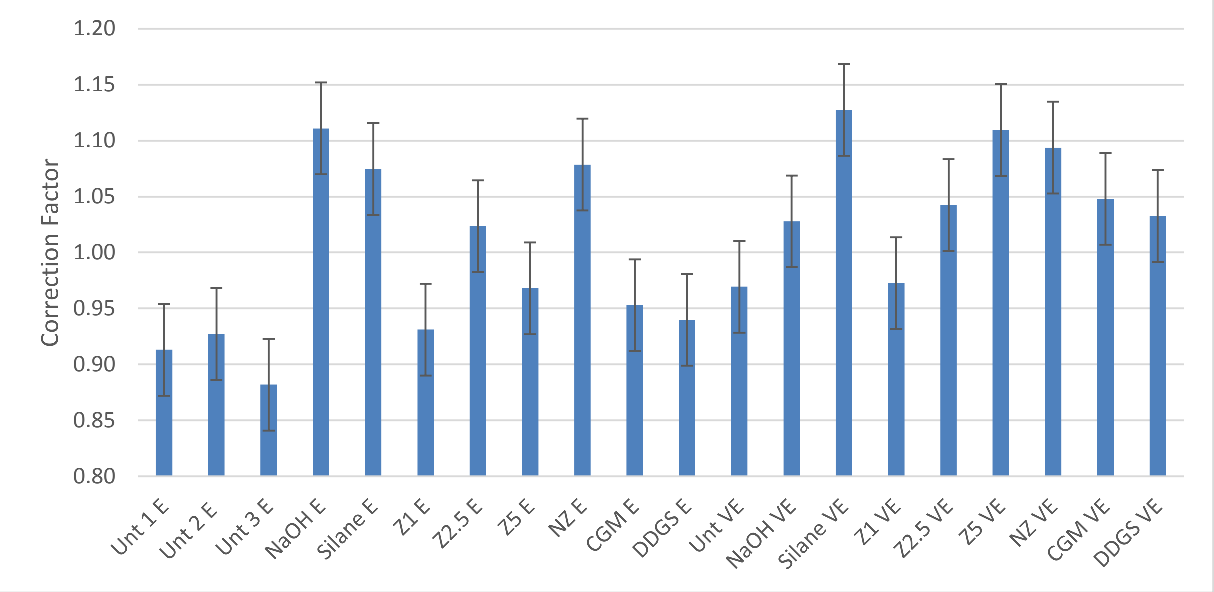

Figure 13: Mechanical properties correction factors.

5.2. Tensile testing results

This section discusses the results of tensile testing in both epoxy and vinyl ester matrices. Tensile strength was investigated to determine if the various zein treatments caused a statistical increase in properties. Both tensile strength and modulus of the materials were investigated.

5.2.1. Epoxy matrix - tensile

The tensile strength test results of the epoxy matrix materials is shown in Figure 14. The error bars in Figure 14 represent 95% confidence interval of the mean. The purple dashed lines represent the 95% confidence interval combining the results of all untreated samples. The order of tensile strength of from high to low is: NZ E, Silane E, Z2.5E, Unt 2 E, NaOH E, Unt 3 E, Unt 1 E, Z5 E, Z1 E, CGM E, and DDGS E. From observing the results, it is difficult to determine any clear trends with varying treatments. Three treatments: Silane E, Z2.5 E, and NZ E show improvement over the untreated samples.

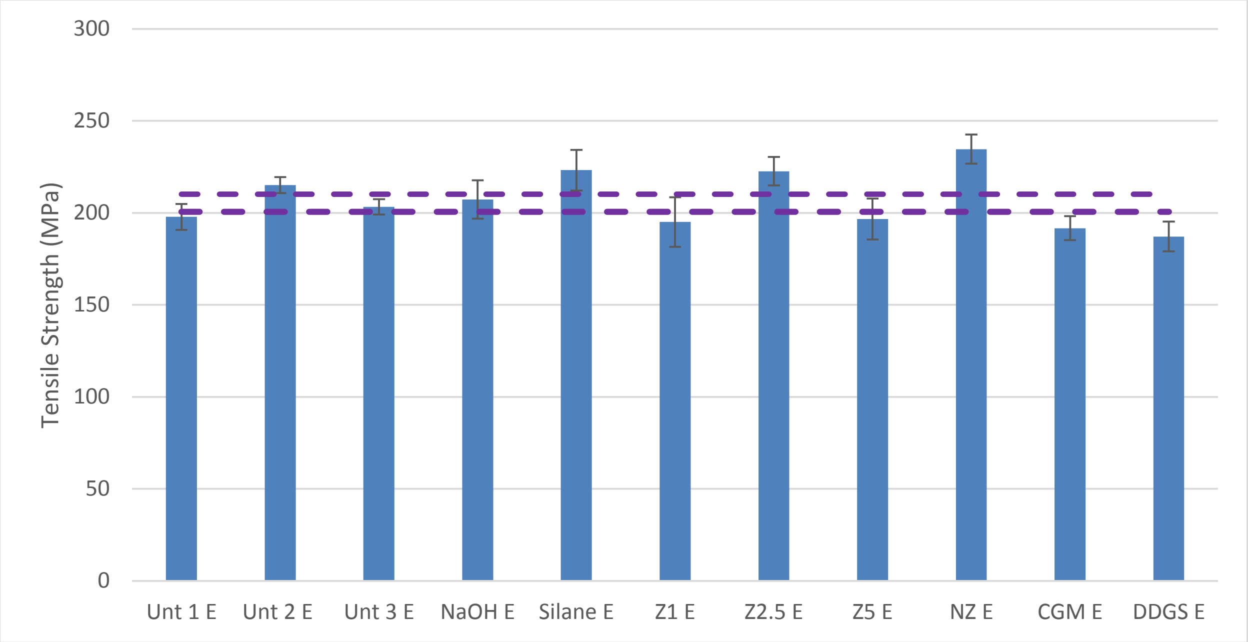

Figure 14: Epoxy matrix normalized tensile strength results.

A correlation in tensile strength vs. pure zein concentration is investigated in

Figure 15. As shown, the least squares fit has a slope of -0.02MPa/%zein. This value shows the treatment concentration of pure zein protein does not correlate with tensile strength in an epoxy matrix. The R^2 value of 0.00 also suggests there is no correlation between these values.

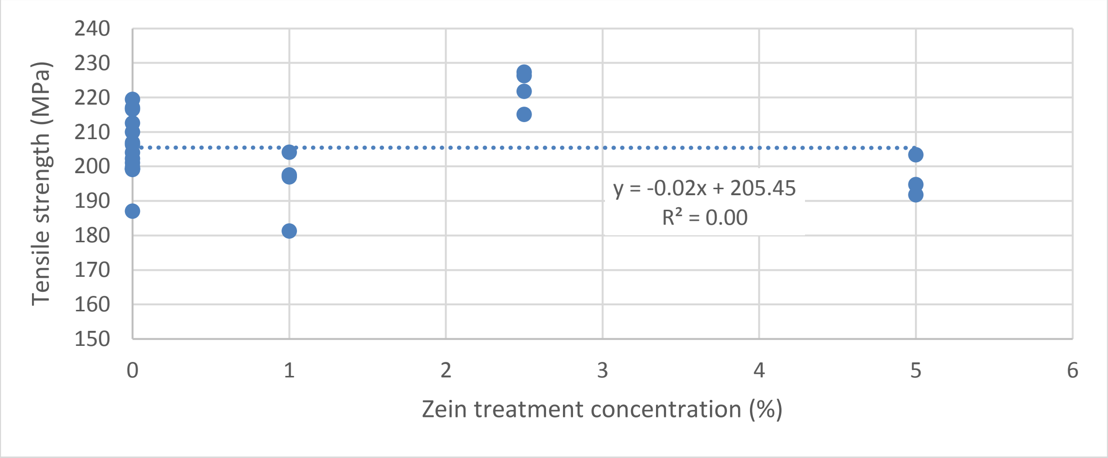

Figure 15: Tensile strength vs. zein treatment concentration.

An ANOVA single factor test was performed to determine if there was a statistical difference between the mean of any two samples. The results of the ANOVA test are shown in Table A1. The resulting P-values of the test is 1.40E-12, which is less than the chosen as $\alpha = 0.05$. This result reveals the null hypothesis is rejected, meaning all of the mean values are not statistically equivalent. The alternate hypothesis must be accepted meaning, at least two samples have statistically different means.

Unfortunately, an ANOVA test does not reveal which samples have different mean values. To answer the question of which samples are statistically different a Pairwise T-test was conducted. The results of the Pairwise T-test are shown in Table 6. Statistically different means ($P > 0.05$) are highlighted in dark blue, while borderline statistically different means (0.1>P>0.05) are highlighted in light blue. One interesting comparison of tensile strengths results is between NaOH E and NZ E. Both these panels incorporated fiber that underwent NaOH treatment. However, the NZ E fiber underwent a zein treatment in addition to the NaOH treatment. The NZ E panel has a tensile strength more than 10% greater than NaOH E which is perhaps due to the presence of the zein protein on the fiber surface. However, since there was no correlation between zein concentration and tensile strength, it difficult to conclude the increase in properties is due solely to the presence of the zein protein and not due to other processing factors as well. It should also be noted the NaOH E sample tensile strength is not significantly different from any of the untreated samples. This is interesting because the NaOH treatment has been shown in literature to be an effective method to increase properties of flax fiber composites.

The overall lack of sensitivity in tensile strength to fiber treatments may be due to the weave of the flax fiber fabric layers. The unidirectional layers are composed of individual twine tows stitched parallel to one another. Each individual tow is made of flax fibers 35 to 50 mm in length twisted together. Perhaps, the mechanical interlocking due to twist in the fiber bundles provides enough frictional force to transfer the tensile load from one fiber to another. Therefore, reducing the sensitivity of fiber matrix interfacial strength on tensile strength results.

Table 6: P-vales for Pairwise T-test of Tensile Strength for Epoxy Matrix

	Unt 1 E	Unt 2 E	Unt 3 E	NaOH E	Silane E	Z1 E	Z2.5 E	Z5 E	NZ E	CGM E	DDGS E
Unt 1 E											
Unt 2 E	0.0010										
Unt 3 E	0.1405	0.0009									
NaOH E	0.0952	0.1313	0.3988								
Silane E	0.0020	0.1374	0.0099	0.0265							
Z1 E	0.6371	0.0029	0.1840	0.1012	0.0042						
Z2.5 E	0.0004	0.0690	0.0018	0.0164	0.9143	0.0043					
Z5 E	0.8059	0.0176	0.1869	0.0951	0.0041	0.7962	0.0044				
NZ E	0.0000	0.0020	0.0002	0.0009	0.0678	0.0009	0.0238	0.0011			
CGM E	0.1312	0.0002	0.0067	0.0158	0.0011	0.5644	0.0001	0.3007	0.0000		
DDGS E	0.0318	0.0004	0.0048	0.0050	0.0008	0.2208	0.0001	0.1044	0.0000	0.2724	

The tensile modulus test results of the epoxy matrix materials is shown in Figure 16. The error bars in Figure 16 represent 95% confidence interval of the mean. The dashed purple lines again show a 95% confidence interval combining all the untreated samples. From investigating Figure 16, it is clear tensile modulus is not sensitive to treatment type. To test this hypothesis, an ANOVA single factor test was performed to investigate if a statistical difference exists. The results of the ANOVA test are shown in Table A2. As shown, the P-values of the test is 0.138. This result reveals there is insufficient evidence to reject the null hypothesis. This result supports the observation that tensile modulus is not sensitive to treatment type. This result is expected since the modulus is measured during the initial elastic loading of the sample. During the initial loading during testing, the fiber matrix interface is not subjected to stress near the point of failure. Therefore, an increase in fiber matrix interface bond strength would not change the modulus of the material. This result also supports the method used for normalizing based on fiber volume fraction. If perfectly normalized, one would expect the modulus values to be constant as was found.

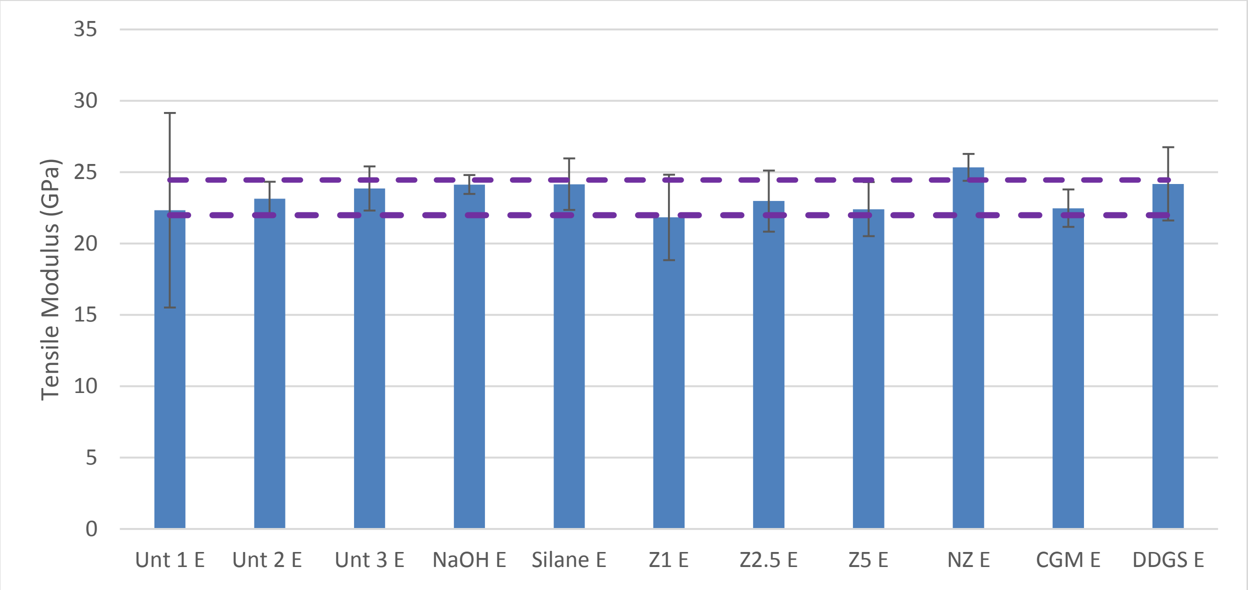

Figure 16: Epoxy matrix normalized tensile modulus results.

Strain to failure was investigated for the epoxy tensile samples. The tensile strain to failure for the epoxy samples is shown in Figure 17. As shown, the untreated samples show a higher strain to failure than the treated samples. This is likely due to fiber pullout experienced by the untreated specimens. The weak interfacial bonding allows fibers to pullout and redistribute the load to other fibers. Therefore, there is a larger strain when catastrophic failure occurs. The NaOH and Silane samples exhibit a low strain to failure. This is likely a result of strong interfacial bonding. The strong bond interface tends for a crack to continue through fiber matrix interfaces.

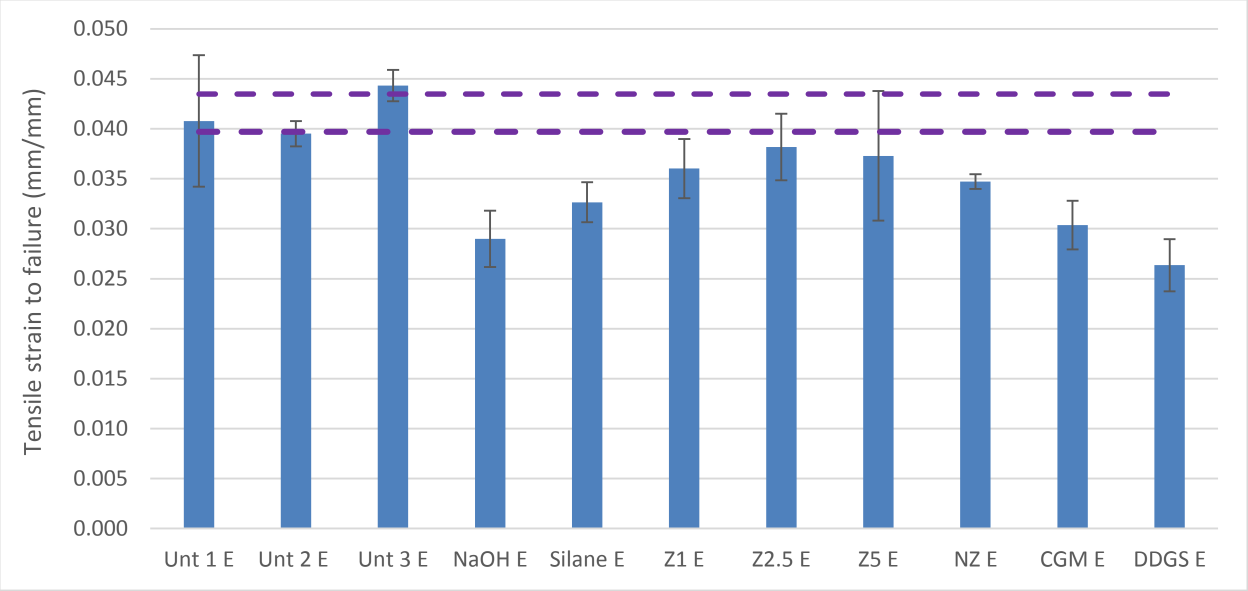

Figure 17: Epoxy matrix tensile strain to failure results.

