USING INSTRUMENTAL AND SENSORY ANALYSIS TO INVESTIGATE THE FLAVOR OF DIFFERENT CUCUMBER VARIETIES AND THE IMPACT ON LEMON FLAVORED WATER

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UIJEONG AN, B.S.

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DEDICATION

To my parents, with love.

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ABSTRACT

UIJEONG AN

USING INSTRUMENTAL AND SENSORY ANALYSIS TO INVESTIGATE THE FLAVOR OF DIFFERENT CUCUMBER VARIETIES AND THE IMPACT ON LEMON FLAVORED WATER

MAY, 2020

Cucumber has unique flavors including green, fresh, fatty, and melon notes. Refreshing concept is trendy in the beverage market, while sugar reduction in processed foods and beverages is a prominent global trend in recent years. However, few studies have focused on the contribution of cucumber flavor to refreshing perception and its role in sugar-reduced beverages. The objective of this study was to investigate cucumber flavor composition and its impact on refreshing perception and other sensory properties of sugar-reduced lemon flavored water. An online survey was conducted to investigate the consumers' expectation and experience in flavored water. Responses from 906 participants indicated flavored water was ranked as the fourth most popular beverage. Lemon flavor was the most popular flavor (52.1%), while sugar was the major concern (47.4%) by consumers. Refreshing perception was selected as the most important function (87.4%). Therefore, sugar-reduced lemon flavored water with refreshing perception was chosen as the thesis research topic. Solid-phase microextraction-gas chromatography-mass spectrometry (SPME-GC-MS) was used to analyze volatiles in four types of cucumbers, resulting in 98 volatiles, with dominant volatiles of (E,Z)-2,6-nonadienal and (E)-2-nonenal. According to the results, a general cucumber flavor was formulated with the 10 most important cucumber volatiles. The impact of cucumber flavor on sugar-reduced lemon flavored water was investigated using descriptive sensory analysis and a consumer test. A total of six lemon flavored water with four sugar levels were formulated: 0% reduction (full sugar: 7%), 50% reduction, 50% reduction with added cucumber flavor, 80% reduction, 80% reduction with added cucumber flavor, and 100% reduction respectively. A total of 12 flavor descriptors were developed by 12 trained panelists. A significant difference (ANOVA, $p < 0.05$) of the sensory attributes across all samples were observed except lemon note. The lemon flavored water (zero reduction) dominated with juicy, lemon, refreshing, lemonade, and sweet flavor notes. The flavor profiles changed along with sugar reduction. The lemon flavored water lost the richness and balance as sugar was reduced, while irritate and bitterness showed up when sugar was reduced by 100%. However, with up to 80% sugar reduction, the flavor was still pleasant. Added cucumber flavor increased fresh, green, cucumber, and refreshing perception in both 50% and 80% reductions while being more effective in the 80% reduction (*t*-test, $p < 0.05$). The acceptance of six lemon flavored waters was evaluated by 100 consumers using 15 questions. Consumers perceived significant differences of all attributors across all samples (ANOVA, $p < 0.05$). The overall liking of five flavored waters, with the exception of the 100% sugar reduction, was a rating above 5 (either like or dislike) out of a 9-point hedonic scale, indicating these formulated flavored waters were acceptable. Adding cucumber flavors had an impact on overall flavor and refreshing intensities, but no impact on hedonic levels (*t*-test, $p < 0.05$). Knowledge gained in this study could provide insight in the potential for sugar-reduced drinks in the beverage industry.

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CHAPTER I

INTRODUCTION

Cucumber (*Cucumis sativus*) is widely cultivated and consumed in the United States. Cucumber originated in India but now grows widely in most countries (Mariod, Mirghani & Hussein, 2017). There are three types of cucumber: pickling, slicing, and seedless. Slicing cucumbers are grown for eating fresh and are generally grown in North America. These cucumbers are longer in shape, smoother in texture, have tougher skin and are more uniform in color. The English cucumber is one of the most popular slicing cucumbers. Pickling cucumbers are grown and bred specially for pickling. Pickling cucumbers are shorter in shape, have bumpy skin and more varieties in color, from yellow to dark green. West Indian gherkin is the most popular pickling cucumber. Other varieties of cucumbers, such as the burpless and seedless cucumbers, are sweeter in taste, have a thinner skin, and are usually grown in a greenhouse. Well-known burpless cucumbers in the U.S. market are Persian and Armenian cucumber (Food and Drug Administration [FDA], 2016).

Cucumbers have a fresh and unique flavor (Chen, Zhang, Hao, & Cheng,2015; Ligor & Buszewski, 2008). A total of 78 volatile compounds have been identified including aldehydes, esters, alkanes, alcohols, and furans (Hao et al., 2013). (E,Z)-2,6-nonadienal and (E)-2-nonenal are the major compounds that contribute to the unique flavor of cucumbers (Hao et al., 2013; Forss, Dunstone, Ramshaw & Stark, 1962; Palma-Harris, McFeeters & Fleming, 2001). Cucumbers have distinctive flavor notes such as aldehydic, green, fatty, floral, melon, and waxy. Nonanal and 2-tridecenal contribute aldehydic flavor, Z-3-hevenol and hexanol contribute green flavor, (E)-geranyl acetone contributes floral flavor, (Z)-6-nonenol and (Z)-6-nonenal contribute melon flavor (Hao et al., 2013; Forss et al., 1962; Palma-Harris et al., 2001). Not every volatile compound from the cucumber contributes to major cucumber flavor. Flavor profiles of cucumbers are impacted by volatile isolation and detection techniques. The different volatile isolation methods that have been used to extract aroma-active compounds in cucumbers include solid-phase microextraction (SPME), microdistillation, aroma extract dilution analysis (AEDA), and liquid-liquid extraction (LLE; Forss et al.,1962; Palma-Harris et al., 2001; Zawirska-Wojtasiak, Gośliński, Szwacka, Gajc-Wolska, & Mildner-Szkudlarz 2009). The most efficient and widely used method for the volatile extraction of fruits and vegetables is SPME because it is very sensitive, rapid, and solvent-free (Palma-Harris et al., 2001; Zawirska-Wojtasiak, et al., 2009). After isolation of the cucumber volatiles, gas chromatography-mass spectrometry (GC-MS) analysis is commonly used for volatile identification. Flavor reconstitution is an approach to verify the composition of the cucumber aroma profile as well.

The *refreshing perception concept* is trendy in the beverage market. According to Merriam-Webster Dictionary & Thesaurus, *refreshing* is defined as serving to restore strength and animation, to revive, to arouse, to stimulate, to run water over or restore water to, with thirst-quenching properties (Labbe et al., 2009 a). Refreshing perception is related to psychological and physiological enhancement including such components as thirst quenching, rehydration, energizing, and mental energy enhancement (Labbe et al., 2009 a).

There are two main characteristics that determine whether a beverage is refreshing, physical and ingredient attributes (Menayang, 2016). Ingredient attributes include fruits, citrus, herbs, botanicals, and spices. Moreover, Firmenich chose cucumber as “flavor of the year” in 2017 (Firmenich, 2016). According to Firmenich, cucumber is a “go-to” flavor for product developers because it drives the refreshment in beverage and sweet goods categories (Firmenich, 2016). Cucumber can be one of the fruits that contributes to refreshing perception.

Sugar is regarded as a major high caloric food ingredient that leads to obesity and overweight children and adults; therefore, sugar-reduced food and beverages are currently the most popular and interesting trend in the food industry. Few studies have been conducted to evaluate the impact of the sugar reduction without maintaining sugar intensity on consumer acceptance (Andersen, et al., 2017; Mielby, Viemose, Bredie, & Hyldig 2016; Wise, Nattress, Flammer, & Beauchamp 2016).

Sensory analysis, utilizing a quantitative descriptive analysis (QDA) and consumer testing to investigate consumers’ sensory perception are efficient methods to understand the link between flavor perception and cucumber flavors of a beverage. For the sensory analysis, different levels of sugar-contained lemon flavored water with cucumber flavor can be used to understand how cucumber flavor affects the sensory profile of sugar-reduced beverages and consumers’ acceptance. One study has been conducted to evaluate different variables of cucumber-based herbal beverage (Heena et al., 2017). Mixtures of different levels of herbal extracts in a cucumber-based beverage containing mint and coriander, 3-9% of total sugar and salt were analyzed for sensory attributes (Heena et al., 2017).

The objective of this study was to investigate cucumber aroma-active volatile compounds and refreshing perception since there are limited studies and a lack of information in this research area. In addition, evaluating the impact of the cucumber flavor on consumer acceptance of sugar-reduced lemon flavored water could provide insight and better understanding of beverages' refreshing perception.

Hypothesis

Cucumber flavor can enhance refreshing sensory perception on sugar-reduced lemon flavored water.

Specific Aims

1. To explore consumers' expectation of flavored water/beverages using an online survey.
2. To identify the volatile compounds in four cucumbers and reconstitute the cucumber flavor.
3. To analyze the sensory profile and consumers' acceptance of sugar-reduced lemon flavored water with cucumber flavors.

CHAPTER II

LITERATURE REVIEW

Cucumber General Characteristics

Cucumber (*Cucumis sativus*) is a member of the family of Cucurbitaceae, which is composed of 90 genera and 750 species (Tatlioglu, 1993). Cucumber is widely grown in all the countries of temperate zones and grows best at temperatures above 20 ℃ (Tatlioglu, 1993). Cucumber is one of the oldest cultivated fruit and originated in India over 5,000 years ago (Wehner & Robinson, 1991). It had spread to China, North Africa, and Southern Europe, and was introduced to Haiti by Columbus in 1494, and brought to the US. soon afterward (Wehner & Robinson, 1991). The U.S. National Plant Germplasm System (NPGS) at Ames, Iowa, is in charge of the cucumber collection in the US. The collections of the NPGS are composed of 1,314 cucumber accessions that represent the primary cucumber gene pool. The collections are primarily comprised of cultivars, land races, and varieties from the world (Wang et al., 2018). The US is the fourth largest producer after China, India, and Russia. Florida is the leading cucumber producing state for all types, while Michigan is the leading state for pickling cucumbers. Cucumber is ranked as the fourth most important U.S. fruit followed by tomato, cabbage, and onion (Tatlioglu, 1993). Cucumber is harvested in the US with around 15,450,000 hundredweight (cwt: 100 pound) harvested from 110,900 acres in 2018 (USDA, 2019b). To meet the demand of the US consumer, Mexico is the primary exporter during the winter season (FDA 2106).

Cucumbers are grown in either fields or greenhouses. Field-grown cucumbers are typically planted as seed and grow on poles or trellises, and are grown for the fresh or sliced market. In contrast, greenhouse cucumbers are typically established as transplants. Greenhouse cucumbers have very large leaves, grown vigorously, and are always grown on a trellis. Those greenhouse cucumbers require close monitoring for good health and productivity (FDA 2016).

The three main types of cucumbers are *slicing, pickling,* and *burpless* (Mariod, Mirghani, & Hussein, 2017). Slicing cucumbers are grown in North America and generally grown for consuming fresh. The features of these cucumbers are longer in shape, softer in texture, tougher in skin and more uniform in color. The English cucumber is one of the most popular slicing cucumbers. Pickling cucumbers are grown specially for uniformity of size and texture for making commercial pickle. Pickling cucumbers are shorter in shape, have bumpy skin and more varieties in color, from yellow to dark green. The most popular pickling cucumber is *West Indian gherkin*. The burpless and seedless cucumbers, are sweeter in taste, have a thinner in skin and are usually grown in a greenhouse. The Persian and Armenian cucumber are well known burpless cucumbers in the U.S. market (USDA, 2016).

The cucumber is commonly consumed fresh in a salad and is valued mainly for its crisp texture and juiciness or fermented (pickled) flavor. Due to the cucumber’s low calories, a cucumber is regarded mainly as a refreshing condiment (Tatlioglu, 1993). One hundred grams of raw cucumber contains 15 kcal energy, 95 g water, 0.65 g protein, 0.11 g fat, 3.63 g carbohydrate, 0.5 g fiber, 0.03 g sugar, 105 IU vitamin A, 2.8 mg vitamin C, 16 mg calcium, and 0.28 mg iron (USDA, 2019a). Cucumber is regarded as a good source of dietary and therapeutic functions including cancer prevention, skin improvement, antioxidant, and anti-inflammatory roles, diabetes prevention, and lowering cholesterol levels (Mukherjee, Nema, Maity, & Sarkar 2013). Thus, cucumber consumption helps in weight management, body hydration, improved skin condition, and prevention of infection (Mukherjee, et al., 2013).

Flavor Components in Cucumber

Cucumber is a widely consumed fruit due to its freshness and distinct flavors (Palma-Harris, et al., 2001). To date, 78 cucumber volatiles have been identified including aldehydes, alkanes, alcohols, esters, and furans (Hao et al., 2013). Studies have investigated the major cucumber volatile compounds and their role during fruit development and the relationship between the volatile compounds and gene expression or enzyme activity (Chen et al., 2015; Ligor & Buszewski, 2008). Lipoxygenase (LOX) and hydroperoxide lyase (HPL) are enzymes from the cucumber that produce its unique aroma. Using the hydro backbone of linoleic and linolenic acid, molecular oxygen at C9 (9-LOX) and C13 (13-LOX) are introduced by the LOX enzymes. Following oxygen introduction, 13-hydroperoxylinolenic acid (13-HPOT) or 9-hydroperoxylinolenic acid (9-HPOT) are formed then cleaved by HPL in different metabolic pathways (Matsui et al., 2006). The main flavor components of the cucumber fruits are C6 and C9 aldehydes and alcohols (Chen et al., 2015). Hexanal, hexanol, (E)-2-hexen-1-ol, and (E)-3-hexen-1-ol are C6 aldehydes and alcohols that contribute grassy and green flavors of the cucumber (Chen et al., 2015). Whereas (E,Z)-2, 6-nonadienal, (E)-6-nonenal, (Z)-6-nonanal, (Z)-6-nonen-1-

ol, (E,Z)-2,6-nonadien-1-ol, and (E,Z)-3,6-nonadien-1-ol are C9 aldehydes and alcohols responsible for melon-like or fatty flavor. (E,Z)-2,6-nonadienal and (E)-2-nonenal have been determined to be the major compounds that contribute to the unique flavor of cucumber (Hao et al., 2013; Forss et al., 1962; Palma-Harris et al., 2001).

Cucumber Flavor Analysis Methods

There are several techniques used to extract cucumber volatiles including SPME, AEDA and LLE (Forss et al., 1962; Palma-Harris et al., 2001; Zawirska-Wojtasiak et al., 2009).

SPME has been widely used to extract volatile compounds from food samples (Vas & Vekey, 2004). SPME is a solid-phase extraction technique where analytes in the sample are directly extracted and concentrated to the SPME fiber from the fruit. It extracts volatile and non-volatile analytes from liquid or gas phase (Mitra, 2004; Spietelun et, al., 2010). After extraction, the SPME fiber is thermally desorbed to the GC-MS for analysis. SPME is considered the most efficient extraction method because of its advantages in sensitivity, time reduction, and lack of solvents (Palma-Harris et al., 2001; Zawirska-Wojtasiak et al., 2009). Other volatile isolation methods such as microdistillation and AEDA have been used to extract and identify cucumber aroma compounds for (Zawirska-Wojtasiak et al., 2009; Schieberle, Ofner, & Grosch, 1990). The microdistillation method provides microscale level distillation by using capillary technique. For cucumber aroma volatile isolation, ester-pentane (1:1, v/v) is used as the extraction solvent. After distillation, samples are injected into a GC-MS for volatile compound analysis. The AEDA is a common odor evaluation method for food as well. AEDA results in combined hedonic response measurement (CHRM) values or flavor dilution (FD) factors of the flavor volatile compounds (Schieberle et al., 1990). The FD factors have been evaluated in cucumber odorants (Schieberle et al., 1990).

Gas chromatography is widely used for aroma compound separation. GC is an ideal piece of equipment to use for aroma studies as it can separate complex mixtures of volatile extracts via high-resolution capillary columns (Pavia, Lampman, Kriz, & Engel 2006). Chemical compounds are sequentially eluted based on the general order of their boiling point and/or polarity. Samples flow through a narrow tube in a gas stream, known as the mobile phase, generally consisting of either helium or nitrogen, allowing different chemical compounds to be absorbed and desorbed by the stationary phase (Grob, 2016). Stationary retention allows different compounds to exit the end the column at a different times based on their retention times. As column temperature increases, the volatile compounds are thermally desorbed from the stationary phases. Mass spectrometry as a detector is widely used in flavor research to identify volatile compounds. MS generally coupled with GC allows for more sensitivity, providing more structural information than any other detectors. For volatile compound identification, the National Institute of Standards and Technology (NIST, USA) mass spectra library of alkane standards which contains C5 to C25 compounds, and a linear retention index (LRI) is the ideal source to use when identifying components of the cucumber volatiles (Chen et al., 2015; Li et al., 2019; Ligor & Buszewski, 2008).

Flavor reconstitution verifies the link of the sensory attributes with aroma-active compounds, while the Odor Activity Value (OAV) could be used to estimate the importance of an aroma compound to the sensory character of a food by calculating the ratio of the concentration of a compound in a food to its sensory threshold in that food. OAV has been used in many studies on foods such as ice wine and tuber fruits (Lan et al., 2019; Liu, Li, Li & Tang, 2012). In order to determine the OAV, each aroma-active compound obtained from volatile quantification using GC-MS is plotted in a calibration curve of reference substances. Individual volatile compounds with an OAV > 1 are blended to create flavor (Liu et al., 2012). Aroma reconstitution was conducted based on the quantitative analytical data of the aroma-active compounds. To date, cucumber flavor reconstitution has not been evaluated.

Sugar Reduction and Refreshing Perception of Beverage

Sugar is considered a major high caloric food that contributes to obesity and overweight children and adults; therefore, sugar-reduced food and beverages are currently popular trends in the food industry. The 2015 Dietary Guidelines for Americans recommends limiting the intake of added sugars in the American diet with American adults now using more low-calorie, reduced sugar, and/ or sugar-free foods and beverages (Millen et al., 2016*)*. Sugar-sweetened beverages are a major source of free sugar intake in both children and adults; however, reducing or removing sugar in the food and beverages is challenging because of its sugar function. Sugar plays an important role in a beverage. It not only contributes to the sweet taste, but also suppresses bitterness and sourness while enhancing the flavor intensity (Ashurst, 2016). The sugar reduction in foods is challenging as it would change flavor and texture balance, food functionality, shelf-life, and cost (Markey, Lovegrove & Methvene, 2015).

A few of the studies have investigated the impact of lowering sugar concentrations and consumer liking across different food categories, such as fruit drinks (Andersen et al., 2017; Mielby et al., 2016; Wise et al., 2016), fruit nectars (Oliveira, Ares, & Deliza, 2018a), and dairy products (Alcaire, Antúnez, Vidal, Giménez, & Ares 2017; Oliveira, et al., 2015; Yoo et al., 2017). Most studies show that reducing the sugar decreases the palatability and consumer acceptance, especially when sugar is reduced rapidly. The impact of sugar reduction on consumer acceptance and preference depends on many factors such as type of products, the level of reduction, aroma, color, product image, package claims, and consumer demographics (Stampanoni, 1993; Wise et al., 2016).

There have been various attempts to reduce sugar consumption including governmental regulation through nutrition labels, application of a sugar tax in several states in the US and voluntary commitment from various ingredient suppliers in the food industry. There are several strategies for the sugar reduction. One strategy has been a gradual sugar reduction (Hutchings, Low & Keast, 2018) through a progressive reduction in sugar over a long period of time to reduce the level of sweetness desired by the consumer (Markey et al., 2015). A slow reduction allows people to slowly adjust to the taste of less sugar. However, this strategy takes a long period of time and is hard to maintain in a lab setting to influence real life consumption. A second strategy is to reduced sugar but maintain sweetness (Hoppert, et al., 2013). This strategy is commonly used in the industry. Without sacrificing sweet taste, sweetness substitutes, flavor enhancers, and the modification of food structure are used to modulate sweetness (Anderson et al., 2017; Ashurst, 2016; Mielby et al., 2016; Ashurst, 2016; Pineli et al., 2016). The last strategy is sugar reduction in a short period of time without maintaining sweetness intensity. To date, few studies have investigated sugar reduction on orange nectars, chocolate-flavored milk, and orange/passion fruit nectars. There are very limited studies on consumer acceptance of flavored water/beverage without maintaining sweetness (Oliveira et al., 2016; Oliveira et al., 2018b; Pineli et al., 2016).

