ABSTRACT

ROSENBERG, LISA BETH. Texture of Pickles Produced from Commercial Scale Cucumber Fermentation using Calcium Chloride instead of Sodium Chloride. (Under the direction of Dr. Suzanne Johanningsmeier).

Pickles are traditionally produced on a commercial scale from cucumbers fermented in 5-8% sodium chloride (NaCl) brines. Although manufacturers recycle brines before discharge, effluent waste streams are high in NaCl. Pickle companies are considering an environmentally friendly alternative method to ferment cucumbers that replaces NaCl with 1.1% calcium chloride ($CaCl_2$), a known firming agent in pickled cucumbers. Fermented cucumber texture is an important factor in consumers' choice of pickles, so the shelf life depends substantially on maintaining desirable texture characteristics of fresh cucumbers through fermentation, bulk storage, processing, and shelf storage of the finished products. It was hypothesized that fermentation of cucumbers in $CaCl_2$ brine combined with higher residual calcium levels in the finished products will result in a longer shelf life of finished products due to slower texture degradation rates. Objectives were to 1) Determine the texture quality of $CaCl_2$ brined, fermented cucumbers during bulk storage and in finished pickle products 2) Determine sensory texture characteristics of both environmentally friendly and traditionally processed fermented cucumber pickles 3) Determine the effects of fermentation brine and residual calcium on the firmness of cucumber pickles during accelerated shelf life testing (ASLT) to predict the end of shelf life for these products and 4) Determine the correlations between the sensory texture characteristics and instrumental measurements to determine the end point for the ASLT model.

Cucumbers were fermented at a commercial facility in 3,000-gallon, open-top tanks with 6% NaCl or 1.1% $CaCl_2$ brines, stored for up to 9 months in bulk storage, desalted,

processed into hamburger dill chips (HDC) and stored at 25, 35, 45, or 55°C. Fermented

cucumber and HDC mesocarp firmness was measured using a puncture test with a 3 mm

diameter probe at a rate of 2.5 mm/sec. Texture degradation rates were determined from the

fitting of a first order kinetic model of time versus mesocarp firmness. Descriptive sensory

analysis was conducted using trained panelists (n=9) to rate the intensity of several key

texture attributes on a 15-point scale.

Brining treatment and storage time in the commercial tankyard had a significant

effect on the mesocarp firmness of processed pickle products $p < 0.0001$ and $p = 0.0002$,

respectively. The processed products fermented with NaCl brine had a higher firmness, $8.4 \pm$

0.1 N compared to 7.8 ± 0.1 N in HDC brined in 1.1% $CaCl_2$ ($p = 0.0005$). HDC produced

from fermented cucumbers stored for only 2 months in the tankyard had a higher firmness

than those from fermented cucumbers stored for 8 months before being processed into

products ($p = 0.0002$), regardless of brining treatment. Although initial firmness values were

lower, the rate of texture degradation for $CaCl_2$ fermented HDC was significantly less than

that of traditional HDC at all storage temperatures. A maximum loss in firmness of $0.35 \pm$

0.9 N/day vs. 0.47 ± 0.2 N/day was found for $CaCl_2$ enhanced HDC vs. traditional HDC

stored at 55^oC. Descriptive sensory texture attributes of crispness and crunchiness were

correlated to the instrumental firmness measured by puncture testing performed on the

processed product stored under ambient and accelerated temperatures ($R^2 = 0.79$ and 0.80).

The cutoff value for mesocarp firmness used to predict the end of shelf life in the ASLT

model was 7 N based on the instrumental and sensory correlations and the ability of sensory

panelists to detect a small change in texture properties. Environmentally friendly $CaCl_2$

fermentations resulted in finished products with suitable texture attributes when the

fermented cucumbers were processed within 2 months of bulk storage, and the proposed

ASLT model will allow pickle processors to more accurately predict the texture quality of

their products during shelf storage.

Texture of Pickles Produced from Commercial Scale Cucumber Fermentation using Calcium Chloride instead of Sodium Chloride

by
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DEDICATION

This work is dedicated to my family for their unconditional love and support throughout all my life's challenges and adventures. Dad, Erica, Marisa, Grandma and Grandpa, Margot, John, Steph, Ben, Renee and Jack; without your love and support I would not be the person I am today. Words could never fully express the amount of love and respect I have for each of you.

BIOGRAPHY

Lisa Beth Rosenberg was born on November 11th, 1988. She was born and raised in Painted Post, New York with her father Richard Rosenberg and her two younger sisters Erica and Marisa. During her childhood, she was involved in many passions including skiing, sailing, tennis, and cooking. While in high school, Lisa was very active in high school, playing varsity tennis as well as performing in the band year round.

Upon completion of high school, Lisa moved to Burlington, Vermont to attend the University of Vermont (UVM). During her tenure as a student, she was a member of UVM sailing team, several outreach organizations, and Alpha Delta Pi Sorority. Lisa also worked several line cook jobs during college, as well as a yearlong internship in quality assurance at Ben and Jerry's in Vermont. During her internship, she realized her desire to pursue further education in food science and attend graduate school following her undergraduate career. In May 2010, Lisa completed her Bachelor's degree in Nutrition and Food Science.

In August 2010, Lisa moved to the south to pursue a Master's Degree in Food Science at North Carolina State University. After a year of mainly class work and working on several projects in the Rheology Lab, she took a 6-month Co-Op with Campbell Soup Company from July to December 2011. In January 2012, she started working under the advisement of Suzanne Johanningsmeier in the USDA-ARS group. Lisa was also an active of the NCSU Food Science Club and served as the 2012-2013 NCSU Food Science Club President. Upon completion on her Master's Degree, Lisa accepted a Product Development Scientist Position with PepsiCo in Valhalla, NY.

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CHAPTER 1

Evaluation of Texture in Fermented Cucumber Products

1.1 Pickle Production and Statistics

Cucumbers, also known as *Cucumis sativus,* have become the fourth largest vegetable crop harvested in the United States after tomatoes, cabbage, and onions (USDA, ERS 2007). These vegetables originated from the country of India and were brought to America by Christopher Columbus in the 15th century (USDA, ERS 2007). The United States currently produces the fourth largest crop of the world's cucumbers annually, ranking behind China, Turkey, and Iran (Lucier and Lin, 2000; FAOSTAT, 2004). Cucumbers that are solely used for pickled products, account for 100,000 to 125,000 acres of land used to produce cucumbers in over 30 states within the United States (Pickle Packers International Inc., 2013). Americans consume approximately 9lbs of pickles per person every year (Pickle Packers International Inc., 2013). Fermentation of cucumbers has existed for thousands of years, and within the United States the retail market has a value of 3.08 billion dollars in sales annually for all types of pickles (Global Industry analysts Inc., 2008). Traditionally, cucumbers are fermented in 5-12% sodium chloride (NaCl) brine and stored for extended periods of time to 1) obtain a final product with desired qualities and 2) to extend the processing season (Fleming and Moore, 1983). The technology of fermenting cucumbers in high salt content has become a challenge when we as a country are becoming more environmentally conscious. As the industry shifts to be more environmentally conscious, fermentation techniques will begin to change with modern technology. Changes in the fermentation brine may affect the texture, flavor, and appearance of the finished products.

Cucumbers are consumed in a variety of ways in both raw and processed forms. Forty percent of the world's cucumber crop is used for fermented cucumber production. Before modern technology, cucumbers were fermented in tanks or containers ranging in size and material (i.e. wooden tanks or barrels). The containers are typically filled with cucumbers and salt brines added to reach an equilibrated salt content of 5-8% (Breidt, 2006; Breidt et al., 2007; Hutkins, 2006). During the winter months, pickle processors use as high as 12-16% NaCl solutions, to prevent the tanks from freezing and allow the fermented cucumbers to remain in the tankyard (Breidt, 2006; Breidt et al., 2007; Hutkins, 2006). The average salt content of finished pickle products has declined over the last decade from 4-6% to the 2-4% (Lucier and Lin, 2000). The overall decline in salt content results from a lower production of fermented cucumbers and an increase in fresh-pack pickle production, which has a lower salt content (Lucier and Lin, 2000). Currently, 35% of the United States cucumber crop is preserved by brine fermentation and storage of the cucumbers in tanks (Fleming et al., 2002). The remaining 65% of cucumbers used to make pickles are immediately processed and preserved when stored in containers with brine.

1.1.1 Pickle definitions: Fermented, Fresh-Pack and Refrigerated

Pickling along with drying and dehydration is one of the oldest food preservation techniques (USDA, ERS 2007). Pickling is defined as a process in which salt and/or vinegar is added in solution as a way to preserve a food (Fleming and Moore, 1983). Although many other vegetables are pickled, cucumbers are the number one pickled vegetable in the United

States (USDA, ERS 2007). Cucumbers are commercially pickled using three major processes: fermentation, acidification with vinegar followed up with pasteurization, and acidifying the brine for cucumbers under refrigeration (Fleming and Moore, 1983).

Today, brining and the fermentation of cucumbers are done for two main reasons: 1) a preservation method that produces finished products with desired qualities of flavor and texture to the consumer, and 2) a way to save energy by preserving during the harvest season and storing the fermented product for periods of time before making them into product. The process of fermenting cucumbers allows companies to produce products all year round. Pickles can be processed into products by three distinct methods: refrigeration, fresh-packing (pasteurized and shelf-stable), and fermentation. This accounts for 25%, 40%, and 35% of each of the three categories for pickled cucumbers respectively (Fleming et al., 2002). A general flow diagram in figure 1.2 shows the process of how pickles are made. Placing fresh cucumbers into jars with an acidified, salty cover liquid produces refrigerated pickles. The jars are not pasteurized, and the equilibrium with the acids and seasonings takes about 8-10 days. These pickles usually have only a 3-4-month shelf life and must be stored in a temperature of 1-4°C.

For fresh-pack pickles, the cucumbers must be harvested and processed within a week, which only allows processors to use cucumbers from the U.S. during late spring to early fall (Communication with Mount Olive Pickle Company, 2013). Fresh pack pickles are made by packing fresh cucumbers into jars and covering them with cover brine consisting of salt, vinegar, color, and spices. The United States Department of Agriculture (USDA)

standards on fresh pack pickles require a minimum content of 0.5% acetic acid (USDA,

1966). Fresh pack pickles are considered an acidified food which requires them to have a pH

lower than 4.6. The pH range for fresh pack products is between 3.5 and 4.0, with an acetic

acid concentration of 0.6% to 2%, and salt concentrations of 2 to 4% (Breidt et al., 2007).

Fresh-pack cucumber pickles go through a pasteurization process to reduce microbial

contamination and inactivate the polygalacturonase enzymes that may contribute to

enzymatic softening. Variables such as pasteurization time and temperature and pH and

acidity have a significant impact on the firmness of fresh pack cucumber pickles (Monroe et

al., 1969).

Fermented cucumbers are produced by placing fresh cucumbers in open-air 8,000 to

10,000 gallon storage tanks made out of fiberglass, wooden or plastic materials and covering

them with a brine containing NaCl and water. The naturally occurring, acid tolerant lactic

acid bacteria present on the surface of the raw cucumber fruits consume the glucose and

fructose that are present in fresh cucumbers and produce lactic acid. The fermentation

reduces the pH below 3.6. Nitrogen or air gas is purged throughout the process of

fermentation to remove carbon dioxide gas to prevent the production of bloated cucumbers.

$CaCl_2$ is added at 0.1 %-0.4 % concentration to maintain the firmness of fermented

cucumbers (Etchells et al., 1977; Fleming et al., 1987; Tang and McFeeters, 1983). The

combination of NaCl, calcium, and acids produced from the lactic acid bacteria results in a

fermented cucumber that possesses a firm, crisp texture that can be stored for a year or more

(Fleming and McFeeters, 1981). Fermented cucumbers are desalted and processed further to

produce shelf-stable pickle products such as hamburger dill chips, genuine dills, relishes, and other products.

1.1.2 Cucumber Microstructure

Pickling cucumber varieties are chosen to have the capacity to produce a firm, crisp-textured, semi-processed, or fermented product (Breene et al., 1972). The microstructure of foods impacts the perceived quality of the product. Cucumbers contain 96% water and the remaining composition includes 2.7% carbohydrates, 0.13% lipids, 0.69% proteins, 0.19% minerals, and the rest is vitamins (USDA, Nutrient Data Base). Cucumber microstructure consists mainly of epithelial cells. During the fermentation process, changes in the size and shape of the epithelial cells occur in the cells walls (Howard et al., 1990). The texture of the pickles change due to several reasons: an increase or decrease in pectic substances, degree of pectin esterification, and the degree of bound cations within the pickles (Tang and McFeeters, 1983). During the fermentation of cucumbers, pectin demethylation occurs at a rapid rate allowing for an elevated binding of cations to the cell wall pectin. The binding of Ca^{2+} with the cells tissue facilitated by fermentation and/or supplementation of $CaCl_2$ into the cover brine is important to prevent tissue softening (Buescher et al., 1979; Tang and McFeeters, 1983; Hudson and Buescher, 1985; Buescher and Hudson, 1986). Maruvada and McFeeters (2009) found that blanching cucumbers prior to low salt fermentations decreased the amount of enzymatic activity present in the product, resulting in a firmer product. The tissues become soft or firm depending on the tissues proximity to the center of the cucumber due to the maturity of the fruit (Walter et al., 1985). In firm tissues, the cell wall consists of

densely packed fibrillar material as the part of its composition, whereas in the soft tissues the cell wall consists of loosely packed material (Walter et al., 1985). However, the sugar composition within the cell walls was not influenced by calcium ions nor did it have an effect in tissue firmness (Tang and McFeeters, 1983).

Texture qualities of pickled cucumbers are influenced by the cucumber size and maturity of the fruit. Influence of cucumber size is due in part to the sugar composition being fully utilized during the fermentation process to retain texture, as well as the maturity of the fruit when harvested (Lu et al., 2002; Fleming and others, 1995). Commercial cucumbers used for pickle production range from 1-6 cm in diameter. The cucumbers are classified into size categories based on diameter: 1 being up to 2.70cm; 2 being - 2.70-3.81; 3 being - 3.81-5.08; and 4 being 5.08-5.72cm in diameter. The size of the cucumber determines the fermentation and texture qualities of the pickle. Texture is often difficult to retain in cucumbers with large diameters that include 3.81 cm or more. When large size pickles are sliced, the locular tissues separate from the pericarp causing softening within the tissue (Hudson and Buescher, 1980). Seed maturity in cucumbers tends to be associated with softening of tissues, since they allow the surrounding tissues to degrade if too mature of a fruit. As the cucumber fruit matures, the seeds become enlarged, causing the surrounding tissues to soften (Hudson and Buescher, 1980). Most pickle producers use cucumbers that are between 2.7 and 4.5 cm in diameter to ensure a quality product.

1.1.3 The role of Salts in Fermentation and Processing of Pickles

Sodium chloride (NaCl), commonly referred to as salt in the food industry, is one of the oldest substances used to preserve foods. The addition of NaCl functions in two ways regarding fermentation of vegetables (Fleming et al., 1992). First, NaCl influences the type of microorganisms that will be active in the fermentation; second, it prevents softening of the vegetable tissues (Fleming et al., 1992). The addition of NaCl to cucumber fermentation aids in the selection of homofermentative lactic acid of bacteria, such as *Lactobacillus plantarum* that are necessary in the fermentation process. By adjusting the NaCl concentration, one can prevent pectinolytic enzymes from softening the vegetable tissues (Bell et al., 1961). When preparing the tanks for fermentation, dry salt is added below the headboards and mixed with water to allow the cucumbers to be submerged in the brine solution (Fleming et al., 1987). The addition of NaCl aids in microbial stability during bulk storage (Fleming et al., 1992; Fleming and others, 1995), and northern commercial producers do not have to worry about freezing in cold weather (Fleming et al., 1987). Other salts, such as $CaCl_2$ and alum, are used in conjunction with NaCl to maintain firmness of cucumber pickles (Buescher and Burgin, 1988; Buescher et al., 2011).

The human body requires calcium as part of a healthy diet. Currently, 75% of calcium intake in the United States diet comes from dairy sources and the remaining 25% comes from vegetables, fruits, and grains (Martin-Diana et al., 2007). Calcium salts are added into foods to extend the shelf life of preserved fruits and vegetables and enhance nutrition of the finished products (Martin-Diana et al., 2007). One benefit of consuming calcium enriched

vegetable products, is that the mineral maintains the texture quality of the produce (Martin-Diana et al., 2007). Calcium ions form cross-links with the free carboxyl groups in the produce to strengthen the cell wall (McFeeters and Fleming, 1989; Howard et al., 1990; Martin-Diana et al, 2007).

Calcium is a divalent cation that prevents plant tissues from softening (McFeeters and Fleming, 1989). However, McFeeters and Fleming (1989) determined that calcium reduces softening in cucumbers by binding to other sites besides pectin and carboxylic groups in the cucumber tissue. Calcium is mainly used in fermented salt-stock pickles at 1100 - 1800 ppm to provide crispness in the pickle before desalting (Buescher et al., 1979; Buescher et al., 2011). During processing of the pickles, the calcium ions are washed out in order to have less residual calcium in the product to meet the federal requirements (Buescher et al., 2011). Calcium use during pickle fermentation has allowed the reduction of sodium concentrations within the fermentation tanks (Buescher et al., 1979, 1981; Hudson and Buescher 1980, 1985, 1986; Buescher and Hudson 1984, 1986; Howard et al., 1990). The salt concentrations used for fermentation and storage of cucumbers is much higher than what is acceptable in finished products made from the cucumbers. A desalting step is used to lower the salt concentrations in fermented cucumbers before they are made into products (Howard et al., 1990). Therefore, the need to reduce chlorides in the tankyard waste streams led to the development of a new process using only $CaCl_2$ as the brining salt in the fermentation of cucumbers (McFeeters and Perez-Diaz, 2010).

McFeeters and Perez-Diaz, (2010) developed an environmentally-friendly fermentation process to replace 6% NaCl with 1.1% $CaCl_2$ as a way to combat the growing environmental issues commercial producers are facing, as well as the pressure from the EPA to prevent the large release of chloride ions into the local waterways. The amount of chloride allowed to be disposed in fresh water is 230ppm (EPA, 1987). If the chloride level in the local water streams reaches or exceeds that critical chloride limit, pickle processors have to be temporarily suspended until under the legal limit. It was determined by laboratory research that using 100mM $CaCl_2$ (1.1%) in the fermentation brine could produce a fermented product that allows pickle processors to eliminate the use of NaCl in the fermentation process and reduce the chloride in the tankyard by 80% (McFeeters and Perez-Diaz, 2010).

Aluminum sulfate, commonly referred to as alum, is another firming agent that is used during the desalting process after fermentation and storage periods (Fabian and Krum, 1949; Etchells et al., 1972). Although alum has been reported to reduce firmness of fresh pack pickles (Etchells et al., 1972), its effectiveness in increasing firmness of fermented-brine cured pickles is well documented (Fabian and Krum, 1949). Fermented cucumbers are usually desalted in water containing alum to aid in firmness while reducing the amount of NaCl in the processed product (Pflug et al., 1975; Buescher and Burgin, 1988). Aluminum binds to the pectic substances to retain the texture of the fruit (Etchells et al., 1972) and plays a central role in enhancing the crispness and firmness of pickles (Etchells et al., 1972).

1.1.4 The Role of Enzymes in Softening of Cucumber Pickles

Polygalacturonases are a type of enzyme found in raw cucumber fruits. The enzyme is typically present during the fermentation process, and may degrade the pectic substances in the cucumber fruit. The polygalacturonase enzymes have been shown to contribute to the gradual softening of pickles, which affects both the quality and market-life of the pickle (McFeeters et al, 1980; Buescher and Hudson, 1986; Cho and Buescher, 2012). Demethylation of cucumber pectin occurs when cucumbers are exposed to brine solutions (Bell et al., 1961; McFeeters et al., 1985). To maintain firmness in fresh-pack or fermented pickles, commercial producers must have little to no polygalacturonase activity and can achieve this result by pasteurizing the finished product before shelf storage (Buescher and Hudson, 1986; Cho and Buescher, 2012). Polygalacturonase activity can also be prevented by the addition of high sodium and calcium chloride to the fermentation brine to prevent the enzyme from softening the tissues of the cucumber fruit (Buescher et al., 1979; Buescher and Hudson, 1984; Fleming et al., 1987). Blanching of cucumbers prior to low salt fermentation was also found to decrease enzymatic activity (Maruvada and McFeeters, 2009). Changes in the composition of the cell wall due to enzymatic activity has mainly been studied in fermented cucumbers, and might be a factor influencing texture properties of both the traditional and environmentally friendly fermented pickle products. In non-blanched, $CaCl_2$ brined, fermented cucumbers, calcium salts along with pasteurization, prevent the polygalacturonase enzymes from being activated and allowed the firmness of the cucumber fruits to be maintained after fermentation was complete (Maruvada and McFeeters, 2009).

However, using calcium salts in the commercial production of fermented cucumbers without NaCl is an area of research that requires further study.

Pasteurization is used in order to inactivate softening enzymes in cucumber fruits and inactivate spoilage microorganisms. This heating step is essential for both fermented and fresh-pack cucumbers in order to prevent as much texture deterioration in the product as possible during shelf storage. The process is commonly used to preserve pickled fruits and vegetables where lower concentrations of sodium chloride and acetic acid are used (Fleming et al., 1993). Many pickles, including both fresh-packed and fermented, undergo this process to assure a long shelf life of the product. Pasteurization consists of using high temperatures to inactivate spoilage organisms without imparting a texture loss on the product. If a pickle remains at an elevated temperature for an extended period of time, then a loss of the cucumber properties will occur (Etchells and Jones, 1944). Pasteurization as a heat process inactivates softening enzymes typically found in the fruit prior to extended shelf storage. A detailed evaluation of processing parameters during pasteurization including temperatures, acid levels, and product pack-out ratios under commercial processing conditions was performed (Monroe et al, 1969). Monroe et al. (1969) established a recommended pasteurization temperature for heating all products to 74°C, at the coldest point of the jar, and holding it for 15 minutes to ensure proper inactivation of microorganisms was achieved. This heat process set the standard for how pickle processers heat their product to produce a shelf-stable product.

1.2 Instrumental and Sensory Analysis of Fermented Cucumbers

1.2.1 Food Texture

Texture of a food product consists of the rheological and structural (geometric and surface) attributes that are perceived by mechanical, tactile, visual, and auditory receptors (ISO, 1981). Szczesniak (1963), a pioneering publication in texture research, classified textural characteristics in three groups: 1) mechanical characteristics perceived by the forces on teeth, tongue and roof of the mouth when the food is stressed, 2) geometrical characteristics related to size, shape and the arrangements of particles in food, 3) other characteristics such as moisture and fat content of food that contribute to mouth-feel attributes.

Many instrumental techniques have been developed to determine the characteristics of a food material. Rheology is defined is the science of the deformation and flow of matter, concerned with forces, deformations and time (Blair, 1958). Rheological tests may be categorized as either empirical or fundamental (Foegeding et al., 2003). Empirical tests are relatively simple tests that typically measure the force of deformation on a sample. The test is typically used to determine basic material texture attributes and used for quality control purposes (Rosenthal, 1999). On the other hand, fundamental tests account for sample volume, shape, and testing conditions. Fundamental tests require the sample to be homogeneous to provide measurements on the physical properties of the food. A puncture test is one type of empirical test that is commonly used to measure the force required to push

a punch or probe into a food. The test is characterized by (a) a force measuring instrument, (b) penetration of the probe into the food causing irreversible crushing of the food, and (c) the depth of penetration is usually held constant (Bourne, 2002).

Compression is a form of a fundamental testing that allows for examination of the structure within a food product and how it resists compression. Compression testing is achieved between two parallel points until the tissue fails (Peleg and Gomez Brito 1977; Diehl et al., 1979). Uniaxial compression gives information about the mechanical and fracture properties of food under large-scale deformation (Luyten et al., 2007). The measurements determined from compression testing allows for several rheological parameters to be evaluated such as stress and strain values (Diehl et al., 1979). The amount of compression force needed to compress at a constant load on a sample was an index of softness and the force required to cause a given deformation within the sample was taken as an index of firmness (Bourne, 2002).

1.2.2 Texture Analysis of Cucumber Pickles

Mechanical texture tests have been performed on fruits, which not only include puncture, compression, and shear tests, but also ultrasonic methods (Abbott, 1999). The first tests performed to determine pickle texture were completed using a fruit pressure tester (FPT). This device measured the amount of resistance from the fruit in relation to the pressure used to puncture through the exocarp with a tipped rod (Magness and Taylor, 1925; Jones et al., 1950). The readings from this test were considered an adequate representation of overall fruit firmness (Jones et al., 1950). Pickles and cucumbers were assigned a value

according to the perceived quality of the fruits, and this scale is still in use today in the picking industry (Jones et al, 1954; Bell et al., 1955). Values above 18 represent very firm; values 14-18 represent fair to good; and values 10-14 represent fair to poor pickle stock (Jones et al, 1950).

Texture Profile Analysis (TPA) was developed in the 1960s and 70s to determine texture parameters in foods, including cucumbers and fermented cucumbers (Szczesniak et al., 1963; Breene et al., 1975). TPA uses force deformation curves from compression testing of standardized samples in order to make correlations between instrumental and sensory analysis. This type of testing showed the differences between brittleness, hardness, and work firmness in the endocarp and mesocarp as determined using an Instron Universal Testing Machine (Jeon et al., 1973). Puncture tests can be performed with several flat tipped plungers in order to see the variation in fracture (Thompson et al., 1982). The puncture test measures the force required to puncture a food product using a punch tip (Bourne, 1982). Thompson et al., (1982) was able to correlate the instrumentally measured firmness of fermented cucumber mesocarp tissue to sensory measurements of hardness.

1.2.3 Descriptive Sensory Analysis

Descriptive analysis is a sensory method by which the attributes of a food are identified and quantified by individuals who have had specific training for this purpose. This sensory technique has been used to establish relationships between sensory and instrumental measures. Sensory evaluation is defined as " the scientific method used to evoke, measure, analyze, and interpret those responses to products as perceived through the senses of sight,

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smell, touch, taste, and hearing" (Stone and Sidel, 1993). Descriptive sensory analysis is a quantitative technique that allows numerical data to be collected to establish relationships between product characteristics and human perception (Lawless and Heymann, 1998). This type of quantitative analysis can measure several parameters of the product including aroma, taste, appearance, and texture. Descriptive sensory analysis testing is useful when detailed information is required about the product. In most textural quality panels, 6-10 individuals are trained. The training of panelists can take from several weeks to several months depending on the attributes being studied (Meilgaard et al., 2007). Descriptive sensory analysis also provides a tool to assess product quality and prototypes of products (Meilgaard et al., 2007). There are two commonly used methods in which to perform descriptive analysis, Quantitative Descriptive Analysis (QDA) and the Spectrum [TM] method.

The Spectrum™ method was designed for industry and expands the texture analysis that was originally based on the texture profile method. The texture profile method was developed by General Foods in the 1960s as a way to define textural attributes in several food products. The Spectrum ™ scale is a refined scale that expands the full spectrum, to analyze attributes in a product. It is a 15-point scale, with 0 corresponding to no sensation and 15 corresponding to the strongest sensation imaginable. This universal scale does not generate a panel-specific vocabulary to describe the sensory attributes of the product, but instead uses a lexicon of standardized terms that are used consistently and have several reference points throughout the scale. These reference anchors allow panelists to scale intensities and are calibrated to make product comparisons. The panelists are provided with lexicons to describe perceived sensations with each sample (Lawless & Heymann, 1998).

Under the Spectrum™ method, panelists usually undergo an intensive training of 50-100

hours by a panel leader. After the intensive training, and after texture attributes are defined

and discussed by the panel, panelists are able to evaluate product characteristics and compare

them to a standardized lexicon (Meilgaard et al., 2007). This method is useful to determine

change in a product over time. As with any method that involves sensory analysis, the

drawback to the spectrum method includes panel training and maintenance.

Early pickle texture research showed that certain texture attributes such as brittleness,

hardness, and cohesiveness could be detected in fermented cucumbers (Breene et al., 1972;

Jeon et al., 1973). Hardness/firmness is one of the most common sensory attributes that is

quantified in texture studies, and it is defined as the perceived force required to fracture a

sample during the first chew of a sample placed between the molars (Meilgaard et al., 2007).

However, these aforementioned texture attributes do not completely describe the texture

properties of a hamburger dill chip. Of particular interest are the auditory textures that are

commonly associated with fruit and vegetable products, crispness and crunchiness.

Consumers want a firm, crispy, and crunchy product when they are consuming fruit and

vegetable products (Kilcast and Fillion, 2001). Foods that produce auditory textures are

usually classified as either crisp or crunchy. Although these two texture terminologies are

strongly related, these descriptions are two distinct terms (Wilkinson, 2000). Crispness is

associated with higher pitch audible sounds (Wilkinson, 2000). Crunchiness is associated

with the lower pitched sounds that typically last longer than crispness sounds (Vickers, 1984;

Fillion and Kilcast, 2002). In order to be classified as either textural term, the food product

must be difficult to deform, but able to fracture under small deformations in order to produce

sound (Luyten et al, 2007). Crisp is one of the most frequently used texture terms to describe food (Szczesniak, 1963), and it is a textural characteristic that is universally accepted as a positive way to describe foods (Szczesniak, 1987). When used to describe food products, it usually signifies freshness and a high quality product. It is a term that is very important to consumers when wanting pleasure out of eating (Szczesniak, 1988). Crunchiness on the other hand, has been difficult for researchers to define in order to perform unified evaluation of food products (Chauvin et al., 2008). Both crispness and crunchiness of foods have a particular role in consumer appetite and continuous eating (Szczesniak, 1988; Chauvin et al., 2008; Fillion and Kilcast, 2002).

1.2.4 Instrumental and Sensory Correlations

Instrumental and sensory analyses of cucumber firmness are a way to assess the quality of pickles. Jeon et al. (1973) were the first to make correlations between fruit pressure tester (FPT) firmness and sensory responses as well as FPT measurements and TPA parameters (Breene et al., 1972; Breene et al., 1974) in both raw and fermented cucumbers. Furthermore, Thompson et al. (1982) found a significant correlation between sensory hardness and mesocarp firmness, defined as the amount of force to rupture the fermented cucumber tissue. When pickles with the skin removed were compressed using texture profile analysis (TPA), there was a high correlation between brittleness and sensory crispiness (Jeon et al, 1975). Buescher et al., (2011) was also able to detect a strong correlation between the amounts of residual calcium in the bulk product and sensory crispness in the finished products.

Relating instrumental and sensory methods of texture measurement is essential for several reasons. The most important reasons to develop objective tests are to substitute for the tedious and time-consuming nature of running sensory tests (Szczesniak, 1987). Instrumental tests would have an advantage of greater speed, accuracy, and standardization to assess quality in a food product. The second reason correlation is important is because the industry wants the ability to predict consumer response and the perceived sensory texture of products (Szczesniak, 1987). Instrumental analysis allows for unbiased results due to the elimination of human error of panelists and creates a common reference for researchers, industry, and consumers (Abbott, 1999). Sensory shelf life is determined by either the consumer or panel refusing to repurchase a product if the expected sensory properties are not fulfilled (Labuza and Schmidl, 1988). Determining the shelf-life by sensory attributes is helpful when differences exist in texture that would cause a loss in acceptance by the consumer. This type of knowledge is useful in either preventing or slowing down these changes to improve the shelf life of a product (Labuza and Schmidl, 1988).

1.3 Accelerated Shelf Life Testing

The UK Institute of Food Science and Technology defines shelf life as "the period of time during which the food product will remain safe, retain its desired sensory, chemical and microbiological characteristics, and comply with any label declaration of nutrition data" (IFST, 1993). Shelf-life is a function of time, environmental factors and susceptibility of product to quality change (Labuza and Schmidl, 1985). Accelerated shelf-life testing (ASLT) is a method that has been widely accepted and used in the food, pharmaceutical, cosmetic

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and other industries that have limited durability (Corradini and Peleg, 2007). The objective of ASLT is to store the finished product under a particular type of abusive condition, and examine the product periodically until end of shelf life occurs, in order to predict shelf life (Labuza and Taoukis, 1990). This particular type of testing provides knowledge on the impact of handling the products' storage distribution, and how shelf life dating is achieved (Mizrahi, 2004). It's a way to be practical when predicting the stability of a product and when it should be pulled off store shelves (Corradini and Peleg, 2007). The ASLT models are used in order to determine the kinetic model under a fast rate of deterioration. Most deterioration when it applies to food quality has been found to fit either a zero or first-order reaction, which is the curve fitting parameter (Labuza and Riboh, 1982). To determine the reaction rate, the data is plotted as the quality factor being measure on the y-axis versus time on the x-axis. The reaction is zero when produced on linear coordinates whereas a first order in linear on a semilog paper (Labuza, 1982). Temperature is the variable most commonly changed during these tests to achieve deterioration in the product. This kinetic approach allows estimations to be made about shelf-life and to compare against current stated shelf-life (Garcia-Garcia et al, 2008). This type of testing allows for an estimation on the degradation rate, to be determined using an Arrhenius equation. A model using a temperature vs. reaction rate relationship allows for shelf-life at normal storage temperature to be predicted (Labuza, 1984).

1.3.1 Arrhenius Equation

The Arrhenius equation is used to describe the rate of a reaction of a sample that uses different temperatures to accelerate deterioration. This kinetic model can be used to extrapolate shelf-life results from ambient to accelerated storage conditions. The general mathematical expression for the Arrhenius equation is as follows:

$$\underline{k = k_0}^{-EA/RT}$$

where k=rate constant for deteriorative reaction at temperature T, k_0 = constant, independent of temperature, E_A = activation energy (J/mole), R = ideal gas constant (8.314 JK^{-1} $mole^{-1}$), and T = absolute temperature (K). The value of E_A is a measure of the reaction rate sensitivity to temperature. The value of E_A is very specific to each system and is used to predict the shelf life at lower temperatures (Labuza, 1982). Each accelerated temperature a product may be subjected to is the E_A/R of the Arrhenius equation. The line predicted from plotting the accelerated temperatures allows for a shelf life to be determined on a sample.