A trend between strain to failure and normalized tensile stress is investigated in Figure 18. The results show there is no correlation between strain to failure and normalized tensile strength. This is because there is no clear trend between treatment type and tensile strength which is likely a results of the frictional fiber interlocking.

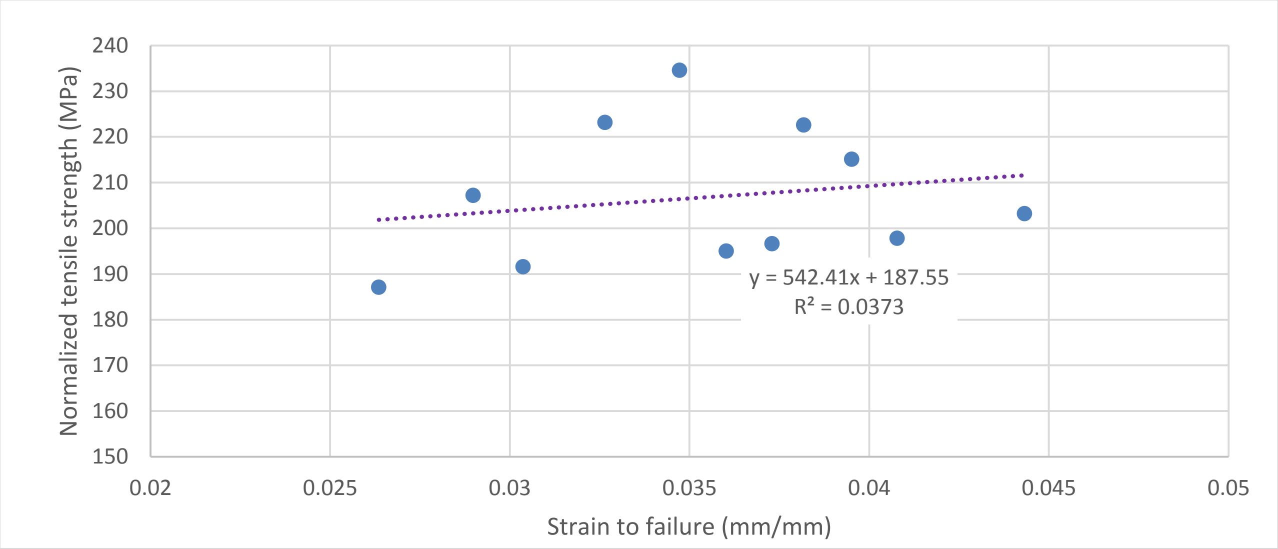

Figure 18: Strain to failure vs. normalized tensile strength for epoxy samples.

5.2.2. Vinyl ester matrix - tensile

The tensile strength test results of the vinyl ester matrix materials is shown in Figure 19. The error bars in Figure 19 represent 95% confidence interval of the mean. The order of tensile strength of from high to low is: Z5 VE, Z2.5 VE, Z1 VE, Unt VE, NZ VE, NaOH VE, Silane VE, CGM E, and DDGS E. The first observation from Figure 19 is a trend for increased tensile strength with an increase in zein treatment concentration. The second observation is the NaOH VE and Silane VE samples show lower tensile strengths than the Unt VE sample. These two commonly used treatments have been shown in literature to increase interfacial bonding. Therefore, it is unexpected to see such a significant drop in properties for these two samples. The last observation is that both CGM VE and DDGS VE perform poorly compared to the Unt VE sample, and much lower than the pure zein treated samples.

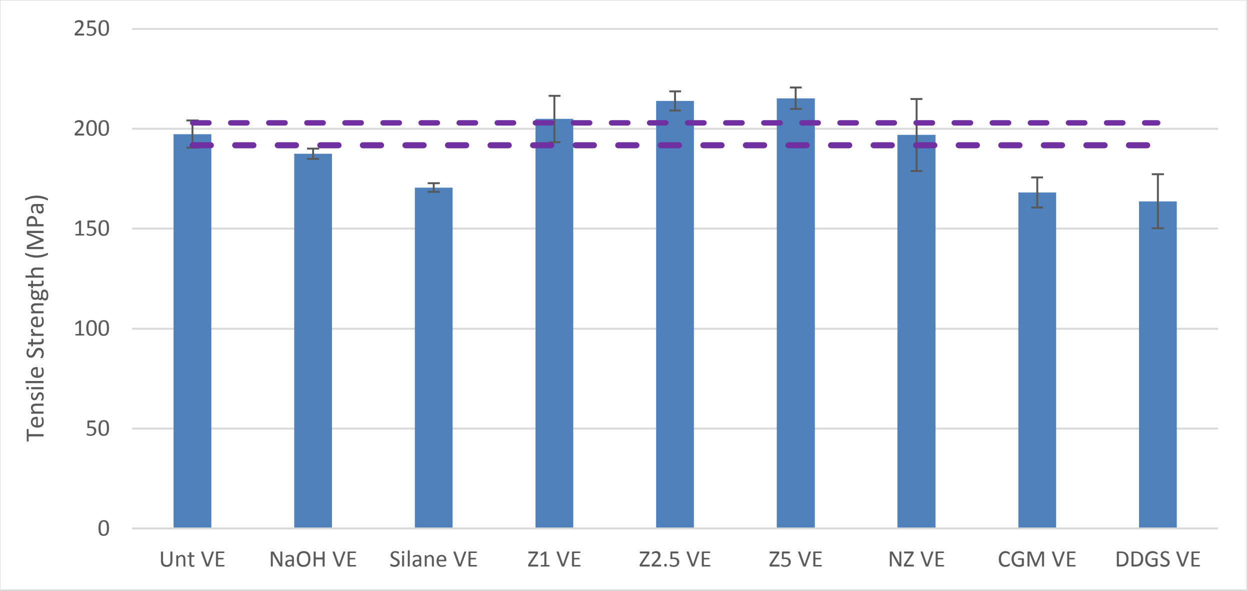

Figure 19: Vinyl ester matrix normalized tensile strength results.

A correlation in tensile strength vs. pure zein concentration is investigated in Figure 20. As shown, the least squares fit has a slope of 3.58 (MPa/%zein) and an R^2 value of 0.57 While this R^2 value does not suggest an excellent correlation, it is great enough to declare there is a positive correlation between tensile strength and zein treatment concentration. By omitting the Z5 E test results, a second model is also shown in Figure 20. The new model results in an R^2 value of 0.61 and a slope of 6.63 (MPa/% zein). It is clear the second model provides a more accurate fit and a more aggressive positive slope. The larger slope shows the zein treatment concentration has more of an effect at the lower concentration range. While the Z5 VE sample was the best preforming, there is evidence of diminishing returns for mechanical property increases beyond the 2.5% zein treatment concentration.

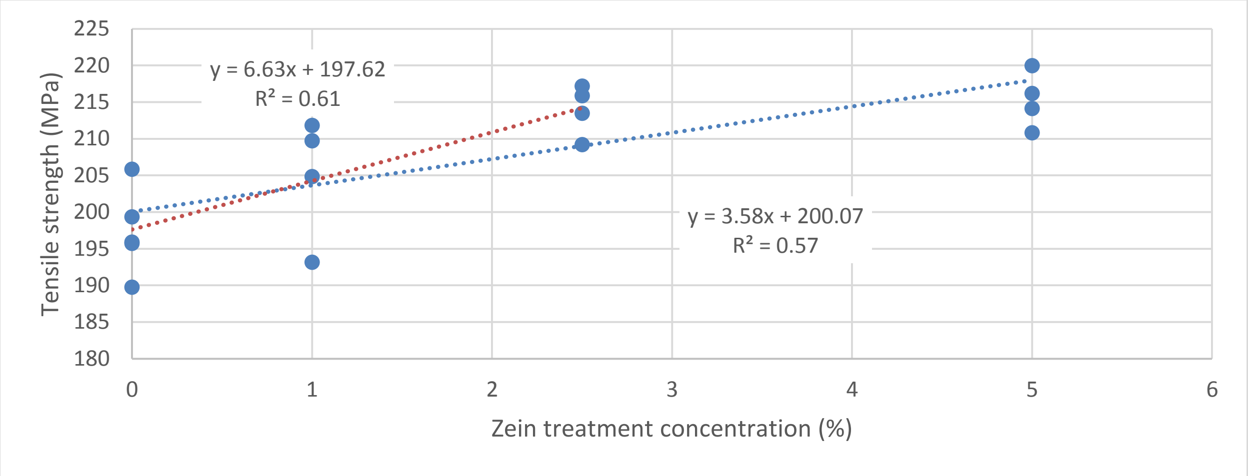

Figure 20: Vinyl ester tensile strength vs. zein treatment concentration.

Again, an ANOVA single factor test was performed to determine if there was a statistical difference between the mean of any two samples. The results of the ANOVA test are shown in Table A3. The test reveals the P-value is 1.43E-11, thus not all samples are statistically equivalent. It is therefore necessary for a Pairwise T-test was conducted. The results of the Pairwise T-test are shown in Table 7. Table 7 shows an interesting result regarding the various pure zein treatments. According to the T-test, Z1 VE, Z2.5 VE, and Z5 VE are statistically equivalent despite showing a strong positive correlation in Figure 20. The samples cannot be deemed statistically different due to the scatter in the data for each panel. A second observation from the T-test is NaOH VE and NZ VE are statistically equivalent. This result shows the zein treatment following the NaOH treatment does not positively influence the tensile strength of the material in a vinyl ester matrix. This may suggest the increase in mechanical properties for the NZ E versus the NaOH E tensile samples was due to processing variation and not the addition of zein protein.

Table 7: P-vales for Pairwise T-test of Tensile Strength for Vinyl Ester Matrix

	Unt VE	NaOH VE	Silane VE	Z1 VE	Z2.5 VE	Z5 VE	NZ VE	CGM VE	DDGS VE
Unt VE									
NaOH VE	0.0168								
Silane VE	0.0002	0.0000							
Z1 VE	0.1860	0.0268	0.0039						
Z2.5 VE	0.0012	0.0002	0.0000	0.1166					
Z5 VE	0.0009	0.0002	0.0000	0.0867	0.6247				
NZ VE	0.9579	0.2459	0.0274	0.3497	0.0855	0.0535			
CGM VE	0.0001	0.0025	0.4592	0.0007	0.0000	0.0000	0.0151		
DDGS VE	0.0018	0.0172	0.2566	0.0007	0.0006	0.0006	0.0064	0.4559	

The tensile modulus test results of the vinyl ester matrix materials is shown in Figure 21. The error bars in Figure 21 represent 95% confidence interval of the mean. From investigating Figure 21, it once again appears the tensile modulus is not sensitive to treatment type.

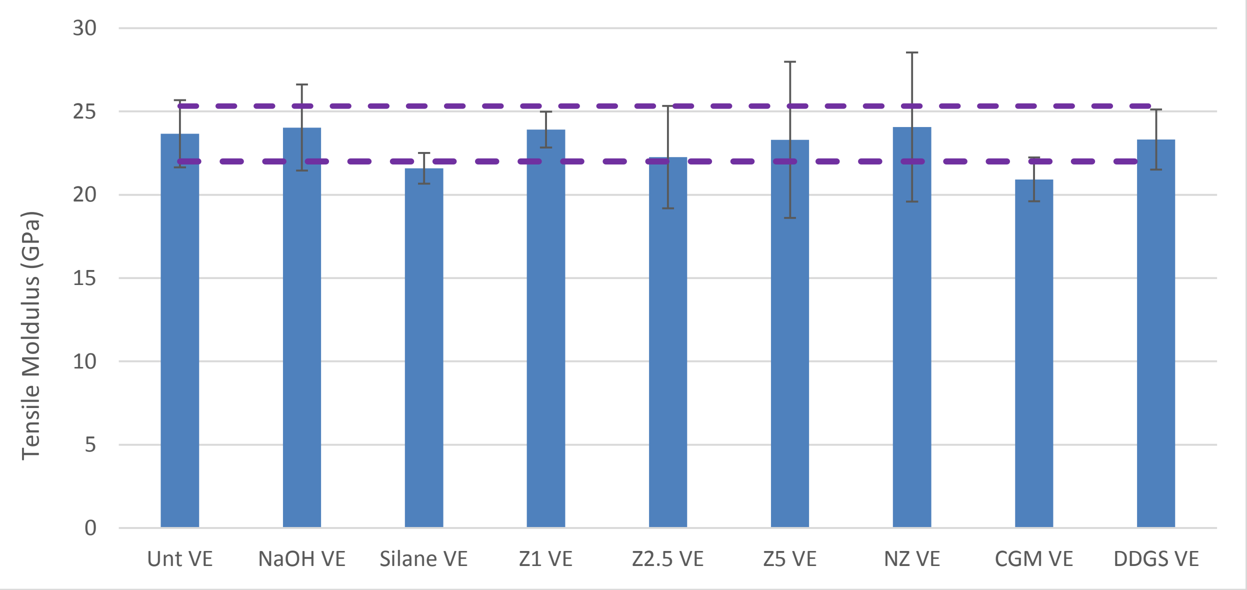

Figure 21: Vinyl ester matrix normalized tensile modulus results.

An ANOVA single factor test was implemented to check for a statistical difference. The results of the ANOVA test are shown in Table A4. The resulting P-values of the test is 0.23 Similar to the epoxy tensile modulus results, this reveals there is not a statistical difference between all samples in the set. This also supports the observation that tensile modulus is not sensitive to treatment type. As previously explained, this result is expected since the modulus is measured during the initial loading of the sample. A statistically equivalent tensile modulus across all samples once again shows the fiber volume normalization method implemented accurately corrects the property values.

Strain to failure was also investigated for the vinyl ester tensile samples as shown in Figure 22. Similar to the epoxy results, the untreated sample shows high strain to failure and the NaOH and Silane treatments show low strain to failure. However, the zein treated samples shown an increase in strain to failure.

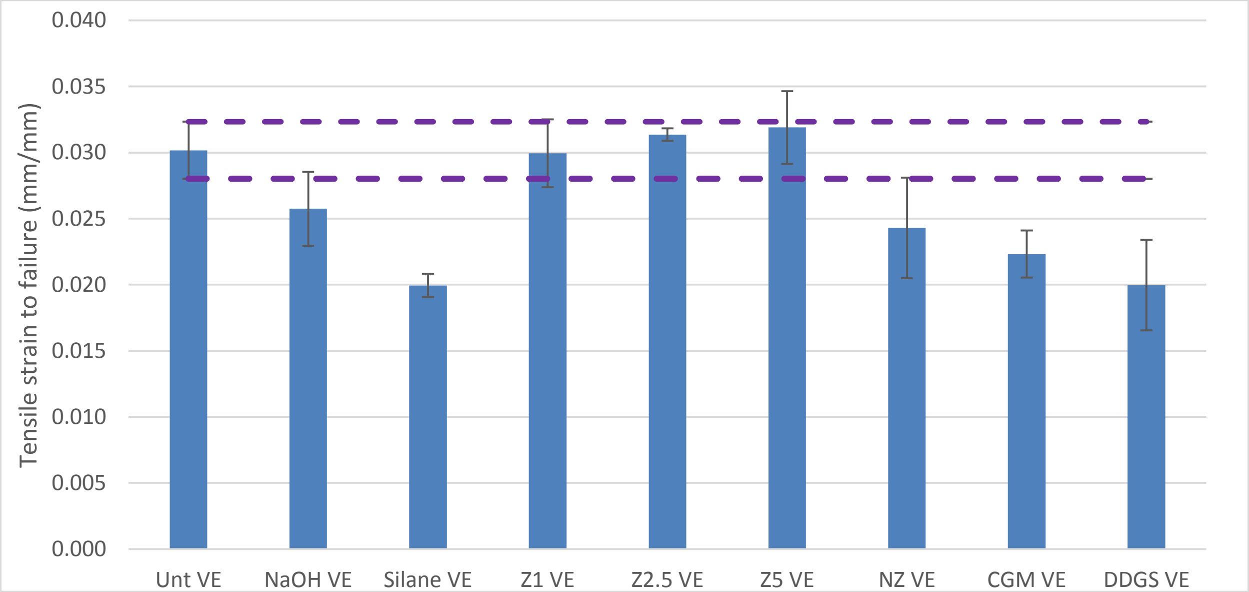

Figure 22: VE matrix tensile strain to failure results.

Figure 23 shows a trend between strain to failure and normalized tensile strength. Unlike the epoxy result, there is a strong correlation between tensile strength and strain to failure. This change is likely a results of the brittle properties of the VE matrix. The inability of the VE matrix to plastically deform prevents the tensile load from redistributing evenly on the fibers. Once one fiber fractures, it causes a stress concentration leading to the surrounding fibers to fracture. The inability to yield causes the crack to quickly propagate in samples with high interfacial bonding.