Refreshing perception is related to psychological and physiological factors including thirst quenching, rehydration, energizing, and mental energy enhancement (Labbe et al., 2009 a). Refreshing is defined as serving to restore strength and animation, to revive, to arouse, to stimulate, to run water over or restore water to, with thirst-quenching properties, according to Merriam-Webster Dictionary& Thesaurus (Labbe et al., 2009a). Specific sensory properties of food and beverages favoring refreshing perception have been identified. There are three stimulations of refreshing perception, trigeminal, taste, and flavor, which include cooling, temperature, $CO_{2,}$ and olfactory stimulated flavors (McEwan & Colwill, 1996; Labbe et al., 2009a; Labbe et al., 2009b). Refreshing effects correlate with multiple sensory attributes such as appearance, odor, taste, texture, or mouthfeel, instead of a single stimulus (Labbe et al., 2009b).

Cucumber Flavor in Sugar Reduced Beverage

Adding secondary ingredients to beverages such as energy, antioxidants, refreshing effect, and thirst quenching are considered beneficial. One study has been conducted to investigate what makes a beverage refreshing. The Symrise research team identified the key driver of refreshing perception in beverages including beer, cider, flavored alcoholic beverages, flavored water, juices, soft carbonated drinks, ready-to-drink tea, and sport drinks (Menayang, 2016). There are two main characteristics that determine whether a beverage is refreshing, physical and ingredient attributes (Menayang, 2016). Physical attributes such as temperature, liquid color, liquid texture, carbonation, and sound are contributors to refreshing, while ingredient attributes include fruits, citrus, herbs, botanicals, and spices.

Lightly sparkling, light-colored, citrusy, and the sounds of opening a can or bottle is considered refreshing to millennial consumers (Menayang, 2016). Moreover, Firmenich chose cucumber as "flavor of the year" in 2017 (Firmenich, 2016). According to Firmenich, cucumber is a "go-to" flavor for product developers that drives refreshment in beverages and sweet goods (Firmenich, 2016).

Sensory Analysis Methods and Application of Cucumber Flavors

Cucumbers have distinct flavors such as aldehydic, green, fatty, floral, melon, waxy, etc. (Chen et al., 2015; Forss et al., 1962; Palma-Harris et al., 2001). Nonanal and 2-tridecenal contribute aldehydic flavor, Z-3-hexenol and t-2-hexenol contribute green flavor, (E)-geranyl acetone contributes floral flavor, while (Z)-6-nonenol and (Z)-6-nonenal contributes melon flavor (Chen et al., 2015; Forss et al., 1962; Palma-Harris et al., 2001). Not every volatile compound from the cucumber is necessary to contribute the major cucumber flavor. Several organoleptic sensory attributes are related to cucumber flavors such as cucumber-like, green, off-odor, cucumber-like taste, sweet taste, and bitter taste for QDA as well as general acceptance and hedonic liking in a consumer test (Zawirska-Wojtasiak, et al., 2009). QDA and consumer tests are appropriate techniques to investigate cucumber aroma-active compounds affecting a sensory profile.

QDA is commonly used in flavor research to investigate the sensory profile of foods and beverages. Generally, 10-12 well-trained panelists, who participated in this consumer test can discriminate differences in sensory properties among various products. Panelists decide the lexicons and standardized vocabulary for products by consensus. By using uninstructed line scale, panelists judge the intensity of descriptors based on reference standards. Moreover, panelists perform independent judgment with results not being consensus derived. Descriptor vocabularies are developed by panelists and with the results not influenced or biased by a panel leader. For the data analysis, spider-web chart and analysis of variance (ANOVA) are used which can differentiate the samples by variances (Meilgaard, Carr, & Civille, 2019).

A consumer test is an appropriate sensory evaluation technique that can be applied to pre-market concept testing, new product prototype explorations, pre-market blind labeled testing, product optimization, assessment of market potential, product improvement, product maintenance, product category review, and support for advertising claims. The consumer test investigates consumers' acceptance (degree of liking) or preference of product. Participants can be employees, local area residents, or the general population all of which can be decided by the research purpose. Generally, 75-100 consumers are recruited per test and screened for product use frequency. For a consumer test, a 9-point hedonic scale and just-about-right scale are commonly used (Meilgaard et al., 2019).

Both sensory analysis and consumer test would investigate the effect of lemon-flavored water incorporated with cucumber flavor with different sugar levels on refreshing perception. To date, only one study has evaluated cucumbers' sensory attributes in products incorporated with herbal-blended extract, different levels of sugar, and salt (Heena et al., 2017). However, the sensory analysis only focused on overall acceptability with different variables to optimize the herbal- blended beverage (Heena et al., 2017).

Summary of Literature Review

Overall, according to the literature, cucumbers are widely consumed fruit in the US that provides beneficial dietary and therapeutic functions. Despite exploration of cucumber in agriculture, comparatively very few studies have been published about the chemical profiles and sensory properties of cucumber and its potential use in the food industry. This thesis research filled a pore of this gap. By evaluating the volatile compounds of cucumber, this research can link the refreshing perception of the cucumber to aroma volatile components. In addition, the QDA and a consumer test were used to investigate cucumber sensory profiles and consumer's preferences of cucumber flavor used in sugar-reduced lemon flavored water.

CHAPTER III

METHODOLOGY

This thesis research included human subjects for an online survey, descriptive sensory analysis, and a consumer test. Institutional Review Board (IRB) application form, consent form, email script, and demographic request were submitted to the IRB at Texas Woman's University (TWU). The IRB applications were approved on October 1, 2018 for sensory analysis and consumer test, and April 19, 2019 for online survey.

Online Survey

To investigate the consumers' expectation in sugar-reduced flavored water/beverages, an online survey was conducted utilizing Google Form. Nine questions related to flavored water beverages and seven demographic questions were included in this online survey (see Table 1). Participants voluntarily completed the survey. The survey was opened to TWU faculty, staff, and students and non-TWU individuals. The survey could be completed in approximately 5-10 minutes. A recruitment email was sent out internal to TWU email and flyers were posted and distributed across TWU Denton campus. After the completion, 10 participants were randomly selected and received a $ 20 Amazon gift card.

SPME-GC-MS for Cucumber Volatile Analysis

Four types of cucumbers including English, American, Kirby, and Persian were purchased from local grocery stores. Whole sized cucumbers were cleaned and skins were peeled off for quality assurance. The peeled cucumbers were cut into pieces and homogenized in a Waring blender. The cucumber puree was transferred to 20 mL SPME vials that were flushed with nitrogen gas for 10 minutes to eliminate extra oxygen. Sodium chloride (NaCl) was added to the cucumber puree to increase the release of water- soluble components for SPME absorption.

SPME fiber coated with divinylbenzene/carboxen/polydimethylsiloxane at a 30/50 μm film thickness was used for volatile extraction at 40℃ for 30 minutes with a SPME auto-sampler following initial sample equilibration at 40℃ for 15 minutes. After absorption, the SPME fiber was thermally desorbed into GC injection port for analysis.

GC-MS (SHIMADZU, GCMS-QP2020) was analyzed using a DB-WAX column that contained polar stationary phase. Samples were desorbed for 1 minute. The initial oven temperature was 40℃, increased to 230℃ at a rate of 5°C/min, and held 3 minutes at the final temperature requiring a total analysis time of 44 minutes. Compound identification was based on matching retention times with standard linear retention indices (LRIs) which were calculated using a series of standard alkanes C5-C25. All samples were analyzed in triplicate.

Reconstitution of Cucumber Flavor

The commonly shared aroma-active volatile compounds were selected according to the data of the volatiles identified in four cucumbers by GC-MS and previously identified in the literature. The average percentage of peak areas of triplicated data of each aroma-active compound was calculated. The major volatiles were selected to reformulate the cucumber flavor. After the volatile compounds were selected, stock solutions for each compound was created. Individual stock solutions were blended into a volumetric flask and diluted with propylene glycol (PG) to create a cucumber stock solution. This reformulated cucumber stock solution was a cucumber flavor solution. Different ratios of cucumber stock solution and water were formulated to mimic best cucumber flavors.

Lemon Flavored Water Formula

All flavored water samples were made in the sensory lab at TWU, Denton, TX. Ingredients were added by an order corresponding to preservatives (sodium benzoate: 0.01%, potassium sorbate: 0.012%) followed by sugar (sucrose: 7%, 3.5%, 1.4% of 0%), acid (citric acid: 0.05%), and then lemon flavors (0.15%). The overall procedure was first water was measured by a graduated cylinder and transferred to a beaker and then preservatives were added including potassium sorbate and sodium benzoate. Preservatives were dissolved completely using a stir plate, then sugar was added. After sugar dissolved completely, acids including citric acid, ascorbic acids, sodium citrate were added. Then flavors were added and mixed completely. After the mixture was ready, a 50 mL portion was transferred to a 300 mL glass bottle with 250 mL of spring water added. The cucumber flavor from Firmenich was added in 50% and 80% sugar-reduced samples with the levels at 15ppm. The prepared flavored water was stored in the refrigerator (4ºC) at least one day to stabilize.

Quantitative Descriptive Analysis

QDA was conducted at in the TWU sensory lab. A total of 12 panelists were recruited, each going through four training sessions. During the first section, panelists were familiarized with six lemon flavored water samples each having four different sugar levels and two having cucumber flavor added (0%, 50%, 80%, 100% reduction, 50% reduction with cucumber flavor, and 80% reduction with cucumber flavor). Panelists shared their opinions about the flavor perception of samples. All samples were given in an ascending order of sugar content so that panelists would taste the least sugar to highest sugar level. Panelists identified all descriptors they could distinguish on a list and wrote down extra descriptors not on their list. The descriptors for lemon flavored waters were finalized, which included eight flavor descriptors and five taste descriptors: refreshing, lemon, fresh, green, juicy, lemonade, cucumber, citrus, sweet, sour, bitter, astringent, and irritate.

During the second section, all standards/references were prepared and given to the panelists. The panelists practiced and kept calibrating themselves with the standards. All reference standards were anchored to an intensity level of five out of ten. After panelists practiced all standards, they were given six lemon flavored water samples to compare side-by-side with standards. Samples were given in a descending order of sugar contents so panelists could taste in an order opposite from the first section. Panelists compared perceptions from the first section as flavor perception was affected by the order of sugar levels.

At the third section, samples were served in random order. By tasting randomly ordered samples, panelists could simulate the real QDA test with a test ballot using the 10-point line scale.

The panelists signed up (consent form and schedule) for three sections of the test, resulting in a triplicate test for each sample by each panelist. All samples were randomized using three-digit codes. All products were served in 2-oz plastic cups covered with lids. Samples were served cold (4ºC). During the test, panelists received samples, a test ballot, a cup of water, and unsalted Saltine crackers as a palate cleanser. The tests were conducted in isolated booths illuminated with incandescent lightning. Panelists were instructed to rinse their mouth between samples with water. $20 cash was provided as sensory incentive after completion.

Consumer Test

For the consumer test, 100 participants were recruited. An internal TWU email was sent to all TWU students, staffs, and faculty. Social Network Services (SNS) including Facebook and Instagram were used to recruit from external sources. Flyers were printed and spread over the TWU Denton campus. In addition, the consumer test was advertised in large sized classes at TWU. To participate in this consumer test, panelists had to pre-register and were selected to take the test based on their eligibility. Panelists were excluded from the test if they consumed flavored water less than 1~4 times a month.

The test ballot was designed including 16 questions of liking, intensity levels, and CATA (Check-All-That-Apply). Overall character liking was asked using a 9-point category scale with the following indicators: extremely dislike, dislike very much, dislike moderately, dislike slightly, neither like nor dislike, like slightly, like moderately, like very much, and like extremely. Overall flavor liking, intensity and flavor balance liking were asked using the same category scale. Other questions included the liking and intensity of lemon, refreshing, mouthfeel, sweetness, and sourness. At the end of the test, demographic questions were added, which identified a panelist's age group, gender, frequency of purchasing flavored water, frequency of drinking flavored water, type of flavor panelists mostly consumed, tendency of seeking new products, allergies, personal health condition, and their Body Mass Index (BMI).

Pre-registered and qualified panelists were invited to participate in the consumer test at their scheduled time. Panelists were asked to fill out the consent form, which included information explanating the purpose of the research, description of procedures, potential risks, and benefit of participation. Panelists' height and weight were recorded to allow for BMI calculation.

Six samples were randomized using a three-digit code. All products were served in the 2-oz plastic cups covered with lids and chilled to 4ºC. During the test, panelists received samples, an iPad with Compusense software, a cup of water, and unsalted Saltine crackers as a palate cleanser. The tests were conducted in isolated booths illuminated with incandescent lightning. Panelists were instructed to rinse their mouth between samples with water. Tests took approximately 20 minutes to complete. The demographic form was collected at the end of each test. Each panelist typed in their BMI from the demographic questionnaire according to the measurements. Participants were compensated with $5 cash and two pieces of chocolate were distributed as an incentive.

Statistical Analysis

All data were collected and analyzed statistically. Mean score and standard error were calculated. An ANOVA was conducted to compare each sample by descriptors. Tukey's honest significant difference (HSD) was performed for the pairwise comparisons with $\alpha = 0.05$ IBM SPSS Statistics Version 25. Principal component analysis (PCA) was performed on the mean data of each descriptors to account for the variation between six samples using XLSTAT 2015.

CHAPTER IV

RESULTS

Online Survey Results

A total of 906 participants took the survey from April 18 to June 16, 2019. Participants were asked to answer questions related to their expectation and experience of flavored water and beverage consumption. A total of 16 questions were included in the survey, including 9 questions related to the expectation of flavored water and beverage and 7 questions for the demographic information (see Table 1). According to survey results, plain water was selected most often by consumers (91.4%), followed by tea (63.5%), coffee (60.5%), flavored water (41.3%), and others (see Figure 1). Flavored water was ranked fourth among the 10 drink and beverage options evaluated. This data indicated flavored water is well accepted and consumed as a regular drink and beverage option.

Consumer expectations of flavored water and beverages, consumers identified a preference for the refreshing effect (87.4%) most often, followed by thirst quenching (73.7%), tastes (65.7%), extra benefits (17.1%), and aroma (11.1%; see Figure 2). These results indicated that refreshing perception, thirst quenching perceptions, and tastes were considered the most important factors to meet consumers' expectation of flavored water and beverages. The expected extra benefits to be added for the flavored water were vitamin and minerals (73.3%), antioxidants (60.8%), energy (53.5%), refreshing (47.4%), less and/or non- sugar products (44%), protein (30.1%), and fiber (20.6%; see Figure 3). These results also identified refreshing is highly expected. These results indicated that refreshing perception was one of the major factors that consumers considered for flavored water and beverage consumption.

When preferred flavor and tastes were evaluated, lemon (52.1%), berry (36.6%), lime (31%), and tropical flavor (28.3%) were the top four flavors consumers preferred, although plain water received a rating of 42.7% (see Figure 4). Other flavors such as watermelon, peach, cherry, grapefruits, herb, orange, strawberry, vanilla, and cucumber were also selected by some participants. This data indicates that flavors from the citric family were more preferred than others. In contrast, all bitter (80.2%), astringent (68.1%), irritate (75.7%), and sour taste (57%) were considered as negative attributes that consumers did not like in flavored water/beverage (see Figure 5).

Other factors determining consumers' choices for flavored water and beverages were also evaluated. Temperature (62.4%) was the highest rated affective factor when consumers choose their beverages, followed by flavors (52.4%), sweet taste (47.4%), carbonation (37.4%), cooling taste (35.2%), and sour taste (8.5%; see Figure 6). Sour taste was least preference by consumers.

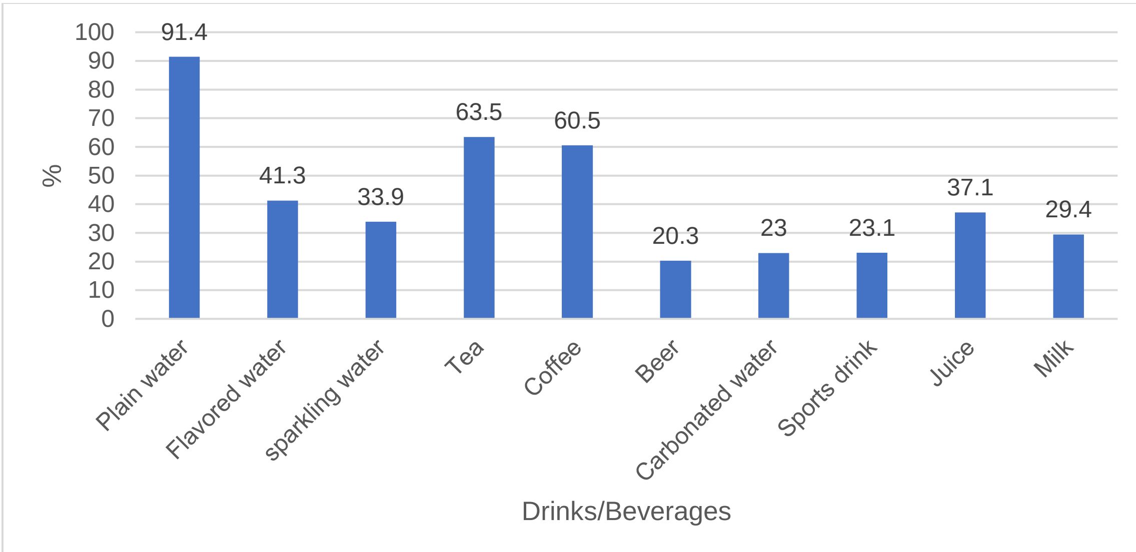
Consumer preferences for sweetness perception of flavored water were evenly distributed. Somewhat sweet (26.8%), little sweet (25.4%), sweet (24.1%) and non- sweet (24.1%) showed similar preference. However, very sweet received 9.8% response while extremely sweet only received 0.8% responses (see Figure 7). These results indicate that consumers preferred the sweet taste over non-sweet to sweet, but no more than very sweet. The specific reasons that consumers' preferred reduced sugar products were taste (59.4%), calories (57.3%), and their health conditions (40.5%; see Figure 8). Consumers preferred sweeteners such as plain sugar (54.2%), Stevia (28.8%), agave syrup (21.1%), and honey (54.2%) over alternative sweeteners such as Splenda (22.4), aspartame (6.4%), and sucralose (4.3%; see Figure 9).

Table 1
Online Survey Questions

Flavored water/beverage questions	
1	What type of drinks/beverages do you usually drink?
2	Which types of flavored water/beverages do you consume the most?
3	Which of the following factors do you consider the most when deciding flavored water/beverage to drink?
4	Which of the following attributes that you do not expect of the flavored water/beverage?
5	How much of sweetness do you like the most for your regular beverage consumption?
6	Which sweeteners do you prefer to eat or drink with?
7	Which effect would you expect the most from the flavored water/beverage?
8	When you choosing sugar-reduced products, what factors do you consider the most important?
9	If you can buy the beverages with additional benefits, what benefits would you like it be added?
Demographic questions	
1	Gender
2	Age
3	Education level
4	Employment status
5	How would you rate your own personal health?
6	How often do you purchase flavored water/beverage?
7	How often do you drink flavored water/beverage?

