RELATIONSHIPS BETWEEN CONSUMER PREFERENCE, FLAVOR ATTRIBUTES AND CHEMICAL COMPOUNDS OF VIDALIA ONION

by

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(Under the Direction of Gabriela Sanchez-Brambila)

ABSTRACT

The chemical compounds and flavor attributes of two Vidalia onion varieties (Nunhems 1006 – sweet variety; Sapelo – hot variety) grown at different fertilizer levels were analyzed to investigate their influence on flavor perception. The first part of this study consisted of the descriptive sensory analysis and chemical analyses of sugars (HPLC), lachrymatory factor (LF; propanethial S-oxide) and methyl thiosulfinates (GC-FID/FPD). The lachrymatory sensation and pungency/aftertaste were significantly correlated with the LF and methyl thiosulfinate content attributing the majority of Vidalia onion flavor, whereas sugars had little impact. In the second study, the relationship between consumer flavor preferences and chemical composition were defined. Samples containing low content of the LF (LF < 1.806 µmol/g) and methyl thiosulfinates were perceived as less pungent, and also preferred by the majority of consumers. Partial least squares (PLS) regression and agglomerative hierarchical clustering (AHC) revealed consumer segmentations with different preference patterns in the population tested.

INDEX WORDS: Vidalia onion, Sensory, Consumer preference, Pungency/aftertaste, Propanethial S-oxide, Methyl thiosulfinates

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DEDICATION

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INTRODUCTION

Onions (*Allium cepa* L.) are grown worldwide and represent the third largest fresh vegetable industry in the United States. Over the past years, onion consumption in the U.S. has considerably increased showing per capita consumption of 20 pounds in 2010 (National Onion Association, 2011). Onions gain popularity partly due to a heavy promotion linking their characteristic flavor with health benefits such as antioxidants and flavonoids (Griffiths, Trueman, Crowther, Thomas, & Smith, 2002). However, some of the strong and unpleasant flavors have deterred many consumers from eating onions despite their health beneficial aspects. Therefore, breeding and producing sweeter and milder onions has become popular to satisfy consumers' demand (Boyhan & Torrance, 2002; Lee, Yoo, Jifon, & Patil, 2009; Vidalia Onion Committee, 2015). Among various sweet onions, Vidalia onion is well-known for its sweetness and less pungency (Sellappan & Akoh, 2002). Under the regulation of the USDA, as well as a state law, Vidalia onions are defined as short-day yellow granex varieties that are grown in the designated areas in Georgia (Vidalia Onions Grown in Georgia, 2015).

Onion flavor is mainly attributed to genetic, environmental and post-harvest factors (Hamilton, Pike, & Yoo, 1997; Randle & Lancaster, 2002; Yoo, Pike, Crosby, Jones, & Leskovar, 2006). It has been reported that the amount of sulfur in soil is the major environmental factor influencing the accumulation of flavor precursors which ultimately generate pungency in onions (Coolong, Kopsell, Kopsell, & Randle, 2005; Freeman & Mossadeghi, 1970; Randle et al., 1995). It is known that the sweeter and less pungent flavor of Vidalia onions come from Georgia's mild climate with abundant rain and sandy soil which allows sulfur to leach out of the root zone.

Until now, there have been numerous attempts to uncover the relationship between onion pungency and volatile sulfur compounds, particularly in terms of the levels of enzymatically generated pyruvic acid (EPY) in onion juice (Schwimmer & Weston, 1961; Terry, Law, Hipwood, & Bellamy, 2005; Wall & Corgan, 1992; Yoo & Pike, 2001). EPY has been used as a standard measure of gross pungency due to its simplicity of assessment. However, this approach have shown some shortcomings because pyruvic acid is not a direct measure of flavor bioactives (McCallum et al., 2005) and meaningful variations in EPY results have been found between different laboratories (Havey et al., 2002). Moreover, due to the fact that pyruvic acid does not give taste by itself, it could not be able to reveal reliable correlation with pungency perceived by human.

Despite all the earlier endeavors, there has been no report that uncovered the relationship between chemical composition of onions and flavor with well-established and systematic sensory analysis techniques. Instrumental analyses provide objective data but only human sensory system can generate true flavor perception; human perception comes from intricate chain of recognition and interpretation of sensory information in brain (Lawless & Heymann, 2010).

Therefore, the aims of current study were (1) to investigate the influence of onion variety and levels of fertilizer on the chemical composition of Vidalia onions; (2) to establish the relationship between chemical composition and the actual flavor perception by trained panel; (3) to define desirable range of chemical compounds' concentration in

Vidalia onion based on consumer preferences; and (4) to gain in-depth insight into Vidalia onion consumers by relating chemical composition with consumer sensory perception and preference.

LITERATURE REVIEW

Onion Flavor Chemistry

The characteristic flavor and aroma of onions are developed through unstable and complicated chain reaction (Yoo, Lee, & Patil, 2012). The flavor profile of onions (*Allium cepa* L.) is mainly determined by the 3 major flavor precursors of S-alk(en)yl-L-cysteine sulfoxides (ACSOs): S-methyl-, S-propyl and S-1-propenyl cysteine sulfoxides. In general, the S-1-propenyl cysteine sulfoxides (PRENCSO) is found in the highest concentration, whereas the S-methyl-cysteine sulfoxides (MCSO) and S-propyl-cysteine sulfoxides (PCSO) are found to have lower concentrations (Lancaster & Boland, 1990). In the process of biosynthesis of ACSOs, sulfur (S) is absorbed by the roots as sulfate (SO_4^{2-}), reduced to sulfide in the plant, and assimilated into cysteine (Randle et al., 1995). Some of the cysteine go through the glutathione cycle and then incorporated into a variety of γ-glutamyl peptides (γ-GPs) which are intermediates in the pathway to ACSOs (Lancaster & Shaw, 1989).

When the onion cells are mechanically ruptured, the enzyme alliinase (E. C. 4.4.1.4) is released from vacuole and hydrolyzes the flavor precursors (ACSOs). This reaction produces pyruvic acid, ammonia and many sulfur volatiles including unstable sulfenic acids (Whitaker, 1976). Sequentially, these acids rapidly react to form methyl thiosulfinates and tear-causing propanethial S-oxides, generally referred to as the lachrymatory factor (LF) (Corzo-Martínez, Corzo, & Villamiel, 2007; Lancaster, Shaw, & Randle, 1998). Methyl thiosulfinates have been reported to be responsible for the characteristic flavors related to fresh onions (Block, 1985; Block, Naganathan, Putman, & Zhao, 1992; Freeman & Whenham, 1976). LF is formed from S-1-propenyl sulfenic acid following PRENCSO hydrolysis, with the action of LF-synthase (Block, Penn, & Revelle, 1979). The LF is responsible for the mouth burn and heat when consuming onions (Kopsell, Randle, & Schmidt, 2002).