This explains why the NaOH and Silane treatments exhibit lower tensile strength in the VE matrix, despite increasing interfacial bond strength. The zein treated samples show higher strain to failure which is likely due to the toughness of the zein layer at the fiber matrix interface.

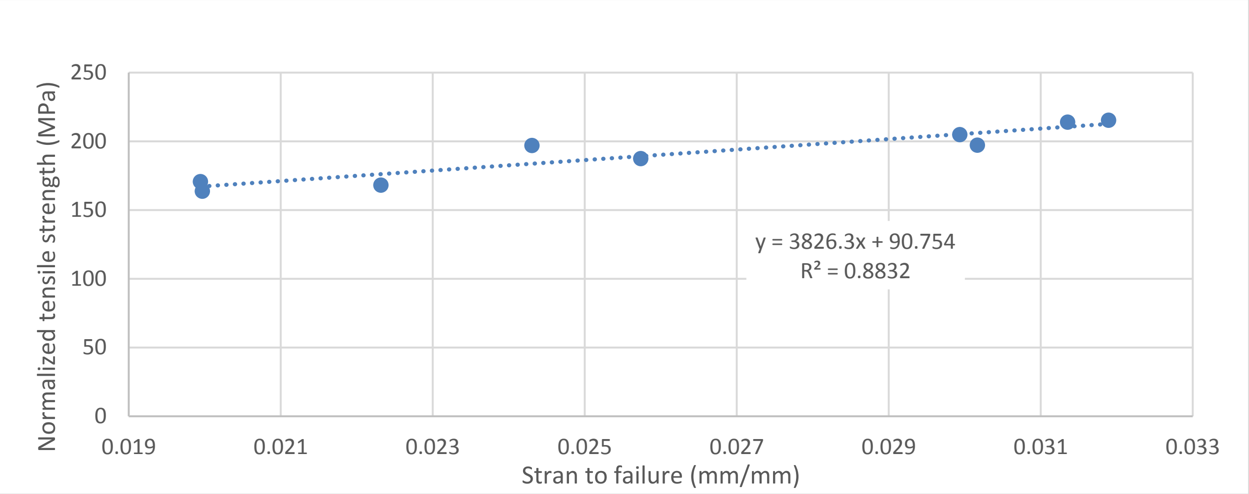

Figure 23: Strain to failure vs. normalized tensile strength.

5.3. Flexural testing results

This section discusses the results of flexural testing in both epoxy and vinyl ester matrices. Flexural strength was investigated to determine if the various zein treatment caused a statistical increase in properties. Both flexural strength and modulus of the materials were investigated using statistical methods. This test is importance when relating material properties to performance in real world applications. The samples undergo tensile, compressive, and shear stresses which is common for real world applications.

5.3.1. Epoxy matrix - flexural

The flexural strength test results of the epoxy matrix materials is shown in Figure 24. The error bars in Figure 24 represent 95% confidence interval of the mean. The order of flexural strength of from high to low is: NaOH E, Silane E, NZ E, DDGS E, Z1 E, Z2.5E, Z5 E, CGM E, Unt 1 E, Unt 2 E, and Unt 3 E. The first observation from investigating Figure 24 is that every fiber treatment shows a mean strength greater than the confidence interval for untreated fabric. The second observation is that all the composites treated with pure zein have statistically equivalent flexural strength about 10% greater than the untreated composite mean. Next, the flexural strength of NaOH E, Silane E, and NZ E achieve a similar strength. Finally, DDGS E preforms the best out the zein based treatments, while the CGM E preformed the poorest of the treated samples.

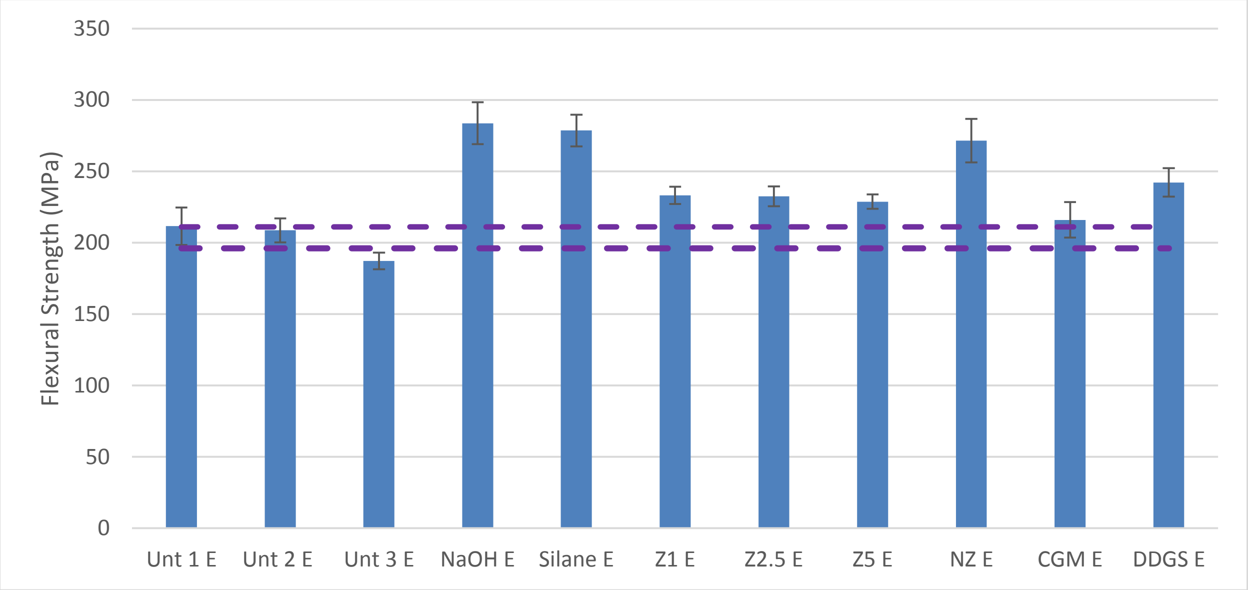

Figure 24: Epoxy matrix normalized flexural strength results.

A correlation in tensile strength vs. pure zein concentration is investigated in Figure 25. As shown, the least squares fit has a slope of 5.38 (MPa/%zein) and an R^2 value of 0.33. This R^2 value implies a weak correlation. A second linear fit is shown Omitting the Z5 E test results from the least squares model in Figure 25. The new model results in an R^2 value of 0.49 and a slope of 12.86 (MPa/% zein). It is clear the second model provides a more accurate fit and a more positive slope. Again, the larger slope shows the zein treatment concentration has more of an effect at the lower concentration range.

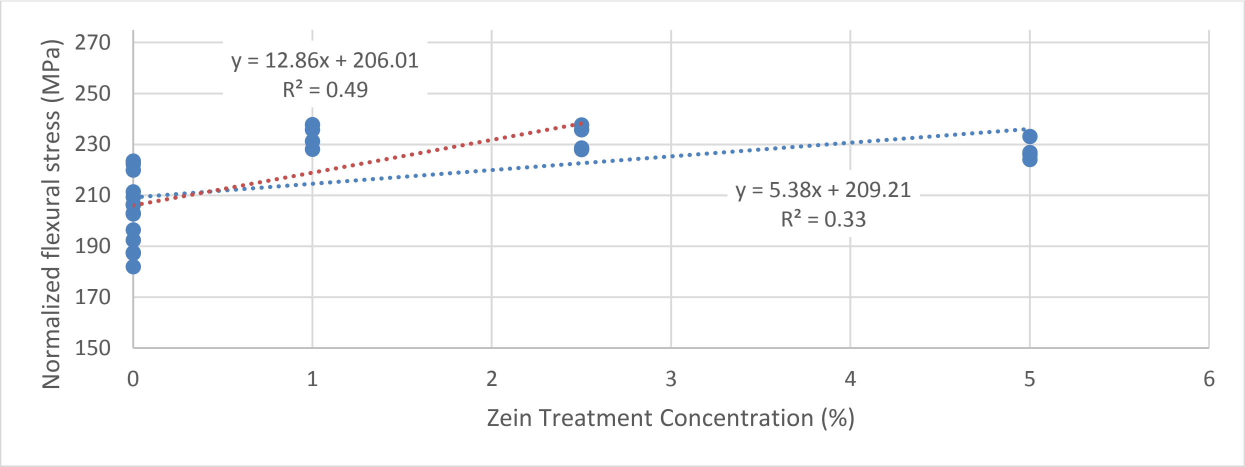

Figure 25: Normalized epoxy flexural strength vs. zein treatment concentration.

An ANOVA single factor test was performed for the epoxy matrix flexural results. The results of the ANOVA test are shown in Table A5. The resulting P-value is 2.43E-20. Thus, it is necessary to conduct a Pairwise T-test. The Pairwise T-test is shown in Table 8. Table 8 supports the observation made that the commercial treatments and NZ E preform statistically equivalent. Also, the table shows chemically pure zein treatments preform statistically equivalent showing a low sensitivity to treatment concentration.

Table 8: P-vales for Pairwise T-test of Flexural Strength for Epoxy Matrix

	Unt 1 E	Unt 2 E	Unt 3 E	NaOH E	Silane E	Z1 E	Z2.5 E	Z5 E	NZ E	CGM E	DDGS E
Unt 1 E											
Unt 2 E	0.6374										
Unt 3 E	0.0069	0.0009									
NaOH E	0.0000	0.0000	0.0000								
Silane E	0.0000	0.0000	0.0000	0.5015							
Z1 E	0.0114	0.0004	0.0000	0.0004	0.0001						
Z2.5 E	0.0102	0.0006	0.0000	0.0004	0.0001	0.8326					
Z5 E	0.0254	0.0011	0.0000	0.0003	0.0000	0.1714	0.2797				
NZ E	0.0001	0.0001	0.0000	0.1774	0.3654	0.0018	0.0017	0.0010			
CGM E	0.5464	0.2442	0.0028	0.0000	0.0000	0.0174	0.0226	0.0578	0.0001		
DDGS E	0.0020	0.0002	0.0000	0.0005	0.0002	0.0908	0.0796	0.0213	0.0043	0.0029	

The flexural modulus test results of the epoxy matrix materials is shown in Figure 26. The error bars in Figure 26 represent 95% confidence interval of the mean. From investigating

Figure 26, it is observed the flexural modulus similar to tensile modulus, is not sensitive to treatment type.

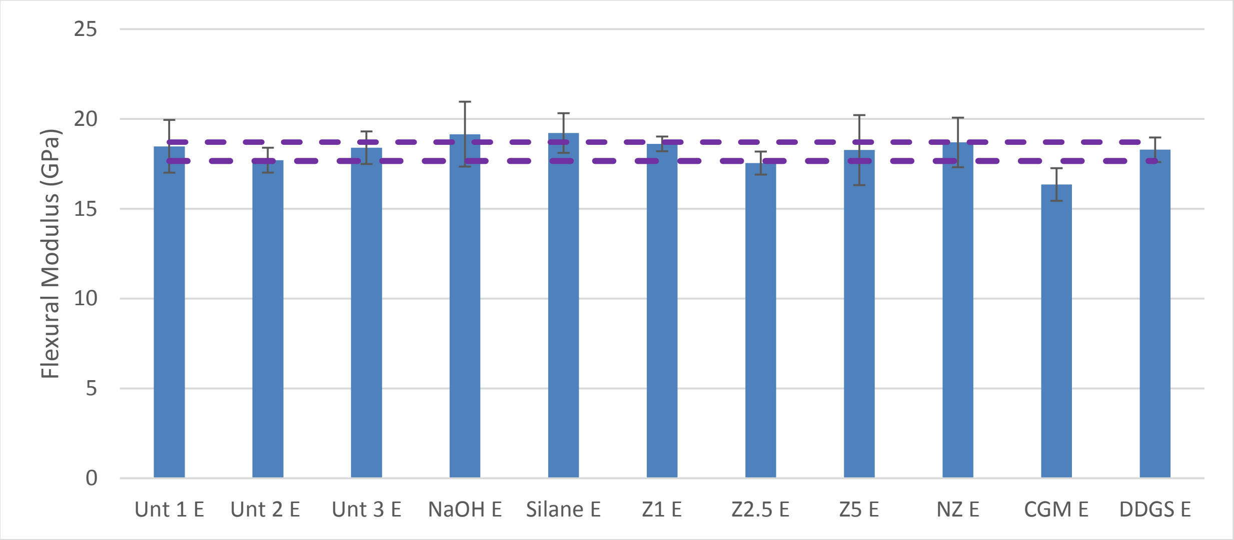

Figure 26: Epoxy matrix normalized flexural modulus results.

An ANOVA single factor test was performed as shown in Table A6, with a resulting P - value of 0.0058. Therefore, contrary to what was observed, a statistical difference exists. A Pairwise T-test was conducted as shown in Table 9. The Pairwise T-test shows the CGM E sample has a statistically different mean than all the other epoxy samples. The only other samples that can be deemed statistically different from some of the samples is the Unt 2 E and Z2.5 E, both of which have slightly lower values than the remaining samples.

The statistically lower modulus values of the CGM E sample may reveal there was a processing defect which resulted in lower modulus values. This would also explain why the CGM E flexural strength was lower than the rest of the treated samples.

Table 9: P-vales for Pairwise T-test of Flexural Modulus for Epoxy Matrix

	Unt 1 E	Unt 2 E	Unt 3 E	NaOH E	Silane E	Z1 E	Z2.5 E	Z5 E	NZ E	CGM E	DDGS E
Unt 1 E											
Unt 2 E	0.2696										
Unt 3 E	0.9106	0.1558									
NaOH E	0.4753	0.1132	0.3683								
Silane E	0.3338	0.0207	0.1740	0.9418							
Z1 E	0.8300	0.0266	0.5908	0.4907	0.2387						
Z2.5 E	0.1928	0.0105	0.0828	0.0830	0.0150	0.0105					
Z5 E	0.8326	0.4038	0.8801	0.4166	0.3183	0.6816	0.4038				
NZ E	0.7875	0.0997	0.6558	0.6199	0.4701	0.8846	0.0997	0.6604			
CGM E	0.0157	0.0290	0.0038	0.0119	0.0009	0.0020	0.0290	0.0617	0.0083		
DDGS E	0.7729	0.0758	0.7984	0.3007	0.1080	0.3302	0.0758	0.9827	0.5218	0.0033	

Strain to failure is compared with normalized flexural strength in Figure 27. As, shown there is positive correlation between strain to failure and normalized flexural strength. This result is different from the epoxy tensile results where no correlation was found. This is because the frictional fiber interlocking cannot support a compressive loading in the twisted fiber bundles. Therefore, the maximum compressive load is limited by interfacial bond strength. It is then expected the strain would correlate with the flexure strength.

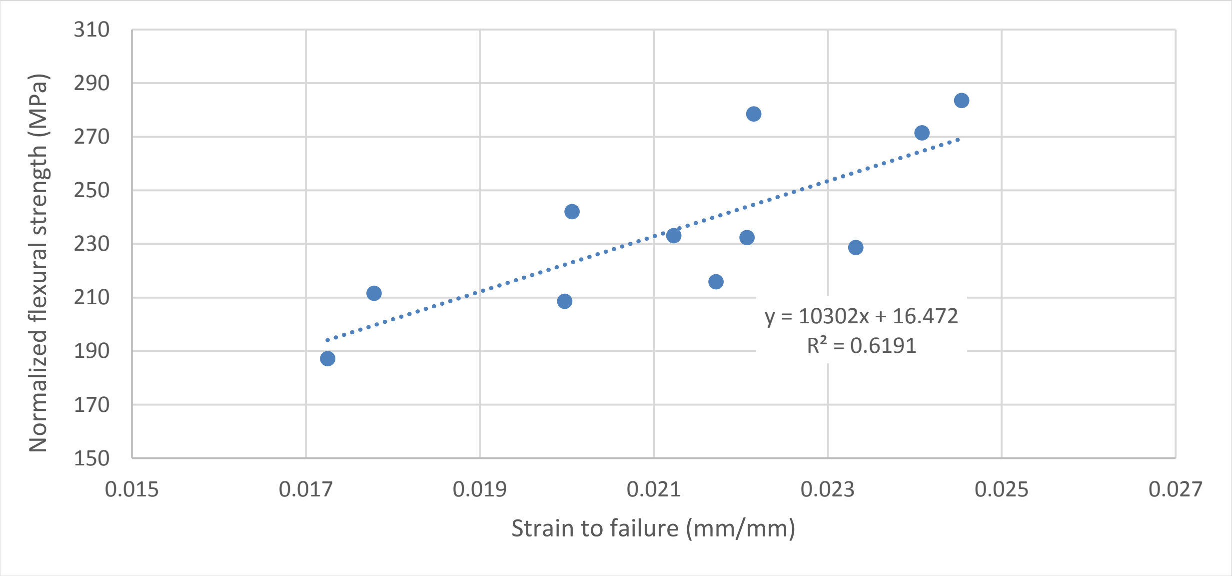

Figure 27: Strain to failure vs. normalized flexural strength for epoxy samples.