1.3.2 Predictive Model in Pickle Products

The pickle industry usually gives their product a 1-2 year shelf-life based on pH, color, taste, and texture (Communication with Mount Olive Representative, 2012). Texture characteristics of pickles are impacted with the amount of storage time on the shelves. Over time, pickles tend to lose their crisp and crunchy textural characteristics. Most pickle processors give a 2-year shelf to their products. However, most consumers typically purchase pickles on a need basis and are typically purchased and consumed every 53 days (Pickle

Packers International, 2013). By using ASLT, higher temperatures can be utilized to observe texture degradation in a shorter period of time. A control at ambient temperatures is usually performed along with the accelerated temperatures to monitor changes during normal conditions. Using both ambient and accelerated temperatures will allow pickle processors a known factor that will allow them to pull products off the shelves if they know the product will no longer be considered acceptable to consumers. Creating pickles with desirable sensory textures that are more environmentally friendly, requires an understanding of factors during the fermentation process as well as what sensory terms describe the products. Continuous changes in texture properties of pickle during tankyard storage, as well as final processed products, made the evaluation of texture a complex process that required a multidimensional approach including assessment of rheological/fracture characteristics, sensory perception, and accelerated shelf life testing.

Hypothesis:

1). Fermentation of cucumbers in calcium chloride brine combined with higher residual calcium levels in the finished products will result in a longer shelf life of finished products due to slower texture degradation rates.

Objectives

1). To determine the texture quality in both bulk stored and finished fermented cucumber products.

2). To determine sensory textural characteristics of both environmentally friendly and traditionally processed fermented cucumbers.

3). To determine the effects of fermentation brine and residual calcium on the firmness of cucumber pickles during accelerated shelf life testing (ASLT) to predict the end of shelf life for these products.

4). To determine the correlations between the sensory texture characteristics and instrumental measurements to determine the end point in the ASLT model.

Significance/Impact:

The instrumental and sensorial characterization of both environmentally friendly and traditionally fermented cucumbers will enhance the knowledge of texture properties of bulk stored and finished products. This knowledge will contribute to pickle processors understanding the changes in texture that occur during fermentation and bulk storage and determine when to process the pickles into finished products.

The development of an accelerated shelf life testing model to predict changes in texture during shelf storage is needed to more adequately determine end of shelf life for fermented cucumber pickles. In this study, different brined and processed fermented cucumbers were created to characterize texture by mechanical and sensory properties. Using a sensory approach to determine the point of quality loss combined with ASLT will enhance pickle processors' knowledge of when end of shelf life occurs. This knowledge will have an impact on the amount of time these products are on store shelves, ultimately resulting in consumers enjoying a firm, crisp fermented cucumber pickle.

References

Abbott, J. A. (1999). Quality measurement of fruits and vegetables. *Postharvest Biology and Technology*, 15(3), 207-225.

Bell, T. A., & Etchells, J. L. (1961). Influence of salt (NaCl) on pectinolytic softening of cucumbers. *Journal of Food Science*, 26(1), 84-90.

Bell, T. A., Etchells, J. L., & Jones, I. D. (1951). Pectinesterase in the cucumber. *Arch Biochem Biophys*, 31(3), 431-441.

Bell, T. A., Etchells, J. L., & Jones, I. D. (1955). A method for testing cucumber salt-stock brine for softening activity. *USDA-ARS*, 72(5), 15.

Bell, T. A., Turney, L. J., & Etchells, J. L. (1972). Influence of different organic acids on the firmness of fresh-pack pickles. *Journal of Food Science*, 37(3), 446-449.

Blair, G. W. (1958). Rheology in food research. *Advances in Food Research*, 8(1-61).

Bourne, M. C. (1982). Measure of shear and compression components of puncture tests. *Journal of Food Science*, 31(2), 282-291.

Bourne, M. C. (2002). Principles of objective texture measurement. Food texture and viscosity: Concept and measurement (pp. 107-189). San Diego, CA: Academic Press.

Breene, W. M., Davis, D. W., & Chou, H. (1972). Texture profile analysis of cucumbers. *Journal of Food Science*, 37, 113-117.

Breene, W. M., Davis, D. W., & Chou, H. (1973). Effect of brining on objective texture profiles of cucumber varieties. *Journal of Food Science*, 38, 210-214.

Breene, W. M., Jeon, I. J., & Bernard, S. N. (1974). Observations on texture measurement of raw cucumbers with the fruit pressure tester. *Journal of Texture Studies*, 5, 317-327.

Breidt Jr., F., McFeeters, R. F., & Díaz-Muñiz, I. (2007). Fermented vegetables. In M. P. Doyle, & L. R. Beuchat (Eds.), *In food microbiology: Fundamentals and frontiers* (3rd ed., pp. 783-793). Washington, D.C: ASM Press.

Breidt, F. (2006). Safety of minimally processed, acidified, and fermented vegetable products. In G. Sapers, J. Gorny & A. Yousef (Eds.), *Microbiology of fruits and vegetables* (pp. 313-335). Boca Raton, FL: CRC Press, Inc.

Buescher, R. W., & Burgin, C. (1988). Effect of calcium chloride and alum on fermentation, desalting, and firmness retention of cucumber pickles. *Journal of Food Science*, 53(1), 296-297.

Buescher, R. W., Hamilton, C., Thorne, J., & Jin Cho, M. (2011). Elevated calcium chloride in cucumber fermentation brine prolongs pickle product crispness. *Journal of Food Quality*, 34, 93-99.

Buescher, R. W., & Hudson, J. M. (1986). Bound cations in cucumber pickle mesocarp tissue as affected by brining and CaCl2. *Journal of Food Science*, 51(1), 135-137.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1979). Inhibition of polygalacturonase softening of cucumber pickles by calcium chloride. *Journal of Food Science*, 44, 1786-1787.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1981). Utilization of calcium to reduce pectinolytic softening of cucumber pickles in low salt conditions. *Lebensm. Wiss. Technology*, 14, 65-69.

Buescher, R. W., & Hudson, M. (1984). Softening of cucumber pickles by cx-cellulase and its prevention by calcium. *Journal of Food Science*, 49, 954-955.

Chauvin, M. A., Younce, F., Ross, C., & Swanson, B. (2008). Standard scales for crispness, crackliness and crunchiness in dry and wet foods: Relationship with acoustical determinations. *Journal of Texture Studies*, 39, 345-368.

Cho, M. J., & Buescher, R. W. (2012). Potential role of native pickling cucumber polygalacturonase in softening of fresh pack pickles. *Journal of Food Science*, 77(4), 443-447.

Civille, G. V., & Szczesniak, A. S. (1973). Guidelines to training a texture profile panel. *Journal of Texture Studies*, 4, 204-223.

Corradini, M. G., & Peleg, M. (2007). Shelf-life estimation from accelerated storage data. *Trends in Food Science and Technology*, 18, 37-47.

Diehl, K. C., Hamann, D. D., & Whitfield, J. K. (1979). Structural failure in selected raw fruits and vegetables. *Journal of Texture Studies*, 10, 371-400.

EPA. (1987). Water quality criteria; availability of document Fed. reg. 52: 37655- 37656. Washington D.C: *Environ. Prot. Agency*.

Etchells, J. L., Bell, T. A., & Fleming, H. P. (1977). Use of calcium chloride to improve the texture of pickles *Advisory statement published and distributed by Pickle Packers International, Inc*. St. Charles, IL.

Etchells, J. L., Bell, T. A., & Turney, L. J. (1972). Influence of alum on the firmness of fresh-pack dill pickles. *Journal of Food Science*, 37, 442-445.

Etchells, J., L., & Jones, I. D. (1944). Pasteurization of pickle products. *Fruit Products Journal*, 21, 330-332.

Fabian, F. W., & Krum, J. K. (1949). The effect of alum on microorganisms commonly found in pickles. *Journal of Fruit Production*, 28, 358.

FAOSTAT. (February, 2004). Food and agriculture organization of the United Nations. *Agricultural Production*, 2013, 2013.

Fillion, L., & Kilcast, D. (2002). Consumer perception of crispness and crunchiness in fruits and vegetables. *Food Quality and Preference*, 13, 23-29.

Fleming, H. P. 1. (1982). Fermented vegetables. In Rose AH (Ed.), *Economic microbiology. fermented foods*, vol. 7 (chapter 7) (pp. 227-258). New York , NY: Academic Press, Inc.

Fleming, H. P., Humphries, E. G., Fasina, O. O., McFeeters, R. F., Thompson, R. L., & Breidt, F. (2002). Bag-in-box technology: Pilot system for process-ready, fermented cucumbers. *Pickle Packers Science*, 8, 1-8.

Fleming, H. P., McDonald, L. C., McFeeters, R. F., Thompson, R. L., & Humphries, E. G. (1995). Fermentation of cucumbers without sodium chloride. *Journal of Food Science*, 60(2), 312-315.

Fleming, H. P., & McFeeters, R. F. (1981). Shelf life of fresh-pack cucumber pickles. St. Charles, IL: Pickle Packers International, Inc.

Fleming, H. P., McFeeters, R. F., & Daeschel, M. A. (1992). Fermented and acidified vegetables. In S. D. Vanderzant C (Ed.), *Compendium of methods for the microbiological examination of foods* (3rd ed., pp. 929-952). Washington, D.C: American Public Health Association.

Fleming, H. P., McFeeters, R. F., & Thompson, R. L. (1987). Effects of sodium chloride concentration on firmness retention of cucumbers fermented and stored with calcium chloride. *Journal of Food Science*, 52(3), 653-657.

Fleming, H. P., & Moore, W. R., Jr. (1983). Pickling. in processing of horticultural crops in the United States. In G. Fuller, & G. G. Dull (Eds.), *In CRC handbook of processing and utilization in agriculture* (Vol. 2 ed., pp. 397- 463.). Boca Raton, Florida: CRC Press, Inc.

Fleming, H. P., Thompson, R. L., Bell, T. A., & Hontz, L. H. (1978). Controlled fermentation of sliced cucumbers. *Journal of Food Science*, 43(3), 888-891.

Foegeding, E. A., Brown, J., Drake, M., & Daubert, C. R. (2003). Sensory and mechanical aspects of cheese texture. *International Dairy Journal*, 13, 585-591.

Garcia-Garcia, P., Lopez-Lopez, A., & Garrido-Fernandez, A. (2008). Study of the shelf life of ripe olives using an accelerated test approach. *Journal of Food Engineering*, 84, 569-575.

Global Industry Analysts Inc. (2008). Pickles- A global strategic business report. Retrieved 06/12, 2013, from http://www.strategyr.com/Pickles_Market_Report.asp

Hamann, D. D., Zhang, J., Daubert, C. R., Foegeding, E. A., & Diehl, K. C. (2006). Analysis of compression, tension and torsion for testing food gel fracture properties. *Journal of Texture Studies*, 37(6), 620-639.

Howard, L. R., & Buescher, R. W. (1990). Cell wall characteristics and firmness of fresh pack cucumber pickles affected by pasteurization and calcium chloride. *Journal of Food Biochemistry*, 14, 31-43.

Hudson, J. M., & Buescher, R. W. (1980). Prevention of soft center development in large whole cucumber pickles by calcium. *Journal of Food Science*, 45, 1450-1451.

Hudson, J. M., & Buescher, R. W. (1985). Pectic substances and firmness of cucumber pickles as influenced by CaCl2, NaCl and brine storage. *Journal of Food Biochemistry*, 9, 211-229.

Hutkins, R. W. (2006). Fermented foods in human history. In R. W. E. Hutkins (Ed.), *Microbiology and technology of fermented foods* (pp. 3-14-246-249.). Ames, Iowa: Blackwell Publishing.

IFST. (1993). Shelf life of foods-guidelines for its determination and prediction. London: *Institute of Food Science and Technology*.

Jeon, I. J., Breene, W. M., & Munson, S. T. (1973). Texture of cucumbers: Correlation of instrumental and sensory measurements. *Journal of Food Science*, 38, 334-337.

Jeon, I. J., Breene, W. M., & Munson, S. T. (1975). Texture of fresh-pack whole pickles: Correlation of instrumental and sensory measurements. *Journal of Texture Studies*, 5, 399-409.

Jones, I. D., Etchells, J. L., & Monroe, R. J. (1954). Varietal differences in cucumbers for pickling. *Food Technology*, 8(9), 415-418.

Jones, I. D., & Etchells, J., L. (1950). Cucumber varieties in pickle manufacture. *Canner*, 110(1), 34, 36, 38, 40.

Kilcast, D., & Fillion, L. (2001). Understanding consumer requirements for fruit and vegetable texture. *Nutrition and Food Science*, 31(5), 221-225.

Labuza, T. P. (1982). *Shelf-life dating of foods* (1st ed.). Westport, CT: Food & Nutrition Press, Incorporated.

Labuza, T. P. (1984). Application of chemical kinetics to deterioration of foods. *Journal of Chemical Education*, 61, 348-358.

Labuza, T. P., & Riboh, D. (1982). Theory and application of Arrhenius kinetics to the prediction of nutrient losses in foods [degradation, keeping, quality, temperature, quality controls, analysis, models. *Food Technology*, 66-74.

Labuza, T. P., & Schmidl, M. K. (1985). Accelerated shelf-life testing of foods. *Food Technology*, 39(9), 57-62.

Labuza, T. P., & Schmidl, M. K. (1988). Use of sensory data in the shelf life testing of foods: Principles and graphical methods for evaluation. *Cereal Foods World*, 33(2), 193-205.

Lawless, H. T., & Heymann, H. I. (1998). *Sensory evaluation of food: Principles and practices*. New York, New York: Kluwer Academic/Plenum Publisher.

Lu, Z., Fleming, H. P., & McFeeters, R. F. (2002). Effects of fruit size on fresh cucumber composition and the chemical and physical consequences of fermentation. *Journal of Food Science*, 67(8), 2934-2939.

Lucier, G., & Lin, B. (2000). Americans relish cucumbers. *Economic Research Service/USDA*, 9-12.

Luyten, H., Lichtendonk, W., Castro, E. M., Visser, J., & Van Vliet, T. (2007). Acoustic emission from Crispy/Crunchy foods to link mechanical properties and sensory perception. *Journal of Food Texture.* 380-392.

Magness, J. R., & Taylor, G. F. (1925). An improved type of pressure tester for the determination of fruit maturity. *U.S Department Agriculture Circulation*, 350.

Martin-Diana, A. B., Rico, D., Frias, J. M., Barat, J. M., Henehan, G. T. M., & Barry-Ryan, C. (2007). Calcium for extending the shelf life of fresh whole and minimally processed fruits and vegetables: A review. *Trends in Food Science and Technology*, 18, 210-218.

Maruvada, R., & McFeeters, R. F. (2009). Evaluation of enzymatic and non-enzymatic softening in low salt cucumber fermentations. *International Journal of Food Science and Technology*, 44, 1108-1117.

McFeeters, R. F. (1992). Cell wall monosaccharide changes during softening of brined cucumber mesocarp tissue. *Journal of Food Science*, 57(4), 937-953.

McFeeters, R. F., Brenes Balbuena, M., & Fleming, H. P. (1995). Softening rates of fermented cucumber tissue: Effects of pH, calcium and temperature. *Journal of Food Science*, 60(4), 786-788-793.

McFeeters, R. F., & Fleming, H. P. (1990). Effect of calcium ions on the thermodynamics of cucumber tissue softening. *Journal of Food Science*, 55(2), 446-449.

McFeeters, R. F., & Fleming, H. P. (1991). pH effect on calcium inhibition of softening of cucumber mesocarp tissue. *Journal of Food Science*, 56(3), 730-732.

McFeeters, R. F., Fleming, H. P., & Thompson, R. L. (1985). Pectinesterase activity, pectin methylation, and texture changes during storage of blanched cucumber slices. *Journal of Food Science*, 50, 201-205.

McFeeters, R. F., Senter, M. M., & Fleming, H. P. (1989). Softening effects of monovalent cations in acidified cucumber mesocarp tissue. *Journal of Food Science*, 54(2), 366-370.

McFeeters, R. F. (1986). Pectin methylation changes and calcium ion effects on the texture of fresh, fermented, and acidified cucumbers. *American Chemical Society*, 217-229.

McFeeters, R. F., & Perez-Diaz, I. (2010). Fermentation of cucumbers brined with calcium chloride instead of sodium chloride. *Journal of Food Science*, 75(3), 291-296.

Meilgaard, M. C., Civille, G. V., & Carr, B. T. (2007). *Sensory evaluation techniques* (4th Edition ed.). Boca Raton, Florida: CRC Press.

Mizrahi, S. (2000). Accelerated shelf-life tests. In D. Kilcast, & P. Subramaniam (Eds.), *The stability and shelf-life of food* (pp. 107-128). Boca Raton, FL, USA: CRC Press.

Monroe, R. J., Etchells, J. L., Pacilio, J. C., Borg, A. F., Wallace, D. H., Rogers, M. P., et al. (1969). Influence of various acidities and pasteurizing temperatures on the keeping quality of fresh-pack dill pickles. *Food Technology*, 23(1), 71-77.

Nicholas, R. C., & Pflug, I. J. (1960). Effect of high temperature storage on the quality of fresh cucumber pickles. *Glass Pack*, 39, 35.

Palnitkar, M. P., & McFeeters, R. F. (1975). Recycling spent brines in cucumber fermentations. Journal of Food Science, 40, 1311-1315.

Peleg, M. (1976). Compressive failure patterns of some juicy fruits. *Journal of Food Science*, 41(1320-1324)

Perez-Diaz, I. M., & McFeeters, R. F. (2011). Preparation of a lactobacillus plantarum starter culture for cucumber fermentations that can meet kosher guidelines. *Journal of Food Science*, 76(2), M120-M123.

Pflug, I. J., Fellers, P. J., & Gurevitz, D. (1975). Diffusion of salt in the desalting of pickles. *Food Technology*, 21, 1634-1638.

Pickle Packers International Inc. (2013). Pickle and pickled pepper fact sheet. Retrieved May 15, 2013, from http://ilovepickles.org/

Rosenthal, A. J. (1999). *Food texture: Measurement and perception* (1st ed.). Gaithersburg, Maryland: Aspen Publishers Inc.

Stone, H., & Sidel, J. L. (1993). *Sensory evaluation practices* (2nd ed.). Orlando, FL: Academic.

Szczesniak, A. S. (1963). Classification of textural characteristics. *Journal of Food Science, 28*(4), 385-389.

Szczesniak, A. S. (1987). Correlating sensory with instrumental texture measurements-an overview of recent developments. *Journal of Texture Studies*, 18, 1-15.

Szczesniak, A. S. (1988). The meaning of textural characteristics-crispness. *Journal of Texture Studies*, 19, 51-59.

Tang, H. C. L., & McFeeters, R. F. (1983). Relationships among cell wall constituents, calcium and texture during cucumber fermentation and storage. *Journal of Food Science*, 48(1), 66-70.

Thompson, R. L., Fleming, H. P., & Hamann, D. D. (1992). Delineation of puncture forces for exocarp and mesocarp tissues in cucumber fruit. *Journal of Texture Studies*, 23, 169-184.

Thompson, R. L., Fleming, H. P., Hamann, D. D., & Monroe, R. J. (1982). Method for determination of firmness in cucumber slices. *Journal of Texture Studies*, 13, 311-324.

Thompson, R. L., Fleming, H. P., & Monroe, R. J. (1979). Effects of storage conditions on firmness of brined cucumbers. *Journal of Food Science*, 44, 843-846.

Truong, V. D., & Daubert, C. R. (2000). Comparative study of large strain methods for assessing failure characteristics of selected food gels. *Journal of Texture Studies*, 31(3), 335-353.

USDA. (1966). In Office of Federal Registry. National Archives and Records Service, General Services Admin (Ed.), Standards for grades of pickles. fed. reg 31:102231-10305. Washington, D.C.:

USDA, E. (2007). Commodity highlight: Pickling cucumbers. Vegetables and Melons Outlook, , 24-30.

Vickers, Z. M. (1984). Crispness and crunchiness: A difference in pitch? *Journal of Texture Studies*, 15, 157-63.

Walter Jr., W. M., Fleming, H. P., & Trigiano, R. N. (1985). Comparison of the microstructure of firm and stem-end softened cucumber pickles preserved by brine fermentation. *Food Microstructure*, 4, 165-172.

Wilkinson, C., Dijksterhuis, G. B., & Minekus, M. (2000). From food structure to texture *Trends in Food Science and Technology*, 11, 442-450.

Yoshioka, Y., Horie, H., Sugiyama, M., & Sakata, Y. (2009). Quantifying cucumber fruit crispness by mechanical measurement. *Journal of Breeding Science*, 59(139-147).

Figures

Fresh Cucumbers Have Propelled Consumption Growth

Lbs. per capita

Pickles

Fresh

Economic Research Service, USDA
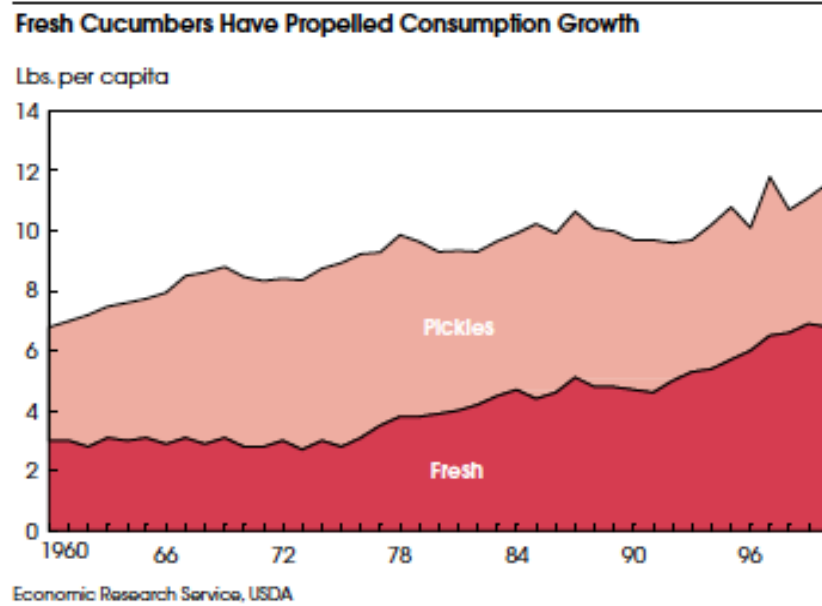
Figure 1.1 Trends in consumption growth for cucumbers and pickles (Reproduced from the works of Lucier and Hwan, 2000).
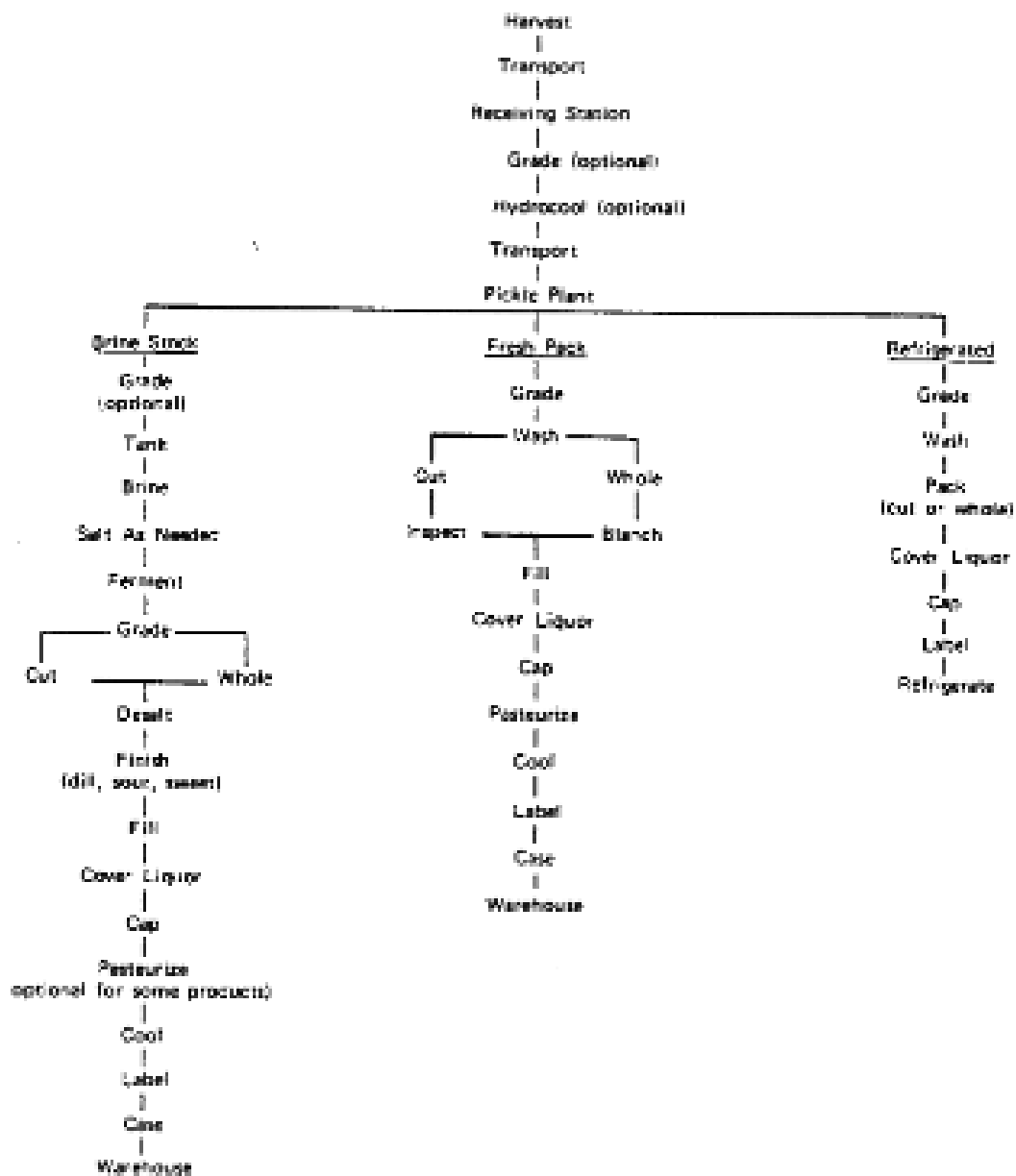
Figure 1.2 Flow Diagram for Cucumber Pickle Processing (Reproduced from the works of Fleming et al., 1983).
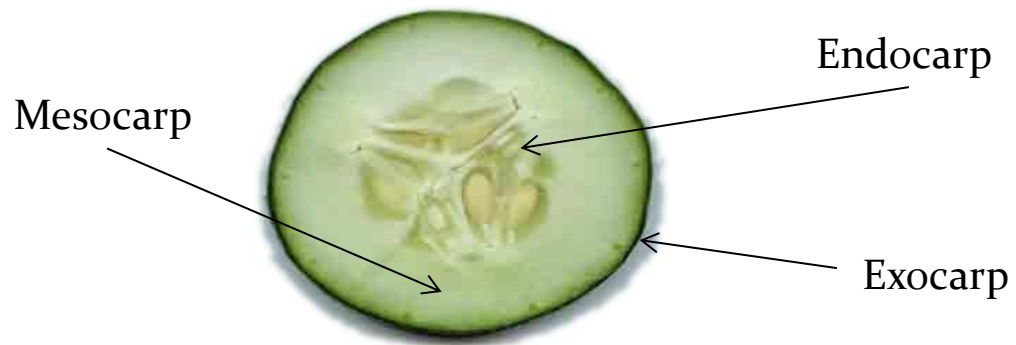
Figure 1.3 A cross sectional view of a cucumber fruit.

Chapter 2

Texture of Pickles Produced from Commercial Scale Cucumber Fermentation using Calcium Chloride instead of Sodium Chloride

2.1 Abstract

The texture of preserved cucumber fruits is a critical quality factor when producing pickles, including pickles made from fermented cucumbers. Pickles are traditionally produced on a commercial scale from cucumbers fermented in 5-8% sodium chloride (NaCl) brines. Although manufacturers recycle brines before discharge, effluent waste streams are high in NaCl. Pickle companies are considering environmentally friendly alternative methods to ferment cucumbers that include using 1.1% calcium chloride ($CaCl_2$), a known firming agent in pickled cucumbers. In this study, the effect of $CaCl_2$ and NaCl brining treatments on fermented cucumber texture properties was evaluated during bulk storage and for the processed pickle products. Cucumbers were fermented at a commercial facility in 3,000-gallon, open-top tanks with 6% NaCl or 1.1% $CaCl_2$ brines and kept for long-term storage to measure firmness retention after fermentation was complete. A portion of the fermented cucumbers were desalted, processed into hamburger dill chips (HDC) and stored at ambient temperature. Pickle mesocarp and exocarp firmness was measured using a TA-XT2 Texture Analyzer with a 3 mm diameter punch probe at a rate of 2.5 mm/sec. Fracture stress and fracture strain were measured on cork bored samples, from the mesocarp tissue, under compression testing to 80% deformation. Descriptive sensory texture attributes of hardness, crispness, crunchiness, and fracturability correlated to peak puncture force of the processed product, $R^2 = 0.70, 0.85, 0.80, 0.59$ respectively. Both brining treatment and bulk storage time had an effect on the overall firmness of the finished products ($p = 0.0007$ and $p < 0.0001$, respectively). The processed products that were fermented with traditional NaCl brine had a higher firmness value of 8.4 ± 0.1 N compared to 7.8 ± 0.1 N in cucumbers

brined in 1.1% $CaCl_2$ (p =0.0005). It was also observed that HDC produced from fermented

cucumbers stored for only 2 months in the tankyard regardless of brining treatment, had a

higher firmness than the HDC samples from the fermented cucumbers stored for 8 months

before being processed into products (p =0.0002). Furthermore, brining treatment and

cucumber size influenced the firmness of the unprocessed fermented cucumbers during bulk

storage for 9 months ($p < 0.0001$). This research demonstrated that commercial processors

can produce pickles that will retain firmness during fermentation using an environmentally

friendly $CaCl_2$ brining method, but should process the fermented cucumbers in 2 months'

time or use size 2B cucumbers in order to ensure the highest quality product for consumers.

2.2 Introduction

Approximately 35% of the pickling cucumber crop in the United States is preserved

in salt brines by fermentation, and can be stored for an extended period of time before being

processed into products. Fresh cucumbers of various sizes ranging between 27-45mm in

diameter are fermented with a cover brine solution containing an equilibrated concentration

of 5-8 % sodium chloride (NaCl), 0.1-0.4 % calcium chloride ($CaCl_2$), and 0.05 % acetic acid

(Breidt, 2006; Breidt et al., 2007; Hutkins, 2006; Buescher et al., 2011). The concentration of

NaCl used in traditional fermentations was selected to inhibit softening enzymes (Bell and

Etchells, 1961) and favor the growth of the naturally occurring lactic acid bacteria. The

natural homofermentative lactic acid bacteria (LAB), that are both salt and acid tolerant,

convert the natural sugars present in fresh cucumbers into acids. The sugars, glucose and

fructose, which are naturally found in the cucumber fruit with levels around 2%, are

converted into an end product of lactic acid, giving a final titratable acidity in the final

product of 0.6 % to 1.2 % (Fleming and others, 1973; Lu and others, 2002). During the

process of fermentation, the pH decreases to a final pH between 3.2 and 3.6 (Breidt, 2006;

Breidt et al., 2007; Hutkins, 2006). The fermentation process allows for cucumbers to be

stored for long periods of time in the tankyard, which allows pickle processors to produce

products year round (Fleming and McFeeters, 1981).

Fermented cucumbers that are made into finished products typically have a one to

two-year shelf life (Communication with Mount Olive Pickle Company Representative,

2013). Typically, cucumbers are fermented in a salt solution of 5-8%. It may be increased to

12-16% during the colder months to prevent freezing in northern tankyards during long-term

storage. NaCl is used in order to retain cucumber texture and prevent spoilage (Fleming,

1982). The addition of $CaCl_2$ to fermentation brines has reduced the amount of salt needed to

retain texture properties by interacting with pectic substances to impede softening of the

fruits (Buescher et al. 1979, 1981; Hudson and Buescher 1980, 1985, 1986; Buescher and

Hudson 1984, 1986; Howard and Buescher, 1990; Fleming et al., 1978; 1987; McFeeters et

al., 1991). The addition of calcium chloride has also been shown to retain the firmness of the

brined cucumbers in the presence of polygalacturonase enzymes (Buescher et al., 1979). The

use of calcium has also been investigated as a firming agent in the desalting process.

Cucumbers that were NaCl brined and processed without calcium had a lower overall

firmness compared to cucumbers that were brined with the addition of calcium chloride, or

when calcium chloride was used in the desalting process (Buescher and Burgin, 1988). This

effect of calcium on retaining firmness in pickle products has also been observed in long-

term storage in the tankyard as well as in finished products during shelf storage (Tang and McFeeters, 1983).

After the fermentation process is complete, the brine is recycled several times before it is discarded. The recycled brine goes through an extensive and expensive waste treatment process before release into the local water systems (Palnitkar and McFeeters, 1975; McFeeters et al., 2010). However, this treated brine contains high levels of sodium chloride; which may be toxic to the plants and wildlife found in the local water systems. Accordingly, the U.S. Environmental Protection Agency (EPA) limits chloride discharge in fresh waters to 230 ppm (EPA, 1987). The pickle industry has had a difficult time meeting the strict requirements set forth by the EPA. This strict chloride limit has led pickle companies to search for alternative methods of fermentation in order to achieve a more environmentally friendly process.

Recently, efforts have been made to reduce the amount of NaCl used in fermentation and storage of cucumbers in order to achieve a fermentation using only calcium chloride, rather than sodium chloride. Buescher et al (2011) determined that even in traditional NaCl fermentations, having a higher amount of calcium in the fermentation process resulting in higher residual calcium levels in the final products retains product crispness in the final product. McFeeters and Perez Diaz (2010) reported that cucumbers could be fermented in cover brine solutions that contain 100 to 200 mM calcium chloride and no NaCl. It was predicted that with concentrations of 1.1% calcium chloride and addition of a *Lactobacillus plantarum* starter culture an environmentally friendly fermentation on a tankyard scale could be achieved, reducing the chlorides by 80% (McFeeters and Perez-Diaz. 2010). Additionally,

calcium salts are not toxic to plants or wildlife, which will make it easier to dispose of these brines in the large quantities necessary for pickle processors. Texture is an important food characteristic that affects consumers' food quality ratings. Consumers prefer a firm, crisp, and crunchy pickle product (Thompson et al. 1982). The objectives of this study were 1) To determine the texture quality of CaCl2 brined, fermented cucumbers during bulk storage and in finished pickle products and 2) To determine the sensory texture characteristics of both environmentally friendly and traditionally processed fermented cucumber pickles.

2.3 Materials and Methods

2.3.1 *Commercial scale fermentations using calcium chloride instead of sodium chloride*

Cucumbers and ingredients for bulk storage of cucumbers and processing hamburger dill chip pickles. Fresh size 2B (31-38mm diameter) cucumbers were obtained from a local source.. Vinegar 200-grain (20% acetic acid), anhydrous $CaCl_2$, yellow # 5 food color, polish dill flavor concentrate, benzoate with color, Antifoam C mixture, NaCl, lids and glass were obtained at a local processor to produce products at a commercial facility.