The intensity of onion flavor can be attributed to genetic, environmental and post-harvest factors (Randle & Lancaster 2002). Earlier studies have shown that the amount of sulfur in soil is the major environmental factor resulting in the accumulation of total onion ACSOs, as well as increase in the ratio of individual ACSOs; thus, high levels of sulfur in fertilizer causes greater flavor intensity (Freeman and Mossadeghi 1970; Randle and others 1995). It has also been demonstrated that the concentration of LF and thiosulfinates increased linearly with the increase of the sulfur rate in fertilizer applied to the soil (Randle, Block, Littlejohn, Putman, & Bussard, 1994).

Sweetness is another important factor in onion flavor. The three main sugars, which are glucose, sucrose and fructose, comprise the majority of the soluble solids of the onion, contributing more than 65% to the dry weight (Crowther et al., 2005). Sugar content influences sweetness and overall onion flavor is determined by the ratio of sugar to pungency (Vavrina & Smittle, 1993). However, some studies suggested that the contribution of sugars to onion sweetness could not be determined precisely, particularly while tasting uncooked or strong onions, due to potential masking effect by other flavor compounds (Crowther et al., 2005; Green, 1996). This is also supported by other studies which identified the masking and inhibitory effects of oral chemical irritation on taste perception, specifically on sweetness, by the irritants causing a burning sensation (Lawless & Stevens 1984; Prescott & Stevenson, 1995).

Methodologies for Consumer Preference Analysis

Understanding which sensory attributes drive consumer preference toward food and food products is important in food industries. In earlier sensory studies, many techniques have been used to relate consumer (hedonic) data with analytical data sets such as sensory descriptive and/or instrumental measurements. Most of the approach was based on the idea of regressing averaged consumer hedonic ratings onto analytical data. However, these analyses failed to capture the inter-individual differences among consumers because the prediction was made on an average consumer. All consumers does not exhibit essentially the identical behavior (perception and preference) and thus, a single mean value cannot represent the entire population (Guinard, Uotani, & Schlich, 2001). Recently, a variety of techniques have been developed to explore the consumer preferences in multidimensional spaces which made it possible to examine interrelationships among products and individual consumers' liking patterns regarding a set of analytical (sensory descriptive or instrumental) data (Guinard, Uotani, & Schlich, 2001; MacFie & Thomson, 1988). These techniques include internal and external preference mapping (Arditti, 1997; Carroll, 1972) and partial least squares regression (Murray & Delahunty, 2000).

Internal preference mapping (MDPREF) is a principal component analysis (PCA) of the covariance matrix of consumers (variables) by products (objects) (Schlich, 1995). The preference map accounts for the variation in the covariance matrix and creates linear combinations of the original variables (Yackinous, Wee, & Guinard, 1999). These new variables are mutually orthogonal and explain decreasing amounts of variance. MDPREF requires consumer hedonic data only, and uses consumer preference in order to locate the products (samples) on the multidimensional space (Meilgaard, Civille, & Carr, 2006; Schlich, 1995). The map represents a summary of the directions of main underlying preferences as vectors, and the consumer segments with similar preference are associated with the preference vectors (Greenhoff & MacFie, 1994). In order to obtain a logical perceptual map, it is required to include at least 6 products evaluated by consumers (Lavine, Jurs, & Henry, 1988).

On the other hand, external preference mapping (PREFMAP) requires both consumer preference and external sources (e. g. sensory descriptive or instrumental data). In this technique, each consumer liking score is regressed onto the principal components (typically first two PCs) obtained from a PCA of the products' sensory attributes derived from sensory descriptive and/or instrumental analysis (Helgesen, Solheim, & Næs, 1997; Schlich, 1995). In other words, external preference mapping gives precedence to external sources to derive the product map, and then consumers' hedonic data are fitted into this space (van Kleef, van Trijp, & Luning, 2006). PREFMAP is fundamentally based on a polynomial regression of individual consumer hedonic scores given to the products onto the independent variables, which are coordinates of the products in the sensory space (Schlich, 1995). To fit the consumer data, four models are generally used: vectorial, circular, elliptical, and quadratic. With the external preference mapping technique, it is possible to reveal the underlying drivers of liking for the products and locate ideal products in consideration of certain consumer segments on the map (Schlich, 1995).

Although these two mapping techniques have been helpful in revealing the drivers of consumer liking and disliking in products, they do not provide exact predictions of optimal product profiles (Plaehn, 2009). Commenting on both internal and external preference mapping, Meullenet, Xiong, and Findlay (2007) wrote "Although these methods (MDPREF and PREFMAP) provide information about the relationship between liking by groups of consumers and sensory attributes, the optimal level of a specific attribute is not necessarily identified. From the standpoint of product development, this causes considerable problems. It is one thing to find through MDPREF or PREFMAP that saltiness drives liking, it is yet another to determine how much salt is enough or how much is too much. This is where we see preference mapping in its original forms fail to provide enough information to the product developer to formulate an optimal product from the sensory standpoint."

As mentioned, internal preference mapping (MDPREF) does not fully characterize the products because the product map is solely generated from consumer hedonic data. Therefore, to gain extended knowledge, MDPREF should be followed by extensive descriptive sensory analysis. External mapping approach also has limitation in that the multidimensional sensory space is generated from external sources alone with no prioritization of the sensory attributes based on their importance to consumers. Therefore, for the PREFMAP to be reasonable, it is essential that the space of external sources should contain dimensions which refer to consumer preferences (Jaeger, Wakeling, & MacFie, 2000).

Partial least squares (PLS) regression has been proved to have some advantages over internal and external preference analyses. The fundamental principle of PLS regression is similar to that of external preference mapping in that it aims at understanding the relationship between X-matrix (descriptive sensory or instrumental data) and Y-matrix (consumer preference) (Martens & Martens, 2001; Mitchell, Brunton, & Wilkinson, 2011). However, differences are in the data treatment processes; in PLS regression, the instrumental and sensory data are used simultaneously to locate products on the preference map by extracting PLS components (latent variables) that represent maximized covariance between linear functions of X- and Y-matrix (Næs, Brockhoff, & Tomic, 2010). Therefore, PLS regression extracts few linear combinations (PLS components) of the X variables that predict as much of the systematic variations in the consumer hedonic data as possible (Hough et al., 1996). Since PLS regression provides solutions to the shortcomings of internal and external preference mapping techniques, it has been used by numerous earlier works to investigate consumer preferences on various foods including olive oil and orange juice (Delgado & Guinard, 2012; di Marzo et al., 2006; Tenenhaus, Pagès, Ambroisine, & Guinot, 2005), and their findings demonstrated its efficacy in consumer studies.

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ANALYSIS OF FLAVOR ATTRIBUTES AND CHEMICAL COMPOSITION OF VIDALIA SWEET ONION (*ALLIUM CEPA* L.): SUGARS, LACHRYMATORY FACTOR (LF; PROPANETHIAL S-OXIDE) AND METHYL THIOSULFINATES[1]

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Abstract

The chemical compositions of two varieties of Vidalia onion (Nunhems 1006 and Sapelo) grown at two different fertilizer levels were studied to investigate their relationship in flavor. Chromatographic analyses (HPLC or GC-FID/FPD) of sugars, propanethial S-oxide (Lachrymatory Factor; LF) and methyl thiosulfinates were conducted to analyze the chemical composition of the 4 onion treatments. Samples were also evaluated for sweetness, lachrymatory sensation (LF-Sens) and pungency/aftertaste by trained sensory panelists. Total sugar content was not significantly different ($p>0.05$) among treatments, while sensory scores were significantly different and inversely correlated to the methyl thiosulfinates content, indicating a possible masking effect on sweetness. Panelists were able to perceive the profiled onion flavors and accurately differentiate between treatments based on the LF-Sens and pungency/aftertaste, which were significantly correlated ($p<0.0001$) to the LF and methyl thiosulfinates content. Therefore, LF and methyl thiosulfinates attributed to the majority of onion flavor perception, while sugar content has little impact.