5.3.2. Vinyl ester matrix – flexural

The flexural strength test results of the vinyl ester matrix materials is shown in Figure 28. The error bars in represent 95% confidence interval of the mean. The order of flexural strength of from high to low is: NZ VE, NaOH VE, Z5 VE, Z2.5 VE, DDGS VE, Silane VE, CGM VE, Z1 E, and Unt VE. The first observation from investigating Figure 28 is once again all treatments out preformed the untreated control samples. The next observation, similar to the vinyl ester tensile strength results, there is a trend of increasing flexural strength with increasing concentrations of pure zein protein. It is also shown the CGM VE and DDGS VE samples show an increase in flexural strength similar to the Z2.5 VE and Z5 VE samples. The last observation of interest is the NZ VE samples out preforms not only NaOH VE, but all other samples as well. This results may support zein is able to improve interfacial bonding between flax fiber and vinyl ester matrix.

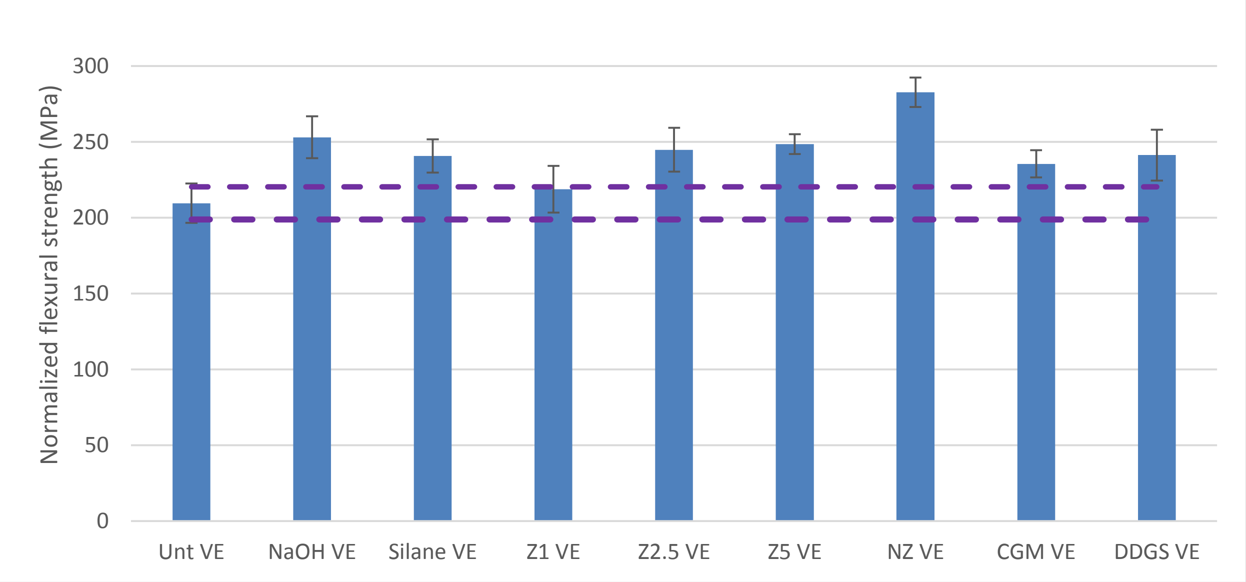

Figure 28: Vinyl ester matrix normalized flexural strength results.

A correlation in tensile strength vs. pure zein concentration is investigated in Figure 29. As shown, the least squares fit has a slope of 8.78 (MPa/%zein) and an R^2 value of 0.65. This R^2 value is large enough to conclude there is a positive correlation between flexural strength and zein treatment concentration. Omitting the Z5 VE test results from the least squares model is also shown in Figure 29. The new model results in an R^2 value of 0.63 and a slope of 14.35 (MPa/% zein). While the R^2 for the second model is no better than the first model, the second model has a much larger slope. The larger slope suggests the zein treatment has a larger effect at concentrations up to 2.5% zein protein concentration and additional zein does not increase properties much. This result again reinforces previous findings that there are greater property gains for the initial increase in zein treatment concentration.

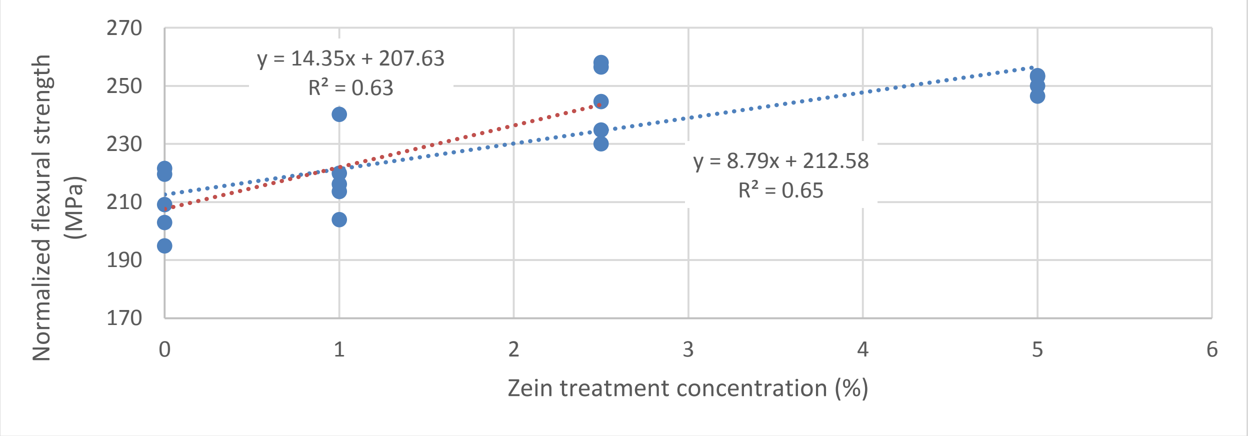

Figure 29: Normalized vinyl ester flexural strength vs. zein treatment concentration.

The results of the ANOVA test vinyl ester matrix flexural strength is shown in Table A7 with a P-values of 1.75E-10. The Pairwise T-test is shown in Table 10. Table 10 show all samples except Z1 VE are statistically different from the untreated sample. The T-test also shows Z2.5 VE and Z5 VE samples are statistically the same supporting the fiber becomes saturated at 2.5% zein treatment concentration. Table 10 also shows CGM VE and DDGS VE are statistically similar to each other and also Z2.5 VE. This result shows CGM and DDGS treatments are viable low cost alternatives to pure zein treatments. NZ VE is also significantly different from NaOH VE supporting zein protein's ability to be a coupling agent in vinyl ester.

Table 10: P-vales for Pairwise T-test of Flexural Strength for Vinyl Ester Matrix

	Unt VE	NaOH VE	Silane VE	Z1 VE	Z2.5 VE	Z5 VE	NZ VE	CGM VE	DDGS VE
Unt VE									
NaOH VE	0.0004								
Silane VE	0.0015	0.1120							
Z1 VE	0.2747	0.0028	0.0202						
Z2.5 VE	0.0016	0.3224	0.5833	0.0131					
Z5 VE	0.0004	0.4876	0.1641	0.0059	0.5590				
NZ VE	0.0000	0.0028	0.0001	0.0000	0.0008	0.0001			
CGM VE	0.0039	0.0301	0.3782	0.0517	0.2068	0.0197	0.0000		
DDGS VE	0.0048	0.2048	0.9403	0.0340	0.6985	0.3460	0.0015	0.4653	

The flexural modulus test results of the epoxy matrix materials is shown in Figure 30. The error bars in Figure 30 represent 95% confidence interval of the mean. From Figure 30, it is observed overall the flexural modulus is not particularly sensitive to treatment type. However, NZ VE has a modulus higher than the untreated confidence interval, and CGM VE has a lower modulus than the untreated confidence interval. An ANOVA single factor test was performed and resulted in a P-value of 0.0058 as shown in Table A8.

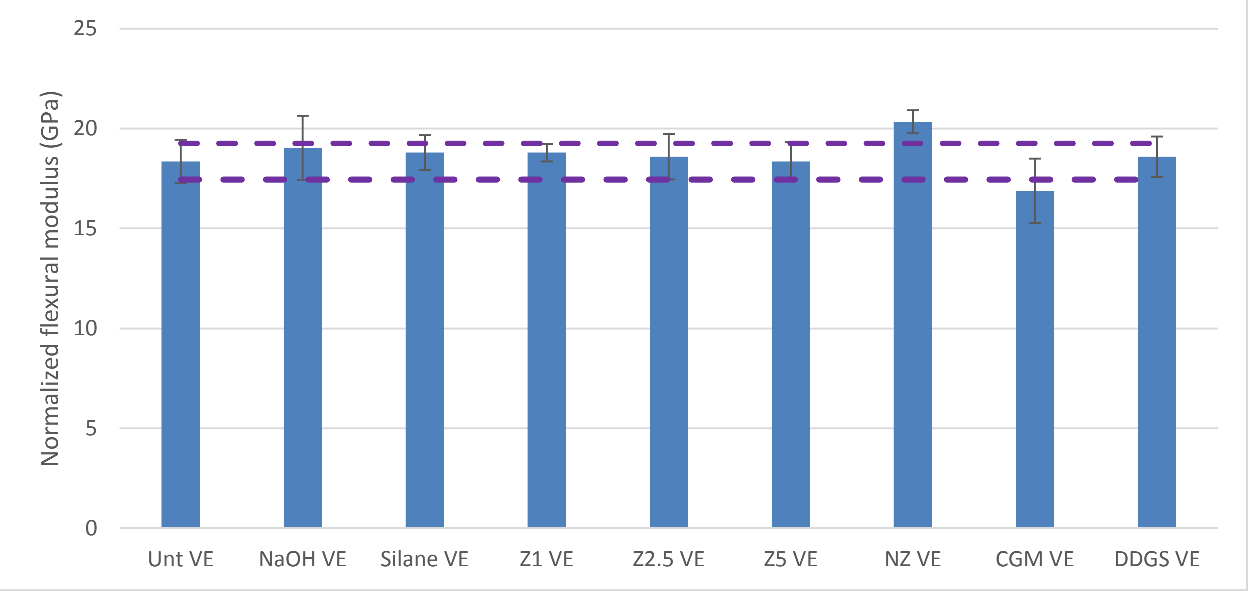

Figure 30: Vinyl ester matrix normalized flexural modulus results.

The Pairwise T-test is shown in Table 11. The Pairwise T-test shows the statistical differences are from the samples NZ VE and CGM VE. The T-test shows NZ VE is statistically the same as NaOH VE. Additionally, the T-test shows CGM VE is statistically different from other samples. This again may be due to processing variation. As expected no conclusions can be made from modulus results since this value is measured at low elastic stress levels when fiber matrix interface is not near its stress limit.

Table 11: P-vales for Pairwise T-test of Flexural Modulus for Vinyl Ester Matrix

	Unt VE	NaOH VE	Silane VE	Z1 VE	Z2.5 VE	Z5 VE	NZ VE	CGM VE	DDGS VE
Unt VE									
NaOH VE	0.3931								
Silane VE	0.4286	0.7503							
Z1 VE	0.3797	0.7172	0.9760						
Z2.5 VE	0.7068	0.5752	0.7123	0.6876					
Z5 VE	0.9959	0.3799	0.4013	0.3318	0.6956				
NZ VE	0.0061	0.1082	0.0068	0.0009	0.0123	0.0027			
CGM VE	0.0920	0.0404	0.0351	0.0318	0.0602	0.0828	0.0034		
DDGS VE	0.6887	0.5632	0.6933	0.6601	0.9979	0.6748	0.0083	0.0533	

Strain to failure is compared with VE flexural strength in Figure 31. Similar to the flexural epoxy results, there is a correlation between strain to failure and flexural strength.

Again, the flexural strength is limited by interfacial bond strength. Since there is greater strain at higher stresses, the samples with the highest failure stress tended to have a high strain at failure.

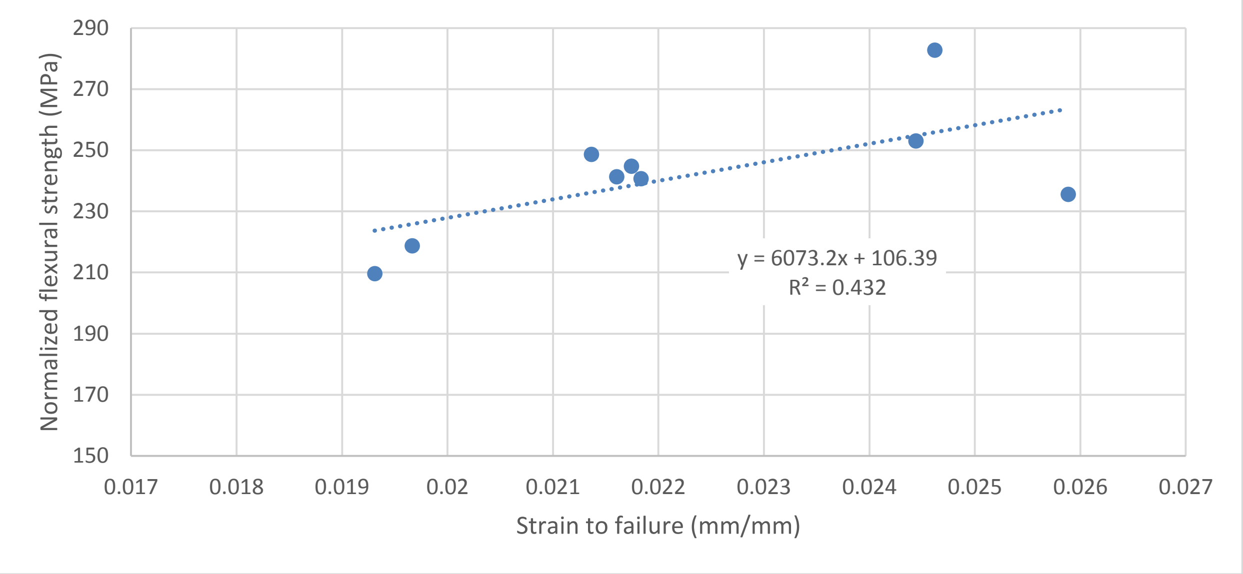

Figure 31: Strain to failure vs. normalized flexural strength for VE samples.

5.4. Short beam shear results

This section discusses the results of short beam shear testing in both epoxy and vinyl ester matrices. Short beam strength was investigated to determine if the various zein treatment caused a statistical increase in shear properties. When investigating interfacial bonding between fiber and matrix, the results of the short beam shear test will be most telling of changes in this critical mechanical property. The short beam shear strength is most closely correlates with interfacial bond strength because of how the shear load is transferred through the material.

5.4.1. Epoxy matrix – short beam shear

The short beam shear strength test results of the epoxy matrix materials is shown in Figure 32. The error bars in Figure 32 represent 95% confidence interval of the mean. The order of short beam shear strength of from high to low is: Silane E, NaOH E, NZ E, DDGS E, Z2.5E, Z5 E, CGM E, Z1 E, Unt 2 E, Unt 1 E, and Unt 3 E. The short beam shear test results have a very similar trends when compared with the flexural strength results, but with more pronounced changes in mean values. Similar to epoxy flexural results, all treatments improve the short beam shear strength when compared to the untreated samples. Again, the synthetic treatments out preform the zein treatments. All three of the synthetic treatments have very similar mean values. Similar to flexural strength results for epoxy samples, the DDGS E sample preforms similar to the Z2.5 samples, while the CGM E sample preformed the poorest for all treated samples. As explained in the epoxy flexural results, the low performance of the CGM E sample is most likely due to variables in the processing since all tested properties preformed low for that sample. However, it is still entirely possible the low properties are also due to the ineffectiveness of the CGM treatment.

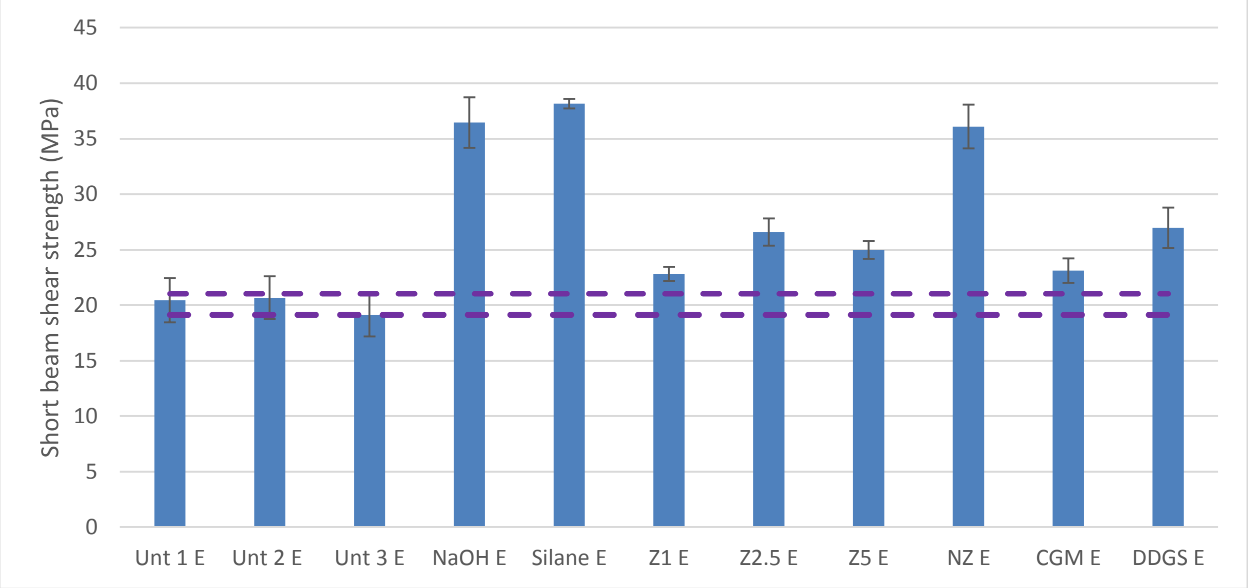

Figure 32: Epoxy matrix normalized short beam shear strength results.