Fermented Cucumbers for evaluation of texture in bulk storage. Fresh size 2B (31-38mm diameter) cucumbers and size 3A (38-44.5mm diameter) were collected from a local processor (Mount Olive Pickling Company, Mount Olive, NC). Eight 3,000-gallon open –air tanks were filled and processed in September 2012. Four of the tanks were each filled with size 2B (31-38mm diameter) and 3A (38-44.5mm diameter) cucumbers respectively. Commercial cucumber fermentations were carried out in 3,000 gallon open –air tanks filled with 50-60% whole cucumbers and 50-40% cover brine solution to achieve a fermented product of 6% NaCl or 1.1% $CaCl_2$. Cucumbers were packed in fiberglass tanks

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and immediately covered with wooden boards to prevent the fruit from floating until equilibrium between the fruit and cover brine components was completed between 2 weeks and 2 months. Each month 20-24 lbs. of fermented cucumbers were removed from the tank to evaluate firmness of the stored product (Figure 2.1).

       Fermentation of cucumbers using different brining treatments. Commercial cucumber fermentations using fresh size 2B (31-45mm diameter) cucumbers were carried out in 3,000 gallon open –air tanks filled with 50-60% whole cucumbers and 50-40% cover brine solution to achieve a fermented product of 6% NaCl or 1.1% $CaCl_2$ brine. Cucumbers were packed in fiberglass tanks and immediately covered with wooden boards to prevent the fruit from floating until equilibrium between the fruit and cover brine components was completed. Primary fermentation of the products is a period of 2 weeks-2 months depending on the packing season (unpublished). For the calcium chloride fermentation to occur a starter culture and preservative were used. The fresh cucumbers and cover brine were inoculated with 10^5-10^6 cfu/ml *Lactobacillus plantarum* LA0445 that was obtained from the culture collection of the U.S. Dept. of Agriculture-Agricultural Research Service, Food Science Research Unit, located in Raleigh, N.C., U.S.A and prepared according to the koshering process described by Perez-Diaz and McFeeters (2011). Different preservatives such as sodium benzoate, fumaric acid, and potassium sorbate were added to the cover brine in varying concentrations in order to prevent spoilage from occurring after fermentation was complete. These fermented cucumbers were in tanks for either 2 or 8-month storage time before being processed in hamburger dill chips.

Desalting of fermented cucumbers for commercial products. Once fermentation was complete and the fermented cucumbers were ready to be made into product, the fermented brine was removed using a purging system that circulates new water with the removal of the fermentation brine to leave behind the desalted fermented cucumbers in the tank. For hamburger dill chips (HDC) the salt content was reduced from 7% to a range between 4.3-4.9% in order for the chips to be at a level in which they could be processed. Alum at a concentration of 0.3% was also added to the desalting water to maintain firmness in the finished products. Once the fermented cucumbers were desalted, they were sliced and packed in 16 oz. jars with a 58: 42 cucumber: cover liquor ratio. All of the treatments were packed out to have a residual calcium level of ~21mM and 2.2% NaCl. Following the packing and brining, jars were sealed before they went through the heating process of pasteurization. Pasteurization was performed on a commercial scale in a tunnel pasteurizer at 145-154°F for 50 minutes at Mount Olive Pickle Company. Once pasteurization was complete, the finished products were stored at ambient temperature (25±2°C), and sampled every 2 months for texture evaluation.

2.3.2 Sensory Analysis of Commercially Processed Fermented Cucumber Pickles

Sensory Panelists. Students and staff (n=10) from North Carolina State University, (Raleigh, North Carolina) Department of Food Bioprocessing and Nutrition Sciences as well as the USDA Food Science Research Unit, (Raleigh, North Carolina) were selected as panelists. The panel was composed of two males and eight females. The panelists were regular pickle consumers. Panelists were selected based upon availability and ability to distinguish texture attributes in fermented cucumber products.

Sensory Panel Training. Approval of the North Carolina State University (Raleigh, North Carolina) Institutional Review Board (IRB # 2734) was obtained for the use of human subjects in this study. Panelists were trained in descriptive sensory analysis using the Spectrum™ method (Meilgaard et al., 2007). Panelists were initially trained (75+ hours) to use a 15-point intensity scale to assess flavor and texture attributes in cucumber products. Seven texture attributes including hardness, crispness, crunchiness, fracturability, skin and seed awareness, and juiciness were evaluated in fermented cucumber products (Table 2.1). During the training, panelists were individually evaluated to determine the overall panel mean and to assure that all panelists were able to scale the attributes of interest. Panelists were trained to rate hardness, fracturability, and juiciness based upon reference samples suggested by the Spectrum™ method. Hardness was described as the force required to compress the sample upon first bite, with anchors between 0 (soft) and 15 (firm). Fracturability was defined as the force in which the sample ruptures when bit down on between the molar teeth at a fast rate. Juiciness was defined as the amount of juice/moisture perceived in the sample. Crunchiness and crispness attributes were based upon reference samples suggested by Chauvin (Chauvin et al., 2008). Crispness was defined as the multiple, higher-pitched sounds produced as the sample is crushed with the molar teeth. Crunchiness was defined as a single lower-toned noise produced with each chew (Chauvin et al., 2008). Crispness and crunchiness intensity scores were evaluated during the first 3 chews. Skin and seed awareness were defined by evaluating several food products by the panelists. Panelists were trained on various foods based upon recommendations of Meilgaard et al., (2007). Following training, (Meilgaard et al., 2007) panelists met as a group to assign a consensus

score for each attribute of the pasteurized hamburger dill chip pickles, which were prepared directly from fermented cucumbers and were provided as a reference sample. Reference pickles were stored at $2^{o}C$ throughout the experiment and maintained their texture attributes, which were determined during the initial training of the panelists.

Sensory analysis. Samples that were evaluated for sensory analysis were presented in 2 oz. plastic cups identified with randomized 3 digit numbers. All samples and the reference pickle were provided at room temperature along with room temperature distilled water, a 2 oz. sample of Muenster cheese, and salt-free saltine crackers to cleanse the palate between samples. Panelists were presented samples in a randomized order to avoid bias based upon the order of sample presentation. Two to four pickle slices were placed in each sample cup. Panelists evaluated a group of 6 to 8 samples per session. They were also given two 2 oz. sample cups identified as the reference sample to calibrate scoring of taste and texture attributes on the 15-point scale. Each panelist was instructed to first taste the reference sample, cleanse his or her palate, and then consume an unknown pickle sample. Panelists were asked to take a two-minute rest period between each sample to avoid tasting fatigue. They were given the option to swallow or expectorate their samples. Three sensory replications were performed on each sample during the study. Sensory analysis was conducted on pickle samples from both NaCl and $CaCl_2$ fermented and processed cucumbers, which were stored at ambient temperature ($25^{o}C \pm 2\,^{o}C$), to determine the effects of brining and storage in the fermented cucumbers.

2.3.3 Evaluation of Instrumental Firmness of Fermented Cucumbers during Bulk Storage

Firmness Measurements of Bulk Storage Fermented Cucumbers. Firmness was measured using a TA.XT2 Texture Analyzer (Texture Technologies Corp, Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) equipped with a 3 mm diameter punch probe. Pickle slices (6 mm thick) were placed onto a base plate containing a 3.1 mm hole. The punch probe moved at a test speed of 2.5 mm/sec into the cucumber slices. Data were collected and analyzed using Texture Expert software (Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK). The force required to puncture the cucumber mesocarp was recorded and expressed in Newtons (N). Newtons is a standard unit of measure of $F = m*a$, where F is the force applied, m is the mass of the object receiving the force, and a is the acceleration of the object. Firmness measurements were done on 15 slices from three jars from each treatment at each sampling time point (Yoshioko et al., 2009; Thompson et al., 1982). Texture was evaluated at ambient temperature (25 ± 2°C) at the sampling times of 2, 4,6,8,10,12, and 18 months from the original packing of the products in July 2011. Mesocarp and Exocarp firmness of the bulk stored fermented cucumbers were also measured using a TA.XT2 Texture Analyzer (Texture Technologies Corp, Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK). The force required to puncture the cucumber mesocarp and exocarp was recorded and expressed in Newtons (N). Firmness measurements were completed on 15 reps of each component of the cucumber, at each sampling time point (Yoshioko et al., 2009; Thompson et al., 1982). Texture was evaluated at ambient temperature (25 ± 2°C) each month from the original packing of the products into the tanks in September 2012.

A fruit pressure tester was used to measure firmness on the bulk-stored fermented cucumbers. The Magness-Taylor Fruit Pressure Tester (FPT) (Ballauf Mfg. Co: Washington, D.C.) fitted with 0.79cm (5/16in.) diameter plunger tip was manually inserted into the fruits from each tank evaluated. All FPT scale readings were estimated at 0.1 lbs. by the same individual who did the all-manual FPT testing. Texture was evaluated at an ambient temperature (25 ± 2°C) each month from the original packing of the products into the tanks in September 2012. Each cucumber was punctured once through the exocarp portion of the cucumber midway between the stem and blossom end. Each puncture test value that was reported is the mean of 10 individual cucumber puncture values.

Compression Measurements of bulk stored fermented cucumbers. Uniaxial compression measurements were taken using a TA.XT2 Texture Analyzer (Texture Technologies Corp, Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) attached with a load cell of 50 N and equipped with a parallel plate with a diameter of 50mm. Cylindrical specimens were extracted from the mesocarp section of the cucumber slice using a cylindrical cork borer. The dimensions of the cylinders were 6mm in diameter and 6mm in height. A one-cycle compression was set for an 80% deformation at a crosshead speed of 2.5 mm/sec. There were two replications of the instrumental analysis conducted on two separate days. For each replication, 15-cored samples of the 8 different tanks were evaluated. Texture was evaluated after storage at ambient temperature (25 ± 2°C) each month from the original packing of the products in September 2012. All samples fractured during compression, therefore force and distance data were used to calculate stress and strain at fracture, as described below (Hamann et al., 2006; Truong and Daubert, 2000). Fracture stress, and

fracture strain were determined from the following equations:

$$\sigma = \frac{F}{A}$$

Where σ = stress (N/mm^2); F= force (N); A is cross sectional area (mm^2) of samples before

compression.

True Compressive Strain (ε_h) was calculated as:

$$\varepsilon_h = \ln\left(1+ \frac{\Delta L}{L_i}\right)$$

ε = hencky strain; ΔL= change in height (mm) of sample after deformation and L_i is initial

height (mm)

Statistical analysis of sensory and instrumental data. All statistical computations

were analyzed using a general linear model from JMP (version 10 SAS® software, SAS

Institute, Cary, NC). Means and standard deviations were calculated for both sensory (n=8)

and instrumental (n=10) data using JMP version 10.1

2.4 Results and Discussion

2.4.1 Evaluation of texture in pickles during storage in the tankyard

Texture of environmentally friendly fermented cucumbers during bulk storage has yet

to be examined until this section. Several forms of texture evaluation were performed to

understand the texture properties of CaCl$_2$ brined, fermented cucumbers during bulk storage

(Figure 2.1). The pickle industry uses a fruit pressure tester to quickly examine the texture of

the cucumbers during bulk storage. This texture measurement was compared to the

instrumental measurements performed on the TA-XT2 texture analyzer. Based on the texture

data collected from the fruit pressure tester, there were no significant differences between the

two brining treatments $p < 0.05$. The average force of the cucumbers both when fresh and

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fermented in either brine was about 16.0 ± 0.2lbs (Figure 2.2). The smaller diameter cucumbers (2B's) had an average mean slightly higher than the 3A size cucumbers. A decrease from initial firmness, regardless of size and brining treatments, due to bulk storage for a long period of time was significant ($p < 0.0001$). Even though the texture decreased, the pickles were still considered to be acceptable by industry standards. The readings from this test were considered an adequate representation of fruit firmness based on the scale developed to rate firmness using a FPT (Jones et al., 1950). Pickles and cucumbers were assigned a value according to the quality of the vegetables firmness (Jones et al., 1954; Bell et al., 1955). Values above 18 represented very firm; values 14-18 represented fair to good; and values 10-14 represented a fair to poor stock of pickles (Jones et al., 1950). In conclusion, the results presented in Figure 2.2 indicate that further investigation is needed to evaluate firmness values in bulk storage, using both fundamental and empirical measurements, since no distinct differences were observed between the different size and treatments of the fermented cucumbers.

Based on the texture data from both the mesocarp and exocarp sections of the fermented cucumbers, which were collected from the same sampling time points, no significant changes in texture occurred during the 9-month storage in the tankyard. The traditional brined and the environmentally brined cucumbers changed from a fresh to a fermented cucumber during the first 30 days within the tank (Figure 2.3). A decrease in texture for both NaCl and $CaCl_2$ size cucumbers that were fermented has been seen since they were first fermented after one month ($p < 0.0005$). However, brining treatment and time in the tankyard had a significant effect on the overall mesocarp texture in the fermented cucumbers

in bulk storage (p =0.0099) (Figure 2.3). The average firmness of size 2B cucumbers (31-38mm) that were brined in 6% NaCl was 11.9 ± 0.1 N compared to a firmness of 10.1 ± 0.1 N in 2B cucumbers brined in 1.1% $CaCl_2$. The average firmness of size 3A cucumbers (38-44.5mm) that were brined in 6% NaCl was 10.0 ± 0.1 N compared to a firmness of 8.8 ± 0.1 N in 2B cucumbers brined in 1.1% $CaCl_2$. The fermented cucumbers, with a large diameter (38-44.5mm), had a significant effect on overall texture in the final product (p=0.0152). The fermented cucumbers that were a size 2B (31-38mm diameter) had an overall instrumental firmness value of 11.0 ± 0.1 N compared to a firmness of 9.4 ± 0.1 N in cucumbers that are sized 3A's (38-44.5mm diameter). An interesting observation noted from Figure 2.3, is that the instrumental firmness values of the $CaCl_2$ fermented HDC size 2B cucumbers was similar to that of traditionally fermented cucumbers with a larger diameter, indicating that the environmentally friendly process could result in a texture similar to larger diameter traditionally fermented cucumbers.

The effects of time, brining treatment, and cucumber size which had an impact on instrumental mesocarp firmness of the fermented cucumbers, was also observed in the exocarp tissue while in bulk storage for 9 months in the tankyard (p =0.0391) (Figure 2.4). Brining treatment and size in the tankyard had a significant effect on the overall exocarp firmness in the fermented cucumbers in bulk storage (p =0.04) (Figure 2.8). The average firmness of size 2B cucumbers (31-38mm) that were brined in 6% NaCl was 24.5 ± 0.1 N compared to a firmness of 20.1 ± 0.1 N in 2B cucumbers brined in 1.1% $CaCl_2$. The average firmness of size 3A cucumbers (38-44.5mm) that were brined in 6% NaCl was 21.5 ± 0.1 N compared to a firmness of 19.3 ± 0.1 N in 2B cucumbers brined in 1.1% $CaCl_2$. The diameter

of the fermented cucumbers also had a significant effect on overall texture in the final product (p=0.0004). The fermented cucumbers that were a size 2B (31-38mm diameter) had an overall instrumental firmness value of 22.3± 0.1 N compared to a firmness of 20.4 ± 0.1 N in cucumbers that were sized 3A's (38-44.5mm diameter).

This is the first report of texture evaluation of commercial scale fermented cucumbers during long term storage in the tankyard. It has been shown that structural characteristics, such as skin thickness and tissue texture, are other factors that influence the texture of pickles (Lu et al., 2002). It has been researched in larger size diameter cucumbers, texture is often difficult to retain (Hudson and Buescher, 1980). When large sized pickles are sliced, the locular tissues separate from the pericarp causing softening within the tissue (Hudson and Buescher, 1980). Most pickle products use cucumbers that are between 2.7 and 4.5 cm in diameter to ensure a quality product. Since the fermented cucumbers are stored as whole cucumbers, the texture deterioration was more apparent and significant after the cucumbers were sliced, so they could be evaluated by instrumental analysis. Mesocarp and exocarp measurements were obtained before desalting, and not taking into account the use of other processing aids such as calcium and alum; that are mainly used to increase the texture properties in the finished processed product.

Similar trends were seen in the compression data of the fermented cucumbers during bulk storage. Fracture stress was significantly affected by the storage time in the tankyard (p <0.0001). During the 9 months of bulk storage, fracture stress decreased regardless of brining and size of the cucumber (Figure 2.5). While fracture stress decreased due to storage time, fracture strain was influence by both storage time (p =0.0029), and diameter size of the

cucumber ($p =$0.0203) (Figure 2.6). The initial higher fracture stress and strain values, for the

traditional fermented cucumbers, are a result of the binding of sodium ions within the tissues

of the fermented cucumber. However, the $CaCl_2$ brined cucumbers maintained their stress

and strain values during bulk storage compared to higher initial values in the NaCl traditional

fermented cucumbers. This could be due to the extra bonds, maintained within the cell wall,

by the calcium ion to prevent the fracture from occurring. Loss of crispness in fermented

cucumbers as a result of brining treatment was reported by Buescher et al., (2011). Higher

residual calcium in the fermented product prevented the tissue from becoming soft,

due to storage or natural enzymes that are found in the fruit. Calcium is a divalent cation and

prevents the plant tissues from softening by binding to other sites besides pectin and

carboxylic groups (McFeeters and Fleming, 1989). Sodium chloride is a natural preservative

that not only allows for the fermentation to occur in pickles, but also prevents softening from

occurring in the cucumber tissue. This was apparent for the first several months of storage,

however as the traditional and environmentally friendly fermented cucumbers are stored for

an extended period of time, the overall firmness in the fermented cucumbers decreased.

*2.4.2 Evaluation of instrumental firmness in processed pickle*s

Fermented cucumbers that are made into finished products typically have a one to

two-year shelf life (Communication with Mount Olive Pickle Company Representative,

2013). Based on the texture data collected from the sampling time points, no significant

changes in texture occurred during shelf storage for 18 months at an ambient temperature

(Figure 2.7). However, fermentation brining treatment appeared to have a significant effect

on the overall texture in the final processed product ($p =$0.0007) (Figure 2.7). The average

firmness of cucumbers that were brined in 6% NaCl was 8.4 ± 0.1 N compared to a firmness

of 7.8 ± 0.1 N in cucumbers brined in 1.1% $CaCl_2$. Texture changes in finished fermented

cucumber products, as a result of brining treatment, was first reported by Buescher et al.,

(2011). Higher residual calcium in the fermented product, during bulk storage, kept the tissue

from becoming soft, due to the natural enzymes that are found present in the fruit. It would

be beneficial for cucumbers to be fermented and processed into products using this

environmentally friendly brining treatment.

The 1.1% calcium chloride brine treatment was investigated to determine if the

texture could be achieved in the final product by doing a 2-desalting step to remove the

excess calcium. A second desalting step would result in a residual calcium level of 21mM in

both the traditionally fermented as well as the environmentally friendly process. Targeting a

residual calcium level of 21mM would substantially reduce the amount of chlorides entering

the waste streams when dumping the treated brine (McFeeters et al., 2010). Despite having a

lower firmness than cucumbers fermented in traditional brine, the environmentally friendly

produced hamburger dill chip products did not have significant loss in firmness during shelf

storage for 18 months ($P > 0.05$) (Figure 2.7). Storage time in the tankyard before being

processed appeared to have a significant effect on firmness of the finished product (p

$=0.0002$). The fermented cucumbers that were stored for 2 months had a higher overall

firmness compared to those stored in the tankyard for 8 months before being made into

finished products. Another interesting thing to point out from Figure 2.7 is that the texture of

$CaCl_2$ fermented hamburger dill chips (HDC) stored in the tankyard for 2 months, was

similar to that of traditionally fermented HDC products that were stored in the tankyard for 8

months before being processed, indicating that the environmentally friendly process can be utilized, but may require a shorter bulk storage time.

2.4.3 Evaluation of sensory texture attributes in processed pickles

Hamburger dill chips that were made from both brine treatments were evaluated for sensory texture attributes 18 months after storage. Similar to the instrumental data, brining treatment and storage in the tankyard had a significant effect on the sensory attributes determined by the panel. Hardness, crunchiness, crispness, and fracturability had significantly lower sensory scores in the hamburger dill chips made from $CaCl_2$-brined cucumbers than NaCl-brined cucumbers (Figure 2.8). The cucumbers that were brined and fermented with NaCl had a crunchiness score one point higher on the Spectrum™ scale than those brined and fermented with $CaCl_2$. Skin awareness, seed awareness, and juiciness were not significantly different between treatments ($p > 0.05$). Storage time in the tankyard before being processed appeared to have a significant effect on sensory attributes of hardness, crispness, crunchiness, and fracturability ($p < 0.0001$). The fermented cucumbers that were held in bulk storage for 2 months had a positive 1-point higher score in hardness, crispness and crunchiness compared to those stored in the tankyard for 8 months before being processed into finished products (Table 2.2). While a descriptive panel does not provide information on a like/dislike scale, the fact that there was a clear difference in the texture attributes of pickles from the traditionally or environmentally friendly processed fermented cucumbers, suggests that future work should be focused on improving the texture during long-term bulk storage.

2.4.4 Correlation between instrumental and sensory analysis in commercially processed pickles

        To determine the best method to evaluate texture in fermented cucumbers, instrumental and sensory correlation was conducted to understand the relationship. Figures 2.9 and 2.10 show the correlations made between the instrumental and sensory analysis performed on the commercial fermented cucumbers stored for 18 months. Overall, crispness correlation had an R^2 value of 0.85 while crunchiness had an R^2 value of 0.80 with instrumentally measured firmness of the mesocarp tissue. Fracturability and hardness were less correlated with instrumental measurements with R^2 values of 0.59 and 0.70, respectively. Ultimately, sensory methods are the only way to truly quantify the sensory texture attributes in fermented cucumbers. However, a strong correlation with an instrumental measurement gives some capability to predict sensory attributes with some degree of confidence (Szczesniak, 1987). Having strong correlations between sensory and instrumental measurements performed on the fermented cucumbers, indicates that the punch method may be a good instrumental test to monitor changes in texture that impact these sensory textures attributes (Szczesniak, 1987). This study is the first examination of sensory and instrumental texture properties of $CaCl_2$ brined fermented cucumbers during bulk storage in a commercial environment.

Conclusion 2.5

        Cucumbers fermented in 1.1% calcium chloride brine, maintained an overall instrumental firmness similar to cucumbers traditionally fermented in NaCl. Cucumbers brined, fermented and stored in 6% NaCl had overall higher instrumental firmness values

than those brined in a 1.1% $CaCl_2$. A storage period of 2 months in the tankyard had an overall higher firmness retention in the finished product regardless of brining treatment when compared to products stored in the tankyard for 8 months before being processed. Calcium chloride brined cucumbers, which were stored for 2 months, had an overall instrumental firmness value similar to those that were traditionally fermented cucumbers stored for 8 months before being processed into products. Hardness, crispness, crunchiness, and fracturability texture attributes were significantly different between the two brining treatments. The sensory scores were all significantly higher in the traditional NaCl brined hamburger dill chips when compared to $CaCl_2$ brined products. A strong correlation between instrumental punch data with crispness and crunchiness texture attributes were found for both fermented cucumber treatments (R^2 = 0.85 and 0.80 respectively).

Calcium chloride fermentations could be performed and would have similar texture to NaCl fermented cucumbers if they were not stored in the tankyard for long storage periods. The mesocarp and exocarp measurements of NaCl fermented cucumbers during bulk storage had overall higher instrumental firmness value than those brined in a 1.1% $CaCl_2$ fermented cucumbers. The size of cucumbers used in making fermented cucumbers also had an effect on the overall texture during bulk storage. Using a smaller diameter cucumber (2B, 31-38mm) with CaCl2 brine, could achieve a similar texture to the traditional brined size 3A (38-44.5mm) fermented cucumbers. The decrease in instrumental firmness for the $CaCl_2$ fermented cucumbers began after two months of bulk storage in all the texture measurements performed on the fermented cucumbers. This research demonstrates that commercial processors can produce pickles that will retain firmness during fermentation using an

environmentally-friendly CaCl$_2$ fermentation method, but should process them within 2 months' time or use size 2B cucumbers to ferment cucumbers in order to ensure the highest quality product for consumers.

References

Abbott, J. A. (1999). Quality measurement of fruits and vegetables. *Postharvest Biology and Technology, 15*(3), 207-225.

Bell, T. A., Etchells, J. L., & Jones, I. D. (1951). Pectinesterase in the cucumber. Arch Biochem Biophys, 31(3), 431-441.

Bell, T. A., Etchells, J. L., & Jones, I. D. (1955). A method for testing cucumber salt-stock brine for softening activity. USDA-ARS, 72(5), 15.

Breidt Jr., F., McFeeters, R. F., & Díaz-Muñiz, I. (2007). Fermented vegetables. In M. P. Doyle, & L. R. Beuchat (Eds.), *In food microbiology: Fundamentals and frontiers* (3rd ed., pp. 783-793). Washington, D.C: ASM Press.

Breidt, F. (2006). Safety of minimally processed, acidified, and fermented vegetable products. In G. Sapers, J. Gorny & A. Yousef (Eds.), *Microbiology of fruits and vegetables* (pp. 313-335). Boca Raton, FL: CRC Press, Inc.

Buescher, R. W., & Burgin, C. (1988). Effect of calcium chloride and alum on fermentation, desalting, and firmness retention of cucumber pickles. *Journal of Food Science, 53*(1), 296-297.

Buescher, R. W., Hamilton, C., Thorne, J., & Jin Cho, M. (2011). Elevated calcium chloride in cucumber fermentation brine prolongs pickle product crispness. *Journal of Food Quality, 34*, 93-99.

Buescher, R. W., & Hudson, J. M. (1986). Bound cations in cucumber pickle mesocarp tissue as affected by brining and CaCl2. *Journal of Food Science*, 51(1), 135-137.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1979). Inhibition of polygalacturonase softening of cucumber pickles by calcium chloride. *Journal of Food Science*, 44, 1786-1787.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1981). Utilization of calcium to reduce pectinolytic softening of cucumber pickles in low salt conditions. *Lebensm. Wiss. Technology*, 14, 65-69.

Buescher, R. W., & Hudson, M. (1984). Softening of cucumber pickles by cx-cellulase and its prevention by calcium. *Journal of Food Science*, 49, 954-955.

Chauvin, M. A., Younce, F., Ross, C., & Swanson, B. (2008). Standard scales for crispness, crackliness and crunchiness in dry and wet foods: Relationship with acoustical determinations. *Journal of Texture Studies*, 39, 345-368.

EPA. (1987). Water quality criteria; availability of document Fed. reg. 52: 37655- 37656. Washington D.C: *Environ. Prot. Agency*.

Fleming, H. P. 1. (1982). Fermented vegetables. In Rose AH (Ed.), *Economic microbiology. fermented foods*, vol. 7 (chapter 7) (pp. 227-258). New York , NY: Academic Press, Inc.

Fleming, H. P., & McFeeters, R. F. (1981). Shelf life of fresh-pack cucumber pickles. St. Charles, IL: Pickle Packers International, Inc.

Fleming, H. P., McFeeters, R. F., & Daeschel, M. A. (1992). Fermented and acidified vegetables. In S. D. Vanderzant C (Ed.), *Compendium of methods for the microbiological examination of foods* (3rd ed., pp. 929-952). Washington, D.C: American Public Health Association.

Fleming, H. P., McFeeters, R. F., & Thompson, R. L. (1987). Effects of sodium chloride concentration on firmness retention of cucumbers fermented and stored with calcium chloride. *Journal of Food Science*, 52(3), 653-657.

Fleming, H. P., Thompson, R. L., Bell, T. A., & Hontz, L. H. (1978). Controlled fermentation of sliced cucumbers. *Journal of Food Science*, 43(3), 888-891.

Hamann, D. D., Zhang, J., Daubert, C. R., Foegeding, E. A., & Diehl, K. C. (2006). Analysis of compression, tension and torsion for testing food gel fracture properties. *Journal of Texture Studies*, 37(6), 620-639.

Howard, L. R., & Buescher, R. W. (1990). Cell wall characteristics and firmness of fresh pack cucumber pickles affected by pasteurization and calcium chloride. *Journal of Food Biochemistry*, 14, 31-43.

Hudson, J. M., & Buescher, R. W. (1980). Prevention of soft center development in large whole cucumber pickles by calcium. *Journal of Food Science*, 45, 1450-1451.

Hudson, J. M., & Buescher, R. W. (1985). Pectic substances and firmness of cucumber pickles as influenced by CaCl2, NaCl and brine storage. *Journal of Food Biochemistry*, 9, 211-229.

Hutkins, R. W. (2006). Fermented foods in human history. In R. W. E. Hutkins (Ed.), *Microbiology and technology of fermented foods* (pp. 3-14-246-249.). Ames, Iowa: Blackwell Publishing.

Jones, I. D., Etchells, J. L., & Monroe, R. J. (1954). Varietal differences in cucumbers for pickling. *Food Technology*, 8(9), 415-418.

Jones, I. D., & Etchells, J., L. (1950). Cucumber varieties in pickle manufacture. *Canner*, 110(1), 34, 36, 38, 40.

Lawless, H. T., & Heymann, H. I. (1998). *Sensory evaluation of food: Principles and practices* (2nd ed.). New York, New York: Kluwer Academic/Plenum Publisher.

Lu, Z., Fleming, H. P., & McFeeters, R. F. (2002). Effects of fruit size on fresh cucumber composition and the chemical and physical consequences of fermentation. *Journal of Food Science*, 67(8), 2934-2939.

Magness, J. R., & Taylor, G. F. (1925). An improved type of pressure tester for the determination of fruit maturity. *U.S Department Agriculture Circulation*, 350.

Maruvada, R., & McFeeters, R. F. (2009). Evaluation of enzymatic and non-enzymatic softening in low salt cucumber fermentations. *International Journal of Food Science and Technology, 44*, 1108-1117.

McFeeters, R. F., & Fleming, H. P. (1991). pH effect on calcium inhibition of softening of cucumber mesocarp tissue. *Journal of Food Science*, 56(3), 730-732.

McFeeters, R. F., Fleming, H. P., & Thompson, R. L. (1985). Pectinesterase activity, pectin methylation, and texture changes during storage of blanched cucumber slices. *Journal of Food Science*, 50, 201-205.

McFeeters, R. F., Senter, M. M., & Fleming, H. P. (1989). Softening effects of monovalent cations in acidified cucumber mesocarp tissue. *Journal of Food Science*, 54(2), 366-370.

McFeeters, R. F. (1986). Pectin methylation changes and calcium ion effects on the texture of fresh, fermented, and acidified cucumbers. *American Chemical Society*, 217-229.

McFeeters, R. F., & Perez-Diaz, I. (2010). Fermentation of cucumbers brined with calcium chloride instead of sodium chloride. *Journal of Food Science*, 75(3), 291-296.

Meilgaard, M. C., Civille, G. V., & Carr, B. T. (2007*). Sensory evaluation techniques* (4th Edition ed.). Boca Raton, Florida: CRC Press.

Palnitkar, M. P., & McFeeters, R. F. (1975). Recycling spent brines in cucumber fermentations. *Journal of Food Science*, 40, 1311-1315.

Perez-Diaz, I. M., & McFeeters, R. F. (2011). Preparation of a lactobacillus plantarum starter culture for cucumber fermentations that can meet kosher guidelines. *Journal of Food Science*, 76(2), M120-M123.

Szczesniak, A. S. (1987). Correlating sensory with instrumental texture measurements-an overview of recent developments. *Journal of Texture Studies*, 18, 1-15.

Tang, H. C. L., & McFeeters, R. F. (1983). Relationships among cell wall constituents, calcium and texture during cucumber fermentation and storage. *Journal of Food Science*, 48(1), 66-70.

Thompson, R. L., Fleming, H. P., Hamann, D. D., & Monroe, R. J. (1982). Method for determination of firmness in cucumber slices. *Journal of Texture Studies, 13*, 311-324.

Thompson, R. L., Fleming, H. P., & Monroe, R. J. (1979). Effects of storage conditions on firmness of brined cucumbers. *Journal of Food Science*, 44, 843-846.

Truong, V. D., & Daubert, C. R. (2000). Comparative study of large strain methods for assessing failure characteristics of selected food gels. *Journal of Texture Studies*, 31(3), 335-353.