1. Introduction

Onions (*Allium cepa* L.) are one of the most important horticultural crops worldwide and a major source of flavoring in many dishes (Griffiths, Trueman, Crowther, Thomas, & Smith, 2002). Onions have a wide range of flavors from very mild and sweet to extremely hot or pungent types. Among various onion types, sweet onions such as Vidalia® (Georgia) and Walla Walla (Washington) are popular in the United States for their characteristically sweet flavor.

The flavor profile of onions is determined by the 3 major flavor precursors of S-alk(en)yl-L-cysteine sulfoxides (ACSOs): S-methyl-, S-propyl and S-1-propenyl cysteine sulfoxides. When the onion cells are mechanically ruptured, these ACSOs are hydrolyzed by the enzyme alliinase producing pyruvic acid, ammonia and unstable sulfenic acids. These acids can rapidly react to form methyl thiosulfinates and tear-causing propanethial S-oxides, commonly referred to as the lachrymatory factor (LF) (Corzo-Martínez, Corzo, & Villamiel, 2007; Lancaster, Shaw, & Randle, 1998; McCallum et al., 2005). Methyl thiosulfinates are responsible for the characteristic flavors related to fresh onions (Block, 1985; Block, Naganathan, Putman, & Zhao, 1992; Freeman, 1976). The LF is produced from S-1-propenyl sulfenic acid through the action of a LF-synthase and is responsible for the mouth burn and heat when consuming onions (Kopsell, Randle, & Schmidt, 2002). According to Randle and Lancaster (2002), these onion flavor intensities can be attributed to genetic, environmental and post-harvest factors. Earlier studies have shown that the amount of sulfur in soil is the major environmental factor influencing the accumulation of total onion ACSOs, and the ratio of individual ACSOs; therefore, high levels of sulfur in fertilizer results in greater flavor intensity (Freeman & Mossadeghi, 1970; Randle et al., 1995). It has also been demonstrated that the concentrations of LF and thiosulfinates increase linearly with increased level of sulfur in fertilizer applied to the soil (Randle, Block, Littlejohn, Putman, & Bussard, 1994).

Sweetness is another important factor in Vidalia onion flavor. The three main sugars which comprise the majority of the soluble solids of the onions are glucose, sucrose and fructose. Although it is known that at least 65% of dry weight of onion is composed of sugars, some studies reported that the sulfur containing volatiles, presumably responsible for the pungency in onions, cause a masking effect on sweetness (Crowther et al., 2005; Green, 1996).

Considerable research has been conducted on the onion flavor, with most of them focused on the measurement of flavor precursors and/or compounds (Lanzotti, 2006). Several studies have been conducted on pyruvate concentration in onions and correlated it with onion flavor (Terry, Law, Hipwood, & Bellamy, 2005; Wall & Corgan, 1992; Yoo & Pike, 2001). However, pyruvic acid itself gives no taste, and has been used as an indirect measurement for overall onion pungency due to its formation alongside sulfur containing flavor compounds during cell damage (Randle, Kopsell, Kopsell, Snyder, & Torrance, 1998). The onion classification based on pyruvic acid content has been commonly used, but meaningful variations among different laboratories (Anthon & Barrett, 2003; Yoo, Lee, & Patil, 2011a; Yoo, Lee, & Patil, 2011b) make it skeptical to be used as a standardized methodology for this purpose (Havey et al., 2002). In addition, the pyruvic acid content does not always match with onion flavor response of pungency meaning that the pyruvic acid is not a good measure of the pungency on all onion varieties (Crowther et al., 2005). These observations indicate that pyruvic acid analysis alone does not fully capture the human perception of onion pungency. Therefore, more reliable methods to measure chemical compounds that are responsible for onion pungency should be investigated, and any potential correlation between those compounds and human flavor perception should be established. Moreover, although the onion LF and methyl thiosulfinates have been reported as the responsible compounds of pungent flavor, the specific relation of these compounds has not been systematically demonstrated with sensory analyses.

The objective of this study was to investigate the relationship between the onion chemical composition and flavor attributes by correlating the content of sugars, LF, and methyl thiosulfinates with sensory attributes (sweetness, lachrymatory sensation and pungency/aftertaste) to define its contribution to the actual flavor perception.

2. Materials and Methods

2.1. Onion Production

Six varieties of Vidalia onions (Savanna Sweet, Nunhems 1006, Sapelo, Isabella, Sweet Harvest, and Candy Kim) were grown at two different fertilizer levels (treatment) at the UGA Vidalia Onion and Vegetable Research Center in Lyons, GA, from November 2013 to April 2014. Based on the preliminary chemical analyses on the LF and methyl thiosulfinates content at harvest, two varieties and two fertilizer treatments that most represented the difference in chemical composition and flavor profile were selected. This provided four onion treatments (Table 1) consisted of Nunhems 1006 (sweet) and Sapelo (hot) varieties with two fertilizer levels (low and high) used in this study.

Table 1. Description of the four onion treatments and samples codes used in the study.

Variety	Fertilizer treatment (kg/ha)			Description	Sample code
	Level	Nitrogen	Sulfur		
Nunhems 1006 (sweet)	Low	106	27	Sweet variety / low fertilization	SLF
	High	179	137	Sweet variety / high fertilization	SHF
Sapelo (hot)	Low	106	27	Hot variety / low fertilization	HLF
	High	179	137	Hot variety / high fertilization	HHF

2.2. Sample Collection

Fourteen onions were randomly collected from a 45 kg bag of each four treatments. Each onion was analyzed for chemical composition simultaneously with individual sensory evaluation session. For every analysis, the basal plate and stem of an onion bulb was removed, and halved vertically through the core. One half of the onion had its outermost and innermost layers removed, leaving the two onion leaves equidistant from the core and skin for chemical analysis. The second half of the onion was packaged in plastic wrapping and immediately transported to the USDA-RRC sensory lab for flavor testing. Sensory analysis was conducted within 2 hours of the initial cut of each onion.

2.3. Sensory Evaluation

2.3.1. Sample Preparation and Presentation

The middle 2-3 layers from the second half of the onion bulb were used for sensory testing, similar to those sections used in the chemical analysis. Each sample was chopped into 1.3 × 1.3 cm square pieces and 4 pieces were served at room-temperature in a 44.3 mL plastic soufflé cup with lid (SOLO, Lake Forest, IL) and labeled with 3-digit random codes. The samples were served to the panel in monadic sequential presentation with the same sequence for every panelists due to time sensitivity of chemical reactions and flavor changes after cutting. A sequence of sample presentation was designed to ensure that each sample was randomly assigned to each of the four possible positions in a balanced manner across the 14 sessions. This was aimed at reducing potential bias introduced by the order effects that might influence the perception of a sample due to the previously tasted onion.