Figure 33 investigates correlation between short beam shear strength and zein treatment concentration. Fitting the data using a least squares method from Unt 1-3 E, Z1 E, Z2.5 E, and Z5 results in a linear model with an R^2 value of 0.54 and a slope of 1.23(MPa/% zein). While this R^2 value does not suggest an excellent correlation, it is great enough to declare there is a positive correlation between short beam shear strength and zein treatment concentration. Omitting the Z5 E test results from the least squares model is also shown in Figure 33. The new model results in an R^2 value of 0.777 and a slope of 2.618 (MPa/% zein). Similarly to all results presented thus far, it is clear the second model is a much better fit to the experimental data.

This better fit from the second model may be due to a saturation of zein protein deposited on the fiber surface. The results suggest this saturation point is between 2.5% and 5% zein protein in a 80% ethanol/20% water mixture. This would explain why the increase in properties does not continue for the 5% zein treatment samples.

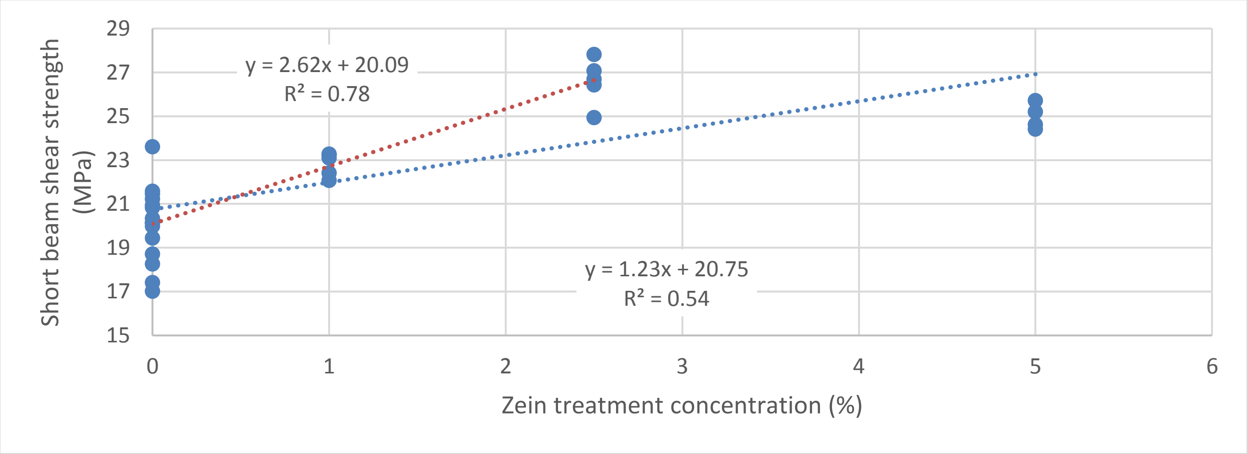

Figure 33: Epoxy short beam shear strength vs. zein treatment concentration.

The results of the ANOVA test for epoxy short beam shear results are shown in Table A9 with a resulting P-values of 3.36E-28. The Pairwise T-test for the data is shown in Table 12. The T-test reveals the three untreated samples are statistically similar. This gives confidence in the T-test's ability to determine if a statistical difference is can be declared. The T-test also supports the observation that the NaOH E, Silane E, and NZ E are statistically equal. It is also shown the Unt 1-3 E, Z1 E, Z2.5 E, and Z5 are all statistically different. This supports the assertion that zein treatment concentration has a direct effect on the interfacial strength between the matrix and fiber surface. The last useful conclusion that can be drawn from the T-test is that the short beam shear strength of the DDGS E sample is statistically equal to the Z2.5E sample. This result suggests that the DDGS solution is an alternative to the high cost pure zein treatment.

Table 12: P-vales for Pairwise T-test of Short Beam Shear Strength for Epoxy Matrix

	Unt 1 E	Unt 2 E	Unt 3 E	NaOH E	Silane E	Z1 E	Z2.5 E	Z5 E	NZ E	CGM E	DDGS E
Unt 1 E											
Unt 2 E	0.8438										
Unt 3 E	0.2503	0.1816									
NaOH E	0.0000	0.0000	0.0000								
Silane E	0.0000	0.0000	0.0000	0.1323							
Z1 E	0.0322	0.0409	0.0053	0.0001	0.0000						
Z2.5 E	0.0003	0.0003	0.0001	0.0001	0.0000	0.0004					
Z5 E	0.0027	0.0030	0.0008	0.0002	0.0000	0.0013	0.0280				
NZ E	0.0000	0.0000	0.0000	0.7558	0.0566	0.0000	0.0000	0.0000			
CGM E	0.0226	0.0289	0.0035	0.0000	0.0000	0.5603	0.0006	0.0085	0.0000		
DDGS E	0.0002	0.0003	0.0001	0.0001	0.0001	0.0025	0.6627	0.0472	0.0000	0.0022	

5.4.2. Vinyl ester matrix – short beam shear

The short beam shear strength test results of the vinyl ester matrix materials is shown in Figure 32. The error bars in Figure 34 represent 95% confidence interval of the mean. The order of short beam shear strength of from high to low is: Silane VE, NZ VE, Z5 VE, NaOH VE, Z2.5 VE, CGM VE, DDGS VE, Z1 VE, and Unt VE. All treatments improve the short beam shear strength when compared to the untreated sample. For the vinyl ester matrix, not all the synthetic treatments out preform the zein treatments. As shown, Z5 VE was the third best treatment narrowly out preforming the NaOH VE sample. All three of the synthetic treatments have similar mean values and represent three of the top four best performers.

Figure 35 investigates correlation between short beam shear strength and zein treatment concentration. Fitting the data using a least squares method from Unt VE, Z1 VE, Z2.5 VE, and Z5 VE results in a linear model with an R^2 value of 0.80 and a slope of 1.66(MPa/% zein). The resulting R^2 value suggest an excellent correlation between short beam shear strength and zein treatment concentration. Omitting the Z5 E test results from the least squares model is also shown in Figure 35. The new model results in an R^2 value of 0.89 and a slope of 2.67 (MPa/%

zein). It is again apparent the second model is a better fit to the experimental data. With a larger slope and R^2 value. Once again, this supports the previous assumption that increasing the zein concentration in the treatment has diminishing returns past 2.5%.

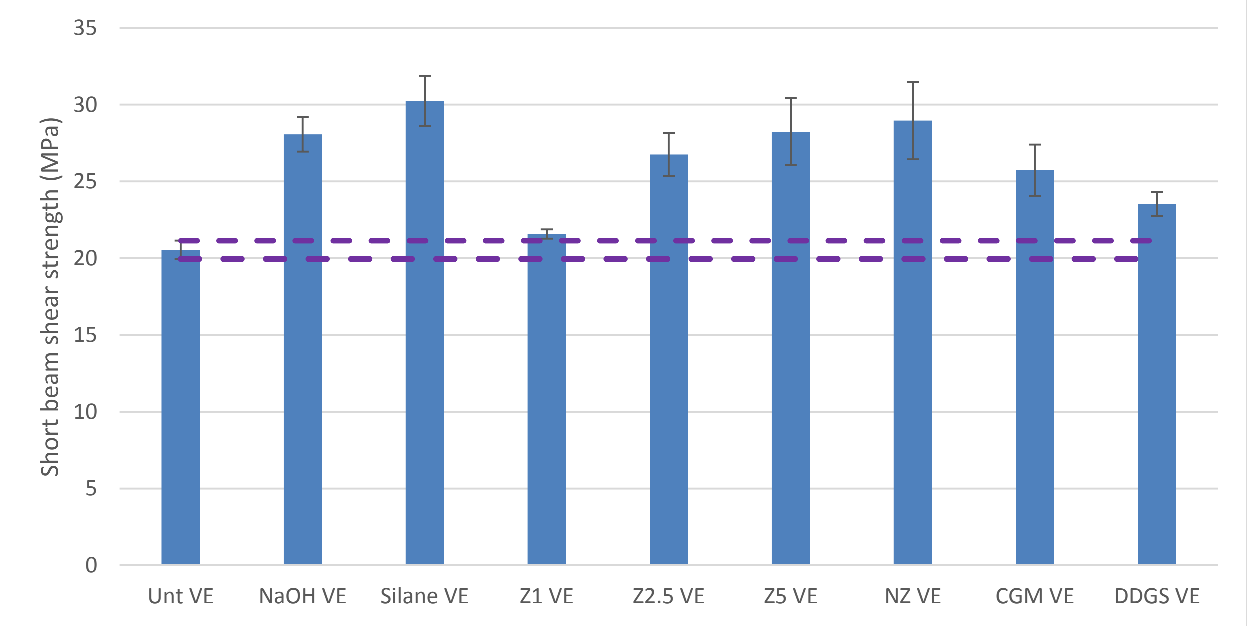

Figure 34: Vinyl ester matrix normalized short beam shear strength results.

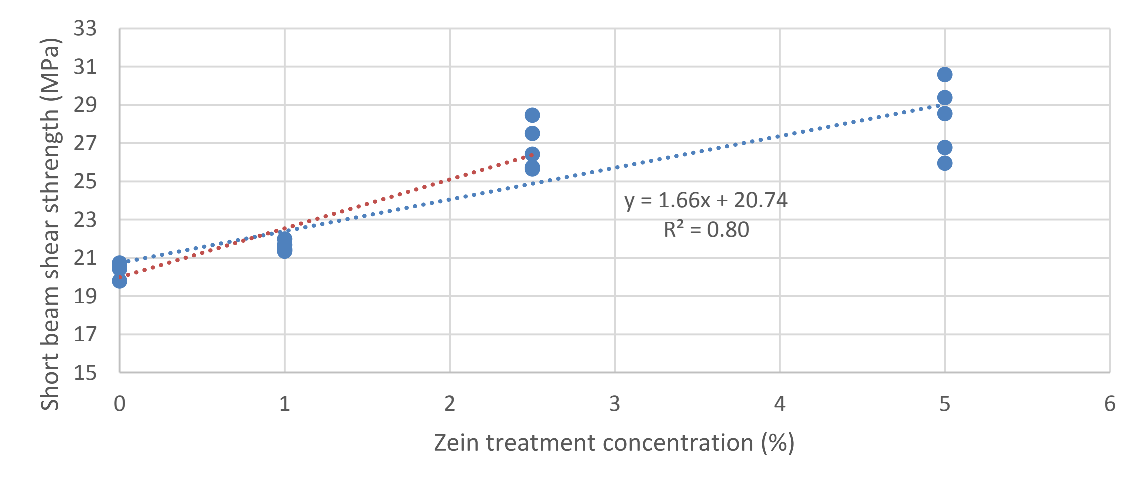

Figure 35: Vinyl ester short beam shear strength vs. zein treatment concentration.

An ANOVA single factor test was performed as shown in Table A10 with a P-value of 5.06E-28. The Pairwise T-test is shown in Table 13. The T-test shows the untreated sample is statistically different from all other samples. Therefore, all treatments have a positive effect. Results, also show the NaOH VE sample is statistically the same as Z2.5 VE, Z5 VE, and NZ VE. This results is encouraging it shows the natural non-toxic zein treatment can match the short beam shear strength increase of the most common synthetic treatment. This result also shows that the additional zein treatment step after an NaOH treatment does not increase properties further. The T-test also shows the Unt VE, Z1 VE, and Z2.5 VE are all statistically different. This supports the assertion that zein treatment concentration has a direct effect on the interfacial strength between the matrix and fiber surface. Z2.5 VE and Z5 VE are not statistically different. This is more evidence to show a saturation point after 2.5% zein treatment concentration. The last useful conclusion that can be drawn from the T-test is that the short beam shear strength of the CGM VE sample is statistically equal to the Z2.5E sample. This result suggests that the CGM solution is an alternative to the high cost pure zein treatment. This result is opposite of the short beam shear results for the epoxy matrix where the DDGS treatment outperformed the CGM in every property.

Table 13: P-vales for Pairwise T-test of Short Beam Shear Strength for Epoxy Matrix

	Unt VE	NaOH VE	Silane VE	Z1 VE	Z2.5 VE	Z5 VE	NZ VE	CGM VE	DDGS VE
Unt VE									
NaOH VE	0.0000								
Silane VE	0.0000	0.0236							
Z1 VE	0.0074	0.0006	0.0002						
Z2.5 VE	0.0001	0.0934	0.0031	0.0007					
Z5 VE	0.0003	0.8606	0.1002	0.0015	0.1814				
NZ VE	0.0011	0.1584	0.3096	0.0017	0.0961	0.5933			
CGM VE	0.0006	0.0223	0.0011	0.0032	0.2625	0.0505	0.0286		
DDGS VE	0.0001	0.0001	0.0001	0.0019	0.0021	0.0034	0.0032	0.0217	

5.5. Summary of mechanical tests results

The summary of the mechanical results is shown below. For the summary, Modulus results have been omitted. As explained in the previous sections, the modulus results either had no statistical differences or few samples fell outside the interval of the other due to processing variation. Therefore, no useful results were derived from the modulus values except to show evidence for the effectiveness of fiber volume fraction normalization. Table 14 shows a summary of order of performance for each of the mechanical test. Examination of Table 14 shows the common synthetic treatments typically are found at the top of the list. However, for four of the six treatment types a pure zein treatment sample is in one of the top three positons, outperforming at least one synthetic treatment. This shows zein protein has the potential to be used in place of common industrial treatment methods. Table 14 also shows the untreated samples regularly appearing at the bottom of the lists. This show in general, all the treatment in this research were to some degree effective in improving the mechanical properties.

Table 14: Summary of Order of Performance for Mechanical Tests

High to low	Epoxy tensile strength	Vinyl ester tensile strength	Epoxy flexural strength	Vinyl ester flexural strength	Epoxy short beam shear strength	Vinyl ester short beam shear strength
1	NZ E	Z5 VE	NaOH E	NZ VE	Silane E	Silane VE
2	Silane E	Z2.5 VE	Silane E	NaOH VE	NaOH E	NZ VE
3	Z2.5 E	Z1 VE	NZ E	Z5 VE	NZ E	Z5 VE
4	Unt 2 E	Unt VE	DDGS E	Z2.5 VE	DDGS E	NaOH VE
5	NaOH E	NZ VE	Z1 E	DDGS VE	Z2.5 E	Z2.5 VE
6	Unt 3 E	NaOH VE	Z2.5 E	Silane VE	Z5 E	CGM VE
7	Unt 1 E	Silane VE	Z5 E	CGM VE	CGM E	DDGS VE
8	Z5 E	CGM VE	CGM E	Z1 VE	Z1 E	Z1 VE
9	Z1 E	DDGS VE	Unt 1 E	Unt VE	Unt 2 E	Unt VE
10	CGM E		Unt 2 E		Unt 1 E	
11	DDGS E		Unt 3 E		Unt 3 E	

Table 15 summarizes the results of most interest in this study. It should be noted for the data measuring percent change, a grey highlight is used to show which samples are statistically equivalent to the sample of comparison. The first row in Table 15 is the slope of the least squares linear model fitting the samples: Unt, Z1, and 2.5. Z5 was omitted because a stronger fit was always found removing the sample. As shown in the table, the addition of zein treatment always resulted in a positive increase for all of the mechanical tests. These results also shows zein treatment has a very similar effect in both matrix materials. The zein treatments show a slightly stronger increase in performance in the vinyl ester matrix composites. However, the difference is so small, and given the inherent uncertainty of the processing and normalization, it is impossible to claim there is a true difference in effect between matrix types.

The second row in Table 15 is the R^2 value for the least squares model. As shown, most of the R^2 values show good correlations between zein treatment concentration and increase in performance except for epoxy tensile strength. The second row shows evidence to support the benefits of zein presented in the first row.

The third row compares the percent change in properties for Z2.5 samples compared to Unt samples. As shown, the Z2.5 Samples preformed at a minimum of 8% greater than the untreated for all tests. Both epoxy and vinyl ester matrix composites show greater than 30% increase in short beam shear strength. Again, short beam shear is the most important property since it most closely correlates with interfacial bond strength. It is also important to note none of the Z2.5 samples are statistically the same as Unt samples. Again, showing zein does in fact have a positive effect on all mechanical properties tested.