Yoshioka, Y., Horie, H., Sugiyama, M., & Sakata, Y. (2009). Quantifying cucumber fruit crispness by mechanical measurement. *Journal of Breeding Science, 59*(139-147)

Figures

Figure 2.1 Instrumental measurements to evaluate texture during bulk storage of cucumbers fermented with 6% NaCl vs. 1.1% CaCl$_2$. 2A = 27mm-31mm diameter, 2B = 31-38mm diameter; 3A = 38-44.5mm diameter, 3B = 44.5mm-51mm diameter.
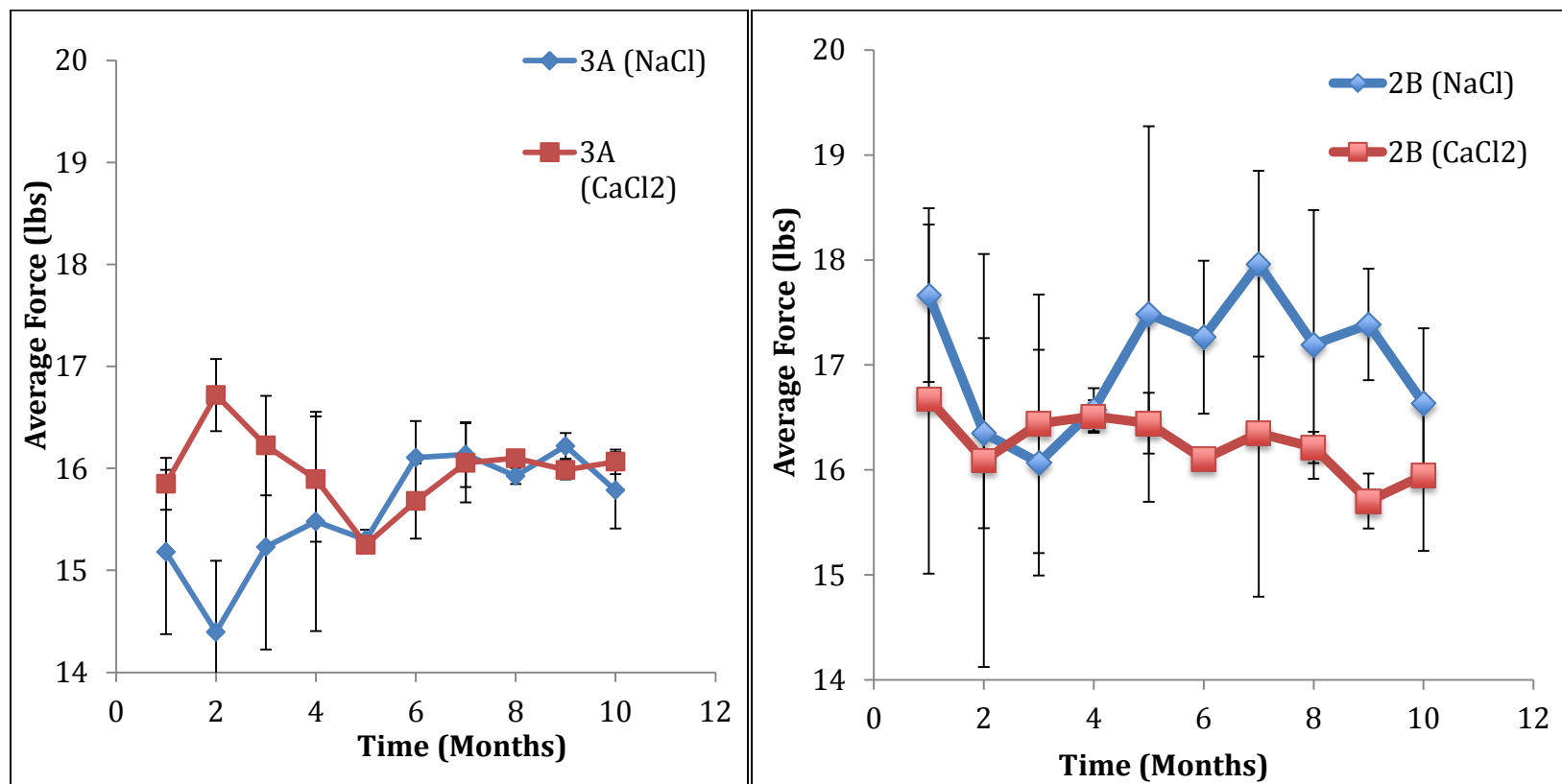
Figure 2.2 Whole fruit firmness of fermented cucumbers during bulk storage determined using a Fruit Pressure Tester. Brined and fermented in 6% NaCl or 1.1% $CaCl_2$. 2A = 27mm-31mm diameter, 2B = 31-38mm diameter; 3A = 38-44.5mm diameter, 3B = 44.5mm-51mm diameter.
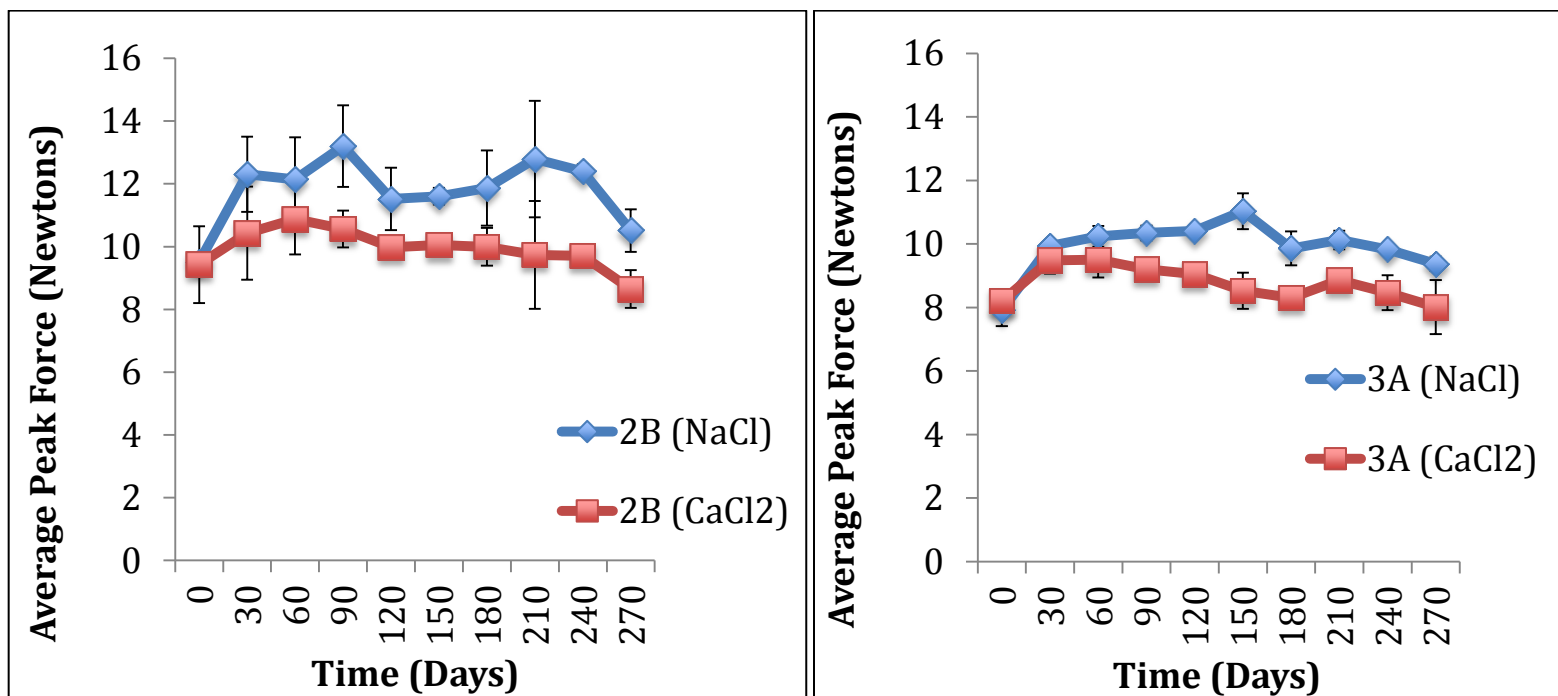
Figure 2.3 Mesocarp firmness of fermented cucumbers during bulk storage. Brined and fermented in 6% NaCl or 1.1% $CaCl_2$. 2A = 27mm-31mm diameter, 2B = 31-38mm diameter; 3A = 38-44.5mm diameter, 3B = 44.5mm-51mm diameter.
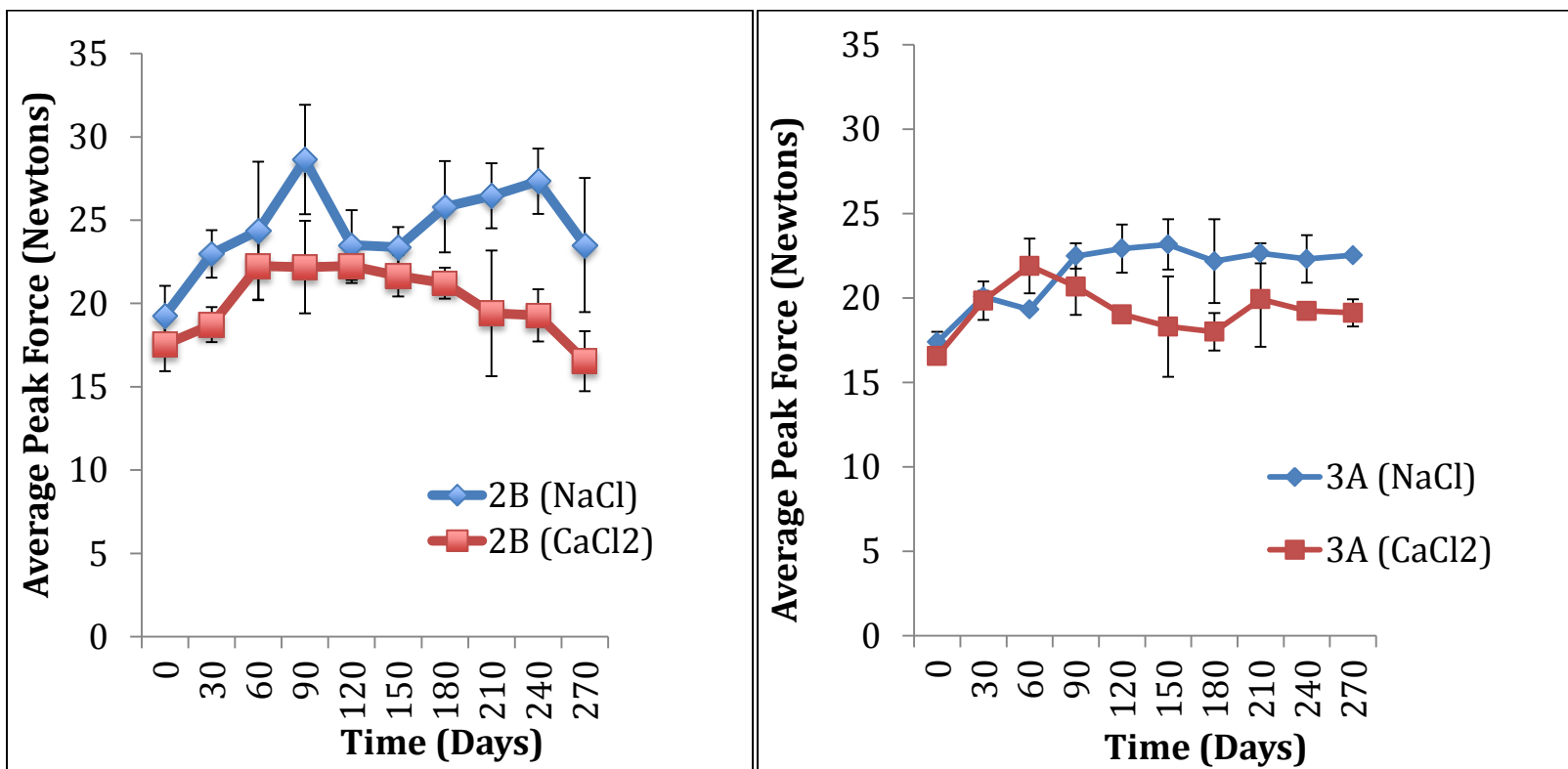
Figure 2.4 Exocarp firmness of fermented cucumbers during bulk storage. Brined and fermented in 6% NaCl or 1.1% CaCl$_2$. 2A = 27mm-31mm diameter, 2B = 31-38mm diameter; 3A = 38-44.5mm diameter, 3B = 44.5mm-51mm diameter.
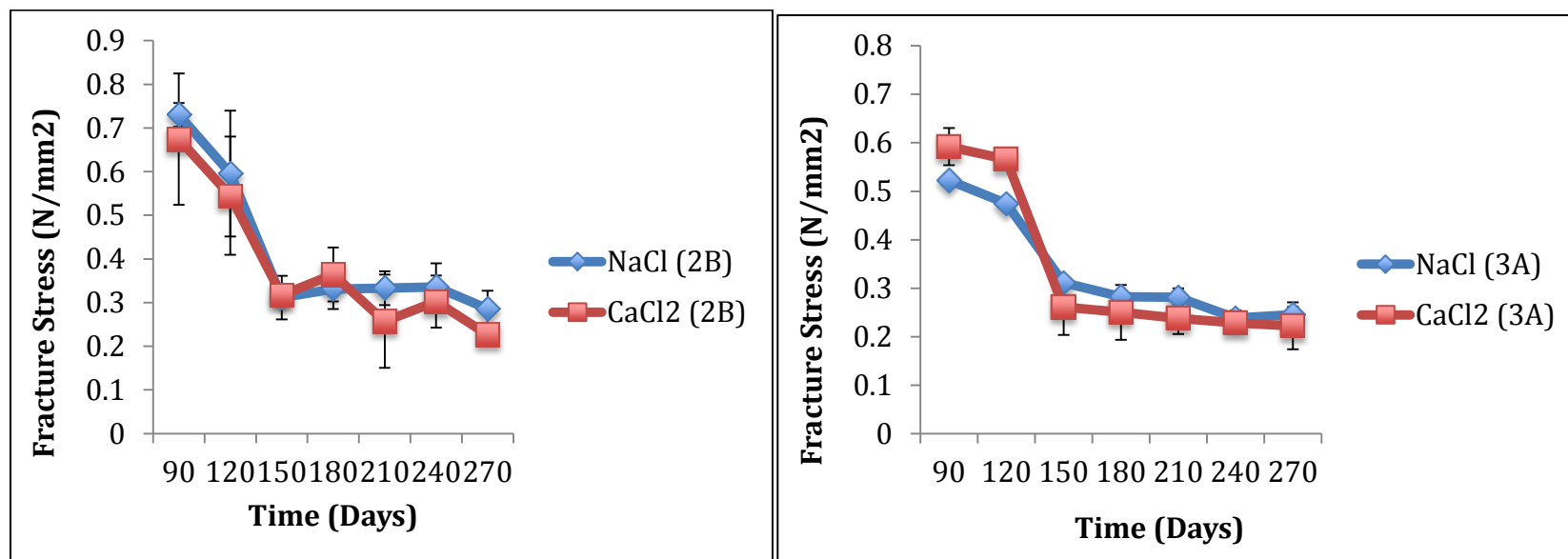
Figure 2.5 Fracture stress of fermented cucumbers during bulk storage. Brined and fermented in 6% NaCl or 1.1% CaCl$_2$. 2A = 27mm-31mm diameter, 2B = 31-38mm diameter; 3A = 38-44.5mm diameter, 3B = 44.5mm-51mm diameter. N=Newtons = kg*(m/s^2) = F= m*a; kg = kilograms, m=meter, s=seconds, F= force, m=mass, a= area.
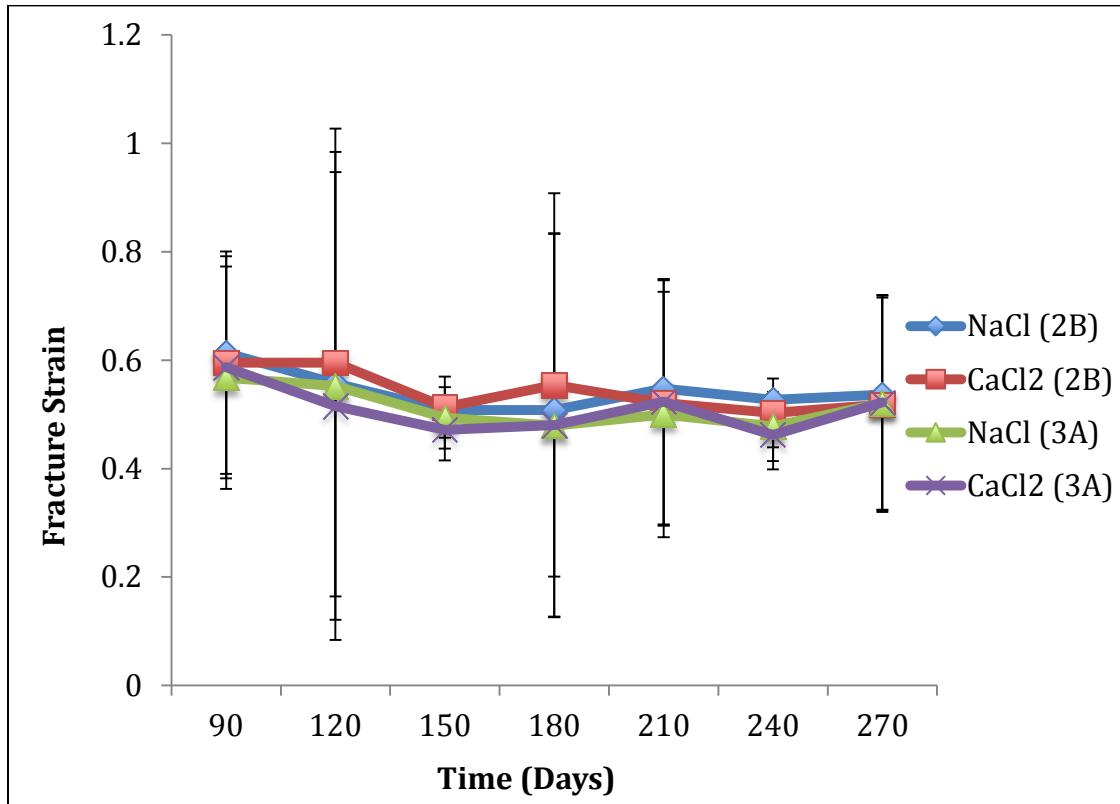
Figure 2.6 Fracture strain of fermented cucumbers during bulk storage. Brined and fermented in 6% NaCl or 1.1% $CaCl_2$. 2A = 27mm-31mm diameter, 2B = 31-38mm diameter; 3A = 38-44.5mm diameter, 3B = 44.5mm-51mm diameter.
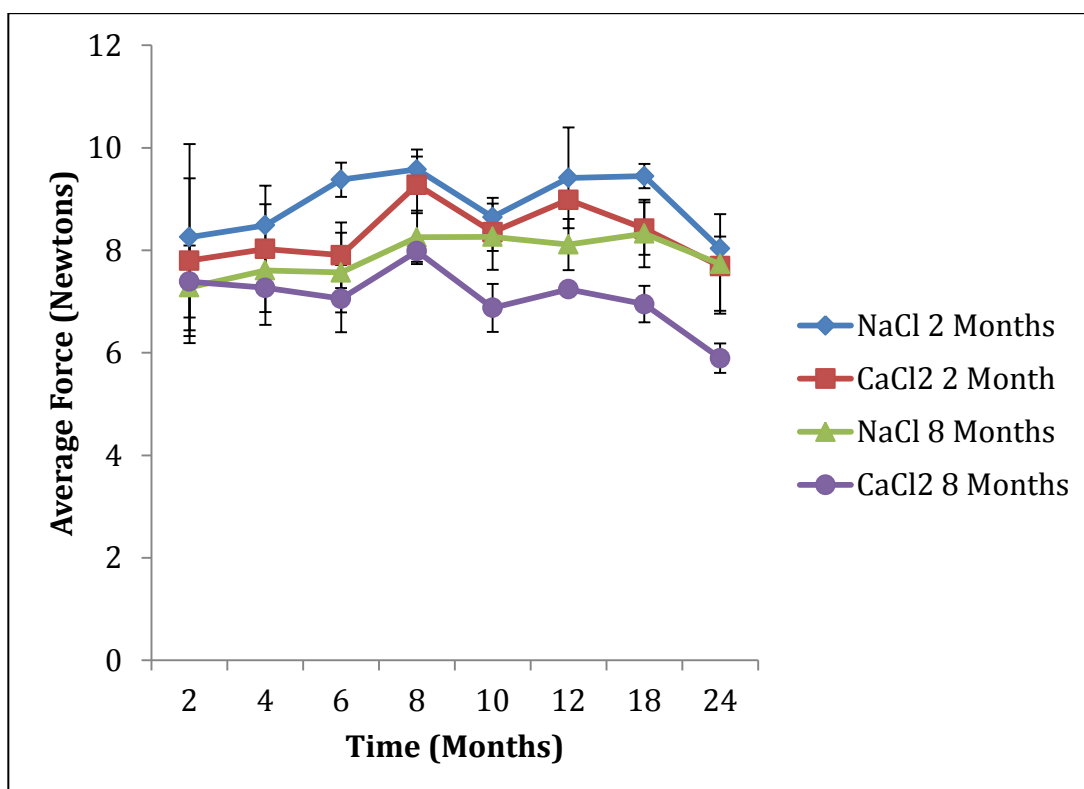
Figure 2.7 Mesocarp firmness in commercially processed hamburger dill chips processed from fermented cucumbers stored for 2 or 8 months before processing into finished products. Brined and fermented in 6% NaCl or 1.1% CaCl$_2$.
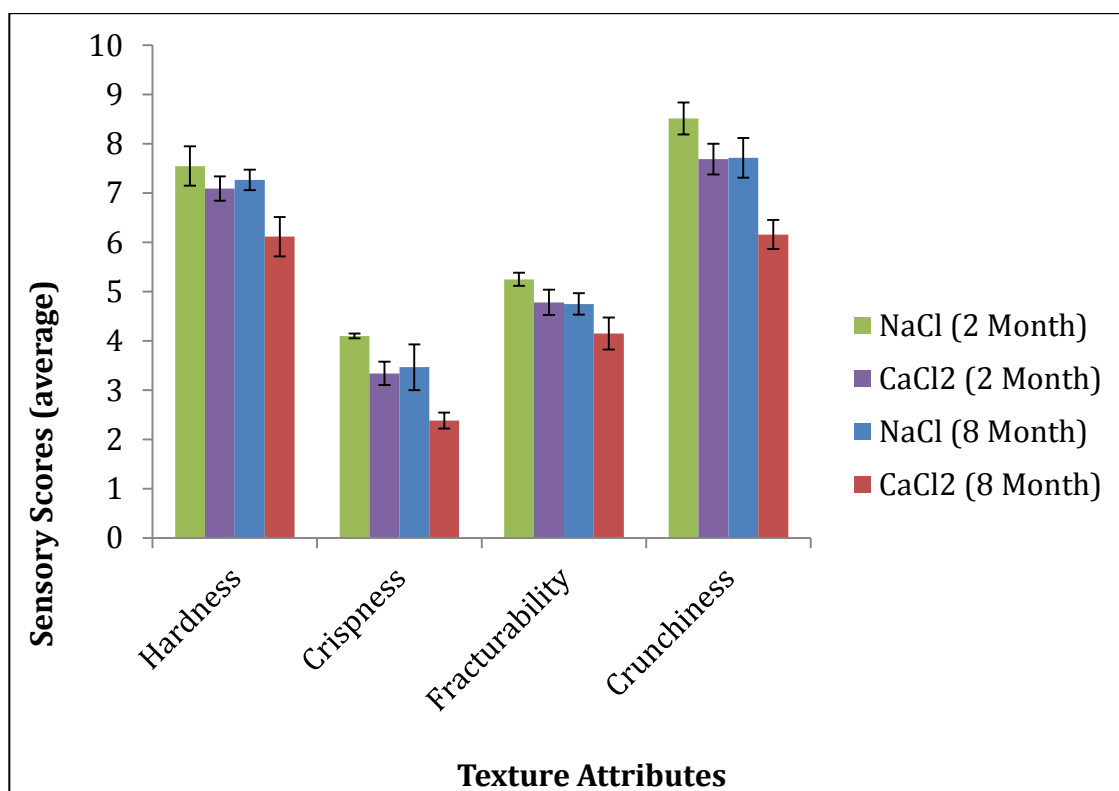
Figure 2.8 Sensory texture attributes of commercially processed hamburger dill chips stored for 2 or 8 months before processing into finished products. Brined and fermented in 6% NaCl or 1.1% CaCl$_2$.
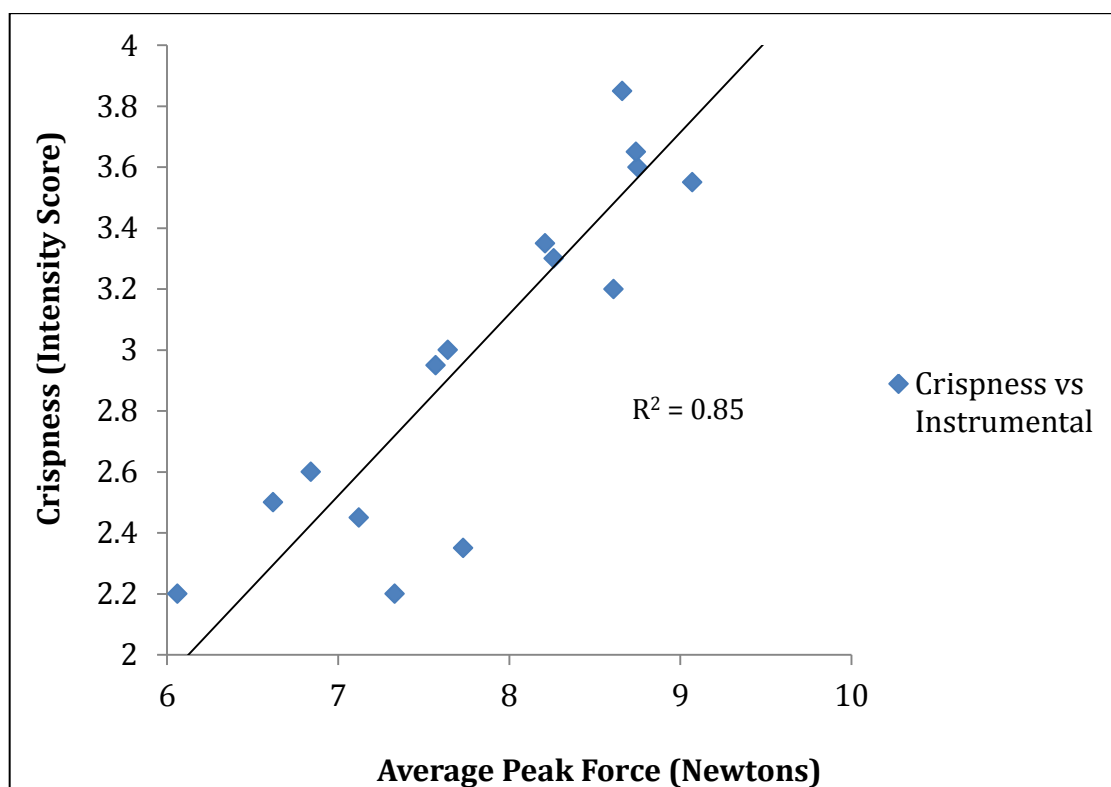
Figure 2.9 Correlation between sensory cripness scores and instrumental measurements of mesocarp firmness.
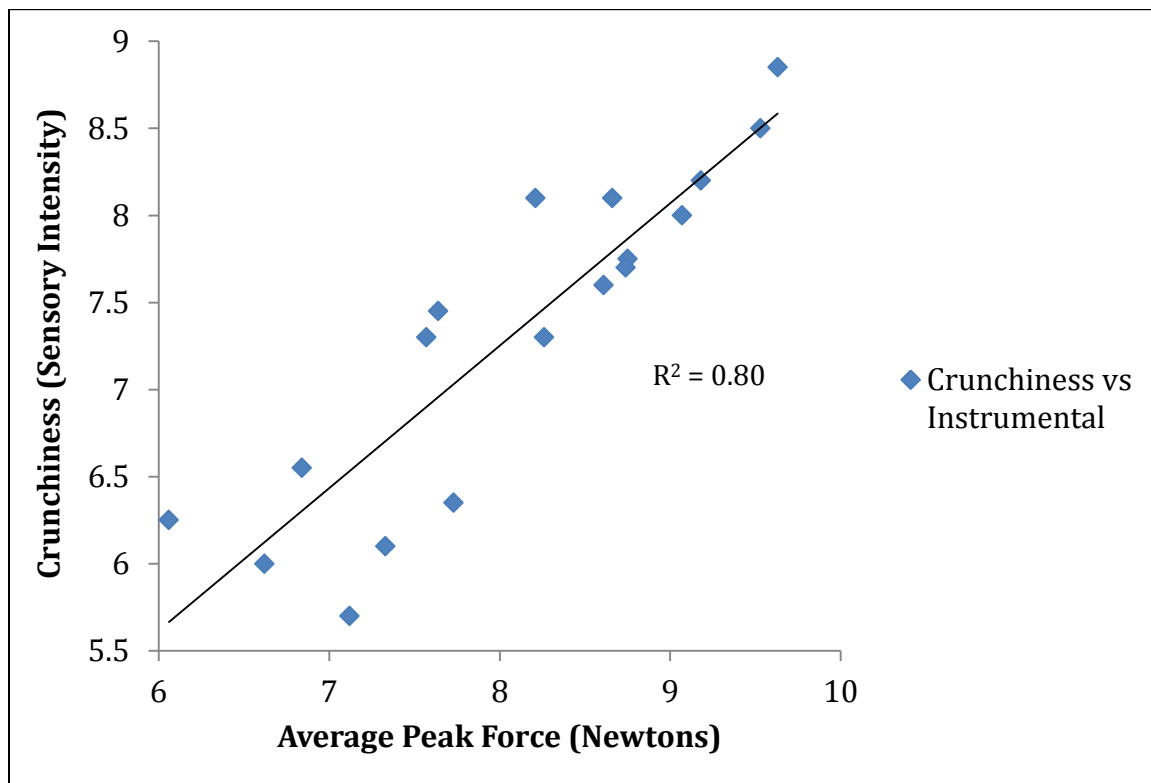
Figure 2.10 Correlation between sensory crunchiness scores and instrumental measurements of mesocarp firmness.

Tables

Table 2.1 Definitions and Techniques used for Descriptive Sensory Analysis

Attribute	Definition	Technique	Anchor(s) on Spectrum™ Scale
Hardness	Force necessary to compress the sample between the molars	Place sample between back molars and compress sample with molars.	Processed Cheese (1/2 in. cube) (4.5) Hebrew National Hotdog (7) Almond (11)
Fracturability	Force with which the sample ruptures when placing sample between molars and biting down completely down at a fast rate	Place sample between back molars and compress sample completely down at a fast rate.	Corn Muffin (1) Ginger Snap (8) Life Saver (14.5)
Crispness	The multiple, higher pitched sounds produced as the sample is crushed with the molar teeth – Evaluate on first 3 chews	Place sample between back molars and compress sample slowly. Repeat compression 3 times with the same sample.	Gala Apple, peeled (1/2in) (4) Water Chestnut (12)
Crunchiness	A single lower-toned noise produced with each chew. Evaluate intensity of crunchiness during the first 3 chews	Place sample between back molars and bite down quickly.	Peanut (4) Carrot (1/2in) (4)
Skin Awareness	The degree to which the outside skin of the product is perceived as intact pieces during mastication	Place sample between back molars and begin the mastication process.	Red Seedless Grape (10)
Seed Awareness	The degree to which the seeds within the product are perceived as intact pieces during mastication	Place sample between back molars and begin the mastication process.	½ cup Tomato Sauce with 1tsp. sesame seeds (4) Pomegranate Seeds (15)
Juiciness	The amount of juice/moisture perceived in the sample	Place sample between the back molar teeth and begin the mastication process.	Banana (1) Sugar Snap Peas (7) Orange Segment (15)

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Table 2.2 Sensory texture attributes of commercially processed hamburger dill chips stored for 2 or 8 months before processing into finished products. Brined and fermented in 6% NaCl or 1.1% $CaCl_2$.

Brining Treatment	TS (mo)	SS (mo)	Hardness	Crispness	Fracturability	Crunchiness
NaCl (6%)	2	18	$7.6^A \pm 0.2$	$4.1^A \pm 0.5$	$5.3^A \pm 0.2$	$8.5^A \pm 0.4$
CaCl₂ (1.1%)	2	18	$7.1^B \pm 0.4$	$3.3^B \pm 0.2$	$4.8^B \pm 0.3$	$7.7^B \pm 0.3$
NaCl (6%)	8	18	$7.3^B \pm 0.4$	$3.5^B \pm 0.1$	$4.8^B \pm 0.1$	$7.7^B \pm 0.3$
CaCl₂ (1.1%)	8	18	$6.1^C \pm 0.4$	$2.4^C \pm 0.4$	$4.2^C \pm 0.4$	$6.2^C \pm 0.4$
Reference Pickle	N/A	N/A	7.4 ± 0.4	3.1 ± 0.1	5.2 ± 0.3	7.8 ± 0.4

*TS= tankyard storage (months), SS= shelf storage (months)

Means in the same column not sharing a common super script are different ($p < 0.05$)

Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 2007).

Attributes not listed were not significantly different among treatments

Chapter 3

An Accelerated Shelf Life Model to Evaluate Texture

Changes in Fermented Cucumber Pickles

3.1 Abstract

Fermented cucumber texture, in conjunction with other sensory properties such as appearance and flavor, is an important factor in consumers' choice of pickles. The shelf life of fermented cucumber pickles depends on several factors including fermentation brining treatment, finished product composition, flavor profile, processing conditions and packaging, and storage temperature. Most pickle companies give their product an 18-30 month shelf life under normal shelf storage conditions. The impact of changes in process on this previously established shelf life is not easy for processors to test. An Accelerated Shelf Life Test (ASLT) was carried out on fermented cucumber pickles to predict the shelf life as a function of mesocarp firmness. Cucumbers were fermented at a commercial facility in 3,000-gallon, open-top tanks with 6% NaCl or 1.1% $CaCl_2$ brines, desalted, processed into hamburger dill chips (HDC) and stored at 25, 35, 45, or 55°C. Pickle mesocarp and exocarp firmness was measured using a TA-XT2 Texture Analyzer with a 3 mm diameter punch probe at a rate of 2.5 mm/sec. Maximum puncture force in Newtons (N) was recorded for 10-15 slices from 2 jars of each replicated fermentation treatment. Fracture stress and strain was measured by compression testing on mesocarp tissue cylinders that were 6mm in diameter by 6mm in height, under 80% deformation using a parallel plate at a rate of 2.5 mm/sec. The texture degradation rates were determined from the fitting of a first order kinetic model of time versus mesocarp firmness. Brining treatment had a significant effect on the overall initial firmness of hamburger dill chips ($p < 0.0001$). Higher levels of residual calcium in the final product, also had a significant effect on the texture degradation rates at all accelerated

storage temperatures ($p < 0.0001$). However, the rate of texture degradation for $CaCl_2$ fermented hamburger dill chips (HDC) that were desalted only once, was significantly less than that of traditional HDC at all storage temperatures. A maximum loss in firmness of 0.35 ± 0.9 N/day vs. 0.47 ± 0.2 N/day was found for $CaCl_2$ enhanced HDC vs. traditional HDC stored at 55°C. Sensory texture attributes of crispness and crunchiness were correlated to the instrumental firmness measured by puncture testing performed on the processed product stored under ambient and accelerated temperatures ($R^2 = 0.79$ and 0.80, respectively). The cutoff value for mesocarp firmness used to predict the end of shelf life in the ASLT model was 7 N based on the instrumental and sensory correlations and the ability of sensory panelists to detect a small change in texture properties. It was determined that HDC containing less than 30 mM of calcium chloride had a shelf life less than what is commonly advertised on the label, whereas the enhanced calcium chloride HDC had a shelf life of greater than 2½ years, when stored at ambient (25 ± 2 $^{\circ}$C) temperature. This research will enable pickle processors to predict the end of shelf life based on instrumental texture measurements that relate to a sensory perception of texture degradation in the product.