Samples were carefully handled so that panelists did not receive samples from the onion areas with previously ruptured cells. Each sample was evaluated within 6 minutes of cutting to minimize changes in the chemical compounds and flavor perception.

2.3.2. Sensory Panel Training and Panel Selection

Eleven potential panelists were selected and screened based on the following criteria: interest, availability during testing period, non-smokers, no health problems that might interfere with testing, favorableness of consuming hot and spicy foods, sensory acuity as well as ability to describe sensory characteristics of food products. The potential panelists received descriptive sensory training in the Sensory Laboratory of USDA-ARS, Russel Research Campus. The panelists were trained using a variety of samples such as white, red, yellow, and sweet onions that ranged in degree of sweetness and pungency. Training was conducted over 9 sessions, each lasting 2 hours.

Three sensory attributes of onions (sweetness, lachrymatory sensation and pungency/aftertaste) were introduced to the panelists and subsequently, they proposed the consensus definition of each attribute through discussion. In previous reports of onion sensory evaluation, pungency in onions was described as the burn produced by the LF and pyruvic acid has been used for measuring this burning sensation of onions (Lee, Yoo, Jifon, & Patil, 2009; Randle et al., 1995; Randle et al., 1998; Schwimmer & Weston, 1961). During the training, however, the panelists described the lachrymatory sensation as the burn in their tongue and mouth associated with heat, while pungency was defined as a strong flavor remaining in the back of the throat after consumption. Therefore, the term ‘pungency’ in the present study was used to describe the strong characteristic onion flavor and aftertaste, rather than the burn caused by onions. In addition, a list of food references with its taste/flavor intensity for sweetness, lachrymatory sensation and pungency/aftertaste was developed as shown in Table 2. To differentiate sensory lachrymatory sensation from the chemical lachrymatory factor compound, 'LF-Sens' was used in this paper to describe the lachrymatory sensation perceived by the sensory panelists.

During the training sessions, panelists were trained: (1) to develop a standardized tasting procedure of onion samples; (2) to get familiar with the intensity scaling on 0-15 point line scale; and (3) to determine effective palate cleansers. The tasting procedures were standardized as follows: panelists chewed an onion piece 3 times (chewing rate = 1 chew/sec) with their mouth closed to rate sweetness, then chewed 7 more times (1 chew/sec) with their mouth open to evaluate the LF-Sens. The intensity of pungency/aftertaste was rated after they expectorated the onion piece and waited for 10 sec. The tasting order of sensory attributes was based on the sequence of chemical reaction in onions, so that the sensory scores can be given when each compound was at its peak concentration limiting interference from other compounds (Fig. 1). Panelists were trained for calibration using food references and the 0-15 point line scale (0 = low; 15 = high) until a consensus within the group was achieved. During training, panelists got feedback after every session to improve their performance. The feedback was aimed at reaching agreement on the definition of three sensory attributes and the panel's ability to use the intensity scale.

Table 2. Definitions for sensory flavor attributes and food references used for sensory evaluation of Vidalia onion samples.

Attribute	Definition	Tasting method	Flavor intensity	Reference [a]	# of chews
Sweetness	Sweet and sugary taste	Chew an onion piece 3 times (1 chew/sec) with mouth closed	2.0	2% sugar solution	-
			5.0	5% sugar solution	-
			10.0	10% sugar solution	-
LF-Sens	A burning, painful and numbing sensation in the tongue and mouth	Chew the onion piece 7 more times (1 chew/sec) with mouth open	2.5	Leek (1/4" slice)	10
			4.5	Mustard sauce (1/2 tsp)	-
			7.0	Ginger paste (1/2 tsp)	-
			10.0	Wasabi sauce (1/2 tsp)	-
			15.0	Ginger root (1/4" slice)	7
Pungency / Aftertaste	Strong and irritating smells and flavors that remain in the aftertaste	Expectorate the sample and wait for 10 sec before evaluation	1.0	Cucumber (1/2" slice)	7
			4.0	Cabbage	9
			6.5	Broccoli	9
			9.0	Radish (1/2 bulb)	11
			15.0	Listerine (15mL)	-

[a] Fresh produces were purchased from local supermarket and stored in 4 ℃ refrigerator and used in 3 days of purchase. Mustard sauce (Dijon mustard, Kroger Co., Cincinnati, OH), Ginger paste (Botanical Food Company Inc., Folsom, CA), Wasabi sauce (S&B Foods Inc., Chuo-ku, Tokyo, Japan), Listerine (McNEIL-PPC, Inc., Skillman, NJ).

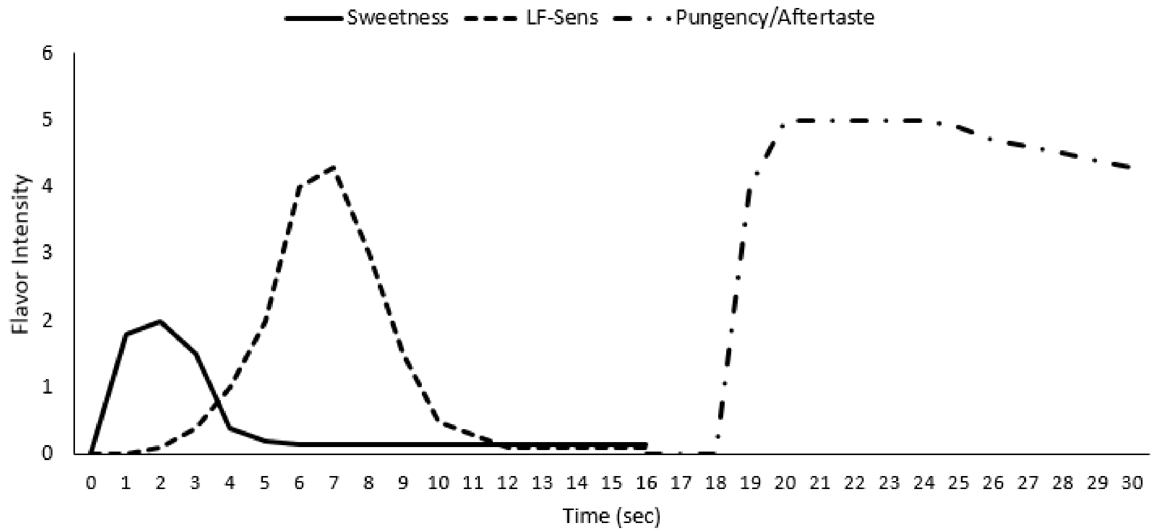

Fig. 1. The principle of flavor intensity evaluation according to the release of onion flavor compounds over time. The highest peak of each sensory attribute within certain time period was reported for the flavor intensity.

After the 8th training session, a performance test was conducted and only the panelists who had good repeatability and reproducibility were selected for the main study (Rossi, 2001). Thus, 9 out of 11 potential panelists were chosen to participate in the main flavor evaluation.