The fourth row compares the percent change in properties for CGM samples compared to Unt samples. CGM samples are shown to be outperformed by pure zein treatments in every test. The treatment shows a decrease in properties for both sets of tensile test data when compared to Unt samples. Also, two of the tests are statistically equivalent to the Unt sample. In summary the

CGM may provide slight gains in properties, but cannot be compared to the level of pure zein treatment.

The fifth row, similar to the fourth row, compares DDGS samples against Unt samples. Similar to the CGM treatment, the DDGS treatment shows a decrease in tensile properties for both sets compared to Unt samples. In the other four test sets, the DDGS treatment appears to trend with the Z2.5 results. The DDGS treatment out preforms the Z2.5 treatment in epoxy flexural strength and epoxy short beam shear strength. However, the difference is below the statistical uncertainty. Therefore, it is difficult to conclude the DDGS treatment is better than pure zein especially when it was detrimental to the tensile properties. There is evidence to suggest a solution derived from DDGS can be used to treat flax fibers to increase properties.

The sixth row show the percent increase of Silane compared to Unt samples. This row was used to compare the effectiveness of Z2.5 with a commercial coupling agent. The Silane treatment is shown to improve short beam shear strength to a much greater extent than seen in the Z2.5 treatments. This shows silane remains a superior coupling agent to zein protein in term of mechanical properties.

The final row compares NZ mechanical properties to NaOH properties. As shown, four of the six tests result in statistically equivalent properties. The other two show a 13% or less increase in properties. From this result it is clear the addition of zein protein on a NaOH treated fiber is not worth the additional time intensive processing step as it cannot be proved there is a benefit.

Table 15: Summary of Mechanical Test Results

	Epoxy tensile strength	Vinyl ester tensile strength	Epoxy flexural strength	Vinyl ester flexural strength	Epoxy short beam shear strength	Vinyl ester short beam shear strength
Zein treatment effect (MPa/%)	5.58	6.63	12.82	14.35	2.62	2.67
R^2	0.21	0.61	0.49	0.63	0.78	0.89
% increase Z2.5 vs. Unt	8%	8%	14%	17%	33%	30%
% increase CGM vs Unt	-7%	-15%	6%	12%	15%	25%
% increase DDGS vs Unt	-9%	-17%	19%	15%	34%	15%
% increase Silane vs Unt	9%	-14%	37%	15%	90%	47%
% increase NZ vs. NaOH	13%	5%	-4%	12%	-1%	3%

5.6. FTIR results

FTIR results for untreated and pure zein treated fibers are shown in Figure 36. The FTIR results show peaks corresponding to prominent chemical groups within the flax fiber. The first distinctive band at 3600 to 2995 cm^{-1} is due to hydrogen bonded –OH stretch within the cellulose of the fiber (34). Second, a pair of peaks at 2906 and 2844 cm^{-1} is due to CH_2 stretching. The CH_2 groups are reportedly due to waxy constituents remaining from the flax plant (35). The pure zein treated samples show large sharp peaks at 1651 cm^{-1} and 1540 cm^{-1}. It is reported these peaks are due to amine groups found within proteins. The 1651 cm^{-1} peak is referred to as the amine I band. This band is due to the presence of the C=O bond found in the carboxylic acids groups of the constituent amino acids of the zein protein (36). Likewise, the 1540 cm^{-1} peak is referred to as the amine II band and is a result of the N-H bending and C-N stretching in the amino acids (36). At 1636 cm^{-1} the broad peak shown in the untreated sample is due to absorbed water within the fiber (37). The large spike at 1161 cm^{-1} is due to the C-O-C ether linkage stretching found in cellulose (35). The small sharp peak at 896 is due to the bending of the same C-O-C linkage (35).

Figure 36 shows FTIR results for untreated and pure zein treated fabrics. From investigation of Figure 36, there are three distinct difference between Z2.5 and Unt fibers. The first difference is the broader peak at 3338 cm^{-1} compared to the sharp peak at this location for the untreated fiber. The broader peak may be due to an additional peak at 3292 cm^{-1} which is due to N-H stretching in the protein amino acids (38). The second difference at 2906 and 2844 cm^{-1} is due to CH_2 stretching. At this location it appears the zein treatments reduce the presence of these peaks. This may be due to either a coating of zein covering the wax, or the boiling temperature of the treatment removing some of the wax from the surface. The final and most important difference seen in Figure 36 is the presence of the amine bands. These bands demonstrate the treatment method was effective in depositing zein on the surface of the fiber. These band are not as apparent in Z1 and they are in Z2.5 and Z5. It can also be seen the size of the peaks are approximately equal in both Z2.5 and Z5. These results further support using a 2.5% zein treatment concentration to achieve full saturation of the zein protein on the surface of the fiber.

Figure 37 shows FTIR results for untreated, NaOH, silane, and NZ treated fibers. Results show a broader peak at 3338 cm^{-1} compared to the untreated sample. Results also show, the pair of peaks at 2906 and 2844 cm^{-1} is due to CH_2 stretching dissapear for the treatments involving an NaOH treatment. This is due to the removal of starches and waxes from the surface of the fiber durring the NaOH treatment. The silane treatment shows a sharp peak at 2900 cm^{-1}. This sharp peak is due to the CH_2 groups on the terminal end of the silane moluecule. The broadened peak at 1580 cm^{-1} is due to the C=C double bonds from the silane molucule. These two observations show silane molecules are bonded on the surface of the fiber. Finally, the NZ treatment shows the set of amine peaks at 1651 and 1540 cm^{-1} showing the NZ fiber has zein deposited on the surface. This is significant because the additional zein did not improve properties after NaOH treatment.

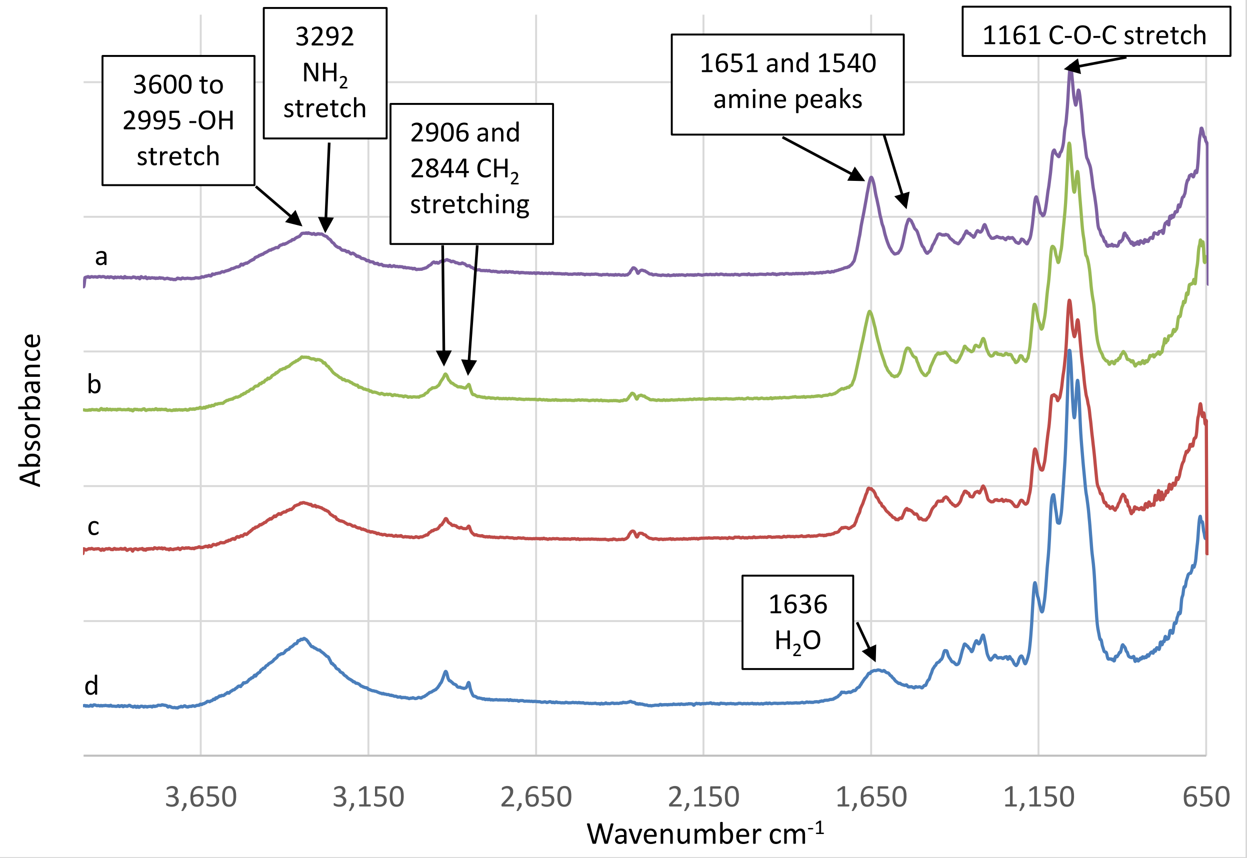

Figure 36: FTIR results of untreated and pure zein treated samples (a) Z5 (b) Z2.5 (c) Z1 (d) Unt.

Figure 38 show FTIR results for untreated, Z2.5, CGM, and DDGS samples. As shown the CGM and DDGS curves appear similar to one another. However, it appears the CGM has a slightly larger amine II band. The CGM and DDGS samples do not show as prominent amine peaks as the pure zein treated samples showed. This may be due to material other than zein protein deposited on the surface of the fiber, thus reducing the prominence of these peaks. The CGM and DDGS both show a peak at 1730 cm^{-1} that is not seen in the Z2.5 sample. The peak at 1730 cm^{-1} is attributed to a carbonyl group (C=O). It is clear the CGM and DDGS treatment deposited material in addition to zein protein, with contains a carbonyl group. This additional material would be the most likely cause for the lower properties seen for CGM and DDGS treatments compared to pure zein treated fibers.

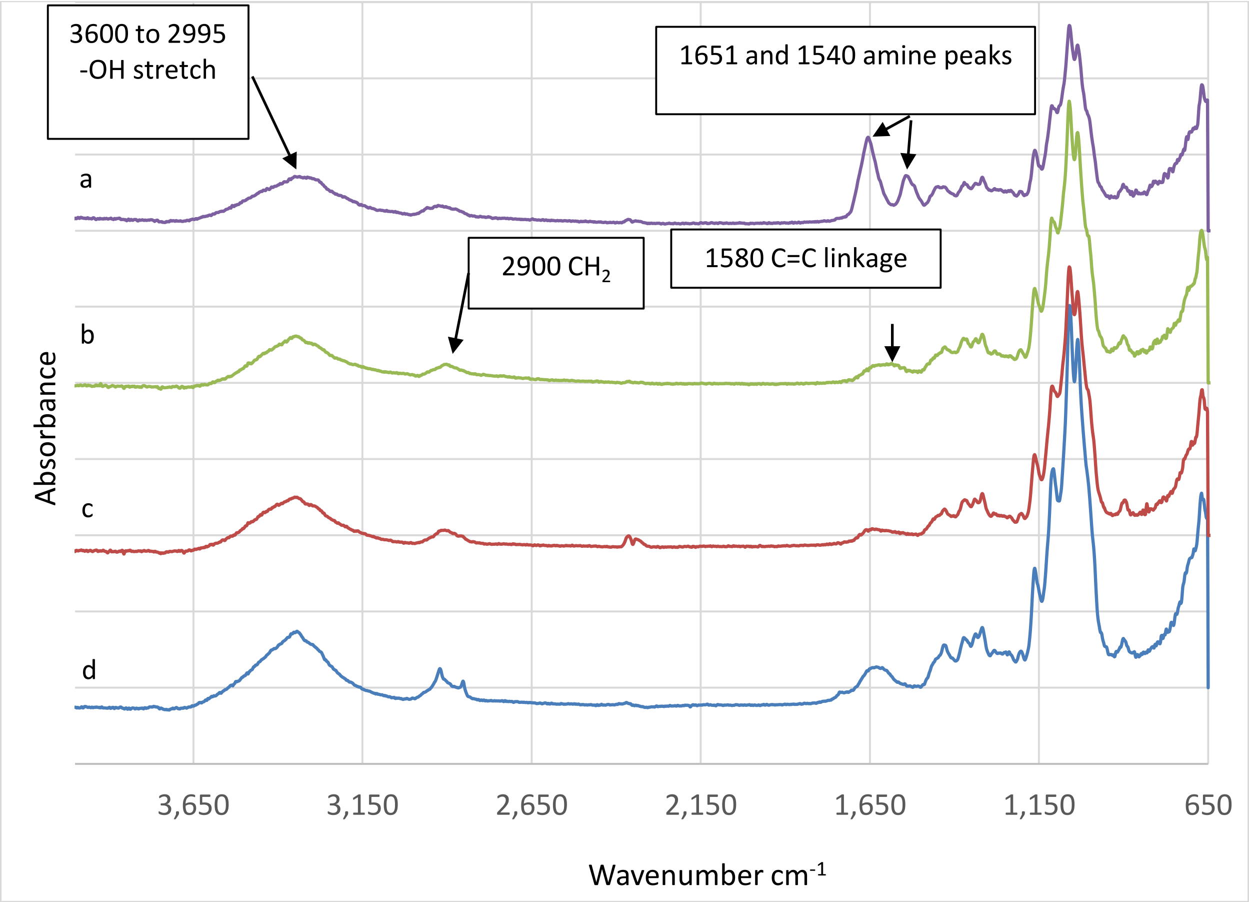

Figure 37: FTIR results of untreated, NaOH, silane, and NZ samples (a) NZ (b) silane (c) NaOH (d) Unt.

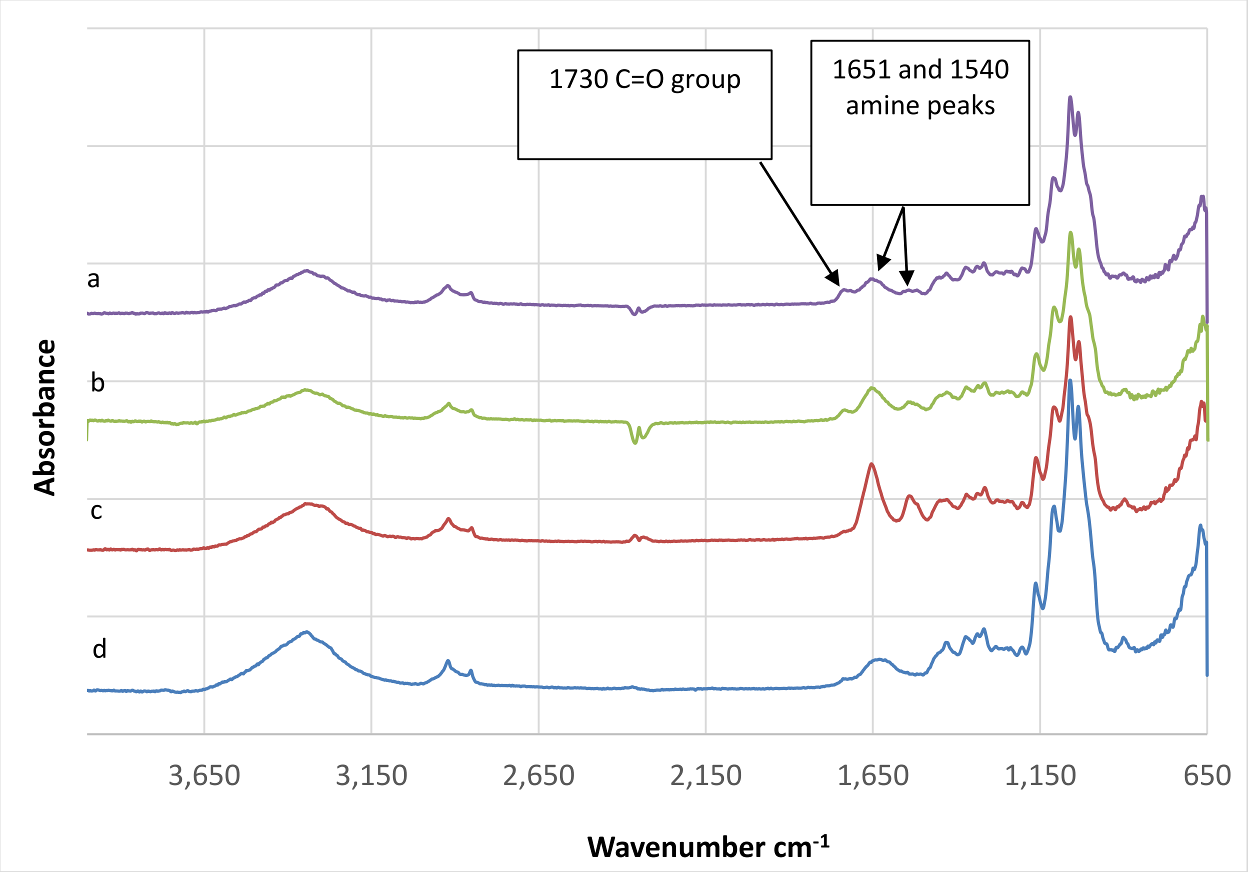

Figure 38: FTIR results of untreated, Z2.5, CGM, and DDGS samples (a) DDGS (b) CGM (c) Z2.5 (d) Unt.