Introduction

Fermented cucumbers usually have a shelf life of 18-30 months due to their combination of salt, calcium, and acids, which results in a firm, crisp texture that can be retained for long periods of time (Fleming and McFeeters, 1981). The UK Institute of Food Science and Technology (IFST) defines shelf-life as "the period of time during which the food product will remain safe, retain it's desired sensory, chemical, and microbiological

characteristics, and comply with any label declaration of nutrition data" (IFST, 1993). Shelf life is a function of time, environmental factors and susceptibility of product to quality change (Labuza, 1982). Objective measurements, such as the chemical and physical changes in a food, can be used to determine end of shelf life (Labuza and Schmidl, 1988). These measurements are usually related to the microbial safety and labeling of a food product (Labuza and Schmidl, 1988). Due to the long shelf life of pickles, several different evaluation techniques, including instrumental and sensory analysis, were applied to understand texture degradation within the product at several storage temperatures. Accelerated Shelf Life Testing (ASLT) is a method used to determine the quality of a food product during storage. The end of shelf life is defined "as the time when the stored samples are perceived as different by a certain amount" (Labuza and Schmidl, 1985). The food industry has a great need to obtain information regarding shelf life of the product in a short period of time, in order to be competitive with other food companies (Mizrahi, 2000). Typically, shelf life storage studies are conducted in a controlled environment and sampled every couple of weeks. Accelerated shelf life storage studies use different variables such as temperature, time, and humidity in order to shorten the shelf life by several months, based on a kinetic model with the following steps (i) selection of the kinetically active factor, (ii) running of a kinetic study at a fast rate of deterioration, (iii) extrapolation of the kinetic parameters to normal storage conditions, and (iv) use of extrapolated kinetic data to calculate the shelf life under real storage conditions (Mizrahi, 2000).

Various methods have been used to understand the texture of fermented cucumbers and the effects of different components such as acids, sugars, and salts, on the finished

product. McFeeters and Fleming, (1989) determined that the addition of calcium to the fermentation brine, reduced softening in the cucumbers by binding to sites in the cucumber tissue other than pectin and carboxylic groups, and, therefore, had a positive effect on texture retention in fermented cucumbers. Howard et al., (1990) looked at the use of calcium chloride, in the brining of cucumbers, for improving texture in the finished product. McFeeters et al., (1991) studied the influence of pH, calcium, and temperature on the softening rates of fermented cucumbers, finding that these effects contributed to a first order kinetic model and allowed for a five-variable equation to be derived. McFeeters and Perez-Diaz (2010) discovered a way to replace 6% NaCl with 1.1% $CaCl_2$ in the fermentation brine, which could reduce the chloride concentration in the waste water by 80% and retain instrumentally measured mesocarp firmness. Garcia-Garcia et al., (2008) also used an accelerated shelf life approach to predict the end of shelf life for ripe olives using quality factors of color and firmness. They determined that the given shelf life of 1080 days was longer than that of the relative quality changes occurring in the product (Garcia-Garcia et al., 2008). However, no such information exists for fermented cucumbers as traditionally brined in sodium chloride, or for the new technology proposed that uses calcium chloride brines and starter culture for the fermentation and bulk storage of cucumbers.

The pickle industry usually gives their product a 1-2 year shelf life based on pH, color, taste, and texture (Communication with Mount Olive Representative, 2012). Texture characteristics of pickles are impacted with the amount of storage time on the shelves. Over time, pickles tend to lose their crisp and crunchy textural characteristics. Most pickle processors give a 2-year shelf to their products. However, most consumers typically purchase

pickles on a need basis; pickles are typically purchased and consumed every 53 days (Pickle

Packers International, 2013). By using ASLT, higher temperatures can be utilized to observe

texture degradation in a shorter period of time. A control at ambient temperatures is usually

performed along with the accelerated temperatures to monitor changes during normal

conditions. Using both ambient and accelerated temperatures will provide pickle processors

with an accurate ASLT model that will allow them to pull products off the shelves when the

products have deteriorated in texture quality. Creating pickles with desirable sensory

textures, that are also more environmentally friendly, requires an understanding of the

fermentation process, as well as the sensory terms that describe the products.

Hypothesis

Calcium chloride brined, fermented cucumber pickles will retain texture longer during

accelerated shelf life testing compared to traditionally fermented cucumber pickles, due to

higher residual calcium levels in the processed fermented cucumbers.

Objectives

1). Determine the effects of temperature and residual calcium on the texture degradation in

both traditional and environmentally friendly fermented cucumber processed pickles during

accelerated shelf life testing to predict the end of shelf life for these products.

2). Determine the correlations between the sensory texture characteristics and instrumental

measurements to determine the end point for the ASLT model.

Significance/Impact

      A fundamental understanding of how accelerated shelf life testing can measure

texture changes during storage is needed to determine end of shelf life for fermented

cucumber products for application in industrial product development cycles. Continuous changes in physical properties of foods during storage, makes the evaluation of texture a dynamic and complex process. In this study, brined and processed fermented cucumbers were created to characterize texture by mechanical and sensory properties. Understanding the reaction rates for texture changes in fermented cucumber pickles, using accelerated shelf life testing, will enhance pickle processors' knowledge of when end of shelf life occurs. Improved knowledge of the actual shelf life of these products will result in products for consumers that are firm, crisp, and desirable.

3.3 Materials and Methods

3.3.1 *Commercial scale fermentations using calcium chloride versus sodium chloride brines.*

Cucumbers and ingredients for pasteurized hamburger dill chip pickles. Fresh size 3A (38-44.5mm diameter) pickling cucumbers, salt, Vinegar 200-grain (20% acetic acid), anhydrous $CaCl_2$, yellow # 5 food color, polish dill flavor concentrate, benzoate with color, Antifoam C mixture, lids and glass were obtained from Mount Olive Pickle Company.

Fermentation of Cucumbers. Fresh size 3A (38-44.5mm diameter) cucumbers were fermented at a commercial facility in 3,000 gallon open–air tanks filled with 50-60% whole cucumbers and 50-40% cover brine solution to achieve a fermented product containing 6% sodium chloride (NaCl) or 1.1% calcium chloride ($CaCl_2$). Cucumbers were packed into fiberglass or plastic tanks and immediately covered with wooden boards to prevent the fruit from floating until equilibrium between the fruit and cover brine components was completed (Breidt, 2006; Breidt et al., 2007; Hutkins, 2006). The fermented cucumbers were stored in bulk in the tanks for a period of 4 or10 months before being processed into products. For the

calcium chloride fermentation to occur a starter culture and preservative were used. The fresh cucumbers and cover brine were inoculated with 10^5-10^6 cfu/ml *Lactobacillus plantarum* LA0445 that was obtained from the culture collection of the U.S. Dept. of Agriculture-Agricultural Research Service, Food Science Research Unit, located in Raleigh, N.C., U.S.A and prepared according to the koshering process described by Perez-Diaz and McFeeters (2011). Different preservatives such as sodium benzoate, fumaric acid, and potassium sorbate were added to the cover brine in varying concentrations in order to prevent spoilage from occurring after fermentation was complete. The preservative potassium sorbate was added to both NaCl and $CaCl_2$ tanks to prevent film yeasts from forming. Sodium benzoate was added as the preservative to $CaCl_2$ tanks in the second replication of the fermentation process.

Desalting of fermented cucumbers for commercial products. Once fermentation was complete and the fermented cucumbers were ready to be made into product, the fermented cucumbers were desalted at a 60:40 cucumber to water ratio in 5 gallon pails in order to desalt the product to a level that can be consumed by consumers. The 5-gallon buckets were rolled and inverted every 4-6 hours in order to ensure equilibrium was complete at ambient temperatures (25 ± 2 ^{o}C). Liquid samples were taken to perform calcium analysis to determine equilibrium between the cucumbers and water. A second desalting of the cucumbers was performed in order to achieve calcium levels equal to what is typically found in traditionally processed pickles. Once equilibrium was reached, the 5-gallon pails were stored in refrigeration until day of processing.

Packing and Pasteurization. The desalted, fermented cucumbers were sliced to a thickness of 6mm using a Hobart food processor (Hobart, Model PF 150, Troy, Ohio) and

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then packed in either 8oz or 16 oz. jars with a 58: 42 cucumber: cover liquor ratio. All of the treatments were packed out to have a residual calcium level of either 21mM or 36mM and 2% NaCl. Jars were heat sealed by placing the lids in boiling water (5-15 seconds) to heat the liner, ensuring a hermetic seal between the lid and the jar. The jars were sealed by hand before pasteurization. The jars from the first replication of accelerated shelf-life testing (ASLT) were pasteurized in a steam-jacketed kettle until they reached an internal temperature of 74°C at the center point, held at that temperature for 15 minutes and then cooled to room temperature steadily, using cold tap water. The pickles for the second replication of ASLT were pasteurized at Mount Olive Pickle Company for 50 minutes at a temperature range of 145-154°F in a tunnel pasteurizer.

3.3.2 Sensory Analysis of Fermented Cucumbers Products

Sensory Panelists. Students and staff (n=10) from North Carolina State University, (Raleigh, North Carolina) Department of Food Bioprocessing and Nutrition Sciences, as well as the USDA Food Science Research Unit, (Raleigh, North Carolina) were selected as panelists. The panel was composed of two males and eight females. Panelists were selected based upon regular consumption of fermented vegetables and the demonstrated ability to distinguish texture attributes in fermented cucumber products.

Sensory Training. Approval of the North Carolina State University (Raleigh, North Carolina) Institutional Review Board (IRB # 2734) was obtained for the use of human subjects in this study. Panelists were trained in descriptive sensory analysis using the Spectrum™ method (Meilgaard et al., 2007). Panelists were initially trained (75+ hours) to

use a 15-point scale to assess the texture attributes of hardness, crispness, crunchiness, fracturability, skin awareness, seed awareness, and juiciness as defined in Table 3.1. During the training, panelists were individually evaluated using food products of established intensities to determine that all panelists were able to scale the attributes of interest. A selection of food product with known values on the Spectrum™ method were used as reference samples for different textures during training.

Panelists were trained to rate hardness, fracturability, and juiciness based upon reference definitions and samples suggested by the Spectrum™ method (Meilgaard et al., 2007). Crunchiness and crispness attributes were based upon reference samples suggested by Chauvin (Chauvin et al., 2008) and modified in this study to develop scales, definitions, and evaluation techniques that facilitated panelists in distinguishing the two auditory attributes. Crispness was defined as the multiple, higher pitched sounds produced as the sample is crushed slowly with the molar teeth. Crunchiness was defined as a singular lower-toned noise produced with each chew. Crispness and crunchiness intensity scores were evaluated during the first 3 chews (Figure 3.2). Skin and seed awareness were defined by evaluating several food products by the panelists (Figure 3.2). Panelists were trained using various foods based upon recommendations for training of Meilgaard et al. (2007).

Following training, panelists' mean scores, for each texture attribute of a standard commercial hamburger dill chip pickle (HDC), were assigned a consensus score which served as a reference value of intensity of the Spectrum™ scale for commercial fermented pickles. Reference HDC were stored at 2ºC and provided as a reference sample for calibration of panelist ratings at each sensory analysis session during the study.

Sensory Analysis. Panelists were presented between 4-8 samples in each descriptive analysis (DA) session in 2oz cups identified with random 3 digit numbers. All samples were presented at room temperature along with distilled water, Muenster cheese cubes and salt-free saltine crackers to cleanse the palate between samples. A randomized order was created to present samples to each panelist, in order to avoid bias in the order of presentation. Each sample cup contained 2 slices**.** Each panelist was also given 2 sample cups of reference fermented hamburger dill chips that had been stored at 2 ^{o}C to maintain flavor and texture attributes during the course of the study. This allowed each panelist to calibrate texture attributes on the 15-point scale. Each panelist was instructed to first taste the reference sample, cleanse the palate before tasting the sample. They were given as much time as needed to taste their samples and a 2-minute rest between samples to avoid fatigue. Each panelist was provided with a spit cup if they preferred to expectorate their samples. Duplicate DA evaluations of each sample were done at each sampling time during the study. Sensory analysis was conducted on pickle samples made from fermented cucumbers that were brined in either $CaCl_2$ or NaCl. This was further broken down into different treatments containing residual calcium levels ranging between 21mM and 36mM (Figure 3.1). The effect of accelerated storage temperature on hamburger dill chips processed from $CaCl_2$ or NaCl fermented cucumbers were evaluated by performing sensory evaluations at the same time points as instrumental evaluation. The descriptive sensory analysis was performed at days 2, 4, 6 for 55°C accelerated fermented cucumbers, 14, 21, 35 days for 45°C, days, 28, 84, 140, 196 days for 35°C and days 56, 112, and 176 for 25°C. These days were selected to capture

the change in texture during accelerated shelf life testing without exhausting the panelists' palates.

3.3.3 Instrumental Analysis of Fermented Cucumber Products

Firmness Measurements. Firmness measurements were taken using a TA.XT2 Texture Analyzer (Texture Technologies Corp, Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) equipped with a 3 mm diameter puncture probe. Pickle slices (6 mm thick) were placed onto a base plate containing a 3.1 mm hole. The puncture probe moved at a test speed of 2.5 mm/sec into the cucumber slices. Data were collected and analyzed using Texture Expert software (Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK). The peak force required to punch through the cucumber mesocarp and exocarp was recorded and expressed in Newtons (N). Firmness measurements were done on 10-15 slices from each of the two jars from each treatment at each sampling time (Yoshioko et al., 2009; Thompson et al., 1982). Firmness was measured immediately after pasteurization was complete to have the initial firmness (time-point zero) before beginning the Accelerated Shelf Life Test. The remaining sample jars were placed into different accelerated temperature storage incubators. Texture was evaluated at accelerated temperature (55^oC) at the sampling times of 2, 4,6,8,10,12, and 14 days. Texture was evaluated at accelerated temperature (45^oC) at the sampling times of 7, 14, 21,28,35,42, and 49 days. Texture was evaluated at accelerated temperature (35^oC) at the sampling times of every 3-4 weeks for 7 months from the original packing date. Texture was evaluated at ambient temperature (25 ± 2^oC) at the sampling times of 2, 4, 6,8,10, and 12 months from the

original packing of the products in April 2012 and at the sampling times of 2, 4, 6 months from the original packing of the products in January 2013.

Compression Measurements. Uniaxial compression measurements were taken using a TA.XT2 Texture Analyzer (Texture Technologies Corp, Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) equipped with a load cell of 50 N and a parallel plate with a diameter of 50mm. Cylindrical specimens were extracted from the mesocarp section of the cucumber slice using a cork borer. The dimensions of the cylinders were 6mm in diameter and 6mm in height. A one-cycle compression was set for an 80% deformation at a crosshead speed of 2.5 mm/sec. There were two replications of the instrumental analysis conducted on two separate days. For each replication, 10-cored samples of the 8 different treatments were evaluated. All samples fractured during compression, so force and distance data were used to calculate stress and strain at fracture, as described below (Hamann et al., 2006; Truong and Daubert, 2000). Fracture stress and fracture strain were determined from the following equations:

$$\sigma = \frac{F}{A} \qquad \text{(Equation 3.1)}$$

Where σ = stress (N/mm^2); F= force (N); A is cross sectional area (mm^2) of samples before compression (A= $\pi r^2 h$) r= radius of mesocarp cylinder; h= height of mesocarp cylinder.

True Compressive Strain (ε_h) was calculated as:

$$\varepsilon_h = \ln(1 + \frac{\Delta L}{L_i}) \qquad \text{(Equation 3.2)}$$

ε = hencky strain; ΔL= change in height (mm) of sample after deformation and L_i is initial height (mm).

pH. Measurements of pH were done at ambient temperature with an Accumet AR25 pH meter equipped with a gel-filled combination pH electrode (Fisher Scientific, Pittsburgh, Pa., U.S.A.) that was calibrated with certified standards of pH 2.00, 4.00, and 7.00 (Fisher Scientific).

Chemical Analyses. The NaCl concentration of the brine was determined by titration with standardized concentration of 0.171N $AgNO_3$ using dichlorofluorescein as an indicator (Fleming and others, 2002). Calcium chloride concentration of the brines was determined by titration with EDTA using Hydroxy Napthol Blue as the indicator dye (Buescher and Burgin, 1988).

3.3.4 Accelerated Shelf Life Testing and Model Estimation

Accelerated Shelf Life Testing Design. To test the effect of accelerated temperature storage on texture degradation in fermented cucumber pickles, glass jars containing pickle chips were placed in different temperature incubators. Pickles were held at temperatures 35, 45, and 55°C in air incubators and at an ambient temperature (25 ±2°C) as control. Sampling was conducted according to Figure 3.1 in order to collect enough sampling time points to show first order kinetics. Replicate samples of each treatment were drawn and firmness and pH were analyzed. New unopened jars were used in the analyses at each time-point. From making adjustments of experimental data to different kinetic models, it was deduced that the changes in firmness followed an apparent first-order kinetic. The influence of temperature on

the reaction rate was used to determine shelf life from the following equation:

$$-\frac{dA}{dt}=k(A)^n$$ (Equation 3.3)

Where A = the quality factor measure in some units of amount, n = the reaction order, and k

= the rate constant, the slope of the plot of the appropriate reaction extent A vs. time t

(Labuza and Schmidl, 1985). Based on the rate constants determined at several different

temperatures, a plot was extrapolated of ln k vs. reciprocal absolute temperature ($^{o}K^{-1}$) to

predict the rate of the reaction at any other temperature (Labuza and Riboh, 1982).

Statistical analysis of sensory and instrumental data. All statistical computations

were analyzed using a general linear model from JMP (version 10 SAS® software, SAS

Institute, Cary, NC). Means and standard deviations were calculated for both sensory (n=10

panelists) and instrumental (n=10-15 pickle slices) data using JMP version 10.1. Analysis of

variance (ANOVA) was performed on mean values. When ANOVA indicated a significant

difference between treatments, Tukey tests were conducted for means separation at 0.05

significance levels. The relation between rheological measurements and sensory attributes

were examined with linear regression correlation coefficients.

 3.4 Results and Discussion

3.4.1 Texture of Fermented Cucumbers during ambient storage

Apart from the varying amounts of residual calcium found in the product, texture

remained constant through one year of ambient storage. The four treatments that were used in

the initial experiment were monitored for a year at an ambient temperature (25 ±2°C), in

order to understand the texture retention with different levels of residual calcium without undergoing accelerated shelf life testing studies (Figure 3.3). For all the samples that were evaluated, normal cucumber fermentation and processing was documented. It was observed that the NaCl brined, fermented cucumbers made into HDC with 36 mM residual calcium, and the $CaCl_2$ one desalted fermented cucumbers made into HDC with 36 mM residual calcium, maintained their firmness at 8.1±0.1N for 1 year of shelf storage. Comparison among the initial firmness values of the different treatments of fermented cucumbers showed that the products were not homogenous (Table 3.2). NaCl fermented cucumbers with additional $CaCl_2$ had the highest values with an initial average firmness of 10.52 ±0.9N, followed by $CaCl_2$ fermented cucumbers processed with a one-desalting step. The initial firmness values for traditional NaCl fermented HDC were extremely different (Table 3.2), compared to the $CaCl_2$ one-desalted fermented cucumber HDC initial firmness values that were lower but relatively consistent. This difference could be due to sampling at different times from the tankyard, or due to fresh cucumbers coming from different lots for the fermentation process. The use of 100 mM of calcium chloride in the fermentation brine resulted in a finished product containing 36 mM after being desalted on a 60:40 cucumber to water ratio. (McFeeters et al., 2010). This number is essential to target in order to be at or below the legal limits of calcium chloride added to vegetable products (21CFR184.1193). By using one-desalting step, the amount of chlorides in the tankyard would be reduced greatly, and the amount of water used to desalt the product would be equivalent to the current process.

Regardless of the temperature used to store the fermented cucumbers, brining treatment had a significant effect on firmness in the product in both the mesocarp and exocarp tissue ($p < 0.0001$). Based on the data (Figures 3.4-3.6), texture degradation was observed to be the slowest for the HDC pickles made from traditional NaCl fermented cucumbers with higher residual calcium, as well as HDC pickles made from $CaCl_2$ fermented cucumbers that were desalted only once. The experiment was designed to determine the rate of texture degradation at various accelerated temperatures, similar to McFeeters et al., (1995) experiment performed to determine the softening rates when looking at temperature, calcium, and pH as variables. This type of analysis was useful in determining the rate of texture deterioration at each temperature. McFeeters et al., (1989) observed softening in the tissues of fermented cucumbers with a high level of variability. This was seen regardless of whether calcium was added to the fermentation brine or there was a variation of temperature. The levels of residual calcium in the product maintained cucumber firmness, due to the binding of the calcium ions to the cell wall polysaccharides (Buescher and Hudson, 1986; McFeeters and Fleming, 1990). Most vegetable products typically contain less then 25mM calcium concentration due to the undesirable chalky flavor that may be detected with high levels of calcium.

These effects that were seen in the mesocarp and exocarp tissue were also seen in the compression data. Fracture stresses after pasteurization were influenced by the brining treatment of the product and time ($p < 0.0001$, Figure 3.7). As stated previously, the initial fracture stress was higher for both NaCl brined fermented cucumbers. Also in Figure 3.7, there was a clear difference of 0.2 N/mm^2 in initial fracture stress values, between the brining

treatment of NaCl and CaCl$_2$. While fracture stress changed as a function of brining treatment and time in accelerated shelf storage, fracture strain for all four treatments remained the same between the range of 0.4 and 0.51, with an average value of 0.46 (Figure 3.8). The levels of residual calcium in the cucumber structure were expected to have an effect on the ability to fracture under uniaxial compression, due to the binding of the calcium ions to the cell wall polysaccharides (Buescher and Hudson, 1986; McFeeters and Fleming, 1990).

The greatest extent of texture deterioration in the fermented cucumbers occurred at the highest accelerated temperatures. At all temperatures, the brining treatment had an effect on texture retention in the final product $p < 0.0001$. Firmness loss as a result of high temperature storage (>30ºC), in shelf stable pasteurized pickles, was first reported by Nicholas and Pflug (1960). Storage temperature has been reported to have the greatest effect on retention of firmness in brined cucumbers, particularly at temperatures above 26.6°C (McFeeters and Fleming, 1989; McFeeters and Fleming, 1990; Nicholas and Pflug, 1960).

3.4.2 Descriptive Sensory Analysis of Fermented Cucumbers during Accelerated Shelf Life Testing

In addition to differences in texture retention, most other sensory attributes were significantly different in fermented cucumber prepared in traditional NaCl brine, compared to pickles prepared from CaCl$_2$ fermented cucumbers (Table 3.3). Hamburger dill chips that were made from both brine treatments, and made into several final processed products with different residual calcium levels, were evaluated for sensory texture attributes during ASLT. For the initial sensory scores for the different treatments, there was a clear difference

between the treatments made from NaCl brined cucumbers and those fermented cucumbers that were brined with $CaCl_2$. The effect of the three storage temperatures ($35^{\circ}C$, $45^{\circ}C$, $55^{\circ}C$) was significant for most of the attributes ($p < 0.0001$). Hardness, crispness, fracturability, and crunchiness, attributes decreased as storage time increased. The fermented cucumbers that were brined and fermented with NaCl, had hardness, crispness, fracturability and crunchiness scores one to two points higher on the Spectrum™ scale than those brined and fermented with $CaCl_2$ (Tables 3.4 -3.7). Texture attribute differences were observed regardless of whether the final product was a traditional NaCl product or one that had a residual calcium level of 36 mM. Skin awareness, seed awareness, and juiciness were not significantly different between treatments and were similar to the reference pickle ($p > 0.05$). There were statistical differences in the brined cucumbers that were fermented in the same treatment style but had different residual calcium levels for the texture attributes of hardness and crunchiness ($p = 0.0419$ and $p = 0.0072$, respectively). Sensory scores for hardness, crispness, fracturability, and crunchiness were a wide range on the Spectrum™ scale for all the treatments. Temperature had a significant effect on the sensory scores perceived during the ASLT ($p < 0.0001$). While a descriptive panel does not provide information on a like/dislike scale, the fact that there was a clear difference in texture attributes made from either the traditional or environmentally friendly fermentation brine at the beginning of the accelerated shelf life testing, suggests that a future goal would be to prevent texture degradation during the bulk storage phase.

3.4.3 Correlation between instrumental and sensory analysis in commercially processed

pickles

Relating instrumental and sensory measurements is important, due to the fact that both are able to describe changes in food texture. Correlations could allow companies to solely use instrumental measurements due to the fact that the characteristics are parallel in sensory scores. Figures 3.9 and 3.10 show the correlations made between the instrumental and sensory analyses. Sensory crispness and crunchiness correlated ($R^2 = 0.79$ and 0.80) with instrumentally measured firmness of the mesocarp tissue for both ambient and accelerated temperatures. Sensory crispness, skin awareness, and crunchiness correlated ($R^2 = 0.62, 0.63$ and 0.63 respectively) with instrumentally measured firmness of the exocarp tissue. As the products went through accelerated shelf life testing, the sensory texture attributes were significantly different depending on the fermentation and bulk storage brining treatment ($p < 0.0001$). Based on the plots of instrumental to sensory scores, a firmness predictive value was determined at the point where the panel detected a change in the texture attributes of the products (Figure 3.9 and 3.10). Based on the instrumental and sensory correlations and the ability of sensory panelists to detect a small change in texture properties, the cutoff value for mesocarp firmness used to predict the end of shelf life in the ASLT model was 7 N.

Relating instrumental and sensory methods of texture measurement is essential for several reasons. The most important reasons to develop objective tests are to substitute for the tedious and time-consuming running of sensory tests (Szczesniak, 1987). Instrumental tests would have an advantage of greater speed, accuracy, and standardization, in order to

better assess quality in a food product. The second reason correlation is important is to better assess the ability to predict the perceived sensory texture in a product (Szczesniak, 1987). Instrumental analysis allows for unbiased results due to the elimination of panelists, and creates a common reference for researchers, industry, and consumers (Abbott, 1999). Strong correlations were observed between the mesocarp firmness using the puncture test and the sensory texture attributes. Crispness and crunchiness are two well-known texture terms that describes the texture of foods. Ultimately, sensory methods are the only way to truly quantify the sensory texture attributes in fermented cucumbers. However, a strong correlation with an instrumental measurement gives some capability to predict sensory attributes with some degree of confidence (Szczesniak, 1987). Having strong correlations between sensory and instrumental measurements performed on the fermented cucumbers, indicates that the punch method may be a good instrumental test to monitor changes in texture that impact these sensory textures attributes (Szczesniak, 1987). This study is the first examination of sensory and instrumental texture properties of CaCl2 brined fermented cucumbers during bulk storage in a commercial environment.

3.4.4 Model Estimation for Fermented Cucumbers

Based on the fact that mesocarp firmness average peak force values had a strong correlation to the sensory textural characteristics of the hamburger dill chip slices, these values were used to determine the end of shelf life. Mesocarp firmness values were plotted as a function of time according to temperature. A first-order kinetic model was applied since it resulted in a better fit to the data. The application of Equation 3.3 to firmness data from each

sample versus temperature (T) and time (t) allowed the rate of the reaction (k) to be determined. The equation and the k value for all the temperature used during the shelf life experiment are shown (Table 3.8 and Table 3.9). Based on the rate constants determined at several different accelerated temperatures (35°C, 45°C, 55°C), (Figures 3.11, 3.12), a plot was extrapolated of *ln* k vs. reciprocal absolute temperature ($^{o}K^{-1}$) to predict the rate of the reaction at ambient storage conditions (Lee et al., 2002, Labuza and Riboh, 1982). From this plot, an Arrhenius shelf life equation was determined using regression analysis so that the shelf life for other temperatures could be predicted (Figures 3.11, 3.12)

The values obtained in the accelerated experiment were used to calculate the shelf life as a function of the relative degradation of firmness. The instrumental mesocarp firmness of 7 Newtons was used as the end of shelf life. This number indicated how sensitive the panel was to determining significant changes within the product under ASLT conditions. Depending on the initial firmness values, a reduction in firmness of 15-30 % was required to reach the 7N cut off value. Shelf life, as assessed by the relative firmness degradation (k), was always longer for fermented cucumbers that contained a residual calcium level above 30mM and had an initial high firmness (Tables 3.8 and 3.9). Thus, softer fermented cucumbers had a shorter shelf life than initially firmer cucumbers. The fermented cucumbers that contained residual calcium level of 36mM had a shelf life of 917 and 808 days respectively for the treatments of NaCl with added calcium and calcium chloride pickles desalted once.

A lack of homogeneity, especially in firmness, has been detected in several other food products besides fermented cucumbers. Garcia-Garcia et al. (2008) determined that a shelf life of 3 years is a reasonable shelf life for high firmness olives, based on the concept that a 20% relative degradation is acceptable amount of texture loss during normal shelf storage conditions. Based on their data, a standard relative degradation of 20% was established to take into account a shorter shelf life for initially softer olives but allow the olives to still be considered acceptable. Garcia-Garcia et al. (2008) also recommended that storage above 30°C should be avoided since the firmness degradation rates increase drastically and diminishes the shelf life for the product.

3.5 Conclusions

A systematic approach was used to develop a predictive model for shelf life of fermented cucumbers. The combined application of both instrumental measurement, as well as descriptive sensory analysis under accelerated shelf life temperature (ASLT) conditions, allowed for the evaluation of different treatments of fermented cucumbers. The ASLT method allowed for the estimation of the texture degradation rate (k) as a shelf life function. However, there is not an established scale of the level of acceptable texture attributes in fermented cucumbers. It was determined that a 15-30% texture degradation range would be considered an acceptable loss of texture in these products during shelf storage conditions. Based on the 7N cutoff point determined from the instrumental and sensory correlations and the ability of sensory panelists to detect a small change in texture properties.

An instrumental method like the punch test, which has strong correlations with the descriptive sensory analysis panels, will allow pickle companies to test the overall texture of any commercial fermented cucumber product. This method also could be used to study additional factors that influence texture in pickles, such as acids, sugar, desalting agents, etc. This method helps further our understanding of the lack of homogeneity in these products, especially texture. There is a lot of variability in texture from jar to jar and slice to slice. Determining a set firmness range for the processed products, and preventing storage in high temperatures, will allow pickle companies to comply with the current shelf life declaration on the labels. This research has allowed for a better characterization of the texture properties of fermented cucumber pickles, and could be used to further characterize other pickled vegetable products.

References

Abbott, J. A. (1999). Quality measurement of fruits and vegetables. *Postharvest Biology and Technology*, 15(3), 207-225.

Blair, G. W. (1958). Rheology in food research. *Advances in Food Research*, 8 (1-61).

Bourne, M. C. (1966). Measure of shear and compression components of puncture tests. *Journal of Food Science*, 31(2), 282-291.

Bourne, M. C. (2002). Principles of objective texture measurement. *Food texture and viscosity: Concept and measurement* (pp. 107-189). San Diego, CA: Academic Press.

Breen, W. M., Davis, D. W., & Chou, H. (1972). Texture profile analysis of cucumbers. *Journal of Food Science*, 37, 113-117.

Breene, W. M., Jeon, I. J., & Bernard, S. N. (1974). Observations on texture measurement of raw cucumbers with the fruit pressure tester. *Journal of Texture Studies*, 5, 317-327.

Buescher, R. W., Hamilton, C., Thorne, J., & Jin Cho, M. (2011). Elevated calcium chloride in cucumber fermentation brine prolongs pickle product crispness. *Journal of Food Quality*, 34, 93-99.

Buescher, R. W., & Hudson, J. M. (1986). Bound cations in cucumber pickle mesocarp tissue as affected by brining and CaCl2. *Journal of Food Science*, 51(1), 135-137.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1979). Inhibition of polygalacturonase softening of cucumber pickles by calcium chloride. *Journal of Food Science*, 44, 1786-1787.

Chauvin, M. A., Younce, F., Ross, C., & Swanson, B. (2008). Standard scales for crispness, crackliness and crunchiness in dry and wet foods: Relationship with acoustical determinations. *Journal of Texture Studies*, 39, 345-368.

Cho, M. J., & Buescher, R. W. (2012). Potential role of native pickling cucumber polygalacturonase in softening of fresh pack pickles. *Journal of Food Science*, 77(4), 443-447.

Civille, G. V., & Szczesniak, A. S. (1973). Guidelines to training a texture profile panel. *Journal of Texture Studies*, 4, 204-223.

Corradini, M. G., & Peleg, M. (2007). Shelf-life estimation from accelerated storage data. *Trends in Food Science and Technology*, 18, 37-47.

Diehl, K. C., Hamann, D. D., & Whitfield, J. K. (1979). Structural failure in selected raw fruits and vegetables. *Journal of Texture Studies*, 10, 371-400.

Etchells, J. L., Bell, T. A., & Fleming, H. P. (1977). Use of calcium chloride to improve the texture of pickles. *Advisory statement published and distributed by Pickle Packers International*, Inc. St. Charles, IL.

Fillion, L., & Kilcast, D. (2002). Consumer perception of crispness and crunchiness in fruits and vegetables. *Food Quality and Preference*, 13, 23-29.

Fleming, H. P., McDonald, L. C., McFeeters, R. F., Thompson, R. L., & Humphries, E. G. (1995). Fermentation of cucumbers without sodium chloride. *Journal of Food Science*, 60(2), 312-315.

Fleming, H. P., McFeeters, R. F., & Daeschel, M. A. (1992). Fermented and acidified vegetables. In S. D. Vanderzant C (Ed.), *Compendium of methods for the microbiological examination of foods* (3rd ed., pp. 929-952). Washington, D.C: American Public Health Association.

Fleming, H. P., McFeeters, R. F., & Thompson, R. L. (1987). Effects of sodium chloride concentration on firmness retention of cucumbers fermented and stored with calcium chloride. *Journal of Food Science*, 52(3), 653-657.

Fleming, H. P., & McFeeters, R. F. (1981). Shelf life of fresh-pack cucumber pickles. St. Charles, IL: Pickle Packers International, Inc.

Foegeding, E. A., Brown, J., Drake, M., & Daubert, C. R. (2003). Sensory and mechanical aspects of cheese texture. *International Dairy Journal*, 13, 585-591.

Garcia Loredo, A. B., & Guerrero, S. N. (2011). Correlation between instrumental and sensory ratings by evaluation of some texture reference scales. *International Journal of Food Science and Technology*, 46, 1977-1985.

Garcia-Garcia, P., Lopez-Lopez, A., & Garrido-Fernandez, A. (2008). Study of the shelf life of ripe olives using an accelerates test approach. *Journal of Food Engineering*, 84, 569-575.

Hamann, D. D., Zhang, J., Daubert, C. R., Foegeding, E. A., & Diehl, K. C. (2006). Analysis of compression, tension and torsion for testing food gel fracture properties. Journal of Texture Studies, 37(6), 620-639.

Howard, L. R., & Buescher, R. W. (1990). Cell wall characteristics and firmness of fresh pack cucumber pickles affected by pasteurization and calcium chloride. *Journal of Food Biochemistry*, 14, 31-43.

Hudson, J. M., & Buescher, R. W. (1980). Prevention of soft center development in large whole cucumber pickles by calcium. *Journal of Food Science*, 45, 1450-1451.

Hudson, J. M., & Buescher, R. W. (1985). Pectic substances and firmness of cucumber pickles as influenced by CaCl2, NaCl and brine storage. *Journal of Food Biochemistry*, 9, 211-229.