2.3.3. Flavor Evaluation

All sensory data were obtained in accordance with the procedures of the University of Georgia Institutional Review Board (IRB) for sensory evaluation, including informed consent and risk/discomfort assessment. Sensory analysis was performed by 8 or 9 panelists (3 male and 6 female) over 14 sessions. Four samples comprised of each of four treatments were evaluated in individual laboratory booths compliant with ASTM international standards (Eggert & Zook, 1986). A hybrid descriptive analysis was used to assess the three sensory attributes of onions (sweetness, LF-Sens and pungency/aftertaste): three known sensory attributes were evaluated by trained panel based on the reference standards using 0-15 point scale (Spectrum®), while each of the attributes was defined by the panelists (QDA®; Quantitative Descriptive Analysis). A tray of references was provided for calibration during evaluation. Considering the relatively strong lingering aftertaste of onions, parsley, lemon sorbet, baking soda as well as unsalted crackers and water were provided for palate cleansing. Panelists were required to use two or more palate cleansers to rinse their mouth between every sample until residual flavor was removed. The effectiveness of provided palate cleansers were tested by the panelists during training sessions. There was a 10 minute break before proceeding to the next sample. Data were collected with Compusense® five (Version 5.4, Compusense Inc., Guelph, Canada).

2.4. Instrumental Analysis

2.4.1. Sample Extraction for Chemical Analysis

The two onion layers selected for analysis were diced and placed onto the press plate of a pneumatic press similar to the one used by Randle and Bussard (1993b). Two stainless steel screens (4 mm and 1 mm mesh size) were placed under the sample, which collected the solid material from the onions, while allowing the juice to flow through the screens to be collected in a beaker below the press.

2.4.2. Chemical Analysis of the Lachrymatory Factor (LF)

Onion LF was analyzed following the procedure by Schmidt et al. (1996), with slight modifications. Thirty seconds after pressing, 4 mL of onion juice was pipetted from the juice collection beaker into a centrifuge tube containing 4 mL of 0.01% m-Xylene as internal standard (>99.0%, Acros Organics, Geel, Belgium. 180862500) in HPLC grade methylene chloride (J.T. Baker, Center Valley, PA, Product #: 9315-33). The solution was vortexed and then centrifuged for 10 minutes using an HN-SII model centrifuge from Damon/ IEC Division (Needham Heights, MA). The organic layer was then extracted into 2 mL target vials and analyzed by gas chromatography with a flame ionization detector (GC-FID), within 30 minutes of extraction. GC analysis was conducted using a 7890A gas chromatograph (Agilent Technologies, Santa Clara, CA) with a DB-1, 30 m × 0.53 mm i.d. capillary column (J&W Scientific, Agilent Technologies, Santa Clara, CA, Part #: 125-1032), and 99.999% He carrier gas, with a flow rate of 8.5 mL/min. Injector temperature was set to 60 ℃, and oven was programed to hold at 60 ℃ for 1 minute, followed by a temperature increase rate of 5 ℃ / minute to 200 ℃. The detector was maintained at 250 ℃. Quantification was conducted by comparing LF peak areas to the area of the internal standard (m-Xylene). The LF peak identity was confirmed by Hewlett-Packard 5890 series II GC coupled to a 5970 mass spectrometer. The chromatographic conditions used were the same as those for the GC-FID procedure, however, the column was a DB-5MS (30 m × 0.25 mm; J&W Scientific, Agilent Technologies, Santa Clara, CA, Part #: 122-5532). The resulting chromatogram and fragmentation pattern for the LF peak were similar to those observed in previous reports (Block, Putman, & Zhao, 1992; Schmidt et al., 1996).

2.4.3. Chemical Analysis of Methyl Thiosulfinates

Methyl thiosulfinates analyses were conducted by extraction of 4 mL of onion juice from the collection beaker into a centrifuge tube containing 4 mL of 0.00025% thioanisole as internal standard (>99%, Fluka Analytics, St. Louis, MO, Cat #: 88470) in

HPLC grade methylene chloride, 2 minutes after onion expression using the pneumatic press. The solution was vortexed, centrifuged for 10 minutes using an HN-SII model centrifuge from Damon/ IEC Division (Needham Heights, MA). The lower organic layer was then extracted into 2 mL target vials for analysis by GC with flame photometric detection (GC-FPD) and a sulfur specific filter (Agilent Technologies, Part #: 1000-1437). A 7890A gas chromatograph (Agilent Technologies, Santa Clara, CA) with a DB-1, 5 m × 0.53 mm i.d. column (J&W Scientific, Agilent Technologies, Part #: 125-100B), and 99.999% He carrier gas, with flow maintained at 8.8 mL/ min. The chromatographic conditions used were the same as those used for the LF analysis method. Quantification was conducted by summing the peak areas associated with the methyl thiosulfinate compounds and then comparing the resulting response to that of the internal standard (thioanisole). Concentrations of individual thiosulfinate were not sufficient enough to be detected using the GC-MS procedure described above; therefore compound identification was done by matching retention times and elution orders of the peaks to those observed in previous reports (Block, Naganathan, Putman, & Zhao, 1993; Schmidt et al., 1996). Use of this procedure in tandem with a sulfur specific detection system further assures accurate identification. Individual compound identification was not considered necessary due to the fact that the signal for the entire class of compounds was summed together in the chromatograms, and those compounds were eluted together within a small retention time range using the chromatographic conditions (Block et al., 1993; Schmidt et al., 1996).

2.4.4. Chemical Analysis of Onion Sugar Content

Sucrose, glucose, and fructose were quantified in onions using a 1260 Infinity HPLC pump equipped with a refractive index detector (Agilent Technologies, Santa Clara, CA). 1 mL of onion juice from the collection beaker was diluted into 4 mL of HPLC grade deionized H_2O. Diluted samples were filtered through a 0.45 µm nylon syringe filter (polypropylene membrane, 25 mm; Whatman plc, Maidstone, United Kingdom, Article #: 28420522), and placed into target vials for HPLC analysis. 20 µL of diluted onion juice solution was injected into the HPLC equipped with a Carbohydrate 700CH column (300 mm × 65 mm i.d.) 10-µm particle size (Alltech, Deerfield, IL), along with an Alltech adsorbosphere XL SCX guard column (7.5 mm × 4.6 mm i.d.) (Hamilton, Pike, & Yoo, 1997; Lee et al., 2009). Column temperature was maintained at 80 °C, with an isocratic flow of 0.5 mL/min, using HPLC grade water as the mobile phase. Quantification was done by comparing the peak areas of the unknown samples to external standard curves for sucrose, glucose, and fructose (≥99.0%, Sigma Aldrich, Taufkirchen, Germany). A certified carbohydrates mix standard (a-D-glucose, fructose, lactose, maltose and sucrose; 1000 µg/mL in Acetonitrile/water (10:90)) from NSI Lab Solutions (FB-CARB, Raleigh, NC, Lot #: 042513) was used as reference, and injected along with each sequence, with an average recovery of 101.16 ± 1.7%.