5.7. Chemical analysis results

Chemical analysis results for all nine fiber types are shown in Table 16. There are several significant results presented in Table 16. The first observation is found in the crude protein column. As shown, the percent of crude protein increases as zein concentration increases. Crude protein is an estimation of protein content based on measured nitrogen levels (39). Unlike mechanical property results, the amount of protein deposited on the fiber does not become constant at greater than 2.5% treatment concentration. In fact, the amount of zein deposited on the fiber appears to be parabolic in nature as zein treatment concentration increases. This trend is shown in Figure 39. This result is significant to reinforce zein treatments of concentration greater than 2.5% does not increase mechanical properties despite a near threefold increase in zein protein deposited on the fiber.

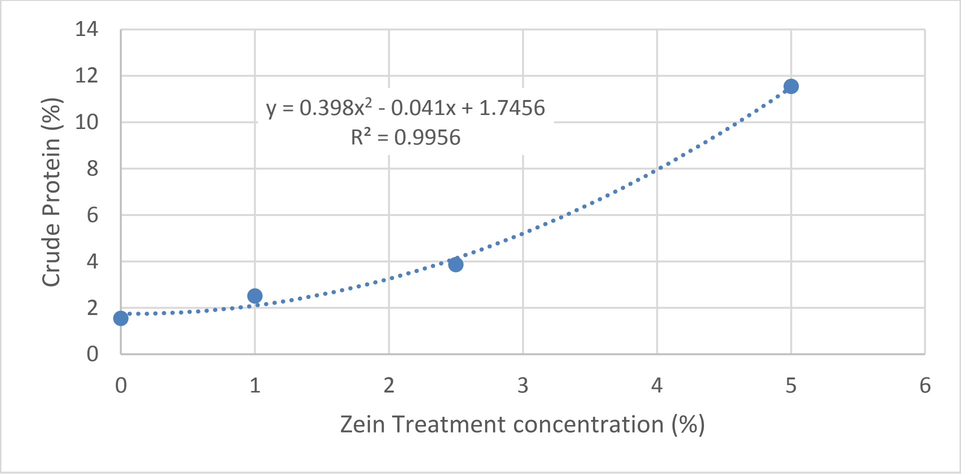

Figure 39: Crude protein vs. zein treatment concentration.

Crude protein results also shows a high level of protein resulting from the CGM treatment. This shows a high level of protein was successfully deposited using the CGM treatment. Likewise, the DDGS treatment shows crude protein deposited on the fiber nearly equal to the amount deposited using 2.5% zein concentration. This shows the concentration of DDGS was correctly chosen to deposit protein at a level equal to 2.5% zein.

The next section of significant results is found in columns labeled: Cellulose, Hemicellulose, and Lignin. The cellulose value is calculated from the reported values of acid detergent fiber percent (ADF) and acid detergent lignin (ADL) percent.

$$cellulose\,\% = ADF - ADL \tag{5.14}$$

The hemicellulose value is calculated from reported values of neutral detergent fiber percent (NFD) and ADF.

$$hemicellulose\,\% = NDF - ADF \tag{5.15}$$

Cellulose results show NaOH treated fiber has higher cellulose content than untreated fiber. The cellulose percent increases because the NaOH treatment removes hemicellulose, lignin, and starch. The cellulose percentage is lower for 5% zein treatment than untreated fiber. This result may be due to the added zein protein deposited on the fiber which drops the percent cellulose with respect to the entire sample. CGM and DDGS samples also shows a drop in percent cellulose compared to untreated sample due to additional material deposited on the fiber surface.

Hemicellulose results show expected decreases for treatments involving NaOH treatments. However, CGM and DDGS treatments report an increase in hemicellulose. These treatments would clearly not increase the hemicellulose content within the fiber. Therefore, the reported increase in hemicellulose must be an error due to the material deposited on the fiber from the treatments.

Similar to hemicellulose results, reported lignin content decreases with treatments involving a NaOH treatment. No other significant observations are found for lignin values.

The reported starch values show a decrease for treatments involving NaOH. It is also shown the 1% zein treated fiber has a lower percent starch than untreated fiber. This may be due to starch being dissolved off the fiber surface during the treatment. Results also show an increase in starch content for the DDGS treatment. It is noted the starch values for the NaOH and Z1 samples are negative. The negative values are a result of measurement error when there was no starch detected. This conclusion is supported since the negative values are very close to zero.

Finally, crude fat values show a decrease for fibers undergoing a NaOH treatment. Crude fat values show an increase for CGM treatment and a much larger increase for DDGS treatment.

This large increase is expected for the DDGS treatment as the grains contain about 10% fat by mass.

Table 16: Chemical Analysis Results

Sample	Dry Matter (100 C), %	Ash %	Crude Protein %	Cellulose %	Hemicellulose %	Lignin %	Starch %	Crude Fat %
Untreated	94.77	1.04	1.55	91.59	5.93	1.04	0.13	0.88
NaOH	95.24	0.68	0.75	96.83	3.43	0.42	-0.03	0.19
Silane	94.27	2.91	6.07	90.94	4.91	0.43	0	0.25
1% Zein	94.98	0.98	2.51	92.49	6.83	1.06	-0.02	0.52
2.5% Zein	94.92	0.96	3.87	84.27	0.09	1.02	0.1	0.44
5% Zein	94.3	0.94	11.54	84.51	6.04	0.91	0.17	0.84
CGM	94.93	1.04	9.55	85.25	7.42	1.05	0.14	1.78
DDGS	94.77	1.6	3.28	90.08	10.94	0.91	0.44	3.39
NZ	93.87	0.63	3.53	95.37	2.96	0.13	0.04	0.26

5.8. SEM results

SEM was used to investigate the fracture surface of tensile specimens for Unt VE, and Z5 VE sample sets. The purpose of using SEM to capture images of tensile specimens was to investigate for visual evidence of increased interfacial bonding. The samples were searched to find an area where fiber matrix interactions could be observed. This location was best found at the edge of the laminate layers. A resin rich area is commonly found between laminate layers because of geometric meshing incompatibilities. In contrast, the areas within fabric layers exhibited much closer packing of fiber because the constituent twine bindles which compose the fabric hold the fibers in close proximity to one another. The edge of a resin rich layer was the best location for an image because the interactions between a fiber and a matrix could be clearly observed. Figure 40 and Figure 41 examine the tensile fracture surface of an untreated sample. In Figure 40 there are several fiber bundles shown which have pulled out from the other half of the sample. This figure also shows holes where fibers have pulled out and remain on the other half of the sample. At the root of the pulled out fiber bundles there is obvious debonding. The debonding is significant enough to cause a crack in the matrix to propagate across the areas containing the other fiber bundles. This is evidence to support the notion untreated flax fibers are unable to form strong bonds with the thermoset polymer matrix. Images were also taken of Unt and Z5 fibers prior to being processed into a composite pane. Also, it can be seen the pulled out fiber surface appears to be clean as there is not matrix material remaining on the fiber surface. This is again evidence for poor interfacial bonding.

Figure 41 shows many of the same features as Figure 40. Again, few fibers have sheared off flush with the matrix fracture but have pulled out. Holes are seen where fibers have pulled out from the matching sample half. Figure 41 also shows a crack propagating between the distances of several fibers. The holes around these fibers are enlarged showing a complete loss of bonding near the fracture surface. Many of the fibers in Figure 41 look clean, in agreement with Figure 40.

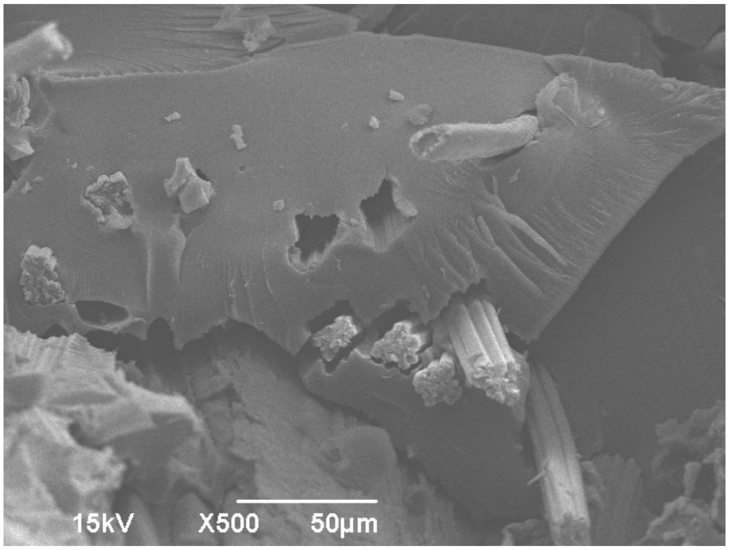

Figure 40: Unt VE tensile fracture surface 1.

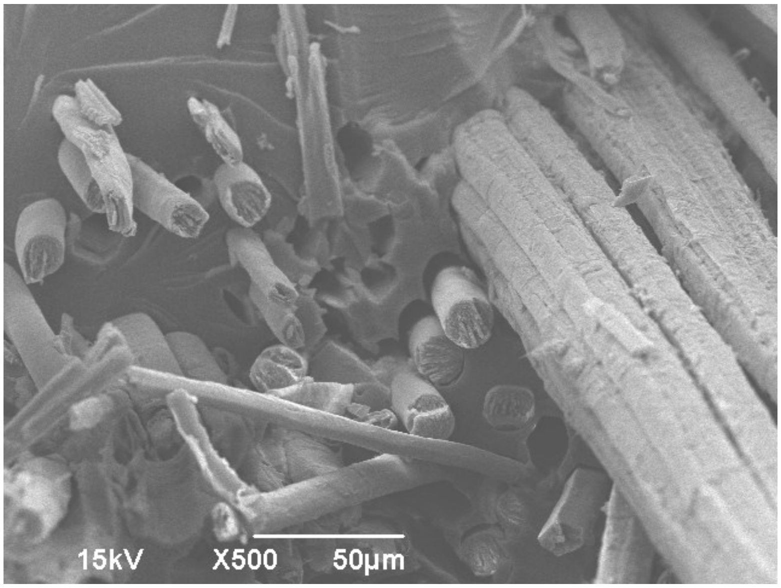

Figure 41: Unt VE tensile fracture surface 2.

In contrast to the two images shown for the Unt VE sample, Figure 42 and Figure 43 show fracture surfaces for Z5 VE samples. From Figure 42 it can first be observed fiber fracture at the location of matrix fracture is more common. While there are some fibers that have pulled out and some holes from fiber that have pulled out, it is not seen to the level shown for untreated samples. For Figure 42, there are also no cracks propagating between fibers. These observations are in agreement with the mechanical test data. The addition of the zein treatment appears to create a stronger interfacial bond at the fiber surface. This increase in bond strength prevents fibers from pulling out of the matrix and results in fiber fracture on the same plane as matrix fracture. It is also apparent the increase in bond strength prevents crack formation caused from fiber matrix debonding. Figure 43 also shows the fracture surface of a Z5 VE tensile sample. This image shows some debonding at the root of the fiber bundle. However, the surface of the fibers appear to have significant amount of matrix material remaining on the fiber evidencing strong interfacial bonding. In addition to the observations above, the images show excellent wetting for the compression molding process used.

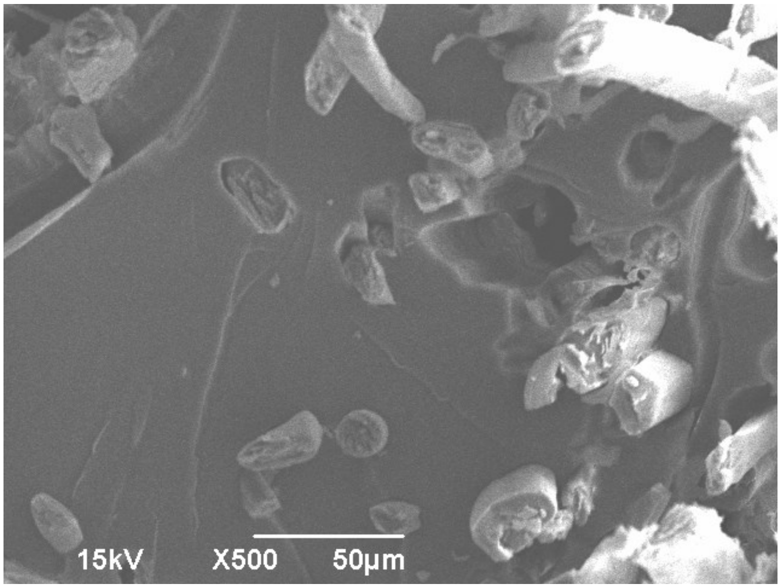

Figure 42: Z5 VE tensile fracture surface 1.

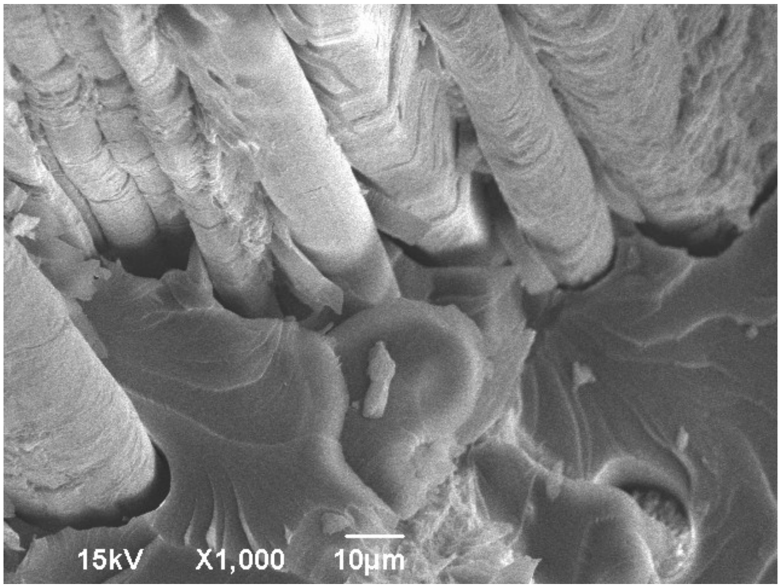

Figure 43: Z5 tensile fracture surface 2.

Figure 44 and Figure 45 compare the surface of a pulled out fiber for Unt VE and Z5 VE, respectively. Figure 44 shows evidence of some matrix material remaining on the fiber surface as well as some area where it appears the outer layer of the fiber has been peeled. However, overall the fiber surface would be characterized as clean. In contrast, Figure 45 shows the surface of a

Z5 VE fiber which has pulled out. The surface of the zein treated fiber shows a lot more material remaining on the surface of the fiber. It is believed the material seen is matrix which has remained attached to the fiber after it has pulled out indicating a stronger interfacial bond than the untreated fiber.

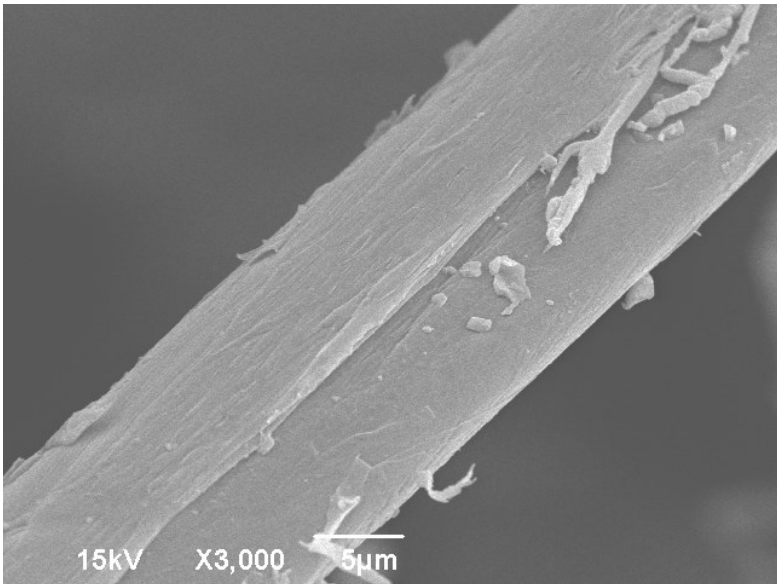

Figure 44: untreated fiber pullout surface.