IFST. (1993). Shelf life of foods-guidelines for its determination and prediction. London: Institute of Food Science and Technology

Jeon, I. J., Breene, W. M., & Munson, S. T. (1973). Texture of cucumbers: Correlation of instrumental and sensory measurements. *Journal of Food Science*, 38, 334-337.

Kilcast, D., & Fillion, L. (2001). Understanding consumer requirements for fruit and vegetable texture. *Nutrition and Food Science*, 31(5), 221-225.

Labuza, T. P. (1982). *Shelf-life dating of foods* (1st ed.). Westport, CT: Food & Nutrition Press, Incorporated,.

Labuza, T. P., & Riboh, D. (1982). Theory and application of Arrhenius kinetics to the prediction of nutrient losses in foods [degradation, keeping, quality, temperature, quality controls, analysis, models]. *Food Technology*, 66-74.

Labuza, T. P., & Schmidl, M. K. (1985). Accelerated shelf-life testing of foods. *Food Technology*, 39(9), 57-62, 64, 134.

Labuza, T. P., & Schmidl, M. K. (1988). Use of sensory data in the shelf life testing of foods: Principles and graphical methods for evaluation. *Cereal Foods World*, 33(2), 193-205.

Labuza, T. P., & Taoukis, P. S. (1990). The relationship between processing and shelf-life. In G. Birch, G. Campbell-Platt & M. G. Lindley (Eds.), *Food for the 90's* (pp. 73-106). New York: Elsevier Applied Science.

Lee, S. -., Guinard, J. -., & Krotchta, J. M. (2002). Relating sensory and instrumental data to conduct an accelerated shelf-life testing of whey-protein coated peanuts. *American Chemical Society*, Chapter 13, 175-187.

Lawless, H. T., & Heymann, H. I. (1998). *Sensory evaluation of food: Principles and practices* (4th ed.). New York, New York: Kluwer Academic/Plenum Publisher.

Luyten, H., Lichtendonk, W., Castro, E. M., Visser, J., & Van Vliet, T. (2007). Acoustic emission from Crispy/Crunchy foods to link mechanical properties and sensory perception. *J. Texture Studies*, 37, 380-392.

Magness, J. R., & Taylor, G. F. (1925). An improved type of pressure tester for the determination of fruit maturity. *U.S Department Agriculture Circulation*, , 350.

Maruvada, R., & McFeeters, R. F. (2009). Evaluation of enzymatic and non-enzymatic softening in low salt cucumber fermentations. *International Journal of Food Science and Technology*, 44, 1108-1117.

McFeeters, R. F. (1989). Function of metal cations in regulating the texture of acidified vegetables. *American Chemical Society*, (Chapter 11), 125-138.

McFeeters, R. F. (2002). Cell wall monosaccharide changes during softening of brined cucumber mesocarp tissue. *Journal of Food Science*, 57(4), 937-953.

McFeeters, R. F., Brenes Balbuena, M., & Fleming, H. P. (1995). Softening rates of fermented cucumber tissue: Effects of pH, calcium and temperature. *Journal of Food Science*, 60(4), 786-788-793.

McFeeters, R. F., & Fleming, H. P. (1990). Effect of calcium ions on the thermodynamics of cucumber tissue softening. *Journal of Food Science*, 55, 446-449.

McFeeters, R. F., Fleming, H. P., & Thompson, R. L. (1985). Pectinesterase activity, pectin methylation, and texture changes during storage of blanched cucumber slices. *Journal of Food Science*, 50, 201-205.

McFeeters, R. F. (1986). Pectin methylation changes and calcium ion effects on the texture of fresh, fermented, and acidified cucumbers. *American Chemical Society*, , 217-229.

McFeeters, R. F., & Perez-Diaz, I. (2010). Fermentation of cucumbers brined with calcium chloride instead of sodium chloride. *Journal of Food Science*, 75(3), 291-296.

Mizrahi, S. (2000). Accelerated shelf-life tests. In D. Kilcast, & P. Subramaniam (Eds.), *Stability and shelf-life of food* (pp. 107-128) Woodhead Publishing.

Nicholas, R. C., & Pflug, I. J. (1960). Effect of high temperature storage on the quality of fresh cucumber pickles. *Glass Pack*, , 39, 35.

Palnitkar, M. P., & McFeeters, R. F. (1975). Recycling spent brines in cucumber fermentations. *Journal of Food Science*, 40, 1311-1315.

Peleg, M. (1977). Compressive failure patterns of some juicy fruits. *Journal of Food Science*, 41(1320-1324)

Szczesniak, A. S. (1963). Classification of textural characteristics. *Journal of Food Science, 28*(4), 385-389.

Szczesniak, A. S. (1988). The meaning of textural characteristics-crispness. *Journal of Texture Studies*, 19, 51-59.

Tang, H. C. L., & McFeeters, R. F. (1983). Relationships among cell wall constituents, calcium and texture during cucumber fermentation and storage. *Journal of Food Science*, 48(1), 66-70.

Thompson, R. L., Fleming, H. P., Hamann, D. D., & Monroe, R. J. (1982). Method for determination of firmness in cucumber slices. *Journal of Texture Studies*, 13, 311-324.

Thompson, R. L., Fleming, H. P., & Monroe, R. J. (1979). Effects of storage conditions on firmness of brined cucumbers. *Journal of Food Science*, 44, 843-846.

Truong, V. D., & Daubert, C. R. (2000). Comparative study of large strain methods for assessing failure characteristics of selected food gels. *Journal of Texture Studies*, 31(3), 335-353.

Yoshioka, Y., Horie, H., Sugiyama, M., & Sakata, Y. (2009). Quantifying cucumber fruit crispness by mechanical measurement. *Journal of Breeding Science*, 59(139-147).

Figures

Collect pickles from CaCl$_2$ Tank 1-17, 1-18 at Mount Olive Pickle Co.	Run Texture	Collect pickles from NaCl Tank 2-21, 1-22 at Mount Olive Pickle Co.

Divide in half

60:40 Cucs:H$_2$O

Wash Once   Wash Twice   Wash Once

Run Texture

Run Chemical Analysis to determine specific Na, Ca, and acetic acid concentrations to calculate the cover brine formulation.

Pack in 16 oz. Jars as Hamburger Dill Chips: 58:42, cucs:cover brine	Pack in 16 oz. Jars as Hamburger Dill Chips: 58:42, cucs:cover brine	Pack in 16 oz. Jars as Hamburger Dill Chips: 58:42, cucs:cover brine	Pack in 16 oz. Jars as Hamburger Dill Chips: 58:42, cucs:cover brine

Add cover brine to obtain equilibrated sodium content of traditional NaCl fermented pickles

Add cover brine to obtain same equilibrated calcium as 2 wash CaCl$_2$ fermentation treatment

Add cover brine to obtain same equilibrated calcium as 1 wash CaCl$_2$ fermentation

Run Accelerated storage study on jars at various temperatures: Evaluate difference of texture after a given amount of time to see if it shows a significant different between temperature as well as treatments

Run Texture after Pasteurization as Day 0 Time point

Run Texture on 25°C Pickles every 8 weeks from Pack Date	Run Texture on 35°C Pickles every 4 weeks from Pack Date	Run Texture on 45°C Pickles every 1 weeks from Pack Date	Run Texture on 55°C Pickles every 2-3 days from Pack Date

Figure 3.1 Experimental design to evaluate texture during accelerated shelf life testing.
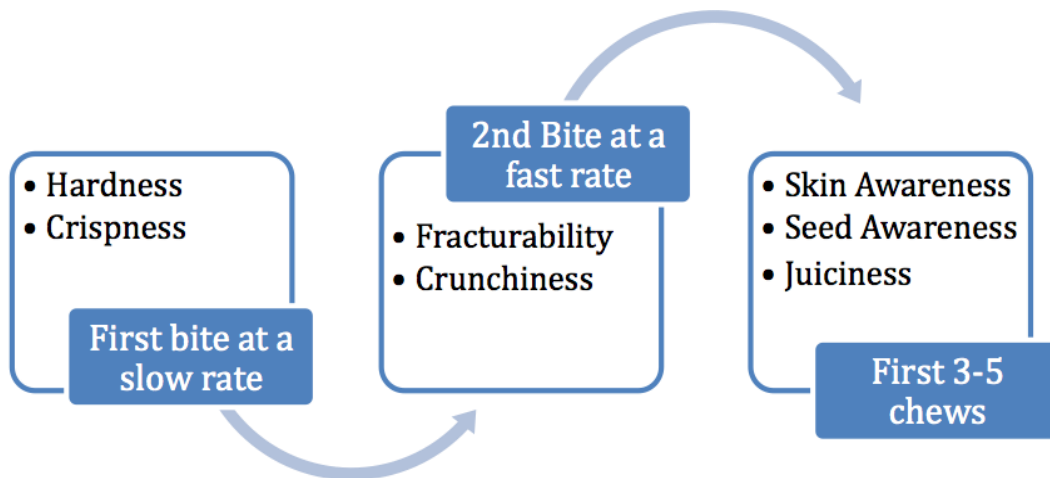
Figure 3.2 Descriptive sensory analysis method to evaluate the texture attributes in fermented cucumber pickle products.
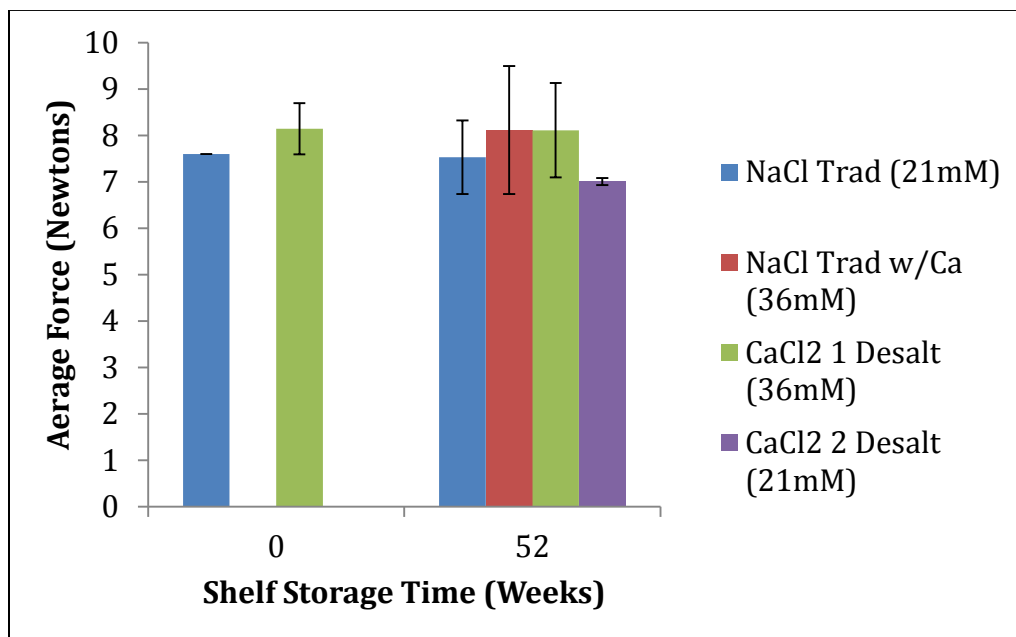
Figure 3.3 Firmness of fermented cucumber pickle products after pasteurization (Week 0) and after 1 year of ambient storage (Week 52). Brined and fermented in 6% NaCl or 1.1% $CaCl_2$ and packed out to different residual calcium levels.
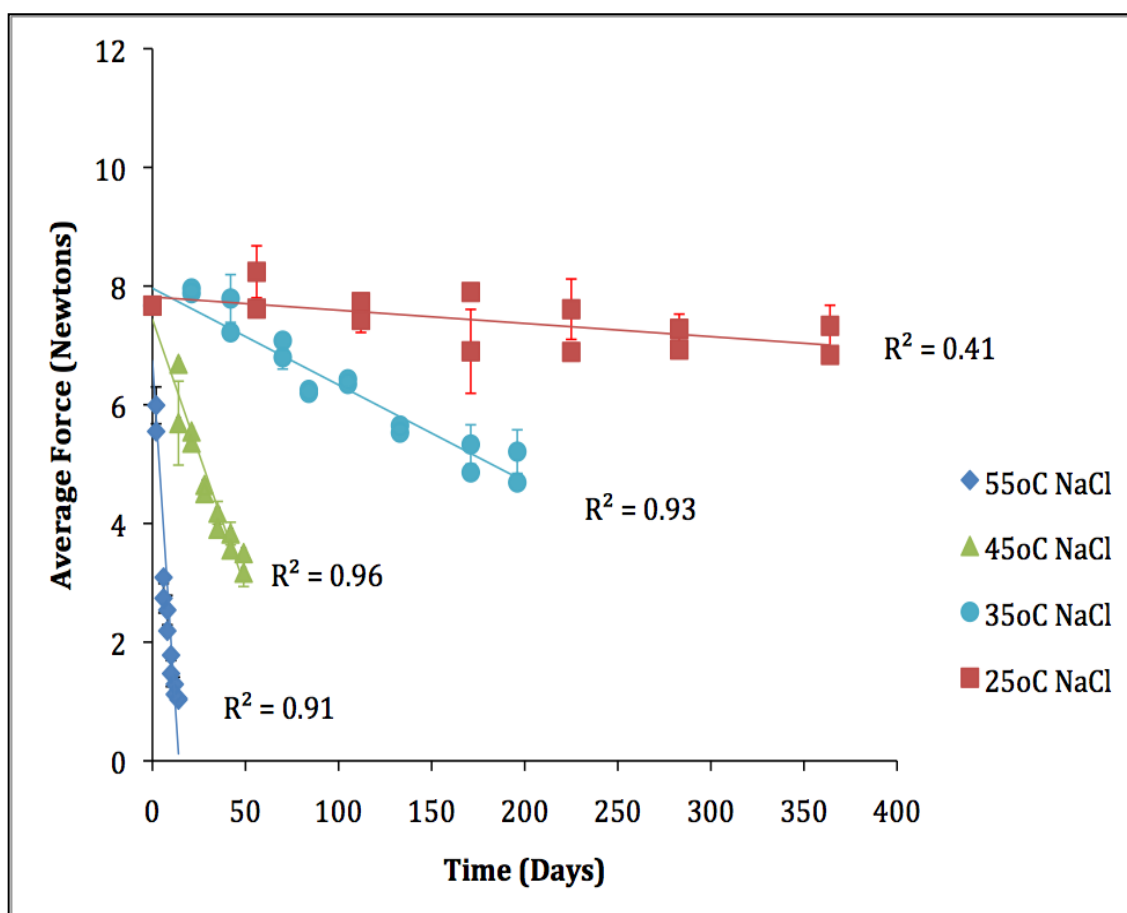
Figure 3.4 Texture degradation during ambient and accelerated temperature storage for traditional NaCl brined and fermented cucumber pickles.
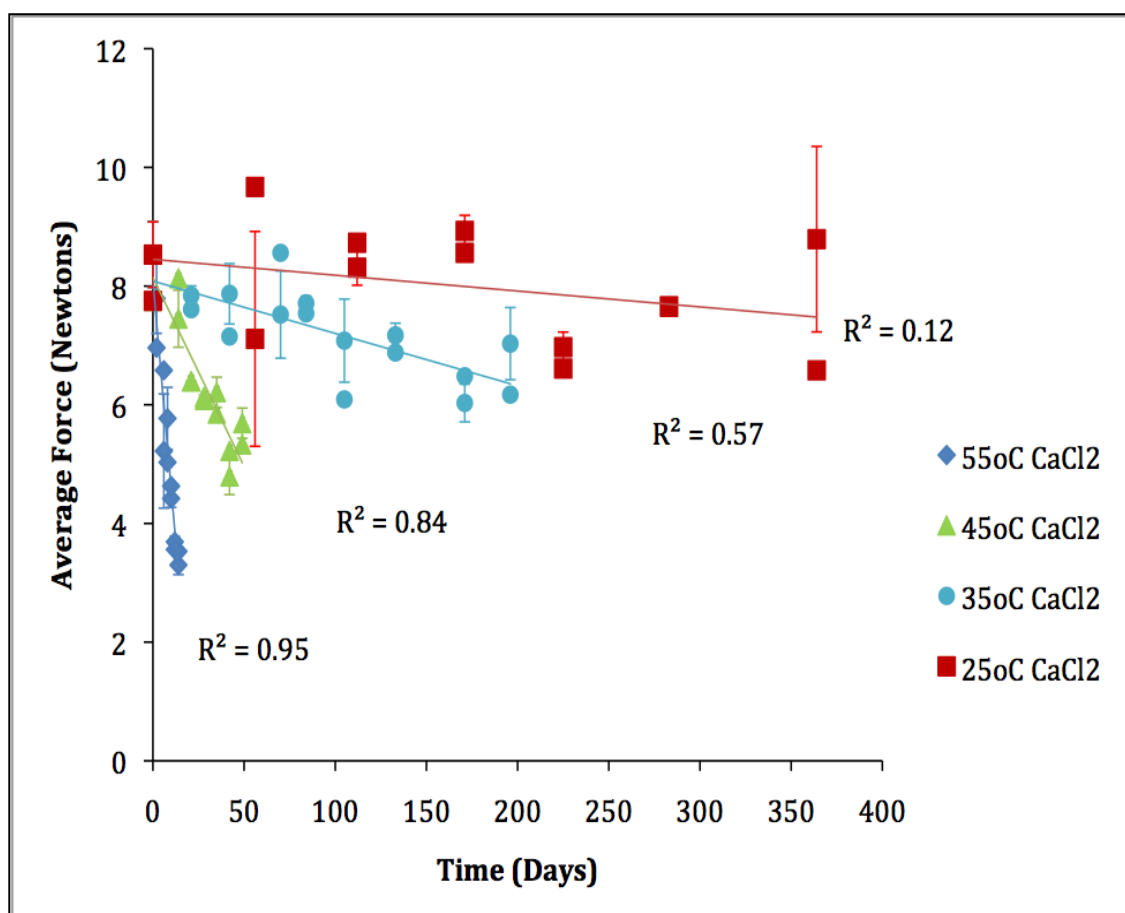
Figure 3.5 Texture degradation during ambient and accelerated temperatures for environmentally-friendly $CaCl_2$ brined and fermented cucumber pickles. Fermented cucumbers were brined with 1.1% $CaCl_2$ and processed to have a residual calcium chloride level of 36 mM.
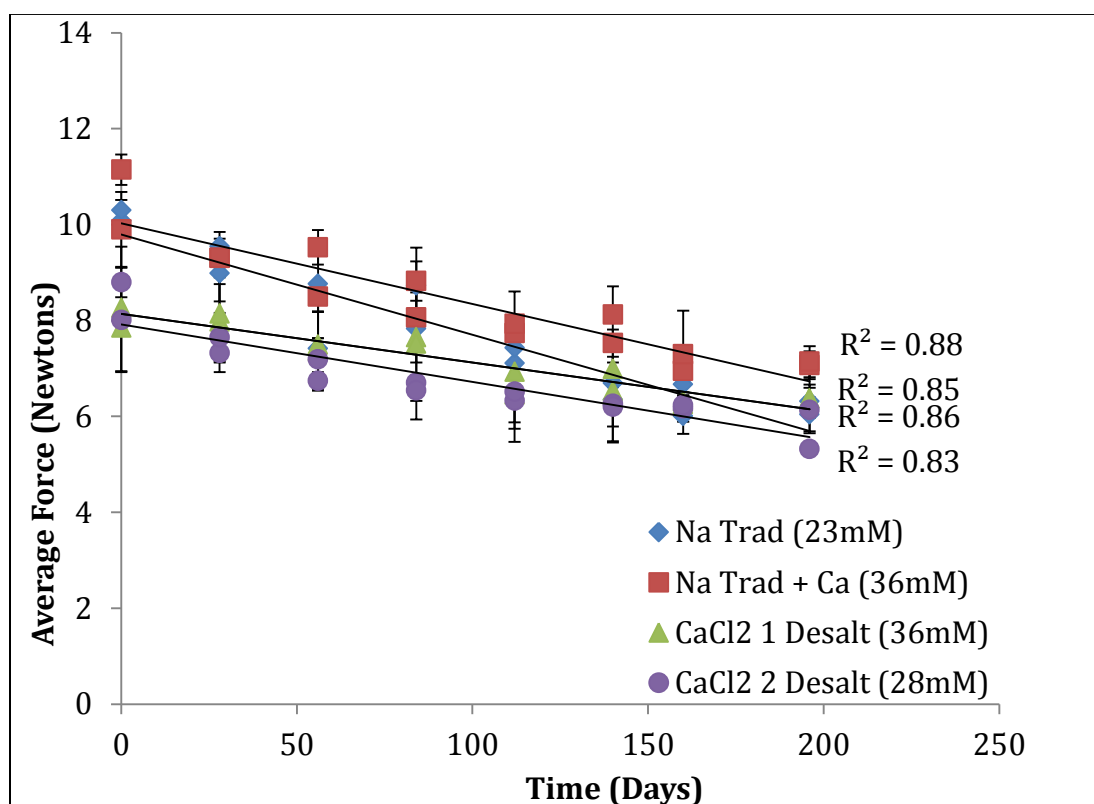
Figure 3.6 Texture degradation during accelerated shelf life testing at 35°C for environmentally-friendly CaCl$_2$ brined and fermented cucumber pickles. Fermented cucumbers were brined with 6% NaCl or 1.1% CaCl$_2$ and processed to have a residual calcium chloride level of 21 or 36 mM.
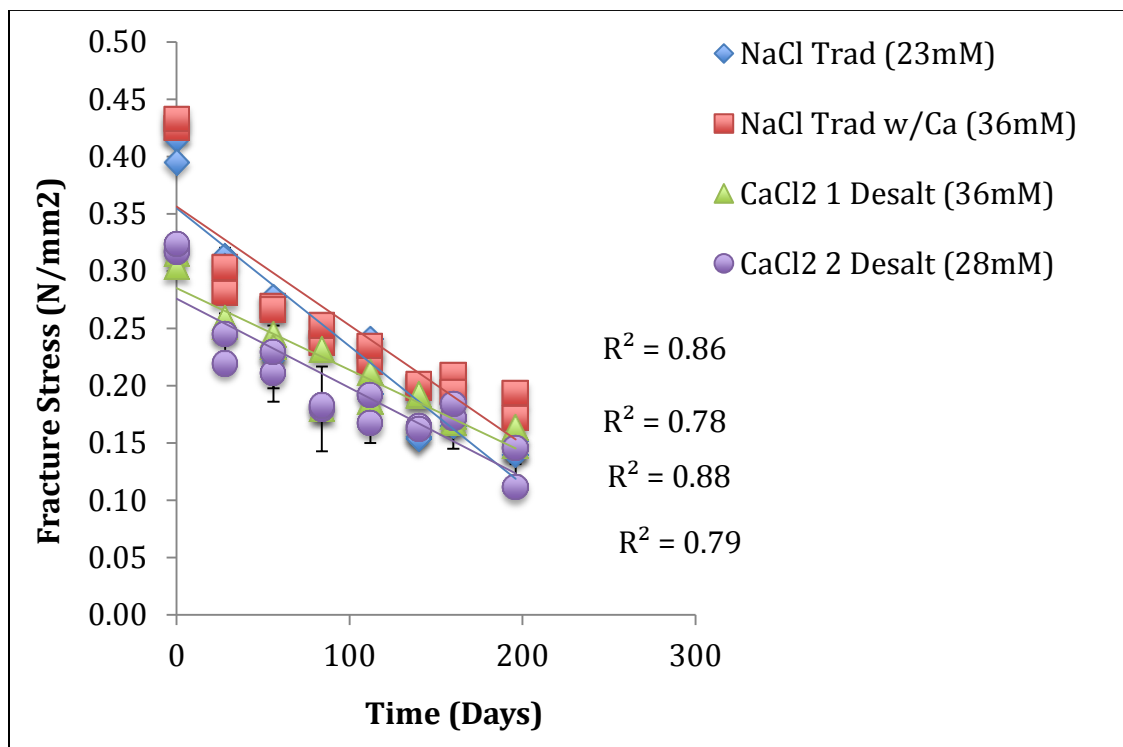
Figure 3.7 Fracture Stress values during accelerated shelf life testing at 35°C for environmentally-friendly CaCl$_2$ brined and fermented cucumber pickles. Fermented cucumbers were brined with 6% NaCl or 1.1% CaCl$_2$ and processed to have a residual calcium chloride level of 21 or 36 mM.
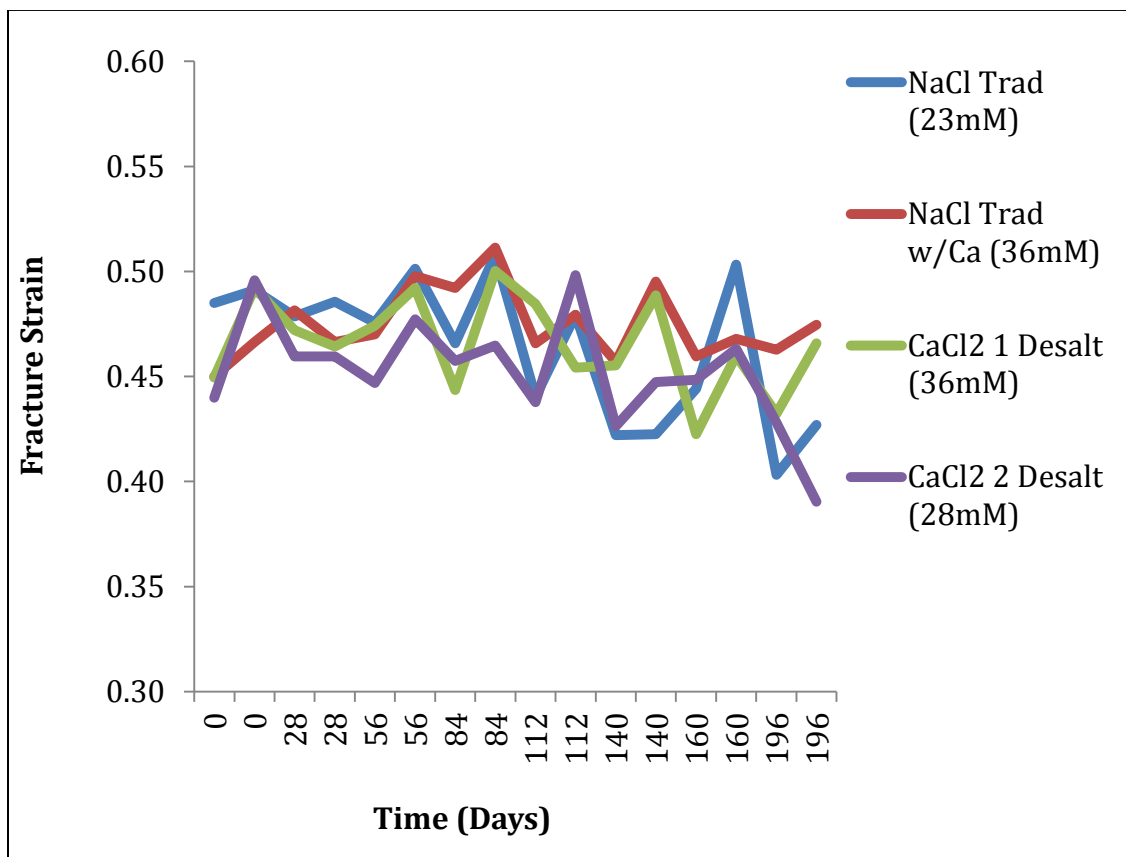
Figure 3.8 Fracture Strain values during accelerated shelf life testing at 35°C for environmentally-friendly CaCl$_2$ brined and fermented cucumber pickles. Fermented cucumbers were brined with 6% NaCl or 1.1% CaCl$_2$ and processed to have a residual calcium chloride level of 21 or 36 mM.
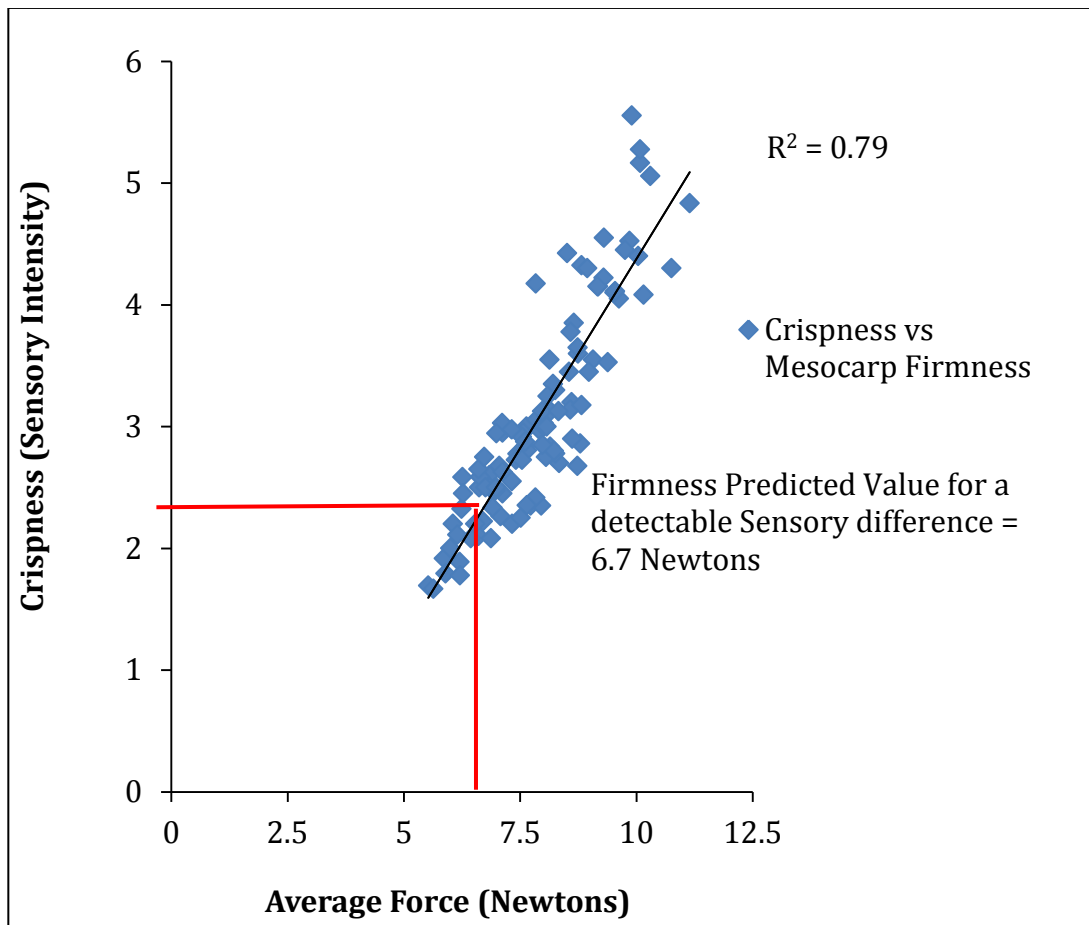
Figure 3.9 Correlation between the instrumenal analysis of texture using a puncture test and the sensory crispness intensity of hamburger dill chip pickles.
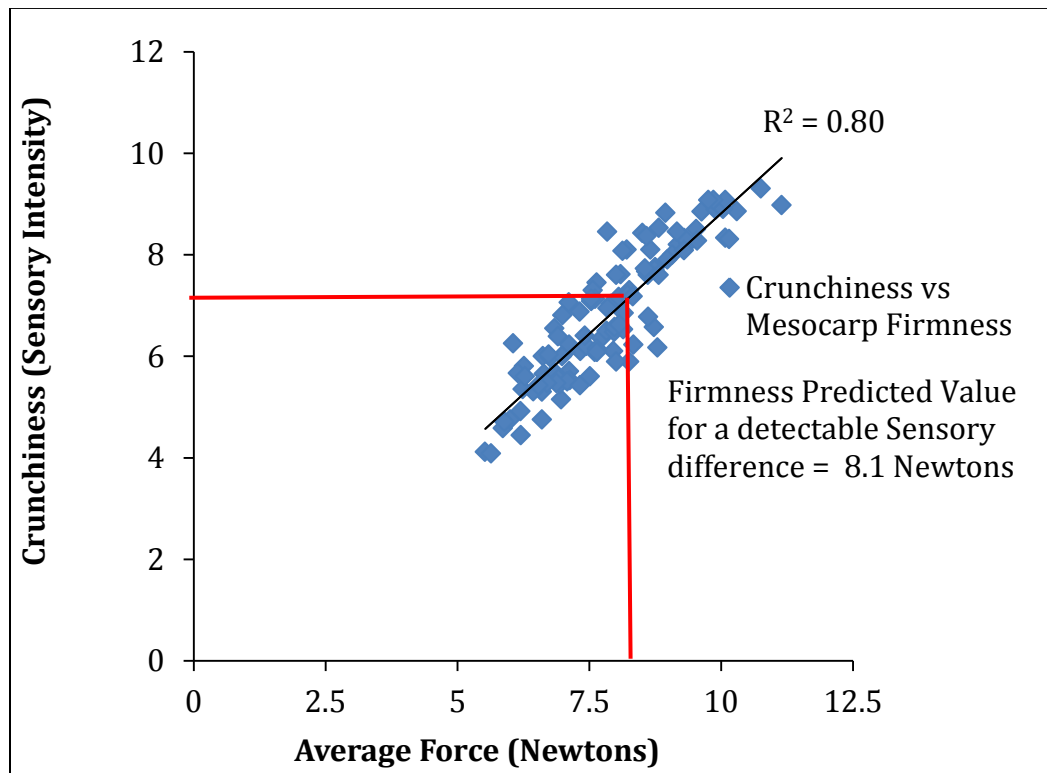
Figure 3.10 Correlation between the instrumenal analysis of of texture using a puncture test and the sensory crunchiness intensity of hamburger dill chip pickles.
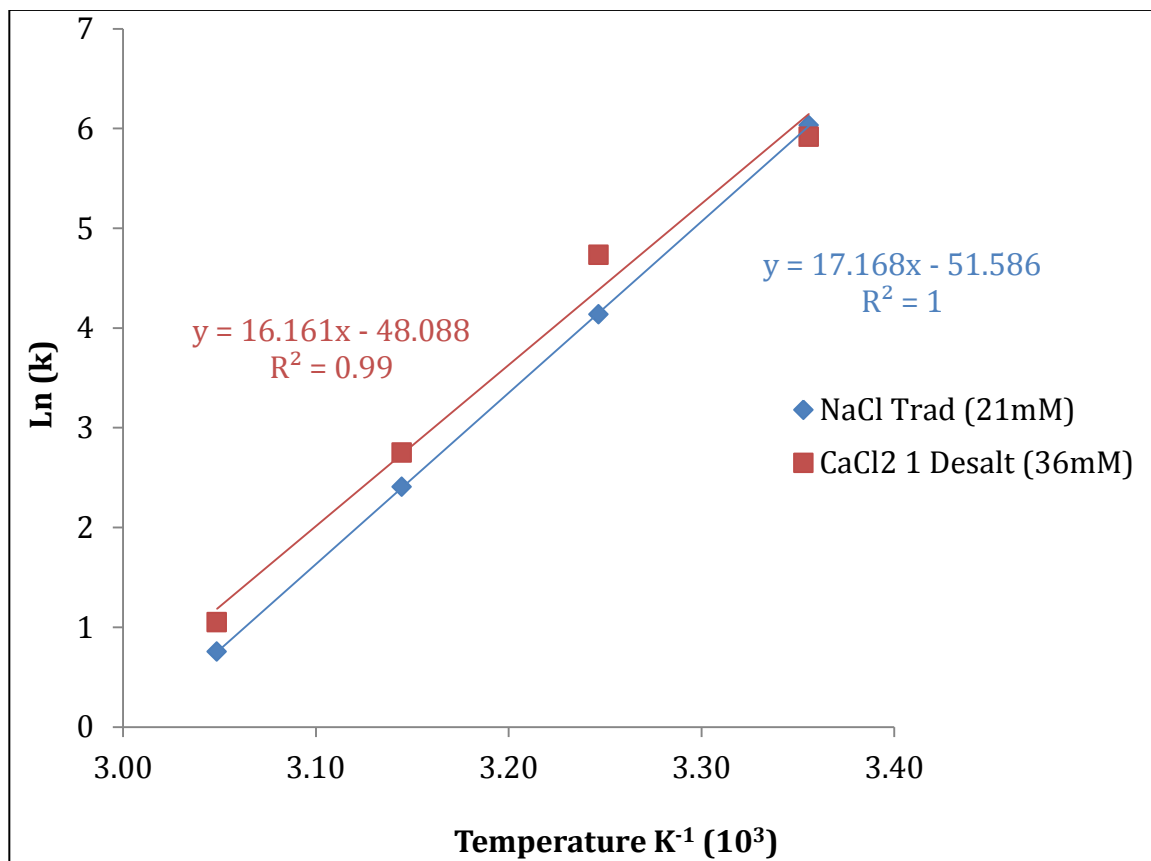
Figure 3.11 Shelf Life Plot of NaCl and $CaCl_2$ brined and processed fermented cucumbers at accelerated temperatures based on values from the mesocarp punch data. Changes in the degradtion rates for firmness as a function of temperature.
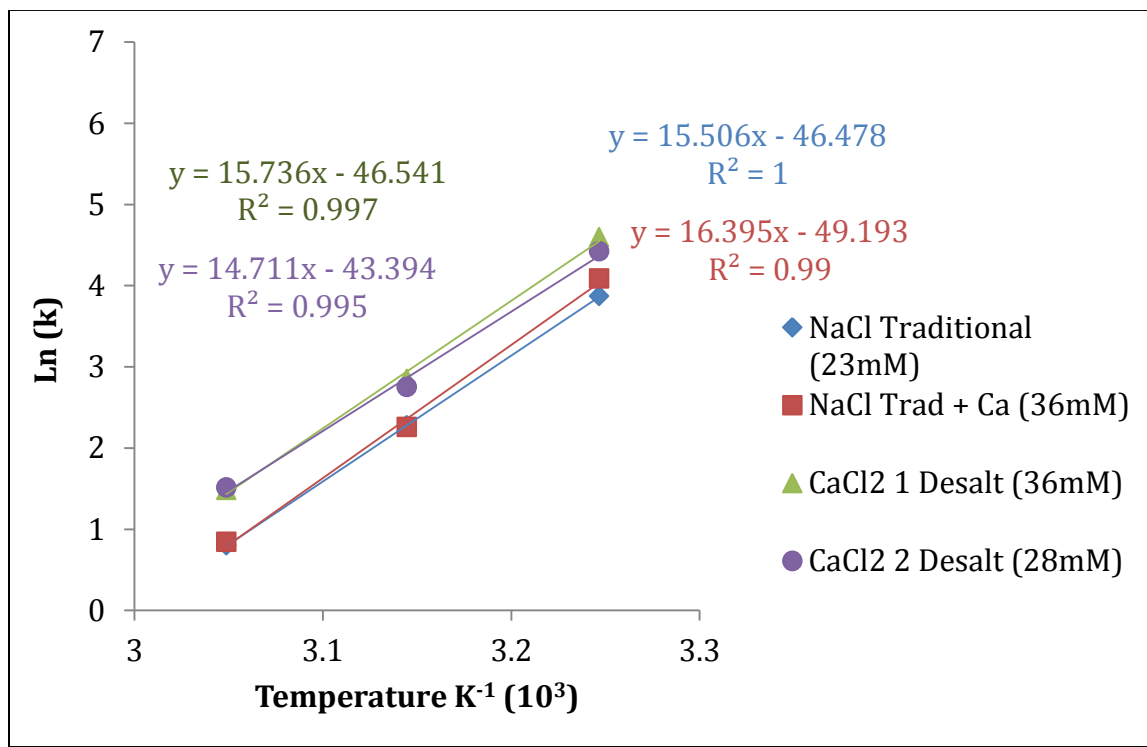
Figure 3.12 Change in the degradation rates for instrumentally measured mesocarp firmness as a function of accelerated temperature for both NaCl and CaCl₂ brined and processed fermented cucumbers.