Figure 1

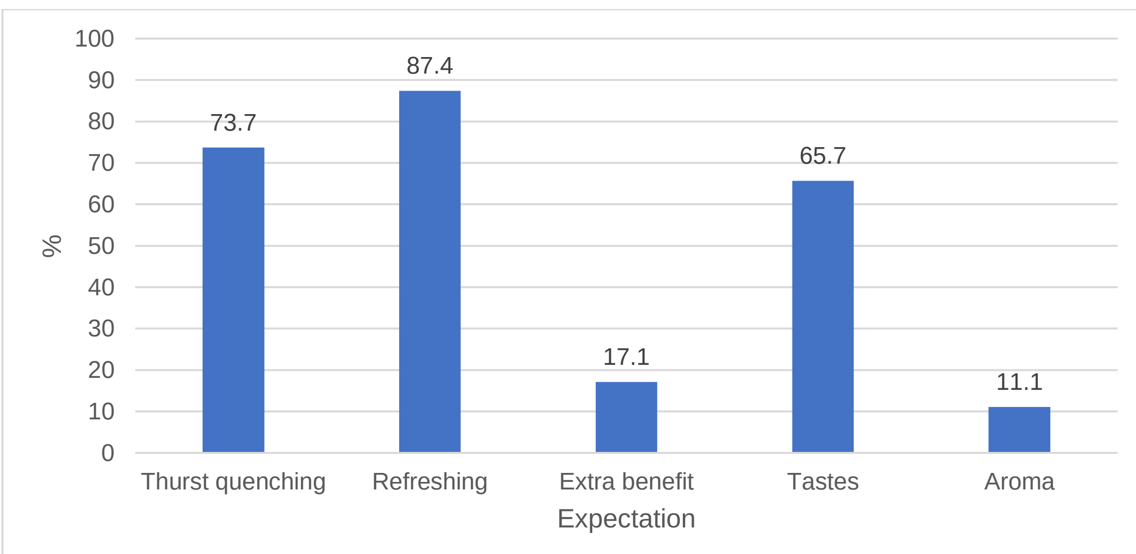
Type of Drinks/Beverage that Consumers Drink the Most



Note. Others: Soda, diet soda, wine, coconut water, kombucha, and non-dairy milk

Figure 2

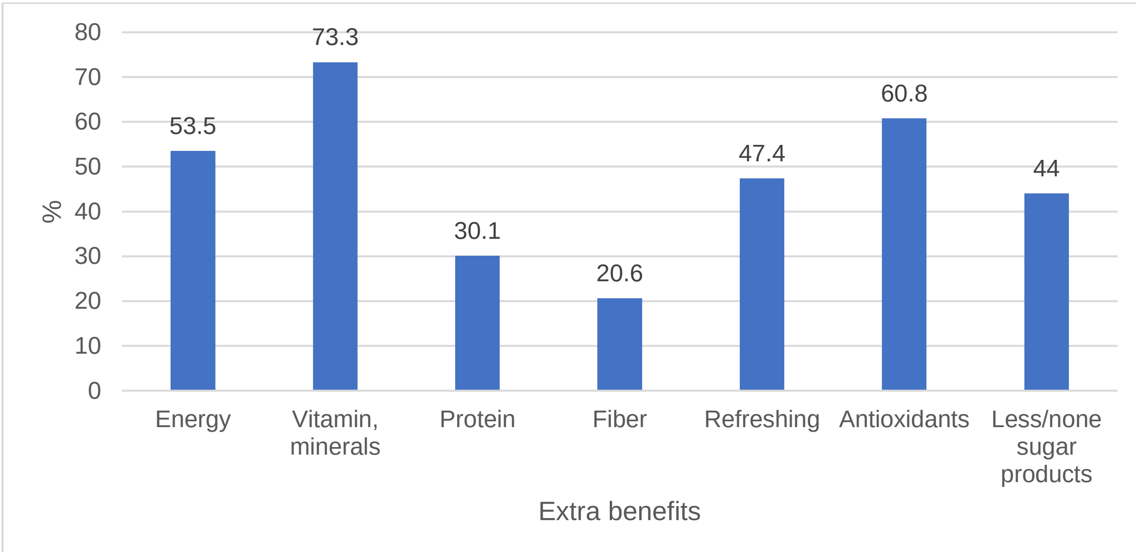
Expectation of Flavored Water/Beverage



Note. Others: Health, low calories, relieves headache, body hydration, and electrolytes

Figure 3

Extra Benefits that Consumers Expect to be Added



Note. Others: Grapefruits, strawberry, orange, passion fruit, acai, coconut, herb, pomegranate, cucumber, vanilla, and ginger

Figure 4

Types of Flavor that Consumers Drink the Most

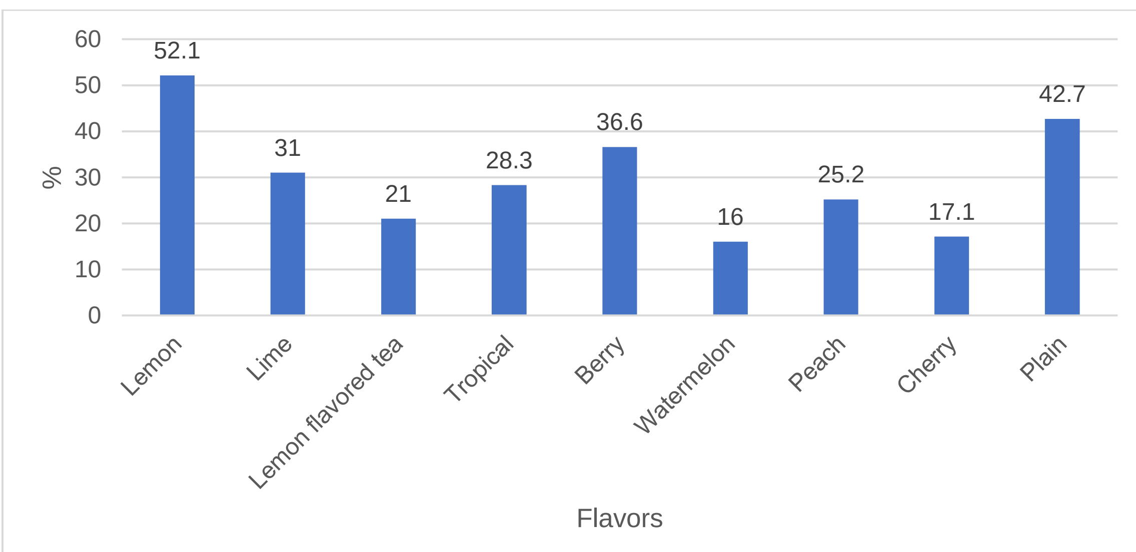
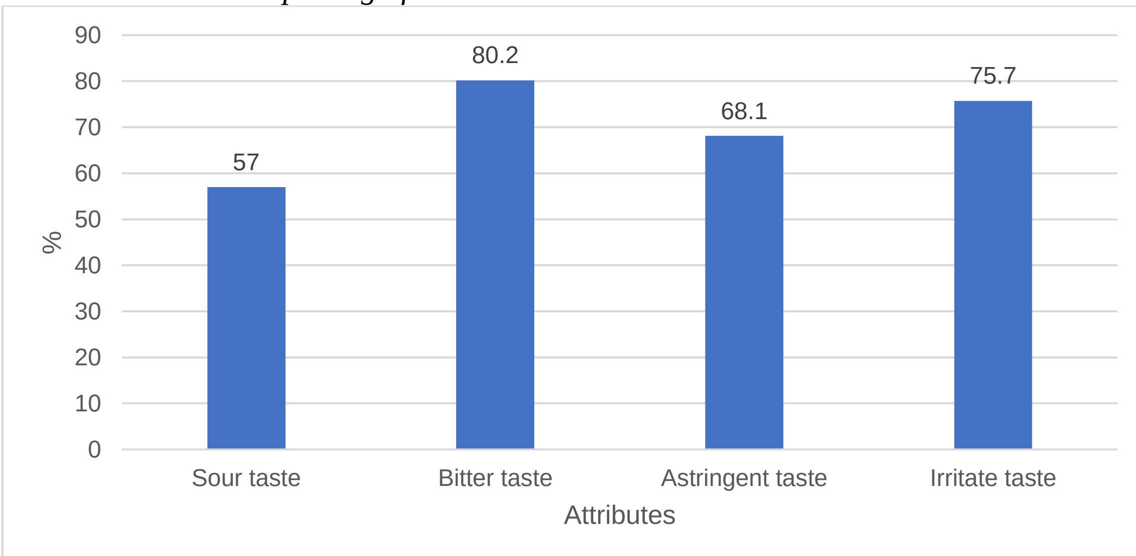


Figure 5

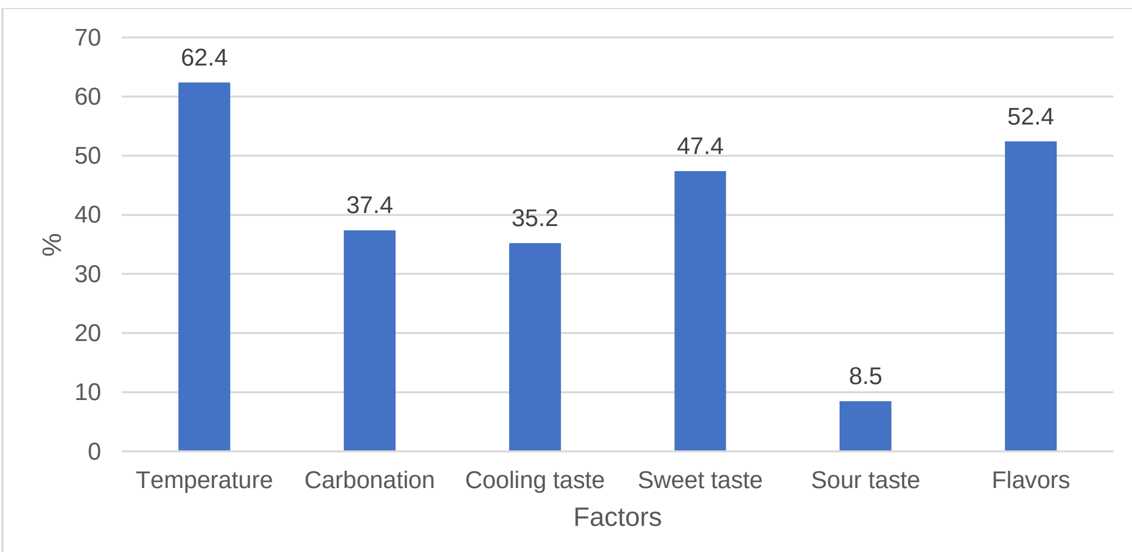
Attributes that Not Expecting of Flavored Water



Note. Others: Carbonation, sweet, artificial flavors, lingering taste, and chemical taste

Figure 6

Factors that Consumers Consider the Most When Deciding Flavored Water/Beverage



Note. Others: Caffeine, sugar free, thirst quenching ability, calories, price, convenience, nutrition facts, artificial taste, health benefits, and organic ingredients

Figure 7

Liking of Sweetness of Regular Beverage

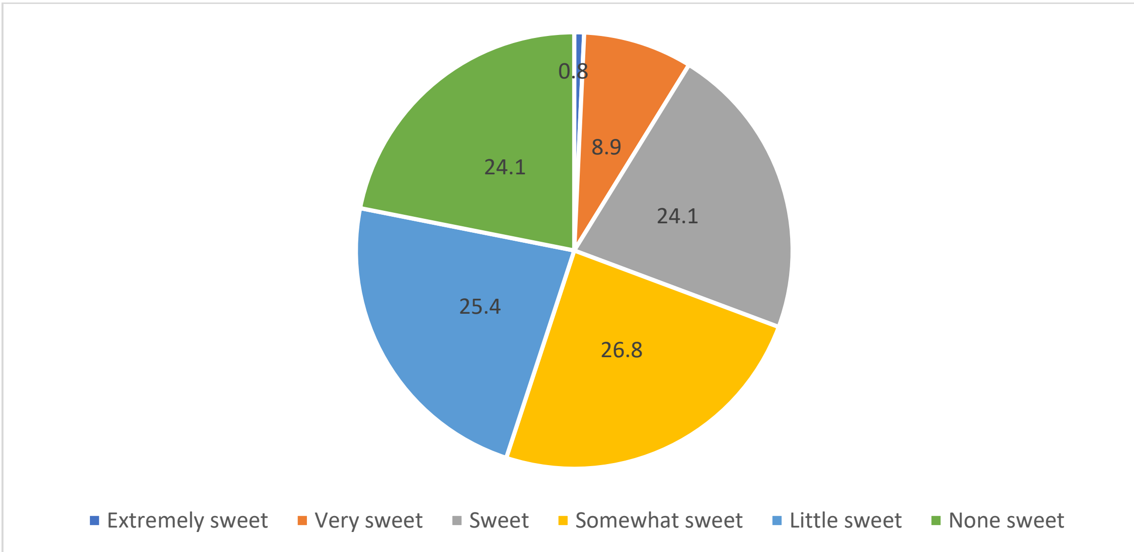
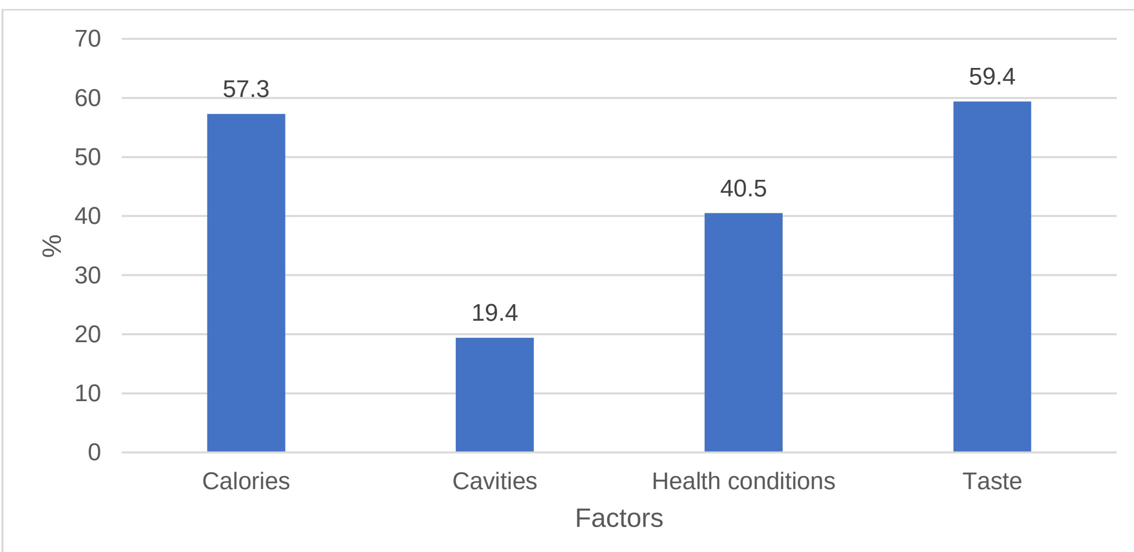


Figure 8

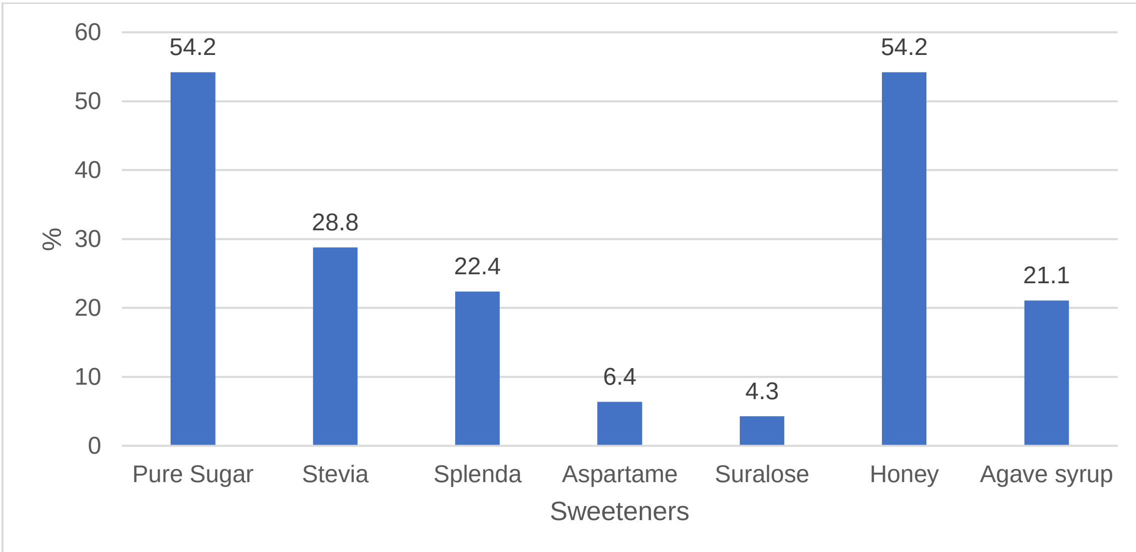
Factors that Consumers Think the Most Important When They Choose Sugar Reduced Products



Others: Alternative sweeteners, natural ingredients (artificial sweeteners), and carbohydrates

Figure 9

Preference of Sweeteners



Note. Others: Fructose, monkfruit, saccharine, high fructose corn syrup, sweet n' low, maple syrup, dextrose, and fruit juice

Online Survey Demographic Information

Demographic information collected at the end of the survey included questions related to gender, age, education level, employment status, health condition, and frequency of flavored water and beverage of purchase and consumption. Among 906 participants, 91.3% were female, with only 8.5% for male (see Table 2). Seven hundred and two (77.5%) of participants were aged from 18- 35 years old, while 22.5% of the participants were over 36 years of age (see Table 2). Six hundred and fifty-five (72.3%) of the participants had an associate degree or higher level degree, which was not surprising since this survey was primarily sent out to university email accounts (see Table 2). In addition, 852 of the respondents (94%) were either full- time or part- time employed or students (see Table 2).

Approximately 50% of the participants answered either healthy or very healthy when assessing their personal health condition (see Table 2).

Participants' frequency of purchase and consumption were evaluated to identify how familiar they are with flavored water/beverage. More than 60% of the participants either purchased or consumed flavored water/beverages more than 1-4 times per month. Approximately 25% of the respondents answered that they purchased or consumed the flavored water/beverages either only occasionally or never (see Table 2).

Table 2

Demographic Information

Sector	Types	Respondents	%
Gender	Male	78	8.6%
	Female	826	91.2%
	Others	2	0.2%
Age	18-25	441	48.7%
	26-30	164	18.1%
	31-35	97	10.7%
	36-40	48	5.3%
	41-45	43	4.7%
	45-50	43	4.7%
	51 and older	70	7.7%
Education level	Less than a high school diploma	1	0.1%
	High school graduate	34	3.8%
	Some college, no degree	216	23.8%
	Associate degree	113	12.5%
	Bachelor's degree	273	30.1%
	Master's degree	185	20.4%

	Doctorate	84	9.3%
Employment Status	Part time	191	21.1%
	Full time	331	36.5%
	Self employed	10	1.1%
	Student	330	36.4%
	Homemaker	11	1.2%
	Unemployed	19	2.1%
	Retired	3	0.3%
	Others	11	1.2%
Health Condition	Very unhealthy	10	1.1%
	Unhealthy	61	6.7%
	Somewhat healthy	371	40.9%
	Healthy	393	43.4%
	Very healthy	71	7.8%
Frequency of purchase of flavored water beverage	Several times a week	227	25.1%
	1~4 times per month	384	42.4%
	Less than once a month	55	6.1%
	Only occasionally	186	20.5%
	Never	54	6%
Frequency of consumption of flavored water beverage	Several times a week	391	43.2%
	1~4 times per month	257	28.4%
	Less than once a month	44	4.9%
	Only occasionally	176	19.4%
	Never	38	4.2%

Volatiles in Four Cucumbers

In this study, volatiles from four different cucumbers, English, American, Kirby, and Persian, were extracted by SPME, with SPME then analyzed using high-resolution capillary GC-MS. Compound identification was conducted based on the National Institute of Standards and Technology (NIST) library and Linear Retention Indices (LRI).

A total of 98 volatile compounds were identified in four different cucumbers (see Table 3). The percentage of peak area of each volatile compound was calculated and HSD was conducted to investigate how volatiles differed in each cucumber. The four cucumbers contained different total volatile compounds: 55 for English, 59 for American, 58 for Kirby, and 67 for Persian. Thirty-two volatile compounds were commonly identified in all four cucumbers. HSD analysis indicated that Kirby and Persian contained a higher level of volatiles, compared to English and American.