2.5. Data Analysis

Two-way (panelist and onion treatment) analysis of variance (ANOVA) with the post-hoc Tukey's HSD test was conducted on sensory data using the PROC GLM procedure of SAS® (Version 9.3, SAS Institute Inc., Cary, NC) to identify the sources of variation and significant difference among the samples at a 5% significance level

($p<0.05$). One-way ANOVA followed by Tukey's HSD test was performed on instrumental data using the same procedure of SAS®. Means of individual sensory attributes and instrumental measurements for each onion sample were computed using the PROC MEAN procedure. The relationship between sensory scores on flavor attributes and contents of chemical compounds was investigated by Pearson's correlation analysis using PROC CORR procedure, and analysis of covariance (ANCOVA) was performed with Akaike Information Criterion (AIC) for model selection.

3. Results and Discussion

3.1. Chemical Composition

The content of total sugar, LF and methyl thiosulfinates were measured in each of the four treatments over 14 replicated sessions. Results of the chemical analyses are summarized in Table 3. The total sugar content of the onions ranged from 6.09 g /100g fresh weight (fw) in the HLF treatment to 6.54 in the SHF treatment. However, there were no statistical differences in total sugar content among 4 treatments ($p>0.05$). In other words, the content of sugars measured in HPLC analyses was consistent in all samples, regardless of different onion varieties and fertilizer levels.

The onion LF concentration showed significant differences ($p<0.05$) among treatments. The hot onion variety with high fertilization (HHF) had the highest LF concentration (6.45 µmol/g), while the sweet onion variety with low fertilization (SLF) had the lowest concentration (1.07 µmol/g). Overall data of this measurement indicate that the LF concentration was higher in hot variety (HLF and HHF) than sweet variety (SLF and SHF), and increased with added sulfur and nitrogen levels in fertilizer.

Results of the methyl thiosulfinates concentration were similar to those of the LF. The HHF treatment was significantly higher ($p<0.05$) in methyl thiosulfinates (1.50 nmol/g) when compared with all treatments, while the SLF treatment was the lowest (0.36 nmol/g). Therefore, the onions that were cultivated with fertilizer containing high levels of sulfur and nitrogen, had higher concentration of methyl thiosulfinates along with the increased LF concentration. Similar results in which onion LF and thiosulfinates responded linearly with sulfur fertilizer applications have been reported previously (Randle et al., 1994).

3.2. Sensory Analysis

The mean scores of sensory analyses are shown in Table 3. Sweetness was not different ($p>0.05$) within the same variety (sweet or hot) and within the same fertilizer level (low or high). However, significant difference ($p<0.05$) in sweetness between the SHF and HLF treatments was found (3.33 and 3.06, respectively).

Interestingly, this was not observed in the results of the chemical analyses of sugar content. These results indicated that panelists were able to detect the differences in sweetness with sensitive perception. Previous reports suggested that sulfur compounds within the onion could diminish the perception of sweetness (Randle & Bussard, 1993a, 1993b). Therefore, these differences in sweetness can be explained by a potential masking effect of LF and/or methyl thiosulfinates in the HLF treatment, which resulted in higher concentration of these compounds.

The LF-Sens scores showed significant differences among all four treatments ($p<0.05$). The HHF had the highest score (4.25), while the SLF was given the lowest score (2.46). It was observed that hot onion variety (HLF and HHF) were stronger in burning sensations than sweet onion variety (SLF and SHF) as shown in Table 3.

Table 3. Mean ± SD [a] of each Vidalia onion treatment for instrumental parameters and sensory scores (n=14).

Treat -ment[b]	Chemical compounds			Sensory scores		
	Total sugars (g/100g fw)	LF (µmol/g)	Methyl thiosulfinates (nmol/g)	Sweetness (0-15)	LF-Sens (0-15)	Pungency/Aftertaste (0-15)
SLF	6.39 ± 0.86 a	1.07 ± 0.48 c	0.36 ± 0.12 c	3.31 ± 0.99 ab	2.46 ± 0.97 d	3.19 ± 1.38 d
SHF	6.54 ± 0.49 a	2.68 ± 1.30 bc	0.51 ± 0.15 bc	3.33 ± 0.98 a	2.96 ± 1.22 c	3.95 ± 1.52 c
HLF	6.09 ± 0.61 a	4.31 ± 1.76 b	0.97 ± 0.43 b	3.06 ± 0.98 b	3.76 ± 1.42 b	4.89 ± 1.58 b
HHF	6.39 ± 0.62 a	6.45 ± 3.38 a	1.50 ± 0.80 a	3.21 ± 1.03 ab	4.25 ± 1.49 a	5.47 ± 1.74 a

[a] Mean values with the different letter within a column are significantly different ($p<0.05$).
[b] SLF = sweet variety with low fertilization; SHF = sweet variety with high fertilization; HLF = hot variety with low fertilization; HHF = hot variety with high fertilization.

Onions treated with high fertilization such as SHF and HHF, were scored higher than those with low fertilization (SLF and HLF). These observations demonstrate that the panelists were able to consistently differentiate the intensity of burning sensation which is associated with the LF concentration based on the different onion variety and fertilizer level.

Sensory evaluation scores for the pungency/aftertaste were significantly different ($p<0.05$) among all treatments in the same manner with the LF-Sens. Again, the HHF was scored with the highest pungency value of 5.47, while the SLF was scored the lowest (3.19). Less pungency/aftertaste was observed in sweet variety as well as in onions with low fertilization. Moreover, as the intensity of perceived LF-Sens increased across onion treatments, so did the intensity of pungent flavor and aftertaste.

Overall, the flavor intensity of LF-Sens and pungency/aftertaste were both distinctive among all treatments while sweetness varied slightly, thus having less effect on the flavor perception. Therefore, it is evident that Vidalia onion flavor was driven more by LF-Sens and pungency/aftertaste which were associated with the concentration of LF and methyl thiosulfinates, respectively.

Furthermore, the sensory analyses suggest that panelists were able to evaluate four onion samples with no carryover effect, thus generating reliable results. Onions can impart relatively strong stimulus for human sensory perception, affecting the ability to discriminate and causing palate fatigue (Bedford, 1984). To assure that panelists were not overwhelmed by the onion flavor or had sensorial fatigue, the number of samples tested in one session was determined through the panel performance test. Bedford (1984) also reported that tasting more than 4 onions at one time was too many for the panel to evaluate these flavor attributes appropriately. In addition, diverse palate cleansers tested during performance test (data not shown) showed to be effective than using water and crackers solely, and may have helped to prevent carry-over effect and/or fatigue.

3.3. Correlation between Chemical Compounds and Sensory Perception

The relationship between the instrumental results and the sensory perceptions of the four onion samples is shown in Fig. 2. Table 4 shows the correlation coefficients of the instrumental and the sensory measurements.

Table 4. Correlation coefficients (r) between chemical compounds and sensory attributes in Vidalia sweet onions [a].

	Total sugar	LF	Methyl thiosulfinates	Sweetness	LF-Sens	Pungency / Aftertaste
Total sugar	1	- 0.1207	- 0.1884	0.1525	- 0.1228	- 0.1931
LF		1	**0.8068**	- 0.1658	**0.7810**	**0.5970**
Methyl thiosulfinates			1	**- 0.3717**	**0.7644**	**0.6189**
Sweetness				1	- 0.1652	**- 0.2619**
LF-Sens					1	**0.8160**
Pungency / Aftertaste						1

[a] Bolded values are significant at $p<0.05$.