Table 3.1 Definitions and techniques used for Descriptive Sensory Analysis.

Attribute	Definition	Technique	Anchor(s) on Spectrum™ Scale
Hardness	Force necessary to compress the sample between the molars	Place sample between back molars and compress sample with molars.	Processed Cheese (1/2 in. cube) (4.5) Hebrew National Hotdog (7) Almond (11)
Fracturability	Force with which the sample ruptures when placing sample between molars and biting down completely down at a fast rate	Place sample between back molars and compress sample completely down at a fast rate.	Corn Muffin (1) Ginger Snap (8) Life Saver (14.5)
Crispness	The multiple, higher pitched sounds produced as the sample is crushed with the molar teeth – Evaluate on first 3 chews	Place sample between back molars and compress sample slowly. Repeat compression 3 times with the same sample.	Gala Apple, peeled (1/2in) (4) Water Chestnut (12)
Crunchiness	A single lower-toned noise produced with each chew. Evaluate intensity of crunchiness during the first 3 chews	Place sample between back molars and bite down quickly.	Peanut (4) Raw Carrot (1/2in) (12)
Skin Awareness	The degree to which the outside skin of the product is perceived as intact pieces during mastication	Place sample between back molars and begin the mastication process.	Red Seedless Grape (10)
Seed Awareness	The degree to which the seeds within the product are perceived as intact pieces during mastication	Place sample between back molars and begin the mastication process.	½ cup Tomato Sauce with 1tsp. sesame seeds (4) Pomegranate Seeds (15)
Juiciness	The amount of juice/moisture perceived in the sample	Place sample between the back molar teeth and begin the mastication process.	Banana (1) Sugar Snap Peas (7) Orange Segment (15)

Table 3.2 Initial values of firmness, pH, and residual calcium for the different treatments of fermented cucumbers used in the accelerated shelf life tests.

Fermentation Brine Treatment	Tankyard Storage Time (mo.)	Finished Product Calcium chloride (mM)	Finished Product pH	Firmness (N)[1]
NaCl (n=1)	9	21 ± 2	3.40 ± 0.05	7.67 ± 0.1
CaCl$_2$ 1 desalt (n=1)	10	36± 2	3.23 ± 0.04	8.14 ± 0.6
NaCl (n=2)	4	23± 2	3.31 ± 0.02	10.19 ± 0.2
NaCl w/Ca (n=2)	4	36± 2	3.23 ± 0.09	10.52 ± 0.9
CaCl$_2$ 1 desalt (n=2)	4	36± 2	3.17 ± 0.03	8.06 ± 0.6
CaCl$_2$ 2 desalt (n=2)	4	28± 2	3.25 ± 0.02	8.40 ± 0.9
Reference	N/A	21 ± 2	3.37± 0.07	8.89 ± 0.9

[1]Values are the average of two replicate jars for each fermentation tank processed, for which the data are, in turn, the average of 10-15 pickle slices

Table 3.3 Initial descriptive texture attribute values for the different treatments during ASLT. Brined and fermented in either 6% NaCl or 1.1% $CaCl_2$ to have different processed residual calcium levels.

Brining Treatment	Product Calcium (mM)	Tankyard Storage (months)	Storage Temp (ºC)	Shelf Storage (days)	Hardness[1]	Crispness[1]	Fracturability[1]	Crunchiness[1]
Reference[2]	21	N/A	4	0	7.5	3	5	8
NaCl 6%	23	4	25	0	7.9[a]±0.2	5.1[a]±0.1	5.7[a]±0.1	8.6[b]±0.4
NaCl 6%	36	4	25	0	8.0[a]±0.0	5.2[a]±0.5	5.6[a]±0.2	8.9[a]±0.1
CaCl₂ 1.1%	36	4	25	0	6.8[d]±0.6	2.9[c]±0.4	4.6[c]±0.2	6.4[c]±0.7
CaCl₂ 1.1%	28	4	25	0	6.5[e]±0.3	2.9[c]±0.1	4.3[e]±0.1	6.0[d]±0.2

[1]Values are the average of the 2 replicates of each sample from 8-10 panelists scoring each attribute.
[2]The panel set 2 Reference pickle DA values that were used to evaluate all pickle samples.
Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 2007).
Attributes not listed were not significantly different among treatments

Table 3.4 Sensory texture attributes for samples stored at 55°C (each data point is the mean of the 2 replicates of each samples from 8-10 panelists scoring each attribute).

Brining Treatment	Product Calcium	Tankyard Storage (months)	Storage Temp (°C)	Shelf Storage (days)	Hardness	Crispness	Fracturability	Crunchiness
NaCl 6%	23	4	55	0	7.9^a±0.2	5.1^a±0.1	5.7^a±0.1	8.6^b±0.4
CaCl$_2$ 1.1%	28	4	55	0	6.5^b±0.3	2.9^c±0.1	4.3^e±0.1	6.0^c±0.2
NaCl 6%	36	4	55	0	8.0^a±0.0	5.2^a±0.5	5.6^a±0.2	8.9^a±0.1
CaCl$_2$ 1.1%	36	4	55	0	6.8^a±0.6	2.9^c±0.4	4.6^a±0.2	6.4^b±0.7
NaCl 6%	23	4	55	2	7.4^a±0.5	3.8^b±0.9	5.1^a±0.8	8.0^a±0.5
CaCl$_2$ 1.1%	28	4	55	2	6.4^b±0.3	2.5^d±0.4	4.2^c±0.5	5.9^c±0.5
NaCl 6%	36	4	55	2	7.6^a±0.2	4.1^a±0.1	5.3^a±0.3	8.4^a±0.1
CaCl$_2$ 1.1%	36	4	55	2	6.4^b±0.2	2.6^d±0.3	4.2^c±0.0	6.1^c±0.0
NaCl 6%	23	4	55	4	6.5^b±0.4	2.8^c±0.2	4.5^b±0.1	6.6^b±0.6
CaCl$_2$ 1.1%	28	4	55	4	6.3^c±0.0	2.3^e±0.3	4.2^c±0.0	5.4^d±0.1
NaCl 6%	36	4	55	4	7.0^a±0.5	3.3^c±0.3	4.8^a±0.5	7.5^a±0.9
CaCl$_2$ 1.1%	36	4	55	4	6.3^c±0.1	2.6^d±0.1	4.4^b±0.0	5.3^d±0.3
NaCl 6%	23	4	55	6	5.8^d±0.4	2.6^d±0.1	4.1^c±0.1	5.9^c±0.3
CaCl$_2$ 1.1%	28	4	55	6	5.6^e±0.0	2.5^d±0.2	3.8^e±0.0	5.1^d±0.4
NaCl 6%	36	4	55	6	6.2^c±0.2	2.7^d±0.3	4.3^c±0.1	6.0^c±0.4
CaCl$_2$ 1.1%	36	4	55	6	6.0^d±0.2	2.5^d±0.0	4.1^d±0.3	5.5^d±0.1

Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 2007).
Attributes not listed were not significantly different among treatments

Table 3.5 Sensory texture attributes for samples stored at 45°C (each data point is the mean of the 2 replicates of each samples from 8-10 panelists scoring each attribute).

Brining Treatment	Product Calcium (mM)	Storage Temp (^{o}C)	Shelf Storage (days)	Hardness	Crispness	Fracturability	Crunchiness
NaCl 6%	23	45	0	7.9^a±0.2	5.1^a±0.1	5.7^a±0.1	8.6^b±0.4
CaCl2_1.1%	28	45	0	6.5^b±0.3	2.9^b±0.1	4.3^d±0.1	6.0^c±0.2
NaCl 6%	36	45	0	8.0^a±0.0	5.2^a±0.5	5.6^a±0.2	8.9^a±0.1
CaCl2_1.1%	36	45	0	6.8^a±0.6	2.9^c±0.4	4.6^d±0.2	6.4^c±0.7
NaCl 6%	23	45	14	6.9^a±0.2	3.1^b±0.2	4.9^b±0.1	7.3^b±0.4
CaCl2_1.1%	28	45	14	6.5^b±0.4	2.7^b±0.1	4.1^e±0.2	5.9^d±0.2
NaCl 6%	36	45	14	7.6^a±0.1	3.7^b±0.2	5.3^a±0.0	8.3^a±0.1
CaCl2_1.1%	36	45	14	6.5^b±0.8	2.7^b±0.3	4.2^d±0.1	6.4^c±0.6
NaCl 6%	23	45	21	6.4^b±0.0	2.5^b±0.3	4.2^e±0.2	6.4^c±0.0
CaCl2_1.1%	28	45	21	6.0^c±0.1	2.3^c±0.2	3.8^f±0.1	5.6^d±0.0
NaCl 6%	36	45	21	7.0^a±0.1	3.0^b±0.1	4.8^c±0.2	7.3^b±0.4
CaCl2_1.1%	36	45	21	6.0^c±0.5	2.3^c±0.3	3.8^f±0.2	5.7^d±0.4
NaCl 6%	23	45	35	5.1^e±0.2	2.0^d±0.1	3.4^g±0.2	4.7^e±0.1
CaCl2_1.1%	28	45	35	4.9^f±0.0	1.7^e±0.0	3.0^h±0.1	4.1^g±0.0
NaCl 6%	36	45	35	5.5^d±0.1	2.0^d±0.1	3.6^f±0.1	5.1^d±0.3
CaCl2_1.1%	36	45	35	5.2^e±0.2	1.8^e±0.0	3.3^g±0.1	4.6^f±0.2

Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 2007).
Attributes not listed were not significantly different among treatments

Table 3.6 Sensory texture attributes for samples stored at 35°C (each data point is the mean of the 2 replicates of each samples from 8-10 panelists scoring each attribute).

Brining Treatment	Product Calcium (mM)	Storage Temp (^{o}C)	Shelf Storage (days)	Hardness	Crispness	Fracturability	Crunchiness
NaCl 6%	22	35	0	7.9[a]±0.2	5.1[a]±0.1	5.7[a]±0.1	8.6[a]±0.4
CaCl2_1.1%	22	35	0	6.5[e]±0.3	2.9[c]±0.1	4.3[e]±0.1	6.0[c]±0.2
NaCl 6%	36	35	0	8.0[a]±0.0	5.2[a]±0.5	5.6[a]±0.2	8.9[a]±0.1
CaCl2_1.1%	36	35	0	6.8[d]±0.6	2.9[c]±0.4	4.6[c]±0.2	6.4[c]±0.7
NaCl 6%	22	35	28	7.4[a]±0.3	3.8[b]±0.5	5.2[a]±0.1	8.1[a]±0.3
CaCl2_1.1%	22	35	28	6.7[b]±0.2	2.9[c]±0.3	4.4[c]±0.1	6.5[c]±0.6
NaCl 6%	36	35	28	7.3[a]±0.1	4.4[a]±0.2	5.3[a]±0.2	8.2[a]±0.2
CaCl2_1.1%	36	35	28	6.5[b]±0.2	3.0[c]±0.2	4.3[c]±0.1	6.5[c]±0.0
NaCl 6%	22	35	84	6.4[c]±0.1	2.5[d]±0.2	4.2[d]±0.1	6.5[c]±0.1
CaCl2_1.1%	22	35	84	5.9[e]±0.2	2.2[d]±0.0	3.7[e]±0.2	5.4[d]±0.0
NaCl 6%	36	35	84	7.1a[b]±0.3	3.0[c]±0.3	4.8[a]±0.4	7.4[b]±0.3
CaCl2_1.1%	36	35	84	6.0[d]±0.0	2.3[c]±0.1	4.0[d]±0.0	5.9[d]±0.4
NaCl 6%	22	35	140	5.9[d]±0.1	2.2[d]±0.0	4.0[d]±0.1	5.2[d]±0.1
CaCl2_1.1%	22	35	140	5.6[e]±0.0	1.9[f]±0.0	3.5[g]±0.0	4.6[f]±0.3
NaCl 6%	36	35	140	6.6[b]±0.1	2.7[c]±0.2	4.5[b]±0.1	6.4[c]±0.6
CaCl2_1.1%	36	35	140	5.9[d]±0.3	2.3[d]±0.3	3.8[e]±0.4	5.0[f]±0.2
NaCl 6%	22	35	196	4.9[g]±0.3	2.1[e]±0.1	2.9[h]±0.4	3.6[h]±0.2
CaCl2_1.1%	22	35	196	4.5[h]±0.3	1.8[g]±0.1	2.6[i]±0.2	3.5[h]±0.3
NaCl 6%	36	35	196	5.5[f]±0.1	2.5[d]±0.0	3.6[f]±0.2	5.1[e]±0.5
CaCl2_1.1%	36	35	196	4.8[g]±0.1	1.9[f]±0.1	2.9[h]±0.1	3.9[g]±0.2

Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 2007).

Attributes not listed were not significantly different among treatments

Table 3.7 Sensory texture attributes for samples stored at 25°C (each data point is the mean of the 2 replicates of each samples from 8-10 panelists scoring each attribute).

Brining Treatment	Product Calcium (mM)	Storage Temp (°C)	Shelf Storage (days)	Hardness	Crispness	Fracturability	Crunchiness
NaCl 6%	23	25	0	7.9[a]±0.2	5.1[a]±0.1	5.7[a]±0.1	8.6[b]±0.4
CaCl2_1.1%	28	25	0	6.5[e]±0.3	2.9[c]±0.1	4.3[e]±0.1	6.0[d]±0.2
NaCl 6%	36	25	0	6.8[d]±0.6	2.9[c]±0.4	4.6[c]±0.2	6.4[c]±0.7
CaCl2_1.1%	36	25	0	6.5[e]±0.3	2.9[c]±0.1	4.3[e]±0.1	6.0[d]±0.2
NaCl 6%	23	25	56	7.6[b]±0.0	4.2[b]±0.1	5.2[b]±0.1	8.5[b]±0.1
CaCl2_1.1%	28	25	56	6.8[c]±0.2	3.0[c]±0.2	4.3[e]±0.1	6.9[c]±0.4
NaCl 6%	36	25	56	7.8[b]±0.0	4.9[a]±0.5	5.4[b]±0.3	9.1[a]±0.0
CaCl2_1.1%	36	25	56	6.9[b]±0.1	2.9[c]±0.1	4.6[c]±0.1	7.0[c]±0.3
NaCl 6%	23	25	112	7.9[a]±0.1	4.4[a]±0.1	5.8[a]±0.2	9.2[a]±0.1
CaCl2_1.1%	28	25	112	6.8[d]±0.4	2.9[c]±0.2	4.5[d]±0.1	6.9[c]±0.4
NaCl 6%	36	25	112	7.9[a]±0.1	4.4[a]±0.1	5.8[a]±0.0	9.2[a]±0.2
CaCl2_1.1%	36	25	112	7.2[b]±0.3	3.2[b]±0.4	4.7[c]±0.3	7.4[c]±0.4
NaCl 6%	23	25	176	7.6[b]±0.1	4.4[a]±0.3	5.4[b]±0.1	8.5[b]±0.1
CaCl2_1.1%	28	25	176	6.1[f]±0.3	2.4[c]±0.3	4.0[e]±0.3	5.6[d]±0.3
NaCl 6%	36	25	176	7.7[b]±0.1	4.6[a]±0.2	5.4[b]±0.2	8.8[a]±0.0
CaCl2_1.1%	36	25	176	6.3[e]±0.4	2.7[c]±0.7	3.8[f]±0.3	5.8[d]±0.7

Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 2007).

Attributes not listed were not significantly different among treatments

Table 3.8 Texture degradation rates and the estimated shelf life for fermented cucumbers. Brined and fermented in 6% NaCl or 1.1% $CaCl_2$ and stored at ambient and accelerated temperatures with varying levels of $CaCl_2$ in finished product.

Brining Treatment	Product Calcium (mM)	Storage Temperature (^{o}C)	Equation	R^2	k (N/day)	Estimated Shelf Life (Days)
NaCl	21	25	y = -0.0024x + 7.86	0.41	0.0024	279
$CaCl_2$	36	25	y = -0.0027x + 8.45	0.12	0.0027	422
NaCl	21	35	y = -0.016x + 7.96	0.93	0.016	42
$CaCl_2$	36	35	y = -0.0088x + 8.08	0.57	0.0088	130
NaCl	21	45	y = -0.09x + 7.44	0.96	0.09	7
$CaCl_2$	36	45	y = -0.064x + 8.14	0.84	0.064	18
NaCl	21	55	y = -0.47x + 6.74	0.91	0.47	1
$CaCl_2$	36	55	y = -0.35x + 8.08	0.95	0.35	3

Notes: k Values are the slopes of the lines fitted to the data. Each data point represents the average of 15 measurements of firmness

Table 3.9 Texture degradation rates and the estimated shelf life for fermented cucumbers. Brined and fermented in 6% NaCl or 1.1% $CaCl_2$ and stored at ambient and accelerated temperatures with varying levels of $CaCl_2$ in finished product.

Brining Treatment	Product Calcium (mM)	Storage Temperature (^{o}C)	Equation	k (N/day)	Estimated Shelf Life (Days)
NaCl w/Ca	36	25	y = -0.0038x + 10.44	0.0038	927
NaCl	23	25	y = -0.002x + 9.74	0.002	1595
$CaCl_2$ 2 desalt	28	25	y = -0.0037x + 8.35	0.0037	378
$CaCl_2$ 1 desalt	36	25	y = -0.0013x + 8.23	0.0013	808
NaCl w/Ca	36	35	y = -0.0168x + 10.026	0.0168	210
NaCl	23	35	y = -0.0209x + 9.7951	0.0209	153
$CaCl_2$ 2 desalt	28	35	y = -0.012x + 7.9169	0.012	117
$CaCl_2$ 1 desalt	36	35	y = -0.0101x + 8.1367	0.0101	104
NaCl w/Ca	36	45	y = -0.1045x + 10.27	0.1045	34
NaCl	23	45	y = -0.1027x + 9.63	0.1027	31
$CaCl_2$ 2 desalt	28	45	y = -0.0638x + 7.83	0.0638	22
$CaCl_2$ 1 desalt	36	45	y = -0.0578x + 8.06	0.0578	18
NaCl w/Ca	36	55	y = -0.43x + 10.06	0.43	8
NaCl	23	55	y = -0.45x + 9.84	0.45	7
$CaCl_2$ 2 desalt	28	55	y = -0.2227x + 8.10	0.227	6
$CaCl_2$ 1 desalt	36	55	y = -0.22x + 8.10	0.22	5

Notes: k Values are the average of the two replicates, for which the data are, in turn, the average of 10 measurements of firmness

APPENDIX

A.1 Abstract: Texture of Fresh-Pack Cucumbers under Accelerated Shelf Life Testing

Appearance, flavor, and texture are the primary attributes that influence a consumer's acceptance and consumption of pickle products. Fresh-pack pickles are traditionally produced using acidification of fresh cucumbers in jars with a cover brine solution containing salt, vinegar, colorants, and acid. Fresh-pack cucumber pickle production uses cover brine and pasteurization as a way of preserving cucumbers without producing large amounts of discharge wastes through bulk storage that is used for fermented cucumbers. The effect of temperature during accelerated shelf life testing on mesocarp firmness in $CaCl_2$ enhanced fresh-pack cucumbers was evaluated. Cucumbers were processed and brined on a pilot plant scale, processed into hamburger dill chip (HDC) and stored at 25, 35, 45, or 55°C. Pickle mesocarp firmness was measured using a TA-XT2 Texture Analyzer with a 3 mm diameter punch probe at a rate of 2.5 mm/sec. Maximum puncture force in Newtons (N) was recorded for 15 slices from 2 jars of each replicated fermentation treatment.

Temperature had an effect on the overall mesocarp firmness in hamburger dill chips containing higher residual calcium in the final product at all accelerated temperatures ($p <$ 0.0001). Residual calcium also had a significant effect on the overall mesocarp firmness in the final product ($p =$0.0103). Fresh-pack cucumbers packed out to have a residual calcium level of 24mM, had higher instrumental firmness value during accelerated storage than traditional packed out cucumbers of 10mM. Rate constants for the different brined fermented cucumbers were determined from the fitting of a first order kinetic model to the experimental data as a function of firmness and time. Based on the texture degradation, pickle processors can determine the appropriate shelf life for each specific storage condition. The current shelf

life on the label (504 days) was higher, when assessed for product firmness. Based on brining treatment and texture degradation kinetics, it was determined that typical fresh-pack dill pickles would have a shelf life of 298 days whereas the enhanced calcium chloride fresh-pack would have a shelf life of 455 days at ambient temperature.

A.2 Introduction:

Fermented cucumbers have been consumed for thousands of years, and with the introduction of fresh-pack pickles in the 1940's, consumption of this particular type of pickles has dominated the United States marketplace. Fresh-pack pickles are fresh cucumbers acidified by a cover brine solution containing salt, vinegar, colorants, and flavorings and heat-treated to prevent spoilage (Fleming et al., 1983). The United States Department of Agriculture (USDA) standards for fresh-pack pickles specify that it must contain a minimum of 0.5% acetic acid (USDA, 1966). The pH for fresh packed cucumber products is usually in the range of 3.5 to 4.0 with acetic acid concentrations of 0.6% to 2% and salt concentrations of 2 to 4% (Breidt et al., 2007). The cucumbers are pasteurized to prevent microbial growth; pasteurization is also responsible for the inactivation of enzymes that typically contribute to fruit softening. Fresh-pack pickle preservation accounts for forty percent of the total fermented cucumber products in the United States.

The addition of $CaCl_2$ to fermentation cover brines within the last 30 years, has reduced the amount of salt concentration needed to retain texture properties by interacting with pectic substances to retard softening in the tissues of the fruit (Buescher et al., 1979, 1981; Hudson and Buescher 1980, 1985, 1986; Buescher and Hudson 1984, 1986; Howard and Buescher, 1990; Fleming et al., 1978, 1987; McFeeters et al., 1991). Firming of the plant

tissue by calcium is due to the interactions with the 2^{++} charge and galacturonans in the cell wall (Tang and McFeeters, 1983). Firming of the cucumber tissue by $CaCl_2$ is both seen in long-term storage in fermented cucumbers stored in tankyard as well as processed products on the shelves (Tang and McFeeters, 1983).

Texture of a food product consists of the rheological and structural (geometric and surface) attributes that are perceived by mechanical, tactile, visual and auditory receptors (ISO, 1981). Texture evaluation in an important step when developing a new food product or optimizing processing variables within a food. Researchers use both sensory evaluation techniques and instrumental measurements to assess texture. Instrumental methods are typically used due to being less costly and less time consuming. Instrumental texture analysis has a good correlation to what is considered consumer acceptance since consumers prefer a firm, crisp, and crunchy product (Thompson et al., 1982). Mechanical texture tests have been performed on fruits, not only include puncture, compression and shear tests, but also ultrasonic methods (Abbott, 1999).

Under the UK Institute of Food Science and Technology, shelf life is defined as "the period of time during which the food product will remain safe, retain its desired sensory, chemical and microbiological characteristics, and comply with any label declaration of nutrition data" (IFST, 1993). Accelerated storage is a method that has been widely accepted and used in the food, pharmaceutical, cosmetic and other industries that have limited durability (Corradini and Peleg, 2007). This particular type of testing provides knowledge on the impact of handling the products' storage distribution, and how shelf life dating is achieved (Mizrahi, 2004). The pickle industry usually gives their product a 1-2 year shelf life

based on pH, color, taste, and texture (Communication with Mount Olive Pickle Company Representative, 2013). Shelf life of pickles has a huge impact on the texture quality of pickles. Over time, pickles tend to lose their crisp and crunchy textural characteristics. Temperature is the variable most commonly changed during these tests to achieve sufficient data. This kinetic approach allows estimations to be made about shelf life and compare against current stated shelf life (Garcia-Garcia et al., 2008). This type of testing allows for an estimation on the degradation rate to be determined using an Arrhenius equation.

Hypothesis

1) Fresh -pack pickle products with higher calcium level in the cover brine will retain a better firmness to those of traditional pickle products under accelerated shelf life testing conditions

Objectives

1) To determine firmness retention of fresh-pack pickles with traditional and elevated calcium levels

2) To determine a predictive model for end of shelf-life for fresh-pack cucumbers

A.3 Materials and Methods

A.3.1 Fermentation of Fresh-Pack cucumbers

Cucumbers and ingredients for pasteurized hamburger dill chip pickles. Freshly-harvested size 2B (31-38 mm diameter) pickling cucumbers, 200 grain vinegar (20% acetic acid), anhydrous $CaCl_2$, yellow #5 food color, Polish dill flavor concentrate, benzoate with color, Antifoam C mixture, lids and glass jars were obtained from a local processor. Morton's pickling salt (NaCl) was purchased from a local grocery store. All the ingredients

and cucumbers were transported to North Carolina State University where the cucumbers

where then washed and sliced. Cucumbers were flat cut sliced with a Hobart food processor

(Hobart, Model PF 150, Troy, Ohio) to a thickness of 6mm.

Preparation of Cover Brine. Cover brine was prepared the day before packing. Food

grade sodium benzoate, anhydrous calcium chloride, FD&C Yellow #5 colorant, Polish dill

flavor concentrate, 200-grain vinegar (20% acetic acid), and antifoam C mixture were

obtained from Mount Olive Pickle Company. Two formulations were used (Table 4.1 and

4.2) to obtain the intended equilibrated concentration of a 1 desalted calcium chloride

cucumber of 36mM and a traditional fresh-pack cucumber with a residual calcium level of

21mM.

Packing and Pasteurization. Slices were then placed into 8oz with a 58:42

cucumber: brine pack out ratio (132 g cucumbers: 95 g cover brine). The lids were heated

and the applied to the jar to create a hermetic seal. The jars were pasteurized in a steam-

jacketed kettle until they reached an internal temperature of 74°C at the coldest point in the

jar. The jars were held at this temperature for 15 minutes to ensure softening enzymes are

deactivated before the jars are cooled down rapidly to room temperature using cold water.

Storage Conditions. Commercially prepared fresh-pack pickles in glass jars are

expected to have at least an 18-month shelf life. Three accelerated temperatures of 35°C,

45°C, and 55°C were used to help predict end of shelf life by accelerating changes in

mesocarp firmness in the product. Ambient temperature (25 ± 2°C) was used as the control

for the accelerated shelf life testing. Figure 4.1 shows how sampling time points were

established to perform texture analysis on the samples. Jars were equilibrated to ambient

temperature (25 ± 2°C) before firmness measurements were taken.

A.3.2 Instrumental Analysis of Fresh-Pack Cucumbers

Firmness Measurements. Firmness was measured using a TA.XT2 Texture Analyzer (Texture Technologies Corp, Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) equipped with a 3 mm diameter punch probe. Pickle slices (6 mm thick) were placed onto a base plate containing a 3.1 mm hole. The punch probe moved at a test speed of 2.5 mm/sec into the cucumber slices. Data was collected and analyzed using Texture Expert software (Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK). The force required to puncture the cucumber mesocarp was recorded and expressed in Newtons (N). Firmness measurements were done on 15 slices from two jars from each treatment at each sampling time point (Yoshioko et al., 2009; Thompson et al., 1982). Texture was evaluated after pasteurization was complete to become time point zero. This would be the starting point for all the time point in accelerated temperatures as well as the control. Texture was evaluated at accelerated temperature (55°C) at the sampling times of 2, 4,6,8,10,12, and 14 days. Texture was evaluated at accelerated temperature (45°C) at the sampling times of 7, 14, 21,28,35,42, and 49 days. Texture was evaluated at accelerated temperature (35°C) at the sampling times of every 3 weeks for 7 months from the original packing date. Texture was evaluated at ambient temperature (25 ± 2°C) at the sampling times of 2, 4, 6,8,10, and 12 months from the original packing of the products in June 2012.

A.3.3 Descriptive Sensory Analysis of Fresh-Pack Cucumbers

Sensory Training. Approval of the North Carolina State University (Raleigh, North Carolina) Institutional Review Board (IRB # 2734) was obtained for the use of human

subjects in this study. Panelists were trained in descriptive sensory analysis using the

Spectrum™ method (Meilgaard et al., 2007). Panelists were initially trained (75+ hours) to

use a 15-point scale to assess the texture attributes of hardness, crispness, crunchiness,

fracturability, skin awareness, seed awareness, and juiciness as defined in Table 4.3. During

the training, panelists were individually evaluated to determine the overall panel mean and to

assure that all panelists were able to scale the attributes of interest. A selection of food

product with known values on the Spectrum™ method ware used as reference samples for

different textures was used for training.

Panelists were trained to rate hardness, fracturability, and juiciness based upon

reference samples suggested by the Spectrum™ method. Hardness was described as the force

required to compress the sample upon first bite with anchors of 0 for soft to 15 for firm.

Fracturability was defined as force with which the sample ruptures when placing sample

between molars and biting down completely down at a fast rate. Juiciness was defined as the

amount of juice/moisture perceived in the sample (Meilgaard et al., 2007). Crunchiness and

crispness attributes were based upon reference samples suggested by Chauvin (Chauvin et

al., 2008). Crispness was defined at the multiple, higher pitched sounds produced as the

sample is crushed with the molar teeth. Crunchiness was defined as a single lower-toned

noise produced with each chew. Crispness and crunchiness intensity scores were evaluated

during the first 3 chews. Skin and seed awareness were defined by evaluating several food

products by the panelists. Panelists were trained on various foods based upon

recommendations of Meilgaard et al., (2007).

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Following training, (Meilgaard et al., 2007) panelists scores as a group were given a mean to assign a consensus score for each attribute to the pasteurized hamburger dill chip pickles prepared directly from fresh-pack fermented cucumbers and stored at 25ºC that was provided as a reference sample for calibration of panelist ratings at each sensory analysis session during the study.