The volatile compounds shared among all four cucumbers were aldehydes, including alcohols, esters, ketones, and furans. The most abundant volatile compounds were C5 (25%) and C9 (25%). The volatile compounds containing 5 carbons were furan, pentanal, penten-3-one, 2,3-Pentanedione, E-2-pentenal, 1-penten-3-ol, and 1-pentanol. C9 compounds included nonanal, 2-nonenal, E,Z-2,6-nonadienal, E,Z-2,6-nonadienol, E,E-2,4-nonadienal, E,Z-3,6-onadien-1-ol, cucumber alcohol, and 4-Oxononanal. Other than C5s and C9s, C8 (18.8%) and C6 (15.6%) compounds were abundant. C8 volatiles included 3-octanone, octanal, 2,6-octadiene, trans-2-(2-pentenyl)furan, oct-(2E)-enal, and oct-(2E)-enal, while 6 carbon containing volatile compounds included hexanal, 2-hexenal, 1-hexanol, 1,3-hexadiene, and cyclohexane.

Table 3

Volatile Compounds Identified in Cucumber

LRI	RT (min)	Compound Name	English		American		Kirby		Persian		*P*-Value
			% of Area	Differences	% of Area	Differences	% of Area	Differences	% of Area	Differences	
	1.63	Acetaldehyde	0.20	A	1.15	B	0.49	A	0.68	A,B	**0.001**
	1.89	Propanal	0.18	A	0.34	A,B	0.46	B,C	0.60	C	**0.000**
	3.08	Furan, 2-ethyl-	0.15	A	0.04	B	0.07	A	0.09	A	**0.001**
	3.39	Pentanal	0.16	A	0.17	A,B	0.12	A	0.20	B	**0.006**
1012	3.98	Penten-3-one	0.20	B	0.08	A	0.09	A	0.07	A	**0.009**
1051	4.66	2,3-Pentanedione	0.03	A	0.04	A	0.05	A	0.10	B	**0.000**
1055	4.72	Butanal, 2-ethyl-3-methyl-	–	–	–		–		0.03	–	
1062	4.84	Acetate <butyl->	–	–	0.02	–	–	–	–	–	
1073	5.03	Hexanal	15.43	B	4.52	A	4.00	A	4.11	A	**0.000**
1087	5.28	Hexanal	0.80	–	–	–	–	–	–	–	
1112	5.79	Acetate <2-methylbutyl->	–	–	0.04	–	–	–	–	–	
1118	5.91	2-Pentenal, (E)-	0.10	A	0.16	A,B	0.19	A,B	0.24	B	**0.032**
1150	6.65	3-Pentanol, 2-methyl-	–	–	–	–	–	–	0.03	–	
1153	6.72	1-Penten-3-ol	0.35	C	0.07	A	0.16	A,B	0.23	B,C	**0.001**
1175	7.22	Heptanal	0.18	A,B	0.19	A,B	0.14	A	0.23	B	**0.010**
1192	7.61	Eucalyptol	–	–	0.18	–	–	–	–	–	
1193	7.63	2,4-Pentadien-1-ol, 3-pentyl-, (2Z)-	–	–	–	–	–	–	0.07	–	
1202	7.83	2-Hexenal, (E)-	0.37	A	1.61	B	0.21	A	3.09	C	**0.000**
1212	8.08	2-Hexenal, (E)-	17.13	–	–	–	8.81	–	–	–	**0.002**

1222	8.33	Furan, 2-pentyl-	1.40	B	0.70	A	0.66	A	0.86	A	**0.002**
1228	8.48	2,4-Nonadienal, (E,E)-	–	–	–	–	–	–	0.03	–	
1242	8.82	1-Pentanol	0.26	B	0.12	A	0.13	A	0.22	A	**0.006**
1245	8.90	3-Octanone	0.21	C	0.12	B	0.07	A	0.18	C	**0.000**
1251	9.06	Cyclohexane, (1-methylethylidene)- (Terpinolene)	–	–	0.02	–	–	–	–	–	
1276	9.69	Acetoin	–	–	0.03	–	–	–	0.05	–	0.189
1277	9.72	Octanal	0.05	A	0.08	B	0.05	A	0.08	B	**0.000**
1286	9.94	2,6-Octadiene, 4,5-dimethyl-	0.08	B	0.03	A	0.07	B	0.07	A	**0.018**
1293	10.12	trans-2-(2-Pentenyl)furan	0.82	B	0.14	A	0.21	A	0.15	A	**0.001**
1307	10.46	2-Penten-1-ol, (Z)-	0.12	–	–	–	–	–	–	–	
1312	10.59	2-Heptenal, (E)-	–	–	0.63	A	0.54	A	0.93	B	**0.001**
1319	10.75	2,3-Octanedione	–	–	0.19	A	0.17	A	0.19	A	0.352
1324	10.88	6-Octen-2-one, (Z)-	–	–	–	–	–	–	0.09	–	
1333	11.11	1,2-Butanediol	–	–	–	–	–	–	0.03	–	
1344	11.38	1-Hexanol	0.84	A,B	0.57	A	1.73	C	1.41	B,C	**0.008**
1374	12.13	3-Hexen-1-ol, (Z)-	0.11	–	–	–	0.06	–	–	–	**0.027**
1376	12.19	2-Nonenal, (E)-	–	–	–	–	–	–	0.06	–	
1386	12.43	Nonanal	0.24	A	1.98	B	1.66	B	3.44	C	**0.000**
1392	12.60	Ethanol, 2-butoxy-	–	–	0.20	–	–	–	–	–	
1395	12.66	2-Hexen-1-ol, (E)-	–	–	–	–	1.24	–	–	–	
1395	12.66	Cyclopentane, 1,3-dimethyl-2-(1-methylethenyl)-, (1.alpha.,2.alpha.,3.beta.)-	–	–	–	–	–	–	0.03	–	
1399	12.76	Oct-3-en-2-one	–	–	–	–	–	–	0.28	–	

1405	12.92	1,3-Hexadiene, 3-ethyl-2-methyl-	0.33	B	0.21	A,B	0.12	A	0.30	A,B	**0.027**
1420	13.28	Oct-(2E)-enal	1.98	C	0.79	B	0.35	A	0.63	A,B	**0.000**
1426	13.42	Pyrazine <2-isopropyl-, 3-methoxy->	–	–	–	–	–	–	0.03	–	
1433	13.61	6-Nonenal, (E)-	–	–	0.20	A	0.17	A	0.21	A	0.865
1443	13.86	6-Nonenal, (Z)-	–	–	2.68	A	3.89	B	4.70	B	**0.002**
1459	14.24	2,4-Heptadienal, (E,E)-	–	–	–	V	–	–	0.92	–	
1464	14.37	2,4-Heptadienal, (E,E)-	0.52	A	0.34	A	0.55	A	0.93	B	**0.000**
1485	14.89	2,4-Heptadienal, (E,E)-	0.68	B	0.28	A	0.52	A,B	–	–	**0.009**
1500	15.25	2-Nonenal, (E)-	0.36	A	0.90	C	–	–	0.69	B	**0.000**
1513	15.51	2-Nonenal, (E)-	12.51	B	27.63	D	0.45	A	21.75	C	**0.000**
1510	15.58	3,5-Octadien-2-one	–	–	–	–	–	–	1.06	–	
1529	15.95	3,5-Octadien-2-one	0.85	A	0.45	A	0.49	A	0.39	A	0.085
1542	16.26	Nona-(2E,6Z)-dienal	2.71	A	–	–	15.72	B	1.38	A	**0.000**
1555	16.58	2,6-Octadienal, 3,7-dimethyl-, (Z)-	–	–	–	–	–	–	1.23	–	
1555	16.58	2,6-Nonadien-1-ol	0.92	–	–	–	–	–	–	–	
1562	16.74	3,5-Octadien-2-one	0.38	–	–	–	0.21	–	–	–	0.149
1567	16.86	Nona-(2E,6E)-dienal	2.71	–	–	–	1.06	–	–	–	**0.001**
1581	17.20	2,6-Nonadienal, (E,Z)-	22.64	A	27.96	B	27.60	B	27.23	B	**0.013**
1591	17.44	Caryophyllene	0.06	–	0.49	–	–	–	–	–	**0.003**
1607	17.81	2-Octen-1-ol, (E)-	–	–	0.06	–	0.03	–	–	–	**0.449**
1610	17.89	2-Decen-1-ol, (E)-	–	–	–	–	0.14	–	0.19	–	**0.011**
1629	18.33	n-Caproic acid vinyl ester	0.10	–	–	–	–	–	0.03	–	0.092
1636	18.50	4-Nonanol	0.63	B	–	–	0.06	A	0.11	A	**0.002**
1652	18.87	n-Caproic acid vinyl ester	0.15	–	–	–	–	–	–	–	

1653	18.88	1-Nonanol	–	–	–	–	0.78	–	1.53	–	0.069
1658	18.99	Humulene <alpha->	–	–	1.20	–	–	–	–	–	
1678	19.46	3-Nonen-1-ol, (Z)-	–	–	0.26	A	0.35	A	0.82	B	**0.000**
1689	19.72	2H-Pyran-2-one, 5,6-dihydro-	0.26	–	–	–	–	–	–	–	
1693	19.81	2,4-Nonadienal, (E,E)-	0.25	B	0.15	A	0.11	A	0.17	A	**0.001**
1700	19.97	(6Z)-Nonen-1-ol	–	–	–	–	–	–	0.10	–	
1709	20.16	2-Octen-1-ol, (E)-	0.28	A	–	–	2.68	B	3.24	B	**0.002**
1709	20.17	2-Nonen-1-ol, (E)-	–	–	1.97	–	–	–	–	–	
1715	20.29	1,8,11,14-Heptadecatetraene, (Z,Z,Z)-	–	–	0.03	–	–	–	–	–	
1724	20.49	Geranial	–	–	–	–	0.07	–	–	–	
1730	20.62	3,6-Nonadien-1-ol, (E,Z)-	0.04	A	0.18	B	0.11	A,B	0.08	A,B	**0.042**
1733	20.68	3,6-Nonadien-1-ol, (E,Z)-	0.07	A	–	–	0.57	B	0.17	A	**0.013**
1739	20.81	3,6-Nonadien-1-ol, (E,Z)-	0.08	–	–	–	–	–	0.36	–	0.442
1746	20.97	3,6-Nonadien-1-ol, (E,Z)-	0.61	–	–	–	–	–	–	–	
1759	21.26	2,4-Decadienal, (E,E)-	0.06	–	–	–	–	–	–	–	
1762	21.32	Cucumber alcohol	0.43	A	1.46	A	5.35	B	4.55	B	**0.001**
1803	22.23	2,4-Decadienal, (E,E)-	0.09	–	–	–	–	–	–	–	
1805	22.26	Tridecanal	–	–	0.24	–	0.27	–	–	–	0.315
1823	22.67	4-Oxononanal	0.46	B	0.47	B	0.13	A	0.13	A	**0.000**
1855	23.42	3-Undecanol, 3-ethyl-	–	–	–	–	0.33	–	0.34	–	0.812
1863	23.60	Furanacrolein <2-, alpha-methyl->	–	–	0.16	A	0.18	A,B	0.22	B	**0.040**
1877	23.93	Cyclohexane, 1-butenylidene-	0.07	A	0.07	A	0.13	A	0.16	A	0.077
1907	24.61	Tetradecanal	0.46	A	1.09	B	1.14	B	0.10	A	**0.001**
1917	24.84	trans-.beta.-Ionone	0.06	A	0.05	A	–	–	0.05	A	0.273

1952	25.65	Caryophyllene oxide	−	−	0.09	−	−	−	0.16	−	**0.001**
1964	25.92	1-Decyn-4-ol	−	−	−	−	−	−	0.03	−	
1993	26.58	3-Decen-1-ol, (Z)-	0.03	−	−	−	−	−	−	−	
2000	26.76	Pentadecanal-		−	6.36	−	5.15	−	−	−	0.171
2012	27.03	Geranyl linalool<(Z,E)>	−	−	0.03	−	−	−	−	−	
2024	27.30	cis-9-Hexadecenal	−	−	0.05	−	0.06	−	−	−	**0.024**
2038	27.63	Octanoic acid	0.19	−	0.06	−	−	−	−	−	0.178
2112	29.32	9,17-Octadecadienal, (Z)-	−	−	−	−	0.04	−	−	−	
2218	31.76	9,17-Octadecadienal, (Z)-	−	−	0.40	A	1.13	B	0.16	A	**0.004**
2276	33.09	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	−	−	0.18	A	1.19	B	0.12	A	**0.000**
	Total area % of each cucumber		**90.38**		**89.94**		**92.50**		**92.15**		

Note. LRI: Linear Retention Index, RT: Retention Time

Cucumber Flavor Reconstitution

Based on the cucumber volatile identification using GC-MS, flavor reconstitution was performed. Nine volatile compounds were selected to reconstitute for a general cucumber flavor. The volatile compounds used for reconstitution were nonanol, trans-2-nonenol, cis-6-nonenol, nonanal, trans-2-nonenal, cis-6-nonenal, E,Z-2-6-nonadienal, E,Z,-2-6-nonadienol, hexanal, and trans-2-octenal. Stock solutions of individual volatile compounds were prepared by weighing each ingredient and dissolving in propylene glycol. Individual volatile compound stock solutions were combined at a particular ratio to make a cucumber flavor stock solution. The reconstituted cucumber flavor stock solution was diluted into water at concentrations of 0.6ppm, 0.8ppm, and 1ppm (see Table 4).

All prepared samples were tasted to find out the best solution to mimic cucumber flavor. One ppm solution was too fatty and soapy, 0.6ppm solution was not balanced well with each flavor ingredient that contributed to a certain chemical compound flavor, with the 0.8 ppm solution finally selected as the best cucumber flavor.

Table 4

Cucumber Flavor Reconstitution

	% of Stock Solution	0.6 ppm solution	0.8 ppm solution	1 ppm solution
Nonanol	1.5	0.009 µL	0.012 µL	0.015 µL
Trans-2-nonenol	2	0.012 µL	0.016 µL	0.02 µL
Cis-6-nonenol	2	0.012 µL	0.016 µL	0.02 µL
Nonanal	1	0.006 µL	0.008 µL	0.01 µL
Trans-2-nonenal	0.2	0.0012 µL	0.0016 µL	0.002 µL
E,Z-2,6-nonadienal	14.5	0.087 µL	0.116 µL	0.145 µL

E,Z-2,6-nonadienol	2.5	0.015 µL	0.02 µL	0.025 µL
Hexanal	0.02	0.00012 µL	0.00016 µL	0.0002 µL
Trans-2-octenal	0.03	0.00018 µL	0.00024 µL	0.0003 µL
Solvent/Solution	100% Propylene Glycol	100 mL Water	100 mL Water	100 mL Water

Qualitative Descriptive Analysis

QDA was conducted to evaluate lemon flavored water's sensory attributes including fresh, green, juicy, lemon, lemonade, cucumber, refreshing, sweet, sour, bitter, astringent, and irritate. Significant difference ($p < 0.05$) for all descriptors (except the lemon note) among six lemon flavored water samples was observed (see Figures 10-11 and Table 5).

Overall, the lemon flavored water had a very positive flavor profile, as all six products received high ratings on fresh, green, lemon and refreshing perceptions, with average scores ranging from 3.0 to 6.2. Negative flavor perception such as bitter, astringent, and irritate were minimum or close to none for all products, even though the score was slightly higher for the 100% sugar reduction level, which was 1.2 for bitter and 1.1 for astringent.

Sweetness level was perceived significantly decreasing in a relationship to sugar reduction. For example, the mean scores for sweetness intensity were rated 7.0 at 0% reduction, 3.5 at 50% reduction, 0.9 at 80% reduction, and 0.1 at 100% reduction. Pearson correlation was analyzed to investigate the relation between sweetness perception and other sensory attributes. Sweetness correlation showed sweetness was positively correlated with juicy, lemon, and lemonade ($p < 0.05$; see Figure 12).

The added cucumber flavor significantly enhanced refreshing perception ($p < 0.05$). A 50% sugar reduction received an average score of 4.8 in refreshing perception but 50% reduction with cucumber flavor added received an average rating of 6. In addition, with an 80% reduction, refreshing scored 4.7 without cucumber flavor, whereas, 6.2 with cucumber flavor added. The refreshing perception was shown to not be consistent with sugar levels. However, refreshing perception was still significantly different by sample ($p < 0.05$). The 100% reduction showed the least refreshing with a score of 4.5. The most refreshing sample was the 50% reduction with a score of 4.8, 80% reduction scored 4.7, and 0% reduction scored 4.6. *T*-test of 50% reduction and 50% reduction with cucumber flavor indicated that green, cucumber, refreshing, and bitter were significantly different ($p < 0.05$). Added cucumber increased green, cucumber, refreshing, and bitter attributes in the to 50% reduction samples (see Table 6). Contrasting 80% reduction and 80% reduction with cucumber flavor, added cucumber significantly increased fresh, green, cucumber, refreshing, sweet, and irritate ($p < 0.05$; see Table 7).

Lemon flavored water QDA principle component analysis (PCA) is presented in Figure 13. The PC1 axis explained 56.82% of the variance alone; while PC2 accounted for 31.40%. PC1 was the major component to differentiate samples by their descriptors. From the left-hand side to right-hand side of the plot, 100% sugar reduction, 80% sugar reduction, and 80% sugar reduction with cucumber flavor were clustered in the left-hand side of the plot corresponding to negative PC1 values, while 50% reduction with cucumber key, 50% reduction, and 0% reduction were clustered in the right-hand side of the plot corresponding to positive PC1 values. Sugar levels were highly correlated to lemonade, sour, sweet, juicy, lemon, and refreshing perception and all were clustered on the right hand side of the PC1 plot. Cucumber, refreshing, fresh, lemon, green, juicy, and sweet were clustered together on the upper side of the plot corresponding to positive PC2 values, while astringent, lemonade, bitter and irritate were clustered together bottom side of the plot corresponding to negative PC2 value. Added cucumber flavor was highly correlated with refreshing, fresh, lemon, and green perceptions (see Figure 13).