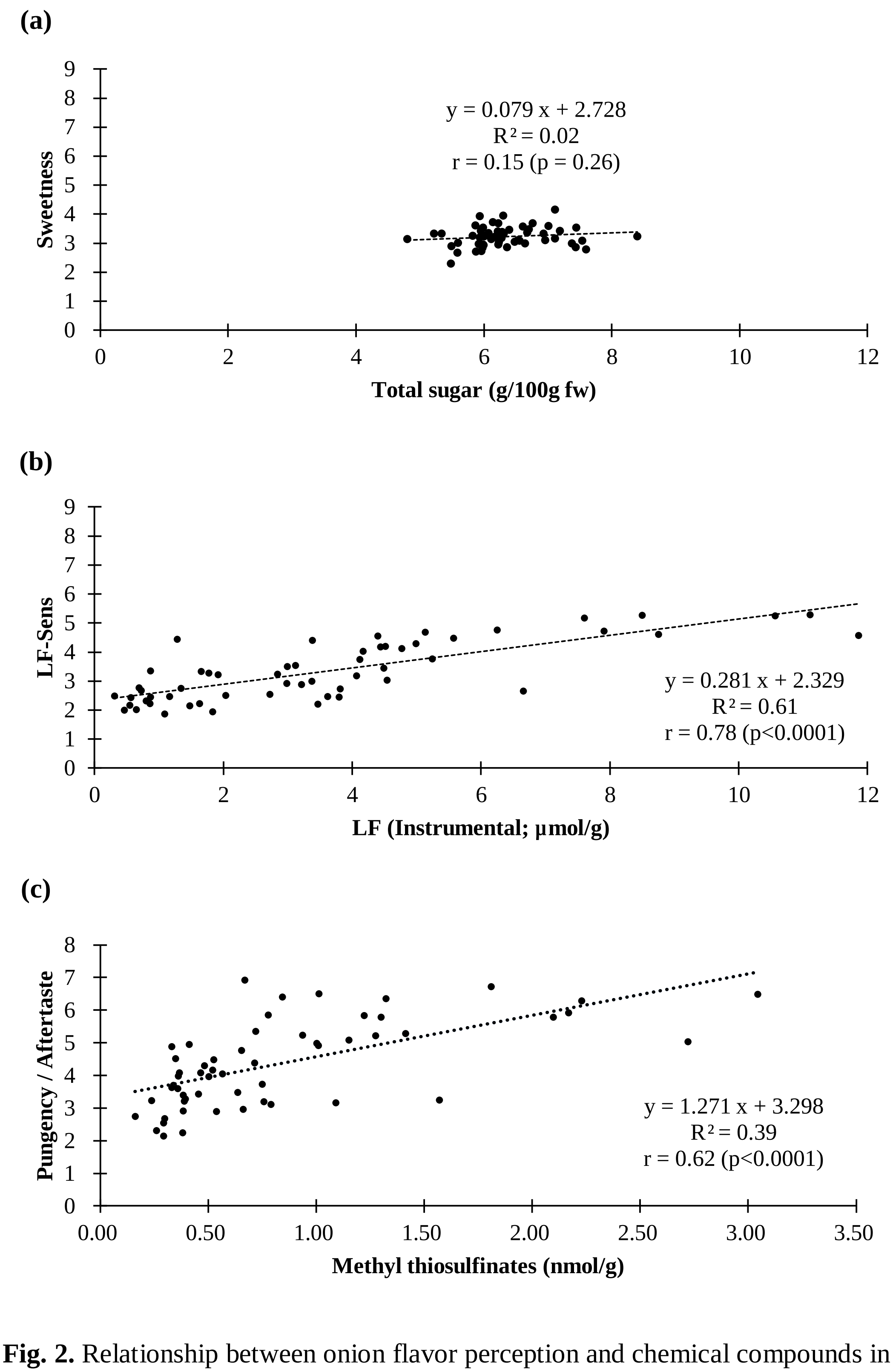

Fig. 2. Relationship between onion flavor perception and chemical compounds in all 14 sessions: (a) Correlation between total sugar content and sensory sweetness; (b) Correlation between the LF content and LF-Sens; (c) Correlation between methyl thiosulfinates content and sensory pungency/aftertaste.

Total sugars (glucose, sucrose, and fructose) measured by HPLC had no correlation to sensory sweetness ($p = 0.262$). Presumably, human perception of sweetness in onions is influenced by factors other than the content of sugars (Crowther et al., 2005). Indeed, the concentration of methyl thiosulfinates showed a negative correlation to sweetness ($r = -0.372$; $p = 0.005$), while there was a weak negative correlation between sweetness and pungency/aftertaste ($r = -0.262$; $p = 0.05$). Therefore, analysis of covariance (ANCOVA) on the sensory scores was performed with treatment as a categorical variable and chemical compounds (total sugars, LF and methyl thiosulfinates) as covariates to investigate the influence of other chemical and/or sensory components on sweetness, besides the total sugars. Best fit model through the stepwise model selection process by Akaike Information Criterion (AIC) was described as following:

$$\text{Sweetness} = \text{Treatment} + \text{Total sugars} + \text{Methyl thiosulfinates} + (\text{Treatment} \times \text{Methyl thiosulfinates}) + (\text{Total sugars} \times \text{Methyl thiosulfinates})$$

Accordingly, it was strongly suggested that methyl thiosulfinates concentration has significant effect on the perceived intensity of sweetness in onions. However, the model explained only about 36% of the variability of sweetness according to the R-squared. Followed by the Fisher's LSD test with a confident interval of 95%, significant differences between the treatments corresponded to the ANOVA results in Table 3 (data not shown). This indicated that a large part of sweetness perception was not clarified, yet the information loss was minimized when the effect by methyl thiosulfinates was included. In order to elucidate the relationship, some effects (e. g. other chemical compounds) that may have influenced the onion sweetness should be analyzed.

On the other hand, significant correlations were found between chemical LF and LF-Sens ($r = 0.781$; $p<0.0001$), and also between methyl thiosulfinates and pungency/aftertaste ($r = 0.619$; $p<0.0001$). Not only were these correlations statistically significant, the coefficient values were substantially high considering the difficulty of identifying onion flavors and controlling variability associated with human sensory perception (Terry et al., 2005). This work shows evidence that the concentrations of LF and methyl thiosulfinates in onions are tightly linked to the burning sensation and aftertaste, and that panelists were able to accurately perceive differences in onions as the concentration of these compounds fluctuate. Given these results, we can conclude that the LF and methyl thiosulfinates concentration play important roles in the sensory characterization of onion flavor. Contrarily, this was not the case with the sugar content in the samples because it did not have correlation with sweetness observed by the trained panelists.