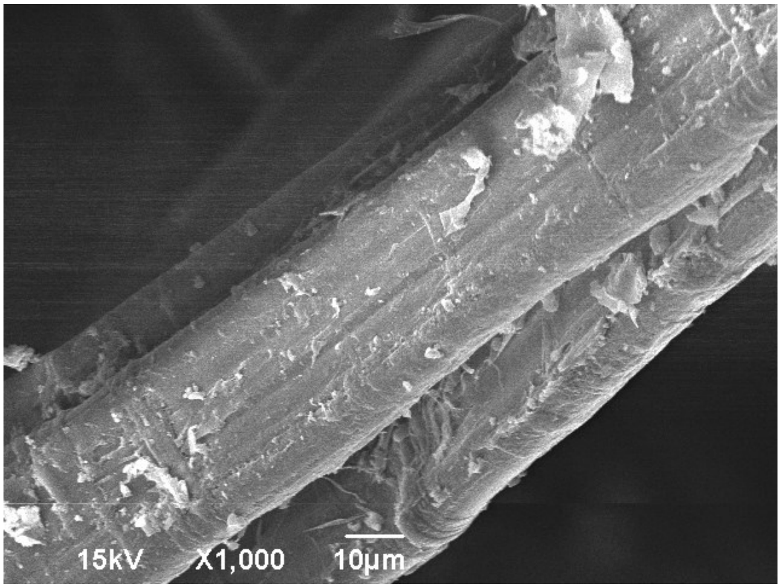

Figure 45: Z5 treated pulled out fiber surface.

Finally, Figure 46 and Figure 47 show untreated and 5% zein treated fiber prior to being processed into a panel. Figure 46 shows a clean surface for the untreated fiber. The surface of the fiber shows the texturing due to the cellulose fibrils present on the surface of the fiber. Figure 47 shows the surface of a 5% zein treated fiber. This image shows the fiber appears clean similar to the untreated sample. However, the texture of the surface is much different from the untreated fiber. The surface of the zein treated fiber does not clearly show the texture due to cellulose fibrils. However, a dimpling texture is present on the surface. This dimpling texture is probably formed by zein coating on the fiber. During the drying step after fiber treatment the ethanol/water solvent must be evaporated to leave the remaining zein coating. These dimples are probably formed during the evaporation of the solvent. This image shows evidence for good coverage of zein on the fiber surface. There appears to be a thin even coating on the surface of the fiber which is desired.

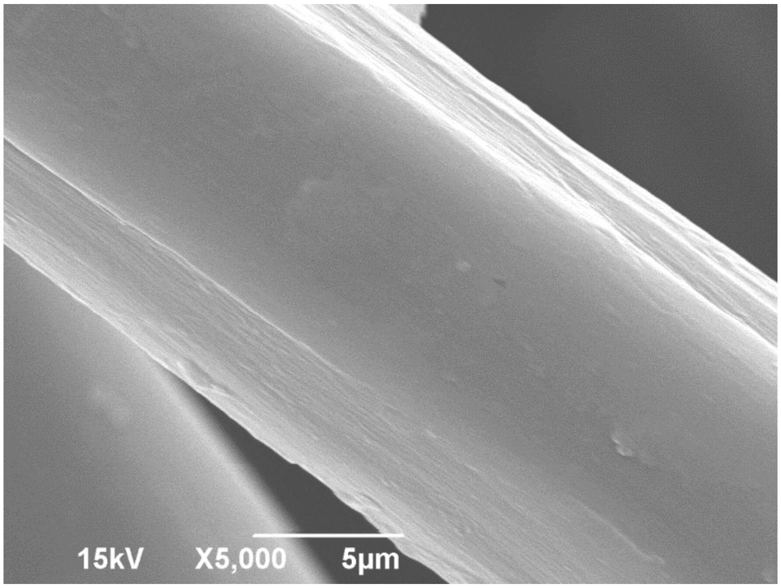

Figure 46: Untreated flax fiber.

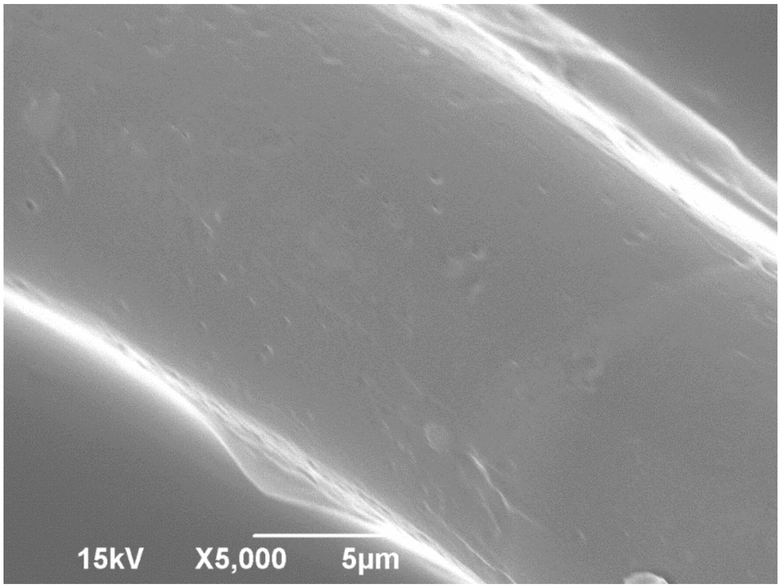

Figure 47: 5% zein treated flax fiber.

CHAPTER 6. CONCLUSIONS AND FUTURE RECOMMENDATIONS

Results from this study show zein protein treatment concentrations reach a peak in mechanical properties near 2.5%. The 5% treatment concentrations do not demonstrate further improvements. For every test that was conducted, this trend was found. There was a near linear relationship between zein treatment concentration and increase in mechanical properties up to 2.5%. Omitting the 5% treated sample always increased the slope of the least squares model and almost always increased the R^2 value. The reason for this drop in effectiveness after 2.5% treatment concentration is most likely due to the fiber surface being saturated at that concentration. Increasing the concentration of the treatment beyond the level necessary to create a thin film of protein only increase the thickness of the coating on the fiber. At that point, the interfacial bond strength is limited to the shear strength of zein protein. The lower concentration is also beneficial from a cost perspective. The chemically pure zein protein is expensive, and using a relatively low concentration of 2.5% can also keep the cost of the treatment low.

This study has shown zein protein does have the ability to improve mechanical properties. The results of this study show zein improved the composite performance in every mechanical property. Results also show zein produces competitive increases in properties in tensile and flexural tests when compared to common industrial treatments. However, the zein treatments were unable to show as large of increase in short beam shear properties. Zein still has potential to be a useful treatment in real world applications since most loading is tensile or flexural. Therefore, these results carry the most importance when assessing uses for real world applications. It also must be stressed the zein protein treatment is entirely nontoxic. The only three ingredients in the treatment are water, ethanol, and zein protein. All of which are completely safe for humans to handle and consume. This treatment strategy is extremely environmentally friendly since the waste products could be disposed without special handling.

The results of this thesis, show that zein is equally effective in both epoxy and vinyl ester matrix materials. Results would suggest perhaps a slightly larger effect in vinyl ester matrix. There tended to be a stronger increase in properties with zein protein in the vinyl ester matrix composites. However, the difference is too small to declare there is an actual difference in the effectiveness between matrix materials.

Test results show both CGM and DDGS were able to increase mechanical properties of the flax fiber composite. The increases however, are not as great as is shown with the pure zein protein. One would have to weigh the increase in properties against the increase in processing cost and time. It must also be noted, the cost of the CGM and DDGS that goes into the treatment solutions are extremely low cost. The most expensive material in the treatment is the ethanol. Greater than material cost, the extra process step and time could offset the performance gains and prevent these from being viable treatments.

Chemical analysis results showed increasing zein treatment concentrations deposited increasing levels of zein on the fiber. This is important to show the level of protein continues to increase while the mechanical properties plateau. Chemical analysis results also showed the NaOH treatment was able to effectively remove hemicellulose, lignin, starch, and fat from the fiber surface. Finally, chemical analysis showed the CGM and DDGS treatments deposited additional fats and starches onto the fiber surface. These unintended products are most likely the reason CGM and DDGS treatments were not able to match the properties of the pure zein treatments.

FTIR results showed zein was successfully deposited on the fiber surface. It also showed equivalent size peaks for Z2.5 and Z5 treatments showing both treatments have fully coated the fiber surface. FTIR was also able to detect peaks due to fats, starches, or sugars, deposited on the fiber during CGM and DDGS treatments.

SEM results showed evidence of increased interfacial bonding for Z5 VE compared to Unt VE. Results showed less fiber pullout for zein treated fibers in addition to less cracking and deboning at the base of pulled out fibers. It was observed there was more material present on the surface of the pulled out fibers indicating higher interfacial bonds. Finally, the surface of a 5% zein treated fiber was compared to an untreated fiber. It appears the fiber is completely coated in zein protein by evidence of pin holes on the surface resulting from evaporation of solvent. These results further reinforce the treatment concentration has been optimized.

This thesis has shown zein protein has the ability to increase mechanical properties in flax fiber thermoset composite materials. There are still more elements of the technology which could be investigated. First, it would be worthwhile to investigate treating flax fiber prior to the textile processing. Zein protein may be suitable to lubricate the fiber during the spinning process in place of the commonly used wheat starch. If this could be implemented, the additional step of zein treatment could be avoided and there would not be an additional cost in treating the fibers with zein. It would also be worthwhile to continue to study optimal concentration of CGM and DDGS treatments. This study shows a zein solution of 2.5% by weight is the optimal concentration for treatment. Given the amount of zein in both CGM and DDGS, the amount added into the solution was selected to achieve 2.5% zein solution based on reported concentrations of zein protein in these byproducts. This concentration is not necessarily the optimal for CGM and DDGS treatments.

There could also be further investigation into treatment additives which could cause more chemical interaction between zein and flax fiber during the treatment stage. For this study the zein was deposited on the surface and bonded by van der Waals forces and perhaps some hydrogen bonding. There could be potential to forming chemical bonds in a manner similar to the mechanism used for the silane coupling agent. This investigation however, would most likely introduce toxic chemicals into the process somewhat defeating the purpose of using zein in the first place.

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APPENDIX

Table A1: ANOVA Single Factor Test of Tensile Strength for Epoxy Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt 1 E	5	989.03	197.81	37.62
Unt 2 E	5	1075.61	215.12	14.14
Unt 3 E	5	1016.05	203.21	13.07
NaOH E	5	1036.14	207.23	81.79
Silane E	4	892.72	223.18	64.24
Z1 E	4	780.04	195.01	94.11
Z2.5 E	4	890.51	222.63	30.94
Z5 E	3	589.92	196.64	36.78
NZ E	4	938.45	234.61	32.68
CGM E	4	766.50	191.63	22.22
DDGS E	4	748.36	187.09	34.12

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	9326.05	10	932.61	22.46	1.40E-12	2.11
Within Groups	1494.99	36	41.53			
Total	10821.04	46				

Table A2: ANOVA Single Factor Test of Tensile Modulus for Epoxy Matrix

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt 1 E	3	67.00	22.33	18.18
Unt 2 E	5	115.67	23.13	1.06
Unt 3 E	5	119.24	23.85	1.80
NaOH E	5	120.66	24.13	0.33
Silane E	4	96.59	24.15	1.69
Z1 E	4	87.30	21.83	4.67
Z2.5 E	4	91.88	22.97	2.37
Z5 E	3	67.21	22.40	1.07
NZ E	4	101.34	25.34	0.46
CGM E	4	89.89	22.47	0.90
DDGS E	4	96.71	24.18	3.46

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	44.16	10	4.42	1.63	0.14	2.12
Within Groups	91.90	34	2.70			
Total	136.06	44				

Table A3: ANOVA Single Factor Test of Tensile Strength for Vinyl Ester Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt VE	5	986.59	197.32	34.68
NaOH VE	4	749.96	187.49	3.28
Silane VE	3	511.76	170.59	1.37
Z1 VE	4	819.55	204.89	69.84
Z2.5 VE	4	855.73	213.93	12.29
Z5 VE	4	861.08	215.27	14.66
NZ VE	4	787.69	196.92	168.69
CGM VE	4	672.87	168.22	29.51
DDGS VE	4	654.84	163.71	95.03

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	11909.99	8	1488.75	30.42	1.43E-11	2.31
Within Groups	1321.38	27	48.94			
Total	13231.37	35				

Table A4: ANOVA Single Factor Test of Tensile Modulus for Vinyl Ester Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt VE	5	118.32	23.66	3.06
NaOH VE	4	96.15	24.04	3.46
Silane VE	3	64.77	21.59	0.25
Z1 VE	4	95.64	23.91	0.61
Z2.5 VE	4	89.03	22.26	4.91
Z5 VE	3	69.89	23.30	6.51
NZ VE	4	96.24	24.06	10.45
CGM VE	4	83.69	20.92	0.91
DDGS VE	4	93.27	23.32	1.69

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	40.29	8	5.04	1.43	0.23	2.32
Within Groups	91.83	26	3.53			
Total	132.12	34				

Table A5: ANOVA Single Factor Test of Flexural Strength for Epoxy Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt 1 E	5	1057.85	211.57	129.00
Unt 2 E	5	1043.01	208.60	52.61
Unt 3 E	4	749.00	187.25	17.91
NaOH E	5	1418.20	283.64	161.86
Silane E	5	1392.92	278.58	92.79
Z1 E	4	932.70	233.17	19.27
Z2.5 E	4	929.78	232.44	24.51
Z5 E	5	1143.49	228.70	19.06
NZ E	5	1357.49	271.50	175.23
CGM E	5	1079.88	215.98	115.57
DDGS E	5	1210.81	242.16	75.57

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	45467.42	10	4546.74	53.69	2.43E-20	2.07
Within Groups	3471.80	41	84.68			
Total	48939.23	51				

Table A6: ANOVA Single Factor Test of Flexural Modulus for Epoxy Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt 1 E	5	92.36	18.47	1.61
Unt 2 E	5	88.54	17.71	0.36
Unt 3 E	4	73.58	18.40	0.43
NaOH E	5	95.74	19.15	2.46
Silane E	5	96.05	19.21	0.91
Z1 E	4	74.42	18.61	0.09
Z2.5 E	5	87.69	17.54	0.31
Z5 E	5	91.32	18.26	2.86
NZ E	5	93.45	18.69	1.44
CGM E	5	81.74	16.35	0.63
DDGS E	5	91.41	18.28	0.35

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	32.52	10	3.25	3.01	5.88E-03	2.06
Within Groups	45.34	42	1.08			
Total	77.85	52				

Table A7: ANOVA Single Factor Test of Flexural Strength for Vinyl Ester Matrix

SUMMARY						
Groups	*Count*	*Sum*	*Average*	*Variance*		
Unt VE	5	1047.85	209.57	126.26		
NaOH VE	5	1264.97	252.99	144.57		
Silane VE	5	1203.64	240.73	91.39		
Z1 VE	5	1093.68	218.74	179.24		
Z2.5 VE	5	1223.94	244.79	158.01		
Z5 VE	5	1243.00	248.60	31.96		
NZ VE	5	1413.48	282.70	72.17		
CGM VE	5	1177.80	235.56	62.06		
DDGS VE	5	1206.67	241.33	212.20		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	17319.13	8	2164.89	18.08	1.75E-10	2.21
Within Groups	*4311.47*	*36*	*119.76*			
Total	21630.60	44				

Table A8: ANOVA Single Factor Test of Flexural Modulus for Vinyl Ester Matrix

SUMMARY						
Groups	*Count*	*Sum*	*Average*	*Variance*		
Unt VE	5	91.77	18.35	0.89		
NaOH VE	5	95.20	19.04	1.95		
Silane VE	5	94.02	18.80	0.56		
Z1 VE	5	93.96	18.79	0.14		
Z2.5 VE	5	92.96	18.59	0.96		
Z5 VE	5	91.78	18.36	0.71		
NZ VE	5	101.69	20.34	0.26		
CGM VE	5	84.43	16.89	1.94		
DDGS VE	5	92.96	18.59	0.76		
ANOVA						
Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	31.70	8	3.96	4.36	9.48E-04	2.21
Within Groups	32.72	36	0.91			
Total	64.42	44				

Table A9: ANOVA Single Factor Test of Short Beam Shear Strength for Epoxy Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt 1 E	5	102.21	20.44	2.97
Unt 2 E	5	103.30	20.66	2.81
Unt 3 E	5	95.54	19.11	2.82
NaOH E	4	145.82	36.45	2.67
Silane E	5	190.84	38.17	0.14
Z1 E	5	114.10	22.82	0.30
Z2.5 E	5	132.97	26.59	1.13
Z5 E	4	99.95	24.99	0.34
NZ E	5	180.46	36.09	2.91
CGM E	5	115.61	23.12	0.90
DDGS E	5	134.90	26.98	2.47

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	2297.15	10	229.71	128.94	3.36E-28	2.06
Within Groups	74.83	42	1.78			
Total	2371.97	52				

Table A10: ANOVA Single Factor Test of Short Beam Shear Strength for Vinyl Ester Matrix

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Unt VE	5	102.74	20.55	0.27
NaOH VE	4	112.28	28.07	0.67
Silane VE	5	151.22	30.24	2.01
Z1 VE	5	107.89	21.58	0.07
Z2.5 VE	5	133.75	26.75	1.48
Z5 VE	5	141.21	28.24	3.58
NZ VE	5	144.82	28.96	4.82
CGM VE	5	128.67	25.73	2.08
DDGS VE	5	117.67	23.53	0.47

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	455.86	8	56.98	32.65	5.064E-14	2.22
Within Groups	61.08	35	1.75			
Total	516.94	43				