Table 5

Mean Scores, Standard Deviation, and Tukey's Honest Significant Difference (HSD) of Lemon Flavored Water QDA

	Fresh*			Green***			Juicy***			Lemon			Lemonade***			Cucumber***		
	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff
0% Reduction	3.40	2.53	A	1.55	1.93	A	4.73	2.71	D	3.36	1.86	A	6.41	1.88	C	0.11	0.21	A
50% Reduction	3.79	2.42	A	1.86	2.11	A,B	3.36	1.71	B,C,D	3.70	1.86	A	3.24	2.29	B	0.49	0.73	A
50% Reduction with Cucumber	4.73	2.28	A	3.22	2.60	B,C	3.78	2.10	C,D	3.85	1.77	A	2.85	2.34	B	4.63	2.78	B
80% Reduction	3.63	2.64	A	2.49	2.27	A,B	2.36	2.11	A,B	3.16	1.97	A	1.38	1.72	A	0.18	0.35	A
80% Reduction with Cucumber	4.94	1.99	A	4.39	2.38	C	3.07	1.88	A,B,C	3.50	1.75	A	1.17	1.53	A	4.62	3.03	B
100% Reduction	4.19	2.52	A	3.39	2.78	B,C	1.90	1.73	A	2.96	2.44	A	0.31	0.45	A	0.46	0.80	A

	Refreshing**			Sweet***			Sour**			Bitter***			Astringent**			Irritate**		
	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff	Mean	SD	Diff
0% Reduction	4.62	2.43	A,B	6.99	2.09	D	2.19	1.49	B	0.06	0.14	A	0.39	0.65	A	0.08	0.22	A,B
50% Reduction	4.75	1.87	A,B	3.51	1.78	C	1.62	1.44	B	0.02	0.07	A	0.30	0.58	A	0.03	0.09	A
50% Reduction with Cucumber	5.96	1.86	A,B	3.57	1.92	C	1.50	1.37	B	0.22	0.39	A,B	0.48	0.73	A	0.01	0.03	A
80% Reduction	4.69	2.76	A,B	0.92	0.84	B	1.19	1.15	A	0.40	0.59	A,B	0.48	0.63	A	0.02	0.05	A
80% Reduction with Cucumber	6.16	2.28	B	1.43	0.92	B	1.25	1.36	A	0.61	0.76	B	0.65	0.75	A,B	0.14	0.27	A,B
100% Reduction	4.53	2.88	A	0.11	0.17	A	0.98	1.05	A	1.19	1.26	C	1.06	1.07	B	0.26	0.49	B

Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Table 6

QDA t-test Results for 50% Reduction With and Without Cucumber Flavor

	t- value	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
Fresh	-1.69	70	0.10	-0.93	0.55	-2.04	0.17
Green*	-2.44	70	0.02	-1.36	0.56	-2.48	-0.25
Juicy	-0.92	68	0.36	-0.42	0.46	-1.33	0.49
Lemon	-0.37	70	0.72	-0.16	0.43	-1.01	0.70
Lemonade	0.71	70	0.48	0.39	0.55	-0.70	1.48
Cucumber***	-8.17	66	0.00	-4.14	0.51	-5.15	-3.13
Refreshing**	-2.75	70	0.01	-1.21	0.44	-2.09	-0.33
Sweet	-0.12	70	0.91	-0.05	0.44	-0.92	0.82
Sour	0.37	70	0.72	0.12	0.33	-0.54	0.78
Bitter**	-2.73	61	0.01	-0.20	0.07	-0.35	-0.05
Astringent	-1.12	66	0.27	-0.18	0.16	-0.50	0.14
Irritate	1.25	56	0.22	0.02	0.02	-0.01	0.06

Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Table 7

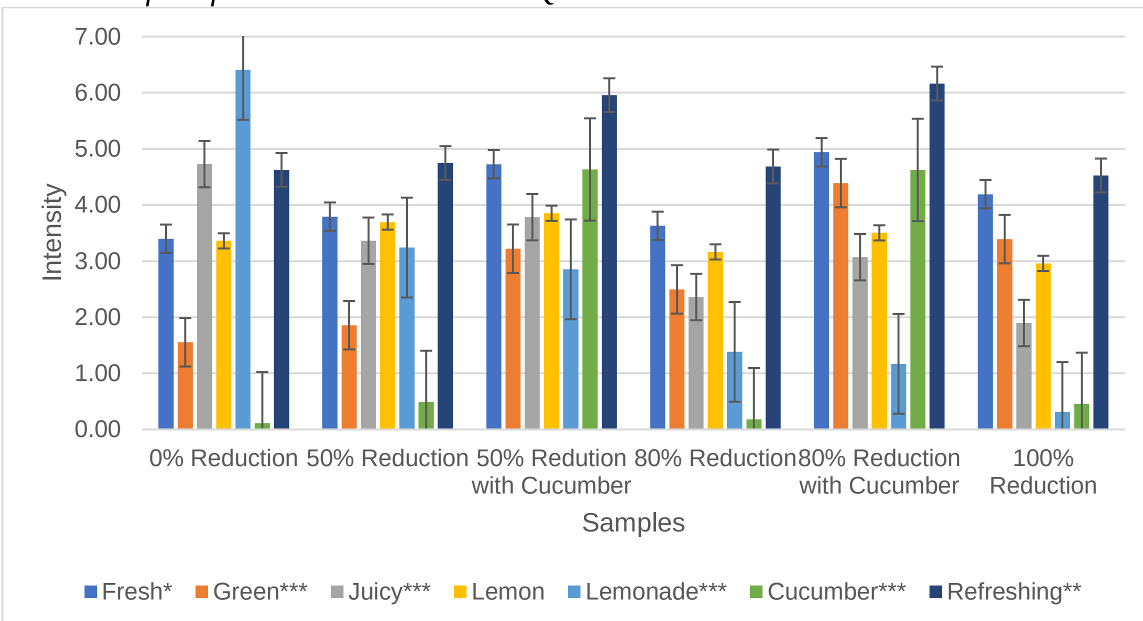
QDA t-test Results for 80% Reduction With and Without Cucumber Flavor

	t- value	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
Fresh*	-2.38	70	0.02	-1.31	0.55	-2.41	-0.21
Green***	-3.46	70	0.00	-1.90	0.55	-2.99	-0.80
Juicy	-1.50	69	0.14	-0.71	0.47	-1.66	0.24
Lemon	-0.77	69	0.45	-0.34	0.44	-1.22	0.54
Lemonade	0.54	65	0.59	0.21	0.40	-0.58	1.01
Cucumber***	-7.97	64	0.00	-4.44	0.56	-5.55	-3.33
Refreshing*	-2.48	70	0.02	-1.48	0.60	-2.67	-0.29
Sweet*	-2.44	69	0.02	-0.51	0.21	-0.93	-0.09
Sour	-0.21	70	0.83	-0.06	0.30	-0.65	0.53
Bitter	-1.31	68	0.19	-0.21	0.16	-0.54	0.11
Astringent	-1.00	68	0.32	-0.17	0.17	-0.50	0.17
Irritate*	-2.35	58	0.02	-0.12	0.05	-0.22	-0.02

Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 10

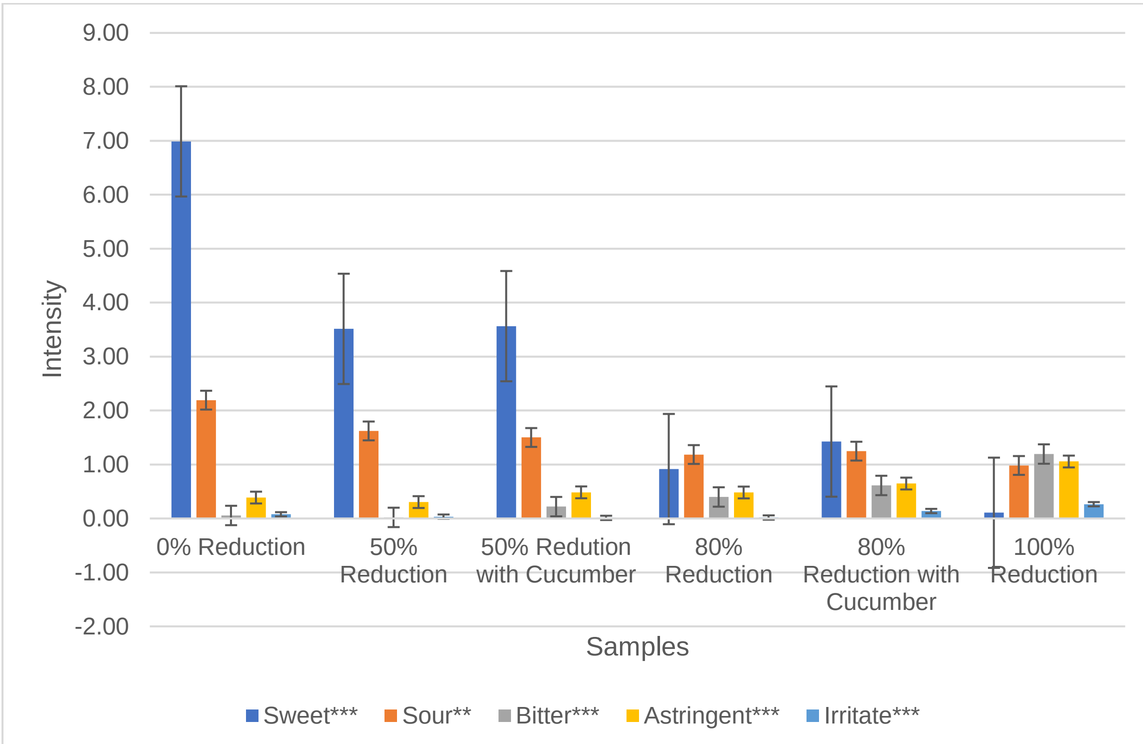
Aroma Profile of Lemon Flavored Water QDA



Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 11.

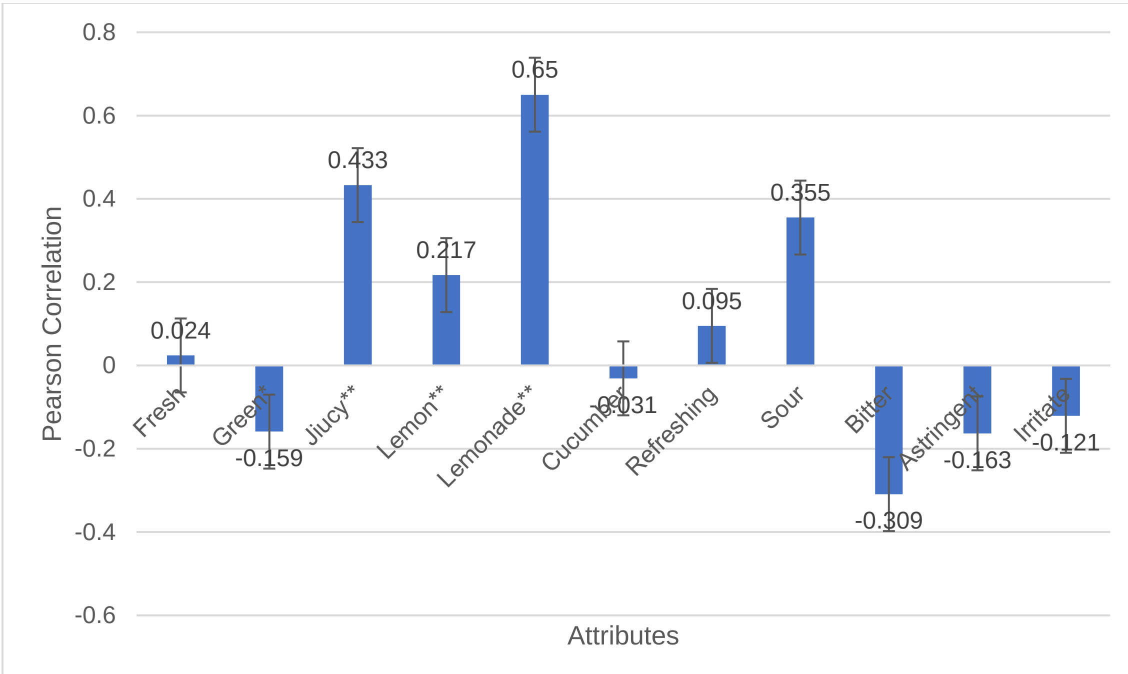
Taste Profile of Lemon Flavored Water QDA



Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 12

Lemon Flavored Water QDA Sweet Correlation with Rest of Attributes



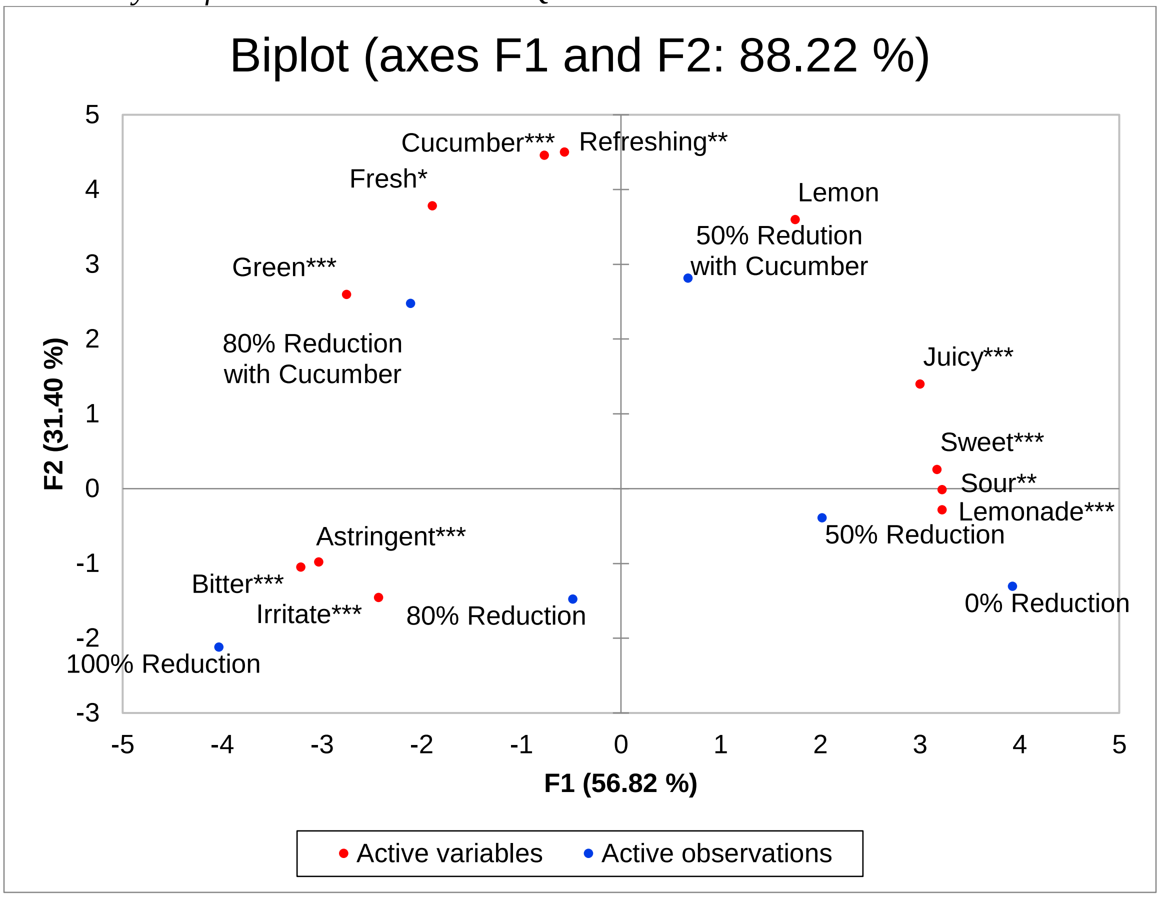
Note.

**: Correlation is significant at the 0.01 level (2-tailed)

*: Correlation is significant at the 0.05 level (2-tailed)

Figure 13

PCA Analysis of Lemon Flavored Water QDA



Consumer Test

A consumer test was conducted to investigate consumers' liking and acceptance of lemon flavored water's sensory attributes including overall character liking, overall flavor liking, and intensity, flavor balance liking, refreshing liking and intensity, lemon flavor liking and intensity, mouthfeel liking and intensity, sweetness liking and intensity, sourness liking, and intensity. Significant difference ($p < 0.05$) for all attributes among six lemon flavored water samples was observed.

Overall character liking, overall flavor liking, overall flavor intensity, and flavor balance were evaluated for lemon flavored water. Zero percent and 50% reductions were grouped together for their higher mean scores of the overall character liking, which were 7 and 6.3, while 100% reduction received the lowest rating at 4.3 (see Figure 14). Overall flavor liking and flavor balance liking indicated consumers were satisfied with up to 50% sugar reduction and 80% sugar reduction with cucumber flavor. Added cucumber flavor in 50% and 80% sugar reductions revealed higher scores of the overall flavor intensity and flavor balance than same sugar reduction levels. The overall flavor intensity scored 0.6 more in 50% reduction with cucumber flavor compared to 50% reduction and 1.2 more in 80% reduction with cucumber key compared to 80% reduction. In addition, flavor balance showed 0.6 more in 80% reduction with cucumber key compared to 80% reduction (see Figure 15). One hundred percent sugar reduction showed the least preference of the overall flavor liking, overall flavor intensity, and flavor balance (see Figure 15). Statistically, all descriptors were shown significantly different ($p < 0.05$).

With cucumber flavor addition, the refreshing liking and intensity were higher than same sugar reduction products in 50% and 80%. Refreshing perception was highest in 80% reduction with cucumber flavor. Lemon flavor liking and intensity were the highest in 0% reduction and decreased as sugar was reduced, and was lowest with the 100% reduction; however, cucumber flavor enhanced lemon flavor intensity in the 80% reduction. In addition, added cucumber flavor enhanced both sweetness liking and intensity. Sourness liking and intensity were higher in 80% reduction with cucumber flavor added but not in 50% reduction (see Figures 16 & 17). Added cucumber flavor increased overall flavor intensity and refreshing intensity in the 50% reduction samples (*t*-test, $p < 0.05$; see Table 8). Added cucumber significantly increased overall flavor intensity, flavor balance, refreshing intensity, and sweetness liking when comparing the 80% reduction and 80% reduction with cucumber flavor (*t*-test, $p < 0.05$; see Table 9).

CATA results are shown in Figure 18. Participants were asked to check all flavors they could perceive which included fresh, green, juicy, lemonade, cucumber, bitterness, astringent, and irritate. The fresh note was perceived in all samples and slightly increased in added cucumber flavor samples. Fourteen more panelists picked the fresh note in 50% reduction with added cucumber flavor and 13 more panelists perceived the fresh note in 80% reduction with added cucumber flavor compared to same sugar reduction levels. Panelists at the similar range regardless of sample perceived astringent. Nine respondents out of 100 perceived astringent in 0% reduction and 14 perceived on 100% reduction. Cucumber note was identified up in added cucumber flavor samples in 50% and 80% reduction: 86 respondents out of 100 detected cucumber flavors in cucumber added samples. The green note was perceived more when cucumber flavor was added. Lemonade and juicy notes were picked up more in high sugar containing samples. Seventy-five panelists could pick up lemonade note from 0% reduction, while only 26 panelists could pick up from 100% reduction. Bitterness was picked up with similar frequency for each sample with an average of 13 panelists perceiving except in the 100% reduction and 0% reduction. Irritate perception was close to zero except with 50% reduction with cucumber flavor added and 100% reduction.

Consumer test PCA correlation is shown in Figure 19. The PC1 axis explained 81.87% of the variance alone; while PC2 accounted for 13.61%. The PC1 was the major component to differentiate samples by their descriptors. From the left-hand side to right-hand side of the plot, 100% sugar reduction and 80% sugar reduction were clustered in the left-hand side of the plot corresponding to negative PC1 values, while 80% reduction with cucumber flavor, 50% reduction with cucumber flavor, 50% reduction, and 0% reduction were clustered in the right-hand side of the plot corresponding to positive PC1 values. Sugar levels were highly correlated to all of the descriptors clustered at the all right hand side of the PC1 plot. Overall flavor intensity, mouthfeel intensity, lemon flavor intensity, and sweetness intensity, sourness intensity, and sourness liking were clustered together on the upper side of the plot corresponding to positive PC2 values. Overall character, overall flavor liking, flavor balance liking, flavor liking, refreshing liking, mouthfeel liking, sweetness liking, and refreshing intensity were clustered together bottom side of the plot corresponding to negative PC2 value (see Figure 19).