Sensory Analysis. Panelists were presented 4 samples at the one year time point from pasteurization in a 2oz cup identified with a randomized 3 digit numbers. All samples were given at room temperature along with distilled water, Muenster cheese cubes, and salt-free saltine crackers to cleanse the palate between samples. A randomized order was created to preset samples to each panelist a different order to avoid bias in the order presentation. Each sample cup contained 2 slices**.** Each panelist was also given 2 sample cups of reference fermented fresh-pack hamburger dill chips that came from a local grocery store. This allowed each panelist to calibrate texture attributes on the 15-point scale. Each panelist was instructed to first taste the reference sample, cleanse the palate before tasting the sample. They were given as much time needed to taste their samples and given a 2-minute rest between samples to avoid tasting fatigue. Each panelist was given a spit cup if the did not want to swallow and expectorate their samples. Duplicate evaluations of each sample were done during the session. Sensory analysis was conducted on pickle samples made from fresh cucumbers that were packed and brined in either traditional NaCl brine or a $CaCl_2$ brine.

A.3.4 Accelerated Shelf Life Testing Model

Accelerated Shelf Life Testing Design. To test the effect accelerated temperature storage on texture degradation in fermented cucumbers during storage, fermented cucumbers

were placed in different temperature incubators to predict a model. Pickles were held at temperatures 35, 45, and 55°C in air incubators and at a temperature of 25 $\pm$2°C as control. Sampling was taken by the Figure 4.1 in order to collect enough sampling time points to show first order kinetics. Replicate samples of each treatment were drawn and firmness and pH were analyzed. Jars used in the analyses were always performed in two new containers. From making adjustments of experimental data to different kinetic models, it was deduced the changes in firmness followed an apparent first-order kinetic. The influence of temperature on the reaction rate can be used to determine shelf life from the following equation:

$$-\frac{dA}{dt}=k(A)^n \qquad\qquad\text{(Equation 4.1)}$$

Where A = the quality factor measure in some units of amount, n = the reaction order, and k = the rate constant, the slope of the plot of the appreciate reaction extent A vs. time t (Labuza and Schmidl, 1985). Based on the rate constants determined at several different temperatures, a plot can be extrapolated of *ln* k vs. reciprocal absolute temperature ($^{\circ}$K^{-1}) to predict the rate of the reaction at any other temperature (Labuza and Riboh, 1982).

Statistical Analysis. All statistical computations were analyzed using a general linear model from JMP (version 10 SAS® software, SAS Institute, Cary, NC). Means and standard deviations were calculated for both sensory (n=8 panelists) and instrumental (n=15 pickle slices) data using JMP version 10.1. Table 4.4 lists the means and standard deviations of sensory scores, respectively. When ANOVA test indicated a significant difference between treatments, Tukey tests were conducted for means separation at 0.05 significance levels. The

relations between Rheology and sensory parameters were examined by correlation coefficients.

A.4 Results and Discussion

A.4.1 *Texture in the Fresh-Pack Cucumbers*

In this study, normal fresh-pack pickle products were produced using a typical formula and one to which additional $CaCl_2$ had been added via an acidified cover brine solution and pasteurization to achieve equilibrium with the fresh cucumbers. Calcium and sodium analysis were performed to determine the amount of salt found in the final product of each treatment (Table 4.3). Regardless of the temperature used to accelerate the degradation in texture in the fermented cucumbers, the firmness of cucumbers with residual calcium of 24mM retained firmness better than the traditional fresh-pack pickles ($p < 0.0001$) (Figures 4.2 and 4.3). Calcium was a significant factor in the overall texture in the final product ($p = 0.0103$). The pickles with calcium levels of 24mM started at a peak force average of 11.5 ± 0.1 N on Day 0 and had decreased to 8.4 ± 0.1 N by Day 365 (Figure 4.3). In contrast, the traditional fresh-pack pickles started at a peak force average of 10.9 ± 0.1 N on Day 0 and had decreased to 6.1 ± 0.1 N by Day 365 (Figure 4.2). There was a 2 N difference between the current treatments of fresh-pack pickles at ambient temperatures. Loss of firmness as a result of high temperature storage (>30ºC) in shelf stable pasteurized pickles was first reported by Nicholas and Pflug (1960). Storage temperature has been reported to have the greatest effect on retention of firmness in brined cucumbers as well; particularly at temperatures above 26.6°C (McFeeters and Fleming, 1989; McFeeters and Fleming, 1990; Nicholas and Pflug, 1960).This trend was observed for the accelerated temperatures of 35,

45, and 55°C. The higher calcium level was tested in order to simulate the amount of residual calcium left in the environmentally friendly fermentation using calcium chloride. These salt-free fermentations when desalted at a 60:40 cucumber to water ratio, would contain 36mM or below of residual calcium, (McFeeters et al., 2010). This number is essential to target in order to be below the legal limits of residual calcium in vegetable products (21CFR184.1193).

Regardless of the temperature used to store the fresh-pack cucumber pickles, brining treatment had an effect on the mesocarp tissue in the finished product ($p < 0.0001$). However, the target calcium in the fresh-pack pickle products was not achieved. The levels of calcium in the product are able to maintain cucumber firmness due to the binding of the calcium ions to the cell wall polysaccharides (Buescher and Hudson, 1986; McFeeters and Fleming, 1990). Most vegetables products typically contain less then 25 mM calcium concentration in the product due to the possibility that consumers may be able to detect undesirable chalky flavors with high levels of calcium.

A.4.2 *Sensory Evaluation of Fresh-Pack Pickles*

In addition to the differences in instrumental texture measurements, most of the sensory attributes were significantly different in the fresh-pack pickles prepared with $CaCl_2$ cover brine higher in calcium compared to a traditional formula for fresh-pack pickles. The sensory attributes hardness, crispness, crunchiness, and fracturability were significantly different between the two treatments ($p < 0.0001$). The pickles that were brined with equilibrated concentrations of calcium of 24 mM had higher sensory intensity scores for hardness, crispness, crunchiness, and fracturability on the Spectrum ™ scale compared to the

pickles brined as traditional fresh-pack pickles (Table 4.4). Seed Awareness, skin awareness, and juiciness were not different between the two treatments. While a descriptive panel does not provide information on a like/dislike scale, the fact that their was a clear difference in texture attributes, suggests that a reduction of salt while maintaining texture in a fresh-pack pickle product may be achieved by addition of higher levels of $CaCl_2$.

A.4.3 Shelf Life of Fresh-Pack Pickles

Firmness data for the different brine fresh-pack pickles were plotted as a function of time. It was determined that a first order kinetic model resulted in a better fit and was applied to all the temperatures used in both pickle products to determine the rate of texture deterioration, *k*. The application of equation 4.1 to firmness data from each sample allowed for the parameters to be estimated. There were significant differences in firmness degradation rates between the two fresh-pack cucumbers. At all the temperature except $55^{o}C$, the rates for the $CaCl_2$ enhanced fresh-pack cucumber were slower than the traditional fresh-pack cucumber pickles (Table 4.5). At $55^{o}C$ the NaCl fresh-pack cucumber had a rate loss of 0.89N/day where the $CaCl_2$ fresh-pack cucumbers had a loss of 1.12 N/day. This is interesting to point out since it has been shown that the presence of calcium and pasteurization reduce the rate of texture degradation (Howard et al., 1990).

The relation between k and temperature for firmness was obtained by graphing ln *k* as a function of temperature (T) (Figure 4.4). Based on the rate constants determined at several different accelerated temperatures ($35^{o}C$, $45^{o}C$, $55^{o}C$), (Figures 4.1, 4.2), a plot was extrapolated of *ln* k vs. reciprocal absolute temperature ($^{o}K^{-1}$) to predict the rate of the reaction at ambient storage conditions (Lee et al., 2002, Labuza and Riboh, 1982). From this

plot, an Arrhenius shelf-life equation was determined using regression analysis and the shelf

life for other temperatures could be predicted Figure (4.4). Shelf life based on relative

firmness degradation for traditional fresh-pack pickles was 298, 99, 22 and 4 days for 25, 35,

45, and 55 °C, respectively. For the $CaCl_2$ enhanced fresh-pack pickles, the shelf life based

on relative firmness degradation for traditional fresh-pack pickles was 455, 121, 27 and 3

days. Thus, traditional fresh-pack cucumbers had a shorter shelf life than $CaCl_2$ enhanced

fresh-pack pickles. This shorter shelf life does not take into consideration that flavor may

deteriorate more quickly than texture in food products. Even though this is an estimation of

shelf-life, the true shelf life of a product is determined by the consumers on whether or not it

is considered an acceptable product.

A lack of homogeneity, especially in firmness, has been detected in several food

products besides cucumber pickle products. Garcia-Garcia et al. (2008) determined that a

shelf life of 3 years is a reasonable shelf life for high firmness olives that considered a 20%

relative degradation as acceptable. By using calcium chloride as an aid in processing of

traditional fresh-pack cucumbers, or solely performing calcium chloride fresh-pack

cucumbers, with a high initial firmness value, will give better texture properties during

normal storage conditions. Even though the texture degradation rate was higher for the $CaCl_2$

fresh-pack cucumbers at 55°C, the cucumbers had a lower texture degradation rate at the

other accelerated temperatures compare to the traditional fresh-pack cucumber. This is in part

to the Ca^{2+} ions binding to the pectic substances that help prevent tissue softening from

occurring (Buescher et al., 1981; Tang and McFeeters 1983; Hudson and Buescher 1985;

Buescher and Hudson 1986). It is possible that the high temperature after pasteurization did

not allow the calcium ions to bind with the tissues of the fresh-pack cucumbers causing a high rate of degradation during ASLT. With higher residual calcium in the final processed product, a better-textured product can be achieved for the consumer.

A.5 Conclusion

Between the different brining and storing treatments for fresh-pack cucumbers, addition of enhanced levels of $CaCl_2$ as a salt maintains a firm, crisp texture that a consumer likes. This higher residual calcium level maintains the overall texture properties at both ambient and accelerated temperatures ($p < 0.0001$) compared to the traditional products. Further research would determine if consumers could accept the taste of fresh-pack cucumbers using higher calcium concentrations in the brine. There is a lot of variability when it comes to texture from jar to jar and slice to slice. Determining a set firmness range for the final processed products, and preventing storage in high temperatures, will allow pickle companies to comply with the current shelf life declaration on the labels. This is also the first documented evidence of evaluating mesocarp firmness of fresh-pack cucumber pickles using ASLT methods. Overall, this has allowed for a better characterization of the texture properties of calcium enhanced fresh-pack cucumber pickles.

Acknowledgements

References

Abbott, J. A. (1999). Quality measurement of fruits and vegetables. *Postharvest Biology and Technology, 15*(3), 207-225.

Breidt Jr., F., McFeeters, R. F., & Díaz-Muñiz, I. (2007). Fermented vegetables. In M. P. Doyle, & L. R. Beuchat (Eds.), *In food microbiology: Fundamentals and frontiers* (3rd ed., pp. 783-793). Washington, D.C: ASM Press.

Buescher, R. W., Hamilton, C., Thorne, J., & Jin Cho, M. (2011). Elevated calcium chloride in cucumber fermentation brine prolongs pickle product crispness. *Journal of Food Quality*, 34, 93-99.

Buescher, R. W., & Hudson, J. M. (1986). Bound cations in cucumber pickle mesocarp tissue as affected by brining and CaCl2. *Journal of Food Science*, 51(1), 135-137.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1979). Inhibition of polygalacturonase softening of cucumber pickles by calcium chloride. *Journal of Food Science*, 44, 1786-1787.

Buescher, R. W., Hudson, J. M., & Adams, J. R. (1981). Utilization of calcium to reduce pectinolytic softening of cucumber pickles in low salt conditions. *Lebensm. Wiss. Technology*, 14, 65-69.

Buescher, R. W., & Hudson, M. (1984). Softening of cucumber pickles by cx-cellulase and its prevention by calcium. *Journal of Food Science*, 49, 954-955.

Chauvin, M. A., Younce, F., Ross, C., & Swanson, B. (2008). Standard scales for crispness, crackliness and crunchiness in dry and wet foods: Relationship with acoustical determinations. *Journal of Texture Studies*, 39, 345-368.

Corradini, M. G., & Peleg, M. (2007). Shelf-life estimation from accelerated storage dat. *Trends in Food Science and Technology, 18*, 37-47.

Fleming, H. P., & McFeeters, R. F. (1981). Shelf life of fresh-pack cucumber pickles. St. Charles, IL: Pickle Packers International, Inc.

Fleming, H.P.; Moore, W.R., Jr. Pickling. In *Processing of Horticultural Crops in the United States.* Fuller, G.; Dull, G.G. Eds.; In *CRC Handbook of Processing and Utilization in Agriculture*, Wolff, I.A. Eds., CRC Press, Inc.; Boca Raton, Florida, 1983; Vol. 2, pp. 397-463.

Fleming, H. P., Thompson, R. L., Bell, T. A., & Hontz, L. H. (1978). Controlled fermentation of sliced cucumbers. *Journal of Food Science*, 43(3), 888-891.

Garcia-Garcia, P., Lopez-Lopez, A., & Garrido-Fernandez, A. (2008). Study of the shelf life of ripe olives using an accelerated test approach. *Journal of Food Engineering*, 84, 569-575.

Howard, L. R., & Buescher, R. W. (1990). Cell wall characteristics and firmness of fresh pack cucumber pickles affected by pasteurization and calcium chloride. *Journal of Food Biochemistry*, 14, 31-43.

Hudson, J. M., & Buescher, R. W. (1980). Prevention of soft center development in large whole cucumber pickles by calcium. *Journal of Food Science*, 45, 1450-1451.

Hudson, J. M., & Buescher, R. W. (1985). Pectic substances and firmness of cucumber pickles as influenced by CaCl2, NaCl and brine storage. *Journal of Food Biochemistry*, 9, 211-229.

 IFST. (1993). Shelf life of foods-guidelines for its determination and prediction. London: *Institute of Food Science and Technology*.

Labuza, T. P. (1982). *Shelf-life dating of foods* (1st ed.). Westport, CT: Food & Nutrition Press, Incorporated.

Labuza, T. P., & Riboh, D. (1982). Theory and application of Arrhenius kinetics to the prediction of nutrient losses in foods [degradation, keeping, quality, temperature, quality controls, analysis, models. *Food Technology*, 66-74.

Labuza, T. P., & Schmidl, M. K. (1985). Accelerated shelf-life testing of foods. *Food Technology*, 39(9), 57-62.

McFeeters, R. F., & Fleming, H. P. (1990). Effect of calcium ions on the thermodynamics of cucumber tissue softening. *Journal of Food Science*, 55(2), 446-449.

McFeeters, R. F., & Fleming, H. P. (1991). pH effect on calcium inhibition of softening of cucumber mesocarp tissue. *Journal of Food Science*, 56(3), 730-732.

McFeeters, R. F., Fleming, H. P., & Thompson, R. L. (1985). Pectinesterase activity, pectin methylation, and texture changes during storage of blanched cucumber slices. *Journal of Food Science*, 50, 201-205.

McFeeters, R. F., Senter, M. M., & Fleming, H. P. (1989). Softening effects of monovalent cations in acidified cucumber mesocarp tissue. *Journal of Food Science*, 54(2), 366-370.

McFeeters, R. F. (1986). Pectin methylation changes and calcium ion effects on the texture of fresh, fermented, and acidified cucumbers. *American Chemical Society*, 217-229.

McFeeters, R. F., & Perez-Diaz, I. (2010). Fermentation of cucumbers brined with calcium chloride instead of sodium chloride. *Journal of Food Science*, 75(3), 291-296.

Meilgaard, M. C., Civille, G. V., & Carr, B. T. (2007). *Sensory evaluation techniques* (4th Edition ed.). Boca Raton, Florida: CRC Press.

Tang, H. C. L., & McFeeters, R. F. (1983). Relationships among cell wall constituents, calcium and texture during cucumber fermentation and storage. *Journal of Food Science*, 48(1), 66-70.

Thompson, R. L., Fleming, H. P., Hamann, D. D., & Monroe, R. J. (1982). Method for determination of firmness in cucumber slices. *Journal of Texture Studies, 13*, 311-324.

Thompson, R. L., Fleming, H. P., & Monroe, R. J. (1979). Effects of storage conditions on firmness of brined cucumbers. *Journal of Food Science, 44*, 843-846.

USDA. (1966). In Office of Federal Registry. National Archives and Records Service, General Services Admin (Ed.), Standards for grades of pickles. fed. reg 31:102231-10305. Washington, D.C.:

Yoshioka, Y., Horie, H., Sugiyama, M., & Sakata, Y. (2009). Quantifying cucumber fruit crispness by mechanical measurement. *Journal of Breeding Science*, 59(139-147)
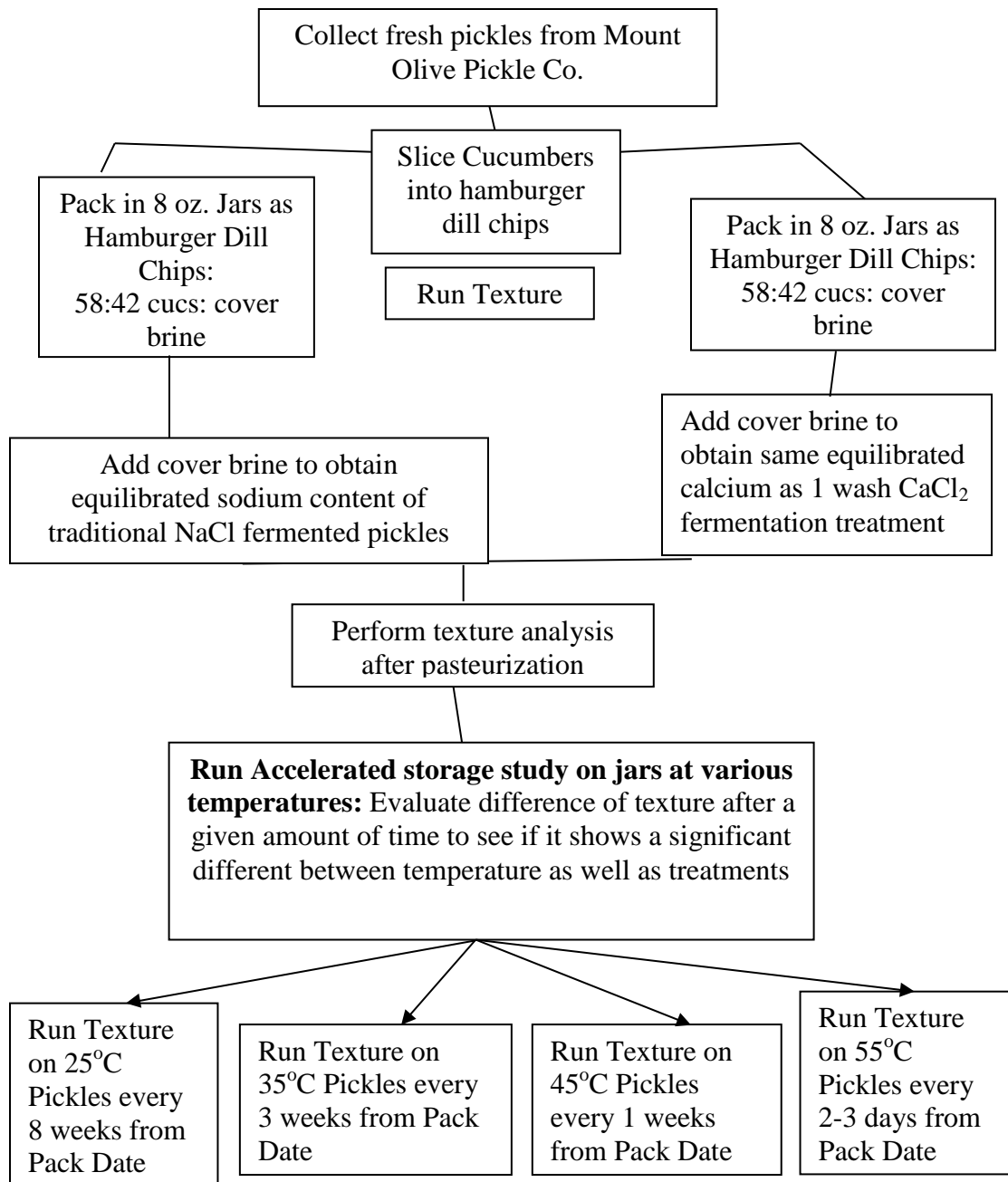
Figures

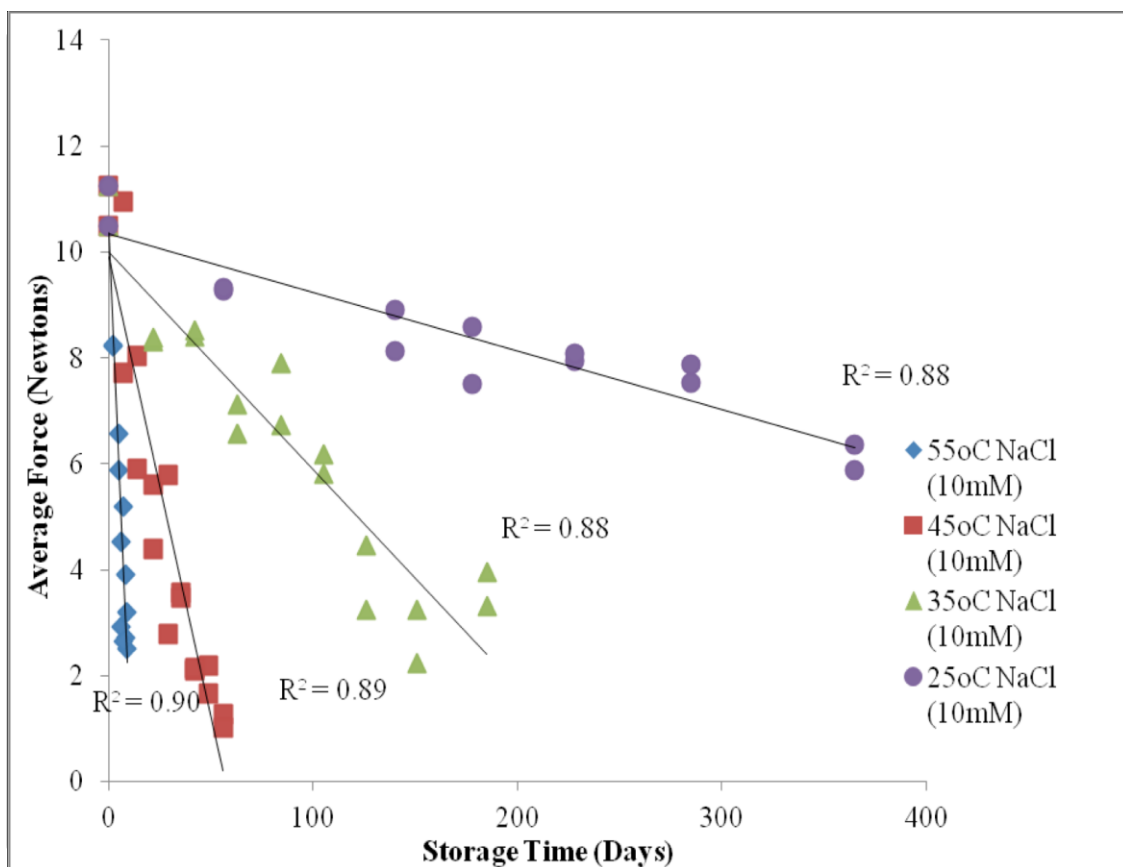Figure 4.1 Experimental design to sample the Fresh-Pack Fermented Cucumbers.

Figure 4.2 Accelerated Shelf Life Testing of Fresh-Pack Pickles that are packed traditionally with a calcium level of 10 mM.
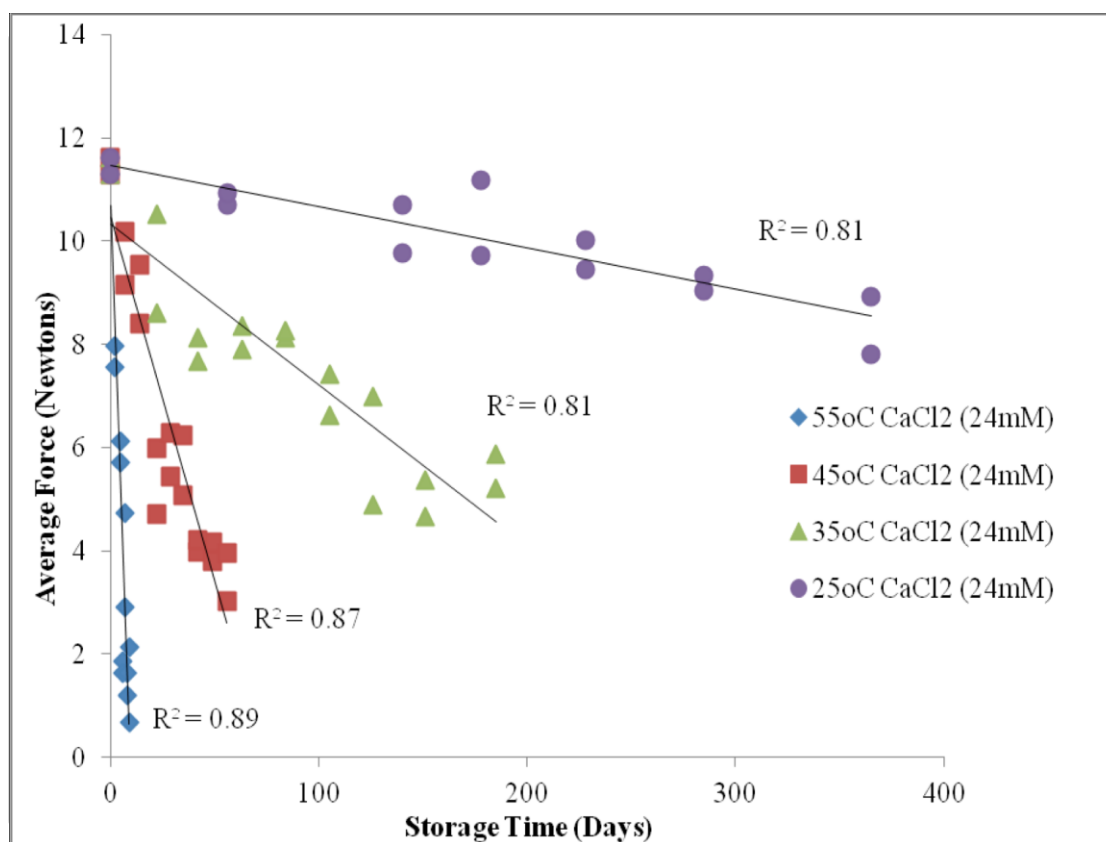
Figure 4.3 Accelerated Shelf Life Testing of Fresh-Pack Pickles that were packed with an enhanced calcium level of 24mM.
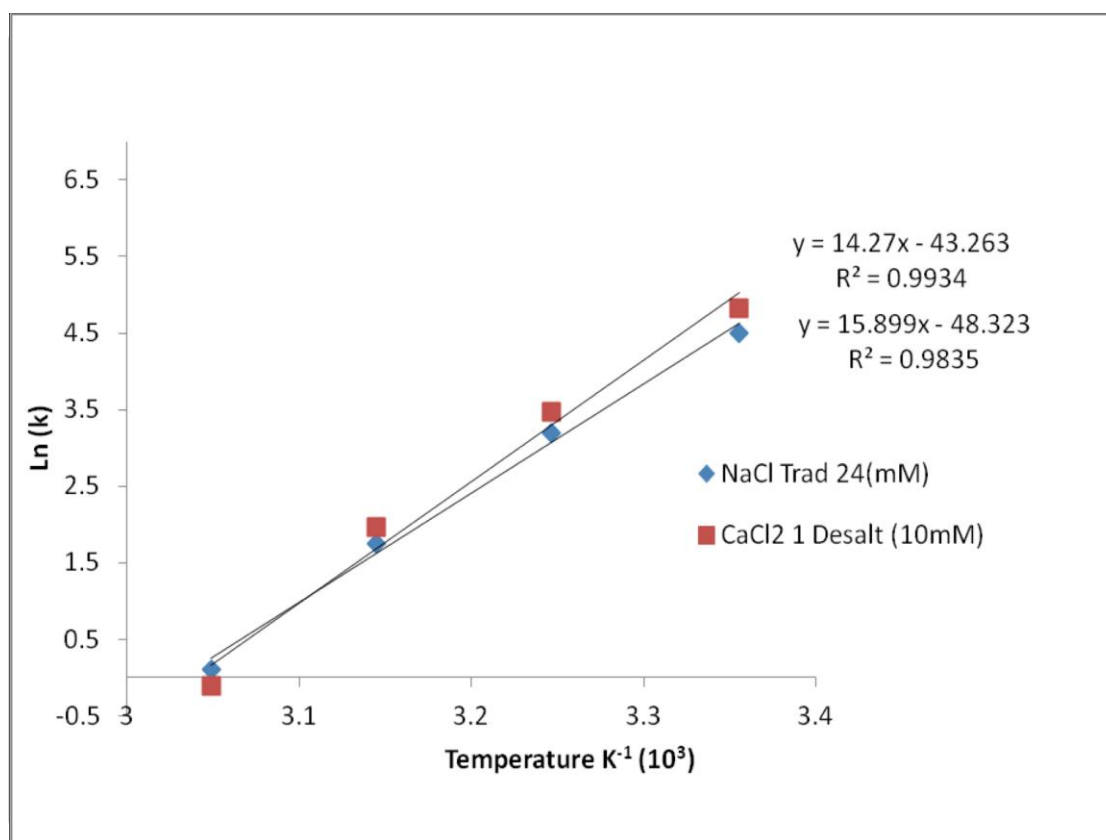
Figure 4.4 Shelf Life Plot of Fresh-pack cucumber pickles with 10 or 24 mM CaCl$_2$ at accelerated temperatures based on values of the mesocarp firmness.

Tables

Table 4.1 Formula for the traditional fresh-pack cucumber with a residual calcium pack out of 10mM.

INGREDIENTS	GRAMS	Percent by lbs (%)
Water	14756.70	90.57
200 Grain Vinegar	1140.07	4.12
Salt	710.76	4.23
Yellow Food Color Mixture	7.89	0.05
Polish Dill Emulsion	11.75	0.10
AntiFoam Mixture	6.55	0.04
Calcium Chloride	47.50	0.28
Benzoate with Color (same as Fresh Pack)	101.71	0.61

Table 4.2 Formula for the traditional fresh-pack cucumber with additional calcium chloride to have a residual calcium pack out of 24mM.

INGREDIENTS	GRAMS	Percent by lbs (%)
Water	14657.90	87.93
200 Grain Vinegar	1132.44	6.80
Salt	706.01	4.24
Yellow Food Color Mixture	7.84	0.05
Polish Dill Emulsion	11.70	0.07
AntiFoam Mixture	6.50	0.04
Calcium Chloride	159.54	0.96
Benzoate with Color (same as Fresh Pack)	101.02	0.61

Table 4.3 Definitions and Techniques used for Descriptive Sensory Analysis.

Attribute	Definition	Technique	Anchor(s) on Spectrum™ Scale
Hardness	Force necessary to compress the sample between the molars	Place sample between back molars and compress sample with molars.	Processed Cheese (1/2 in. cube) (4.5) Hebrew National Hotdog (7) Almond (11)
Fracturability	Force with which the sample ruptures when placing sample between molars and biting down completely down at a fast rate	Place sample between back molars and compress sample completely down at a fast rate.	Corn Muffin (1) Ginger Snap (8) Life Saver (14.5)
Crispness	The multiple, higher pitched sounds produced as the sample is crushed with the molar teeth – Evaluate on first 3 chews	Place sample between back molars and compress sample slowly. Repeat compression 3 times with the same sample.	Gala Apple, peeled (1/2in) (4) Water Chestnut (12)
Crunchiness	A single lower-toned noise produced with each chew. Evaluate intensity of crunchiness during the first 3 chews	Place sample between back molars and bite down quickly.	Peanut (4) Carrot (1/2in) (4)
Skin Awareness	The degree to which the outside skin of the product is perceived as intact pieces during mastication	Place sample between back molars and begin the mastication process.	Red Seedless Grape (10)
Seed Awareness	The degree to which the seeds within the product are perceived as intact pieces during mastication	Place sample between back molars and begin the mastication process.	½ cup Tomato Sauce with 1tsp. sesame seeds (4) Pomegranate Seeds (15)
Juiciness	The amount of juice/moisture perceived in the sample	Place sample between the back molar teeth and begin the mastication process.	Banana (1) Sugar Snap Peas (7) Orange Segment (15)

Table 4.4 Sensory texture attributes of fresh-pack pickles after one year of storage at ambient temperature.

Treatment	CaCl$_2$ (mM)	%NaCl	ST (^{o}C)	SS (days)	Hardness	Crispness	Fracturability	Crunchiness
Typical Calcium	9.9	2.0	25	365	6.1[a](2.0)	2.7[a](1.2)	3.8[a](1.5)	6.1[a](1.5)
Enhanced	23.5	2.1	25	365	7.6[b](1.4)	4.2[b](1.8)	4.8[b](1.6)	8.1[b](2.5)

ST= storage temperature (^{o}C), SS= shelf storage (days)

Means in the same column not sharing a common super script are different (p<0.05)

Intensities were scored on a 0 to 15-point universal scale where 0=none and 15=very high intensity (Meilgaard et al., 1991).

Values in parentheses are standard deviations.

Attributes not listed were not significantly different among treatments

Table 4.5 Texture degradation rates and shelf life for Fresh-Pack cucumber pickles stored at ambient and accelerated temperatures with varying levels of $CaCl_2$ in finished product.

Treatment	Temperature (^{o}C)	Equation	R_2	Slope	Estimated shelf life (days)
NaCl	25	y = -0.0111x + 10.346	0.88	-0.0111	298
NaCl	35	y = -0.0411x + 10.008	0.88	-0.0411	99
NaCl	45	y = -0.173x + 9.9009	0.89	-0.173	22
NaCl	55	y = -0.9021x + 10.359	0.9	-0.9021	4

Treatment	Temperature (^{o}C)	Equation	R_2	Slope	Estimated shelf life (days)
$CaCl_2$	25	y = -0.008x + 11.464	0.81	-0.008	526
$CaCl_2$	35	y = -0.0312x + 10.336	0.81	-0.0312	139
$CaCl_2$	45	y = -0.1403x + 10.462	0.87	-0.1403	31
$CaCl_2$	55	y = -1.1174x + 10.702	0.89	-1.1174	3

Notes: k Values are the average of the two replicates, for which the data are, in turn, the average of 15 measurements of firmness