Table 8

Consumer Test t-test Results for 50% Reduction With and Without Cucumber Flavor

	t- value	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
Overall character	0.73	198	0.47	0.21	0.29	-0.36	0.78
Overall flavor liking	0.45	198	0.65	0.13	0.29	-0.44	0.70
Overall flavor intensity*	-2.23	198	0.03	-0.63	0.28	-1.19	-0.07
Flavor balance	0.11	198	0.92	0.03	0.28	-0.52	0.58

Refreshing liking	-0.66	198	0.51	-0.19	0.29	-0.76	0.38
Refreshing intensity***	-3.18	198	0.00	-0.81	0.26	-1.31	-0.31
Lemon liking	1.63	198	0.11	0.42	0.26	-0.09	0.93
Lemon intensity***	3.25	198	0.00	0.88	0.27	0.35	1.41
Mouthfeel liking	0.28	198	0.78	0.08	0.28	-0.48	0.64
Mouthfeel intensity	-0.08	198	0.94	-0.02	0.26	-0.54	0.50
Sweetness liking	-0.13	198	0.89	-0.04	0.30	-0.63	0.55
Sweetness intensity	-1.11	198	0.27	-0.30	0.27	-0.84	0.24
Sourness liking	1.33	198	0.19	0.30	0.23	-0.15	0.75
Sourness intensity	1.43	198	0.15	0.41	0.29	-0.16	0.98

Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Table 9

Consumer Test t-test Results for 80% Reduction With and Without Cucumber Flavor

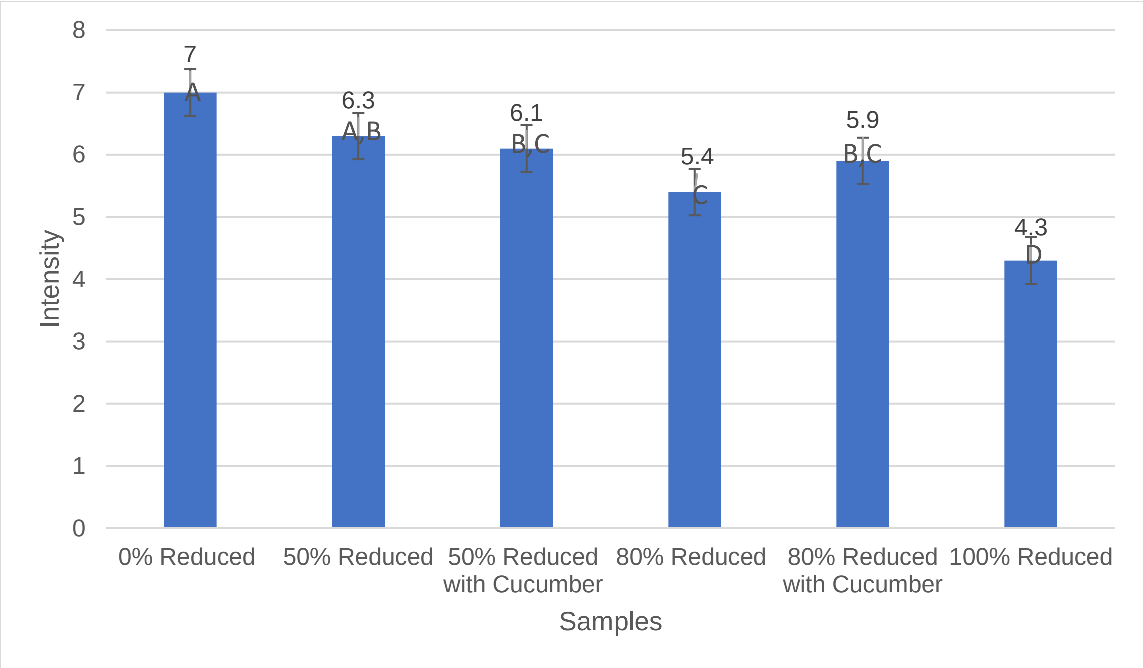
	t- value	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
						Lower	Upper
Overall character	-1.66	198	0.10	-0.52	0.31	-1.14	0.10
Overall flavor liking	-1.63	198	0.11	-0.52	0.32	-1.15	0.11
Overall flavor intensity***	-4.09	198	0.00	-1.21	0.30	-1.79	-0.63
Flavor balance*	-1.95	198	0.05	-0.62	0.32	-1.25	0.01
Refreshing liking	-1.23	198	0.22	-0.35	0.29	-0.91	0.21
Refreshing intensity***	-3.02	198	0.00	-0.86	0.29	-1.42	-0.30
Lemon liking	-0.25	198	0.81	-0.07	0.28	-0.63	0.49
Lemon intensity	-0.68	198	0.50	-0.21	0.31	-0.82	0.40
Mouthfeel liking	-0.45	198	0.66	-0.12	0.27	-0.65	0.41
Mouthfeel intensity	-0.32	198	0.75	-0.08	0.25	-0.58	0.42

Sweetness liking*	-2.09	198	0.04	-0.65	0.31	-1.26	-0.04
Sweetness intensity	-1.15	198	0.25	-0.32	0.28	-0.87	0.23
Sourness liking	-1.53	198	0.13	-0.38	0.25	-0.87	0.11
Sourness intensity	-0.11	198	0.92	-0.03	0.28	-0.59	0.53

Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 14

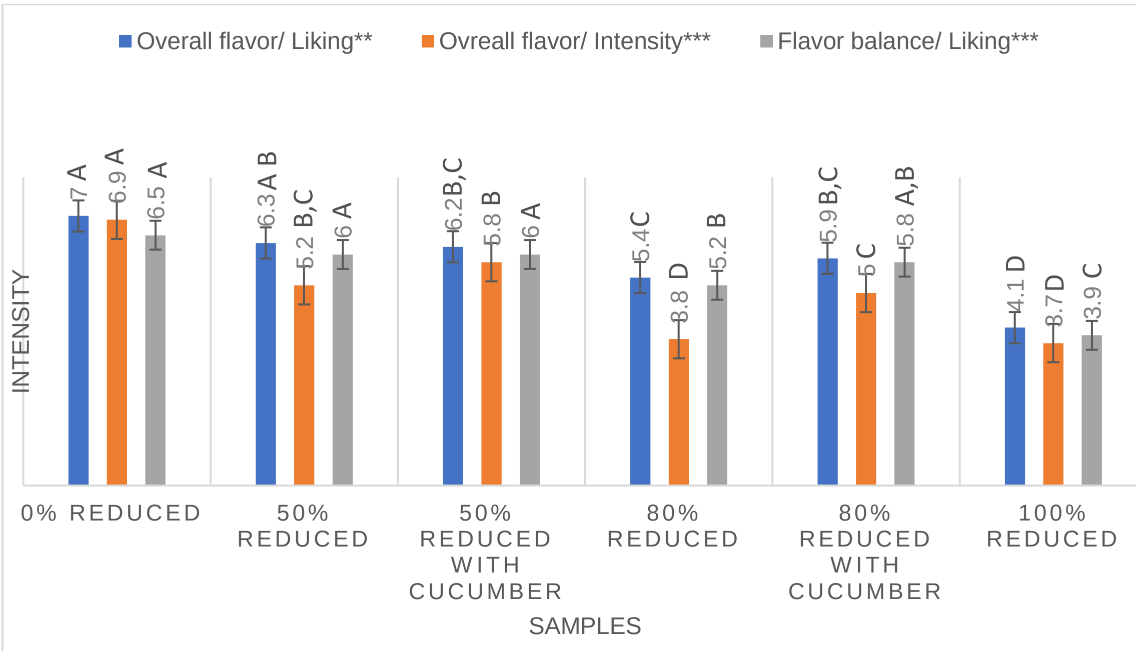
Overall Character Liking of Lemon Flavored Water Consumer Test



Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 15

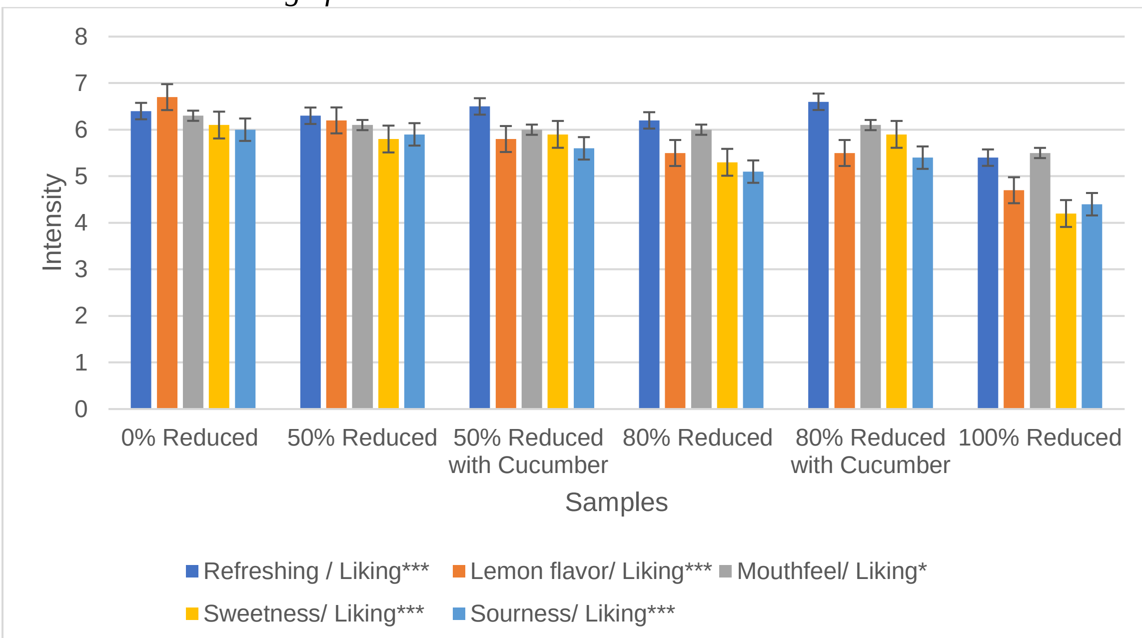
Overall Flavor and Flavor Balance Liking and Intensities of Lemon Flavored Water Consumer Test



Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 16

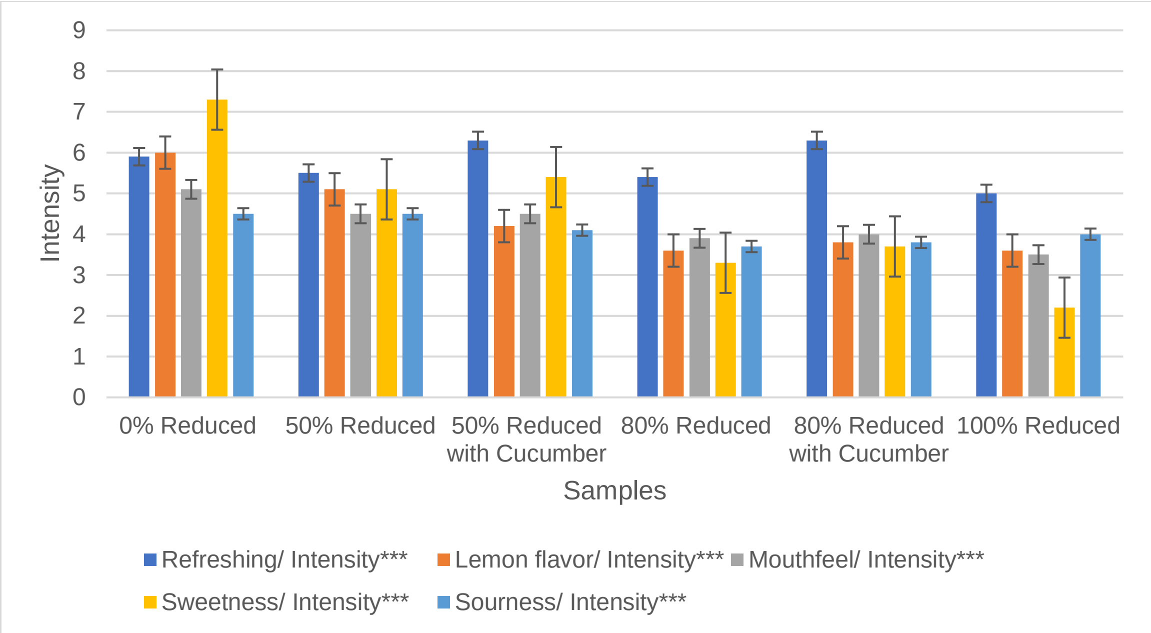
Flavor Attribute Liking of Lemon Flavored Water Consumer Test



Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 17

Flavor Attribute Intensities of Lemon Flavored Water Consumer Test



Note. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$

Figure 18

Check-All-That-Apply Chart of Lemon Flavored Water Consumer Test

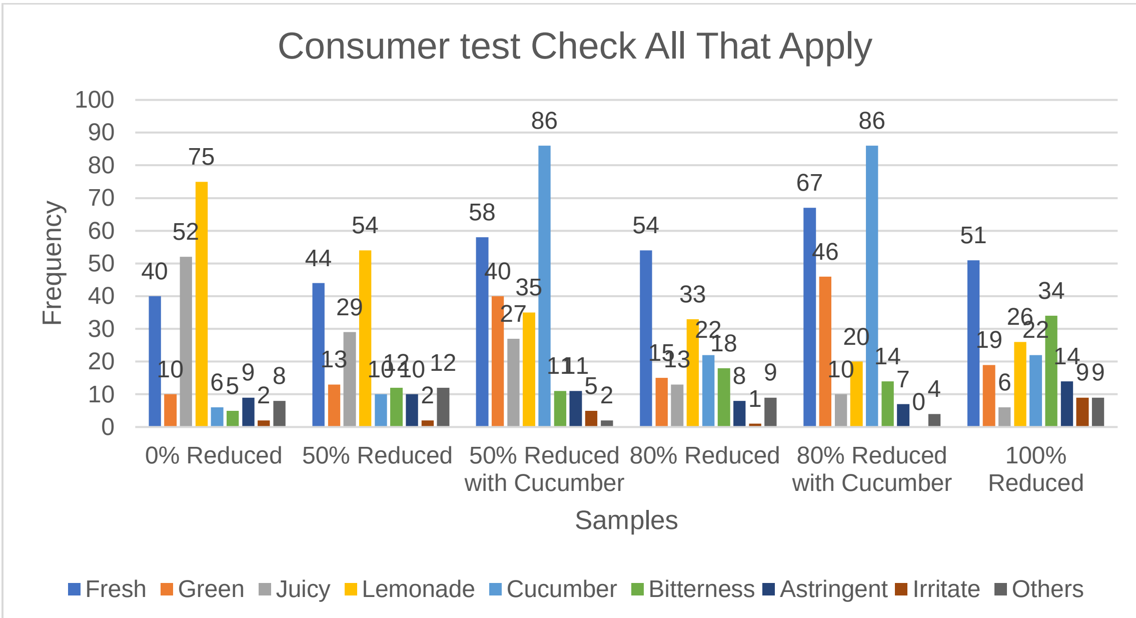
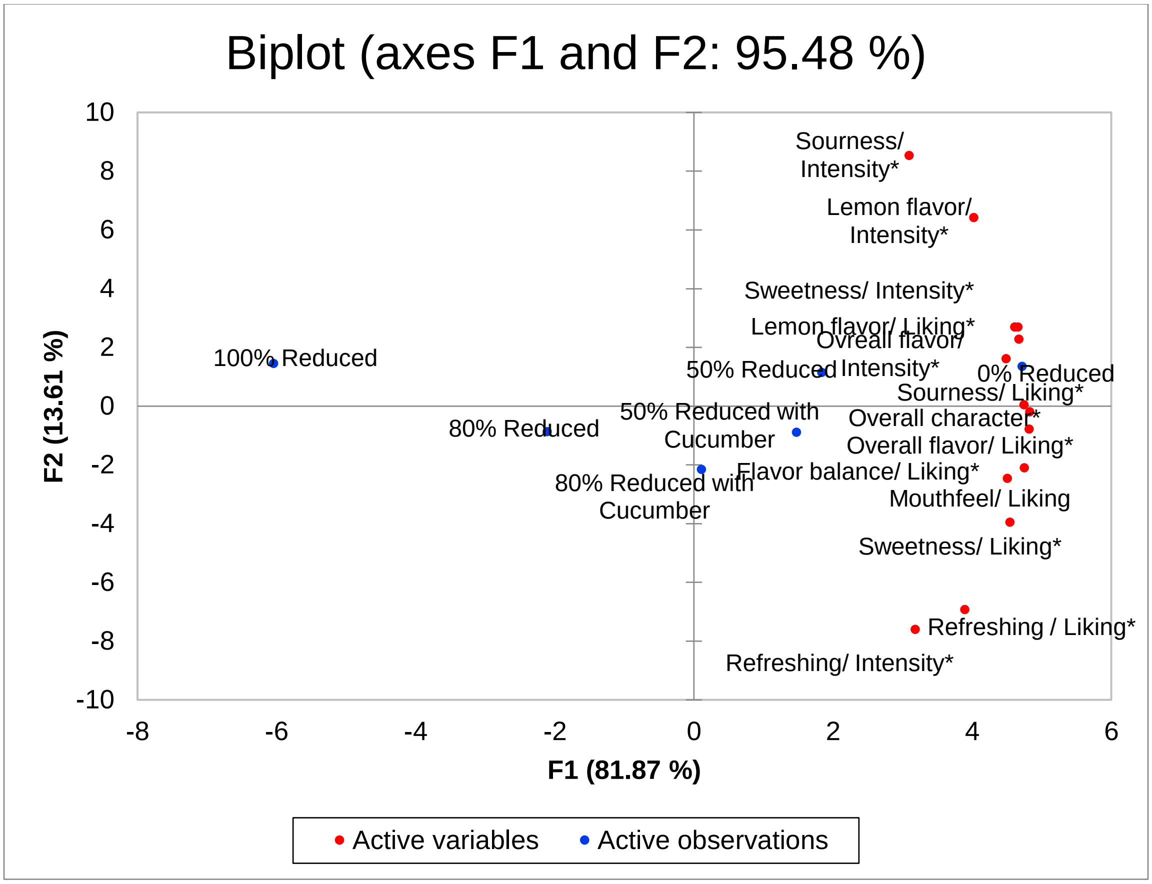


Figure 19

PCA Analysis of Lemon Flavored Water Consumer Test



Consumer Test Demographic Information

A total of 100 panelists participated in the lemon flavored water consumer test. All participants were pre-screened based on their frequency of consumption. All participants consumed the type of flavored water or/and carbonated water at least 1-4 times per month. The gender ratio was 85 females and 15 males (see Table 10). Age was broken into 3 groups, 18-25, 26-35, and 36-50 years old (see Table 10). Each participant's body mass index was calculated by measuring height and weight. BMI categories were divided into four groups which included underweight (<18.5), normal weight (18.5-24.9), overweight (25-29.9), and obesity (>30; see Table 10).

To evaluate the relationships of those demographic factors, PCA correlation analysis was performed. The loading values of refreshing liking and sweetness liking, gender (male, female), age (18-25, 26-35, 36-50), and BMI (underweight, normal weight, overweight, obesity) were used to calculate the score values for all 6 samples. Demographic PCA plot is shown in Figure 20. The PC1 axis explained 71.93% of the variance alone, while PC2 accounted for 13.38%. The PC1 was the major components to differentiate samples by their factors. From the left-hand side of the plot to right-hand side of the plot, 100% reduction and 80% reduction were clustered in the left-hand side of the plot corresponding to positive PC1 values; while 50% reduction, 50% reduction with cucumber, 80% with cucumber, and 0% reduction were clustered in the right-hand side of the plot corresponding to negative PC1 values. All refreshing and sweetness liking of female, obesity, 26-35 years old groups were clustered together in upper side of the plot corresponding positive PC2 values; while male, overweight, normal weight, underweight, 18-25 years old groups together in bottom side of the plot corresponding to negative PC2 values. Refreshing liking from all variances, except over 36 years old and obesity group, clustered together close to the 80% reduction with cucumber and 50% reduction with cucumber samples. Cucumber flavor is highly related to refreshing liking perception (see Figure 20).

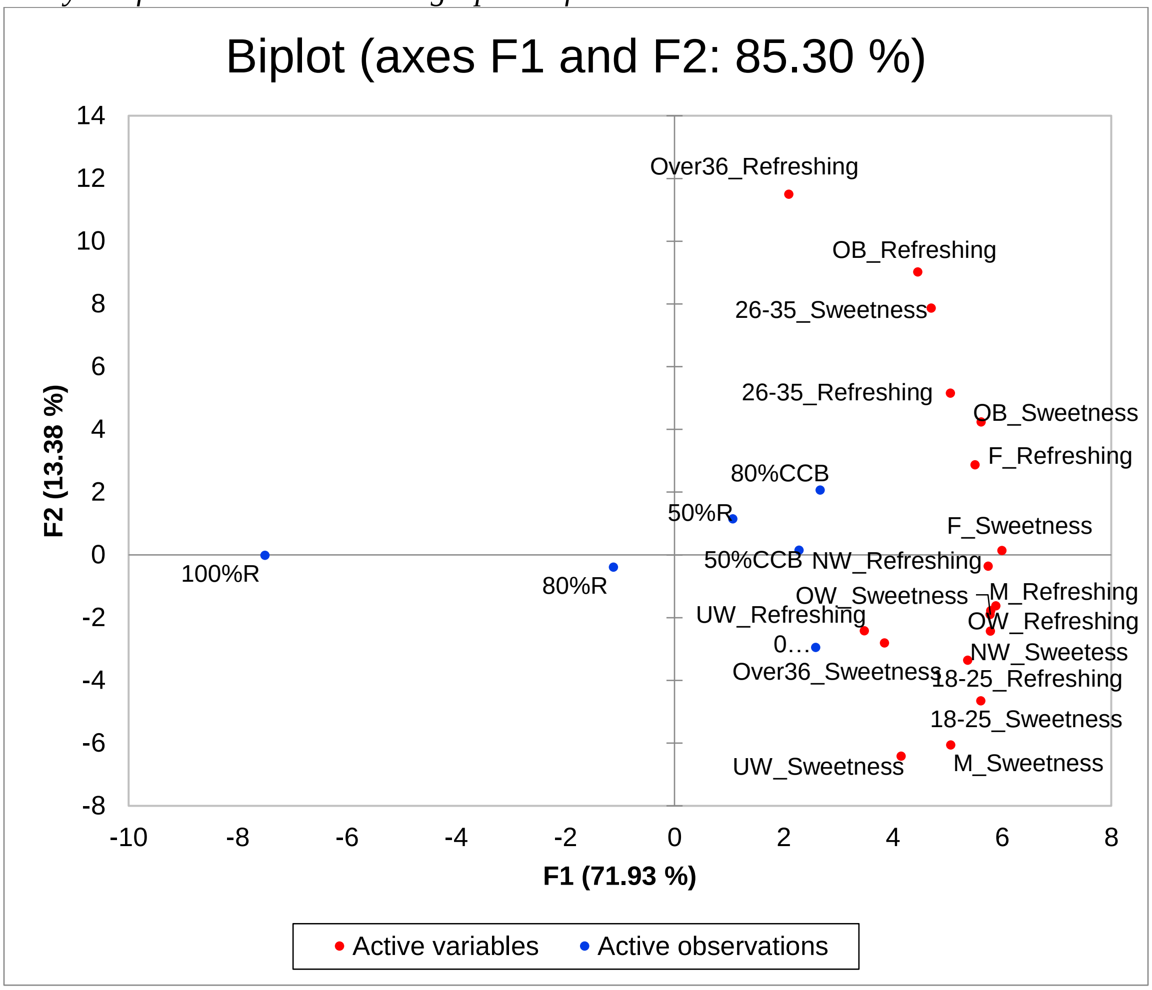
Table 10

Consumer Test Demographic Information

Gender	Population	Age	Population	BMI	Population
Male	15	18-25	70	Underweight	2
		26-35	26	Normal weight	56
Female	85	36-50	4	Overweight	23
				Obesity	19

Figure 20

Analysis of Consumer Test Demographic Information



CHAPTER V

DISCUSSION

Online Survey

According to the online survey results, flavored water was identified in the top four consumed drinks and beverages from the respondents. It indicated that flavored water was an important and major product in the beverage market (Ashurst, 2016).

This online survey indicated that consumers are seeking refreshing perception for their flavored water and beverage consumption. The results were coincident with another study that identified refreshing perception as the key factor of consumers’ flavored water and beverage choice (Menayang, 2016). According to this study, for flavored water and beverages being refreshing, there are many factors that affect its refreshing perception such as temperature, liquid color, liquid texture, carbonation, the sound, fruits, citrus, herbs, botanicals, and spices (Menayang, 2016). This online survey showed that citrus and fruity flavors were mostly preferred and temperature was the top consideration for the flavored water and beverage choice.

Beverage trends in 2019 showed that consumers are looking for protein beverages, nootropic beverages that can enhance brain performance and health, collagen/beauty beverages, beverages that help gut health, and functional waters with added vitamin, mineral, herbs, and fruit (Marci, 2018). This online survey showed similar trends where participants expected vitamins, minerals, and antioxidants as the extra benefit. Even though there were no choices or questions related to the brain, gut health or collagen/beauty beverages in this survey, this online survey was still reliable in identifying the consumers' expected benefits in flavored water and beverage selections.

Consumers are seeking reduced sugar or non-sugar foods or beverages as they recognize over consumption of sugar leads to negative health outcomes such as obesity, cardiovascular disease, diabetes, and cavities (Hutchings, Low, & Keast, 2018). This online survey results showed that flavored water choice and beverages were affected by well-being and health concerns. This tendency led to participants' preference for less or non-sugar products and natural sweeteners such as, pure sugars, honey, or agave syrup. These results were consistent with market trends that indicate that the flavored water and beverage market are growing and consumers are looking for new, natural, and healthy foods and beverages. Consumers are more concerned with natural ingredients on food products and the long-term health effect of added artificial ingredients and additives.

This online survey indicted the consumers' expectation and experience with flavored water and beverages. However, this online survey had limitations in that the results would be female biased since 91% of respondents were female. However, 76.4% of the participants were shown to drink flavored water/beverage at least once per month so the results are more related to regular consumers of these products.

Cucumber Volatile Analysis and Flavor Reconstitution

A total of 98 volatile compounds were identified with the major volatile compounds being aldehydes, alcohols, ketones, esters and furans. The most abundant carbon groups were C5 and C9 aldehydes and alcohols. C6s, C8s, were the rest. In the literature, it has been shown that C9 and C6 aldehydes and alcohols are identified as important volatile components in cucumbers (Chen et al., 2015). According to the finding from this research, C6 aldehydes and alcohols, hexanal, hexanol, (E)-2-hexen-1-ol, and (E)-3-hexen-1-ol contribute grassy and green flavors of cucumber. Hexanal and hexanol were identified in all cucumber varieties. Hexanal is the ingredient that contributed green and grassy flavors of cucumber. Other research studies also investigated the melon-like or fatty flavor that is associated with C9 aldehydes and alcohols (Hao et al., 2013; Forss et al., 1962; Palma et al., 2001).

Results of this study indicated that (E,Z)-2,6-nonadienal was identified as the most abundant compounds with its content in the four cucumbers at English (22.64%), American (27.96%), Kirby (27.60%), and Persian (27.23%). (E)-2-nonenal was the second most abundant compounds in the cucumbers at 12.51% in English, 27.63% in American, 0.45% in Kirby, and 21.75% in Persian. Therefore, (E,Z)-2,6-nonadienal and (E)-2-nonenal were regarded as the major cucumber volatile compounds.

The cucumber reconstitution was performed to mimic cucumber flavor. The cucumber flavor reconstitution has not been evaluated in the literature. Based on different dilutions of cucumber stock solution, samples were perceived with different characteristics. 0.6 ppm, 0.8 ppm, and 1 ppm dilution samples were created and evaluated. In samples tasted by trained panelists, the best cucumber solution was the 0.8 ppm. 0.6 ppm was found to be too weak to blend of each ingredients well and 1 ppm was perceived as too fatty and soapy since its major chemical compound gives fatty flavors. This fatty flavor was contributed by (E,Z)-2,6-nonadien-1-ol and (E,Z)-2,6-nonadienal and consisted 17% of cucumber stock solution. As (E,Z)-2,6-nonadien-1-ol and (E,Z)-2,6-nonadienal increased, fatty and soapy flavor were revealed. Therefore, 0.8 ppm was selected as the best solution.

The results indicate that each cucumber contained different aroma-active compounds and had unique flavors based on its chemical composition. The reconstituted cucumber flavor was well accepted by the panelists to represent its unique cucumber flavor. The limitation of this study was that only cucumber volatile identification analysis had been conducted. In future studies, volatile quantification and gas-chromatography-olfactometry (GC-O) would identify each volatile compounds' flavor profile in the cucumber.

Flavored Water QDA and Consumer Test

Few sensory related studies have investigated the impact of sugar on beverages and flavored water (Andersen et al., 2017; Mielby et al., 2016; Wise et al., 2016,). Sugar not only contributes to the sweet taste, but also can suppress bitterness and sourness and enhance the intensity of flavor (Ashurst, 2016). As shown in this study, sugar not only played a role as sweetener but also as sugar decreased, bitterness and astringent intensity increased but lemon and lemonade flavor intensity decreased. The results indicated that sugar played in important role in masking off-flavors such as bitterness and astringent while boosting the flavor intensity in beverages.

No research has been reported evaluation cucumber flavor impact on lemon flavored water sensory properties, to the best of our knowledge. However, the results from this study including flavored water sensory analysis (QDA) and consumer tests indicated that adding cucumber flavor enhanced the refreshing perception in 50% and 80% sugar reductions. The QDA result showed cucumber flavor increased its refreshing perception intensity 1.2 more at 50% sugar reduction and1.5 more at 80% reduction, respectively. Consumer tests also indicated that refreshing liking mean score was the same at a 50% sugar reduction but slightly increased 0.4 more at 80% reduction when cucumber flavor was added.

Refreshing perception is related to psychological and physiological enhancement such as thirst quenching, rehydration, energizing, and mental energy enhancement (Labbe et al., 2009a). Even though there is no sensory research investigating the relationship between refreshing perception and added cucumber flavor or sugar level, one study has investigated the impact of thirst quenching perception in water based products as affected by the impact of temperature, flavor, and sugar content. The study indicated that there was no significant difference in thirst quenching in regular and sugar-reduced popsicles. However, in this thesis it was found that 100% sugar-reduced lemon flavored water was identified as the least refreshing (4.53), compared to 4.7 at 80% reduction, 4.8 at 50% reduction, and 4.6 at 0% reduction. In addition, refreshing in lemon and raspberry flavor popsicles did not differ significantly. However, mint flavor increased flavor intensity, thirst quenching, and refreshing perception. Even though this thesis study only focused on lemon flavored water, refreshing perception was shown to differ significantly. Moreover, cucumber flavor enhanced the refreshing perception in both 50 and 80% sugar reductions.

No research has evaluated on consumers' liking and acceptance for lemon flavored water yet. This thesis study showed that 0% sugar reduction, 50% reduction, 50% reduction with cucumber flavor and 80% reduction with cucumber flavor were acceptable. In addition, cucumber flavor could enhance overall flavor intensity, refreshing, and sweetness in both 50% and 80% reduction. Overall flavor intensity increased significantly *(*$p < 0.05$*)* 0.6 in 50% reduction and 1.2 in 80% reduction with cucumber flavor added. Refreshing and sweetness significantly *(*$p < 0.05$*)* increased 0.2 and 0.1 in 50% reduction and 0.4 and 0.6 in 80% reduction. Even though 80% reduction was not acceptable because of its low sugar content, cucumber flavor helped to compensate for sweetness. Cucumber flavor worked better at 80% reduction than 50% reduction for consumers' liking and acceptance.

Regarding consumer test data collection, even though 100 participants are enough to be reliable to analyze consumers' liking and preference, it was still limited to validate demographic data. There were only 2 people in the underweight group and only 4 people participated in the 36-50 years old group.

CHAPTER VI

CONCLUSION

Flavored water was ranked the fourth most popular drink according to an online survey. Temperature, flavor, and sweet taste were the top three factors that consumer considered when they chose flavored water. Regarding flavor, lemon was the most popular flavor and consumer watched for sweetness (sugar), while bitterness was least preferred. Regarding the functional properties of flavored water, refreshing perception was rated the highest. Therefore, flavored water with a focus on lemon flavor, sugar reduction, and refreshing perception was chosen as this thesis research topic.

Cucumber has unique flavors composed of a mixture of green, grassy, melon-like, and fatty flavors. Cucumber flavors mostly consisted of C6 and C9 aldehydes and alcohols. (E,Z)-2,6-nonadienal and (E)-2-nonenal were determined to be responsible for the unique cucumber flavor. Cucumber flavor reconstitution study verified this volatile analysis results.

The contribution of cucumber flavor to flavored water has never been investigated. In this thesis study, the functions of cucumber flavor added to sugar-reduced flavored water was tested by both QDA and consumer study. Overall, according to QDA and consumer test results, 50% sugar reduction was a cut-point for sugar reduction without maintaining sweetness intensity in flavored water. Adding cucumber flavor to sugar reduced flavored waters increased refreshing and sweetness perception in both 50% and 80% sugar reductions. Cucumber flavor was highly correlated to liking perception of consumer acceptance including overall character, overall flavor liking, flavor balance liking, refreshing, mouthfeel, and sweetness. Cucumber flavor was also positively correlated in green, fresh, refreshing profiles of the lemon flavored water. It would be worth to developing and launch less sugared flavored water with more refreshing effect containing cucumber flavors.

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APPENDIX A

Institutional Review Board Approval Letter

Institutional Review Board
Office of Research and Sponsored Programs
P.O. Box 425619, Denton, TX 76204-5619
940-898-3378
email: IRB@twu.edu
https://www.twu.edu/institutional-review-board-irb/

DATE: October 9, 2018

TO: Dr. Xiaofen Du
Nutrition & Food Sciences

FROM: Institutional Review Board (IRB) - Denton

Re: Exemption for Exploring Consumer Liking for Sugar Reducing without Maintaining Sweetness Intensity in Flavored Water and the Impact of Beverage Secondary Functions (Protocol #: 20261)

The above referenced study has been reviewed by the TWU IRB (operating under FWA00000178) and was determined to be exempt from further review.

If applicable, agency approval letters must be submitted to the IRB upon receipt PRIOR to any data collection at that agency. Because a signed consent form is not required for exempt studies, the filing of signatures of participants with the TWU IRB is not necessary.

Although your protocol has been exempted from further IRB review and your protocol file has been closed, any modifications to this study must be submitted for review to the IRB using the Modification Request Form. Additionally, the IRB must be notified immediately of any adverse events or unanticipated problems. All forms are located on the IRB website. If you have any questions, please contact the TWU IRB.

cc. Dr. Shane Broughton, Nutrition & Food Sciences

TWU

Uijeong An <uan@twu.edu>

IRB-FY2019-143 - Initial: Exempt Letter

irb@twu.edu <irb@twu.edu>
To: uan@twu.edu, xdu@twu.edu

Wed, Apr 10, 2019 at 10:43 PM

Texas Woman's University
Institutional Review Board (IRB)
irb@twu.edu
https://www.twu.edu/institutional-review-board-irb/

April 10, 2019

Ujieng An
Nutrition and Food Sciences

Re: Exempt - IRB-FY2019-143 Online survey for flavored water/beverage

Dear Ujieng An,

The above referenced study has been reviewed by the TWU IRB - Denton operating under FWA00000178 and was determined to be exempt on April 9, 2019. If you are using a signed informed consent form, the approved form has been stamped by the IRB and uploaded to the Attachments tab under the Study Details section. This stamped version of the consent must be used when enrolling subjects in your study.

Note that any modifications to this study must be submitted for IRB review prior to their implementation, including the submission of any agency approval letters, changes in research personnel, and any changes in study procedures or instruments. Additionally, the IRB must be notified immediately of any adverse events or unanticipated problems. All modification requests, incident reports, and requests to close the file must be submitted through Cayuse.

Approval for this study will expire on April 8, 2020. A reminder of the study expiration will be sent 45 days prior to the expiration. If the study is ongoing, you will be required to submit a renewal request. When the study is complete, a close request may be submitted to close the study file.

If you have any questions or need additional information, please contact the IRB analyst indicated on your application in Cayuse or refer to the IRB website at http://www.twu.edu/institutional-review-board-irb/.

Sincerely,

TWU IRB - Denton

APPENDIX B

Institutional Review Board Consent Form

TEXAS WOMAN'S UNIVERSITY
CONSENT TO PARTICIPATE IN RESEARCH

Title: Exploring Consumer Liking for Sugar Reducing without Maintaining Sweetness Intensity in Flavored Water and the Impact of Beverage Secondary Functions.

Investigator: Xiaofen Du, PhD .... Xdu@twu.edu 940-898-2667
Graduate Assistant: Uijeong An .... UAn@twu.edu 940-898-2663

Explanation and Purpose of the Research

You are being asked to participate in a research study conducted by Dr. Xiaofen Du and MS Uijeong An at Texas Woman's University. The purpose of this research is to (A) investigate the impact of reduced sugar (0%, 30%, 50%, 80%, and 100%) on flavored water with lemon, lemon tea, tropical and mixed berry flavors, respectively and acceptance of those products, and(B) investigate consumer liking on the four different flavored water with reduced sugar at either 50% or 80% and the addition of two secondary flavor functions (such as refreshing and thirst quenching).

Description of Procedures

In order to be a participant in this study, you must be 18-50 years of age and consume flavored water regularly. You must be not allergic to lemon, tropical fruit, or berries. In this study, sensory test will be separated into two; (1) Quantitative Descriptive Analysis, (2) Consumer test. The overall procedure will be:

(1) Quantitative Descriptive Analysis

Flavored water will be tested in this study formulated from Firmenich. Descriptive Analysis will be carried out in the sensory lab at Texas Woman's University. Panelists will taste flavored water to develop descriptive lexicons, along with defin tions and references. The panel will then be trained over several sessions to practice rating the intensity of the attributes. All products will be served in 2 oz plastic portion cups covered with a plastic lid. The tests will be conducted in isolated booths. The order of the presentation of the products will be randomized across subjects. Panelists will rinse their mouths between samples with bottled unflavored spring water. Each product will be evaluated in duplicate. It will take a maximum of 40-60 min per section, and 6-10 hours maximum cumulative time.

(2) Consumer test

Flavored water will be tested in this study formulated from Firmenich. Consumer test will be carried out in the sensory lab at Texas Woman's University. Panelists will drink all the samplers to rate their degree of acceptance and preference according to the questionnaire which include hedonic questions and Just-About-Right scale question. All products will be served in 2 oz plastic portion cups covered with a plastic lid. The tests will be conducted in isolated booths. The order of the presentation of the products will be randomized across subjects. Panelists will rinse their mouths between samples with bottled unflavored spring water. After evaluation, panelists will complete the exit survey. The session will be run through the whole day, from 10 AM to 5 PM.

____
Initials

Potential Risks

Allergens, as with all food products, may be a concern for consumers allergic to lemon, tropical fruit, or berries. All participants will be verbally screened for allergens prior to participate in the taste-testing. The procedures and flavored water in this experiment post no additional risks when compared to foods normally eaten by consumers.

A loss of confidentiality is always a possibility. Facts to reduce issues with confidentiality will include no linking of subject to scientific raw data. Consent forms will be retained in a locked cabinet and destroyed and shredded at study termination and manuscript submission.

There is potential risk of loss of confidentiality in all email and downloading. Confidentiality will be protected to the extent that is allowed by law.

The researchers will try to prevent any problem that could happen due to this research. You should let the researcher know at once if there is a problem with food allergies so they can assist you. However, TWU does not provide medical services or financial assistance for injuries that might happen because you are taking part in this research.

Participation and Benefits

Your involvement in this study is completely voluntary and you may withdraw from the study at any time. Following the completion of the study you will receive a $10 gift card for your participation. If you would like to know the results of this study we will mail them to you.*

Questions Regarding the Study

You will be given a copy of this signed and dated consent form to keep. If you have any questions about the research study you should ask the researchers; their phone numbers are at the top of this form. If you have questions about your rights as a participant in this research or the way this study has been conducted, you may contact the Texas Woman's University Office of Research and Sponsored Programs at 940-898-3378 or via e-mail at IRB@twu.edu.

____ ____________

Signature of Participant Date

____

Initials

This survey is approved by the Texas Woman's University Institutional Review Board. Your participation is voluntary and input is confidential.

Potential Risks

Completion of the survey will provide no more than minimal risk, such as loss of time. The survey is administered online; there is a potential loss of confidentiality in all email, downloading, and Internet transactions.

Entering the gift card drawing requires that you provide your email address. This means that your responses will no longer remain anonymous. However, your responses will not be shared or used for purposes other than this research project. Entry is not required.

The return of your completed questionnaire constitutes your informed consent to act as a participant in this research.

- ❑ I accept

APPENDIX C

Institutional Review Board Script

Email Script

(Quantitative Descriptive Analysis)

Research Study is Open to TWU and Non-TWU Individuals (DENTON CAMPUS LOCATION)

Research volunteers needed for sugar reduction on flavored water panel! Overconsumption of added sugar has been linked with negative healthy outcomes. Do you regularly drink flavored water? If so you may be eligible to participate in a trained taste panel for sugar reduction. If you are between 18-50 years old and otherwise healthy you may qualify. Participants will smell and taste four sets of different levels of sugar contained flavored water, with maximum consumption of 2 ounce each section. Participants will participate in descriptor training section and then evaluate flavored water. It requires 40-60 min per session; 6-10 hours maximum cumulative time. A taste ballot including no more than 20 questions will be signed and finished during the taste section.

Benefits include: Experiencing smell and taste of different levels of sugar contained flavored water. Upon completion, you will receive compensation via a $10 gift card for your participant.

If interested, please contact Dr. Xiaofen Du at xdu@twu.edu or 940-898-2667.

There is a potential risk of loss of confidentiality in all email, downloading, electronic meetings, and internet transactions.

Email Script

(Consumer Test)

Research Study is Open to TWU and Non-TWU Individuals (DENTON CAMPUS LOCATION)

Research volunteers needed for sugar reduction on flavored water panel! Overconsumption of added sugar has been linked with negative healthy outcomes. Do you regularly drink flavored water? If so you may be eligible to participate in consumer test for sugar reduction. If you are between 18-50 years old and otherwise healthy you may qualify. Participants will smell and taste four sets of different levels of sugar contained flavored water, with maximum consumption of 2 ounce each section. Participants will participate in tasting flavored water. The session will be run through the whole day, from 10 AM to 5 PM. A taste ballot including no more than 20 questions will be signed and finished during the taste section.

Benefits include: Experiencing smell and taste of different levels of sugar contained flavored water. Upon completion, you will receive compensation via a $10 gift card for your participant.

If interested, please contact Dr. Xiaofen Du at xdu@twu.edu or 940-898-2667.

There is a potential risk of loss of confidentiality in all email, downloading, electronic meetings, and internet transactions.

Purpose

Under the TWU Flavor Chemistry program, research is being conducted on the concept of Exploring Consumer Expectation of Flavored Water/Beverage.

Procedure

You are invited to complete a questionnaire to help us gather preliminary information on the sugar reduced flavored water. The survey is administered online.

The survey will take approximately 10 minutes to complete. You will have the option to stop at any time. Click the following link to participate:

TEXAS WOMAN'S UNIVERSITY

Exploring Consumers' Expectation of Flavored Water Survey

Survey Link

Compensation

Upon completion you can enter for a chance to win Amazon gift cards. (valued at $20 each).

Contact Information

Please contact me (UAn@twu.edu) or my faculty advisor Dr. Du (XDu@twu.edu) if you have any questions about this project.

Thank you for your assistance.

APPENDIX D

Panel Recruitment Flyer

TEXAS WOMAN'S
UNIVERSITY

Consumer Test

Sugar-Reduced Lemon Flavored Water

Research Study

To investigate consumer liking of sugar-reduced flavored water

Products

Lemon flavored waters with reduced sugar level are formulated in food production lab, Nutrition and Food Sciences.

When

- 3/21 (Thursday): 9:00am~ 1:00pm; 1:00pm~ 4:00pm
- 3/25 (Monday): 9:00am~ 1:00pm; 1:00pm~ 4:00pm
- 3/27 (Wednesday): 9:00am~ 1:00pm; 1:00pm~4:00pm

Where

- Woodcock Hall 113, Denton campus, Texas Woman's University

- **Please Pre-Register:** https://goo.gl/forms/MVNGVGxoRRg9N6ZZ2

The test will last 15-20 min

Participants will received $5 after completion of the test

Participation is voluntary and you can withdraw at any time

If you have any questions, please contact

either Dr. Xiaofen Du at xdu@twu.edu (940-898-2667) or Uijeong An at UAn@twu.edu

There is a potential risk of loss of confidentiality in all email, downloading, electronic meetings, and internet transactions.

TEXAS WOMAN'S UNIVERSITY

Exploring Consumer Expectation of Flavored Water/Beverage Survey

Research Study

Under the TWU Flavor Chemistry program, research is being conducted on the concept of Exploring Consumer Expectation of Flavored Water/Beverage.

Procedures

You are invited to complete a questionnaire to help us gather preliminary information on the sugar reduced flavored water. The survey is administered online.

The survey will take approximately 5~10 minutes to complete. You will have the option to stop at any time. Click the following link to participate.

Survey Link

forms.gle/FdAmWsT2VLFKLSp99

You have chance to win **$20 Amazon gift cards**

If you have any questions, please contact Uijeong An at UAn@twu.edu or Dr. Xiaofen Du at XDu@twu.edu (940-898-2667).

There is a potential risk of loss of confidentiality in all email, downloading, electronic meetings, and Internet transactions.

APPENDIX E

Quantitative Descriptive Analysis Test Ballot

Name: QDA

Flavored Water QDA Test

You are participating in a study to evaluate different kinds of flavored water.

- **If you know you are allergic to lemon, tropical fruit, or berries, please withdraw from the panel.**
- **If you are or think you are pregnant, if you are nursing, or if you are immune compromised, please withdraw from the panel.**

The sensory evaluation

- **Please taste the samples in the order given (left to right) and rate the intensity of each attribute. Make sure you are taking the correct sample number.**
- **Rinse your mouth with water between samples and take some crackers to clean your pallet.**
- **Keep calibrating yourself with the standards during the panel.**

THANK YOU!!!

Lemon

Name: QDA

Smell and Taste

Overall Flavor Intensity

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Flavor Balance

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
off med highly

Refreshing

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Lemon

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Fresh

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Green

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Juicy

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Lemonade

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Cucumber

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Citrus

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Lemon

Name: QDA

Sweet

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Sour

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Bitter

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Astringent

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Irritate

0_______1_______2_______3_______4_______5_______6_______7_______8_______9_______10
low med high

Overall Comments

Lemon

APPENDIX F

Consumer Test Ballot

Welcome to the sugar reduction panel!

You are participating in a study to evaluate flavored water.

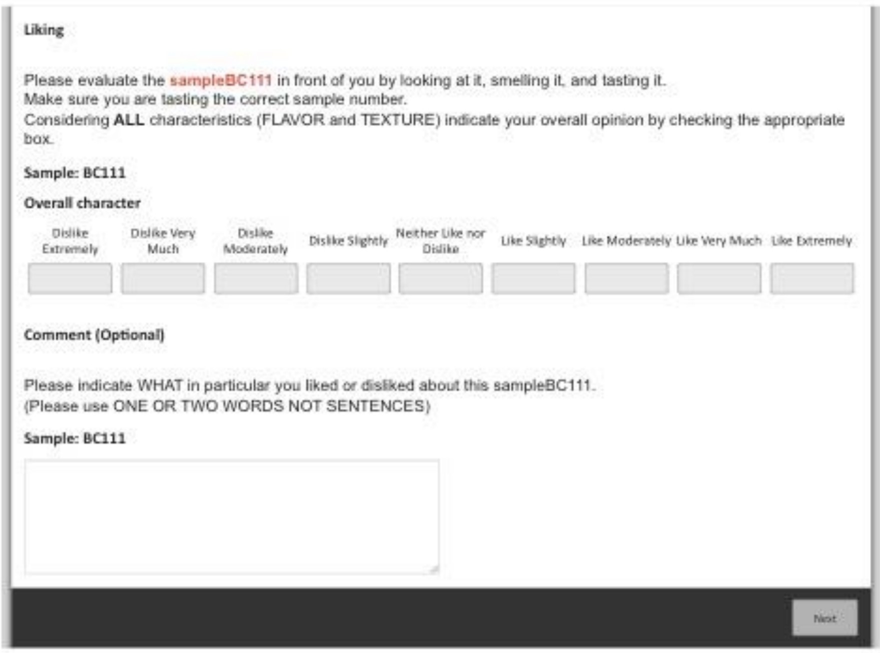
If you know you are allergic to lemon, tea, tropical fruits, or berries, please withdraw from the panel.

If you are or think you are pregnant, if you are nursing, or if you are immune compromised, please withdraw from the panel.

Click the *next* button to begin

Next

1/20


Liking

Please evaluate the **sampleBC111** in front of you by looking at it, smelling it, and tasting it.
Make sure you are tasting the correct sample number.
Considering **ALL** characteristics (FLAVOR and TEXTURE) indicate your overall opinion by checking the appropriate box.

Sample: BC111

Overall character

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Comment (Optional)

Please indicate WHAT in particular you liked or disliked about this sampleBC111.
(Please use ONE OR TWO WORDS NOT SENTENCES)

Sample: BC111

Next

2/20

Category- Liking

Please re-taste the **sampleBC111** as needed and indicate how much you **LIKE or DISLIKE** the following. Check the box that represents your response.

Sample: BC111

Overall flavor/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Ovreall flavor/ Intensity level

Extremely Light	Very Light	Moderately Light	Slightly Light	Neither Strong nor Light	Slightly Strong	Moderately Strong	Very Strong	Extremely Strong

Flavor balance/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Next

3/20

Category- Liking

Please re-taste the **sampleBC111** as needed and check the box for your response for both questions (**LIKING** and **INTENSITY LEVEL**) for each characteristic.

Sample: BC111

Refreshing perception/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Refreshing perception/ Intensity level

Extremely Weak	Very Weak	Moderately Weak	Slightly weak	Neither Strong nor Weak	Slightly Strong	Moderately Strong	Very Strong	Extremely Strong

Lemon flavor/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Lemon flavor/ Intensity level

Extremely Weak	Very Much Weak	Moderately Weak	Slightly Weak	Neither Strong nor Weak	Slightly Strong	Moderately Strong	Very Much Strong	Extremely Strong

Mouthfeel/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Mouthfeel/ Intensity level

Extremely Thin	Very Much Thin	Moderately Thin	Slightly Thin	Neither Thick nor Thin	Slightly Thick	Moderately Thick	Very Much Thick	Extremely Thick

Sweetness/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Sweetness/ Intensity level

Extremely Weak	Very Much Weak	Moderately Weak	Slightly Weak	Neither Strong nor Weak	Slightly Strong	Moderately Strong	Very Much Strong	Extremely Strong

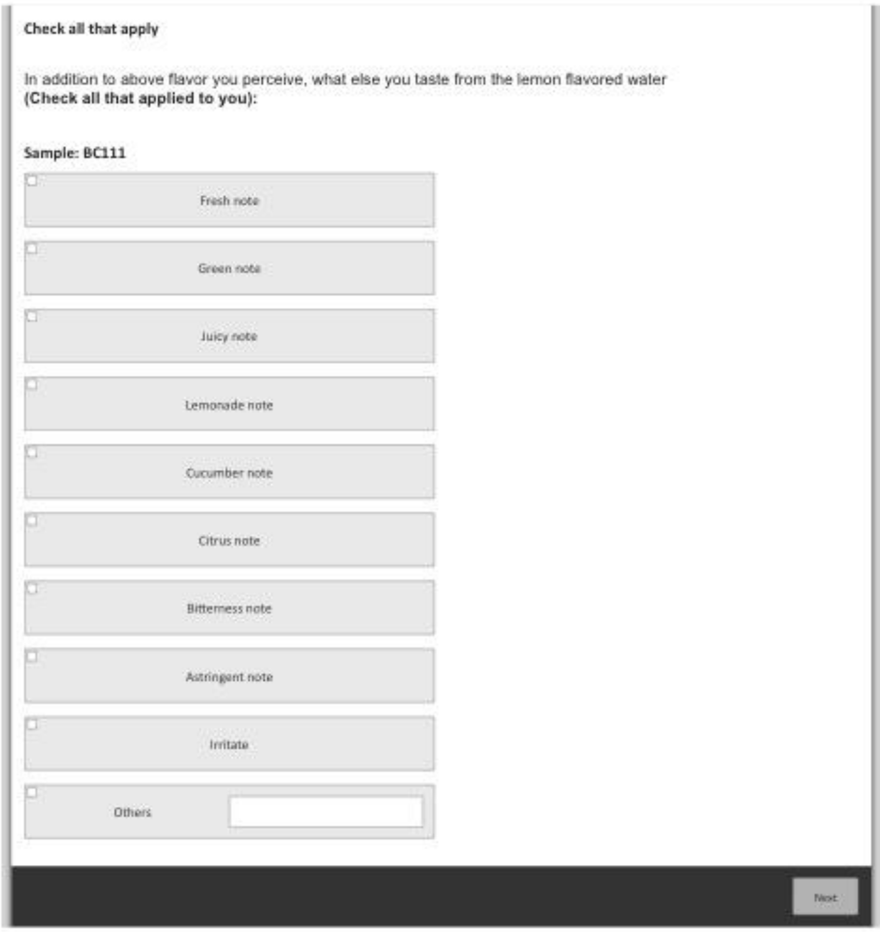
Sourness/ Liking

Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely

Sourness/ Intensity level

Extremely Weak	Very Much Weak	Moderately Weak	Slightly Weak	Neither Strong nor Weak	Slightly Strong	Moderately Strong	Very Much Strong	Extremely Strong

Next



Check all that apply

In addition to above flavor you perceive, what else you taste from the lemon flavored water
(Check all that applied to you):

Sample: BC111

- [ ] Fresh note
- [ ] Green note
- [ ] Juicy note
- [ ] Lemonade note
- [ ] Cucumber note
- [ ] Citrus note
- [ ] Bitterness note
- [ ] Astringent note
- [ ] Irritate
- [ ] Others

Next

6/20


Comment (Optional)

Your Overall Comments

Next

7/20


Overall differneces
Overall differences within all given samples
Extremely Different
Very Much Different
Moderately Different
Slightly Different
Neither Not Different nor Different
Slightly Not Different
Moderately Not Different
Very Much Not Different
Extremely Not Different
Next

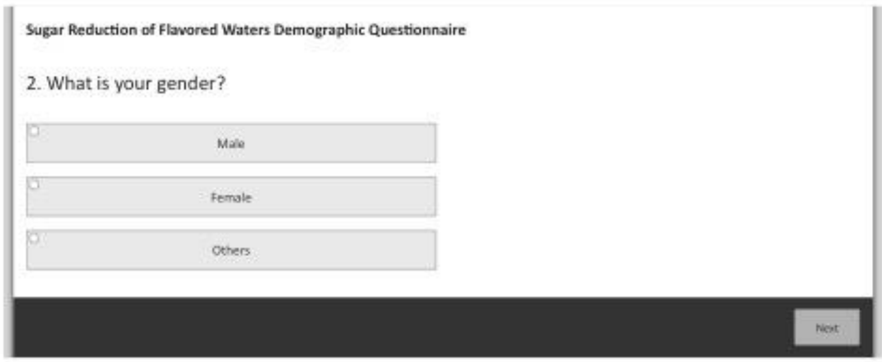
8/20

Sugar Reduction of Flavored Waters Demographic Questionnaire

1. What is your age group?

- 18~25
- 26~30
- 31~35
- 36~40
- 41~45
- 45~50

Next


Sugar Reduction of Flavored Waters Demographic Questionnaire

2. What is your gender?

Male

Female

Others

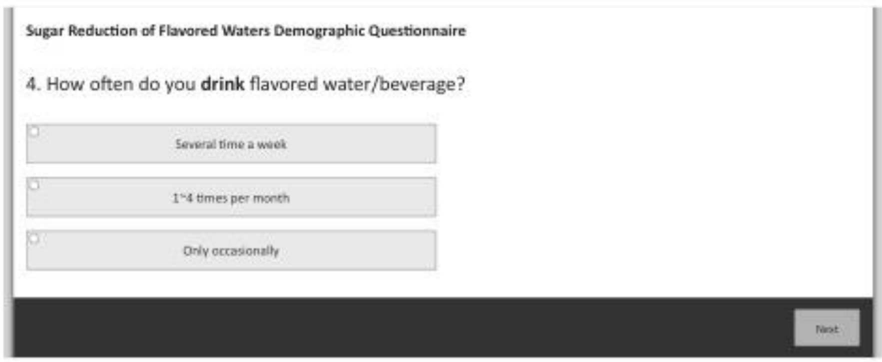
Next

Sugar Reduction of Flavored Waters Demographic Questionnaire

3. How often do you **purchase** flavored water/beverage?

- Once a week or more often
- Every 2~4 weeks
- Less than once a month
- Never

Next


Sugar Reduction of Flavored Waters Demographic Questionnaire

4. How often do you **drink** flavored water/beverage?

- Several time a week
- 1~4 times per month
- Only occasionally

Next

Sugar Reduction of Flavored Waters Demographic Questionnaire

5. Which type of flavored water/beverage do you consume most frequently

- ☐ Lemon
- ☐ Lemon flavored tea
- ☐ Tropical
- ☐ Mixed berry
- ☐ Others ______

Next

Sugar Reduction of Flavored Waters Demographic Questionnaire

6. How often do you look for new type of flavored water/beverage in the supermarket?

- Several times a week
- 1~4 times per month
- Only occasionally
- Never

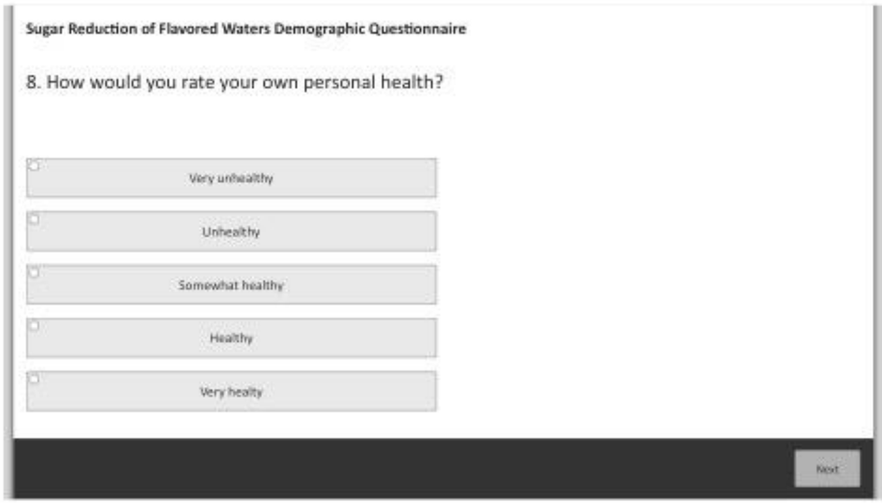
Next

Sugar Reduction of Flavored Waters Demographic Questionnaire

7. Are you suffering any food allergies?

- No
- If yes, please specify ____

Next



Sugar Reduction of Flavored Waters Demographic Questionnaire

8. How would you rate your own personal health?

- Very unhealthy
- Unhealthy
- Somewhat healthy
- Healthy
- Very healty

Next

Sugar Reduction of Flavored Waters Demographic Questionnaire

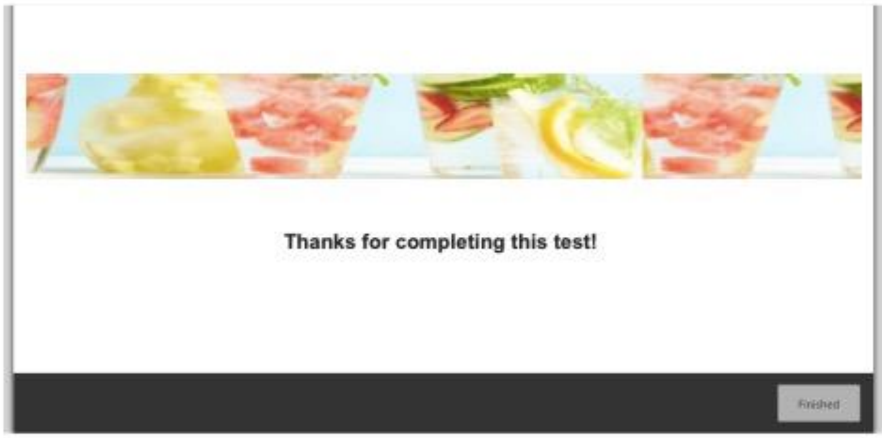
9. What is your BMI?

Type your BMI

Next

Please drink water or eat cracker to clean your palate!

Next


Thanks for completing this test!
Finished