In addition, it was found that all of the LF content, LF-Sens, methyl thiosulfinates content and pungency/aftertaste had strong positive correlations to each other ($p<0.0001$). This indicates that the onion which gives a strong burning and numbing sensation will also have a strong aftertaste with unpleasant smells and flavors to human perception. This study shows that if onion flavor is evaluated based on chemical composition, either the measurement of the LF or methyl thiosulfinates content may provide an accurate estimation of human perception of both burning sensation and pungency/aftertaste. Nevertheless, while the LF was a single compound, methyl thiosulfinates was measured as a group of compounds, and that may have a making effect on sweetness. In order to fully understand the effect of methyl thiosulfinates in onion flavor, future studies should focus on how the flavor notes of the individual methyl thiosulfinate compounds influence onion flavor perception. Accordingly, the analysis and interpretation of the LF measurement could be much easier than that of the methyl thiosulfinates, providing the most efficient and accurate results with a known sensory perception.

According to the industry and previous studies (Crowther et al., 2005; Green, 1996; Wall & Corgan, 1992; Yoo & Pike, 2001), the analysis of pyruvic acid concentration is most commonly used for assessment of flavor quality in onions based on pungency characteristic. However it has been reported that this method has some limitations: (1) Pyruvic acid is not a direct flavor compound of pungency and is flavorless by itself; (2) Currently, there is no standardized methodology that limits the variation between results among different laboratories (Havey et al., 2002). Therefore, it is necessary to assess the onion pungency characteristic with more reliable and repeatable method, for example, by measuring the compound that directly causes pungent flavor, such as the LF as demonstrated in this work.

The intention of this work was to demonstrate the relationship of the chemical compounds in Vidalia onion and their most relevant flavor characteristics (sweetness, LF-Sens and pungency/aftertaste) to define the contribution of individual chemical compound to the human perception of Vidalia onion flavor. Considering the significant correlations, the sensory analytical protocol used in this work was convincing for the identification of these flavor characteristics in terms of; (1) systematic sensory analyses for the identification and definition of the flavor descriptors (attributes) of Vidalia onion; (2) elucidating the order of appearance of flavor attributes (sweetness, lachrymatory sensation and pungent/aftertaste) during tasting experience for a proper sensory evaluation; and (3) standardization of a tasting methodology for onion sensory evaluation.

4. Conclusion

The study revealed that the hot onion variety (Sapelo) had the higher content of LF and methyl thiosulfinates than the sweet variety (Nunhems 1006), and those two compounds increased with added fertilizer. Total sugar content was not influenced by the onion variety and levels of fertilizer. Therefore, the onions that were cultivated at high levels of sulfur and nitrogen had higher content of methyl thiosulfinates along with the increased LF content. The content of LF and methyl thiosulfinates in Vidalia onions were highly correlated to the burning sensation and pungency/aftertaste perceived by sensory panels. However, there was no significant correlation between sugar content and sweetness. Given these results, we can conclude that the LF and methyl thiosulfinates content play important roles in the sensory characterization of Vidalia onion flavor. Based on the accuracy and convenience of the methodology used in this study, the measurement of LF content can be used for industry application to anticipate flavor characteristics of lachrymatory and pungency intensity in Vidalia onions.

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RELATIONSHIP BETWEEN CONSUMER PREFERENCE AND FLAVOR COMPOUNDS OF VIDALIA ONIONS [2]

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Abstract

A consumer study was conducted to evaluate flavor perception and preference in two varieties of Vidalia onions (Nunhems 1006 and Sapelo) grown at three levels of fertilizer (low, medium, and high) to better understand consumer acceptability in relation to the chemical composition of Vidalia onions. The content of sugars, lachrymatory factor (LF; propanethial S-oxide) and methyl thiosulfinates were measured using HPLC, gas chromatography-flame ionization detector (GC-FID) and GC-flame photometric detector (GC-FPD). The LF and methyl thiosulfinates in Vidalia onion showed higher concentrations as the fertilizer level increased, regardless of different onion varieties. The study showed that the majority of consumers preferred onions with less intensity of sharp/pungent/burning sensation (SPB) which corresponded to low LF and methyl thiosulfinates concentration. There was no specific tendency in consumer preference found in relation to the sugar content. Canonical correlation analysis (CCA) revealed that the concentration of LF and SPB intensity perceived by consumers are the key attributes influencing the overall onion flavor, and ultimately consumer liking. LF content of 1.806 μmol/g was recommended for desirable Vidalia onion flavor. Partial least squares (PLS) regression identified significant negative relationships between consumer liking and high concentration of the LF and methyl thiosulfinates. Agglomerative hierarchical clustering (AHC) analyses of the consumer ratings revealed segmentation of preference patterns.

1. Introduction

Over the past years, the consumption of onions (*Allium cepa* L.) has considerably increased in the United States with the heavy promotion of associating its flavor with health benefits (Griffiths, Trueman, Crowther, Thomas, & Smith, 2002). However, consumers have shown reluctant behaviors toward eating onions due to some of the unpalatable flavors such as strong pungency and lingering aftertaste. Recently, the interest in consumption of sweet or mild onions has been growing in the U. S., thus the breeding and producing these types of onions have become popular (Boyhan & Torrance, 2002; Lee, Yoo, Jifon, & Patil, 2009; Vidalia Onion Committee, 2015). Among various onion types, sweet onions such as Vidalia® (Georgia) and Walla Walla (Washington) onions are well-known for their distinctively sweet flavor. In particular, Vidalia onions are very popular among consumers and commercially important in Georgia (Sellappan & Akoh, 2002).

The flavor profile of onions is determined by chain reactions which involve complicated and unstable compound productions (Yoo, Lee, & Patil, 2012). In *Allium* species, three major flavor precursors of S-alk(en)yl-L-cysteine sulfoxides (ACSOs) are found: S-methyl-, S-propyl and S-1-propenyl cysteine sulfoxides. These ACSOs are hydrolyzed by the enzyme alliinase once the onion cells are mechanically ruptured. Through this reaction, pyruvic acid, ammonia and numerous volatile sulfur compounds including sulfenic acids are produced. Because these acids are chemically unstable, they rapidly react to form methyl thiosulfinates and tear-causing propanethial S-oxides, commonly referred to as the lachrymatory factor (LF) (Corzo-Martínez, Corzo, & Villamiel, 2007; Lancaster, Shaw, & Randle, 1998; McCallum et al., 2005). Methyl thiosulfinates are mainly responsible for the characteristic flavors related to fresh onions (Block, 1985; Block, Naganathan, Putman, & Zhao, 1992; Freeman & Whenham, 1976). The LF is produced from S-1-propenyl sulfenic acid by the LF-synthase and is responsible for the mouth burn and pungency when consuming onions (Kopsell, Randle, & Schmidt, 2002).

The intensity of onion flavor is attributed to genetic, environmental and post-harvest factors (Hamilton, Pike, & Yoo, 1997; Randle & Lancaster, 2002; Yoo, Pike, Crosby, Jones, & Leskovar, 2006). It has been reported that the amount of sulfur in soil is the major environmental factor resulting in the accumulation of total onion ACSOs, and the ratio of individual ACSOs; therefore, high levels of sulfur in fertilizer generates greater pungency (Coolong, Kopsell, Kopsell, & Randle, 2005; Freeman & Mossadeghi, 1970; Randle et al., 1995). Additionally, it has been demonstrated that the increased sulfur in fertilizer led to linear increase of the LF and thiosulfinate concentrations, thus giving more pungency in onions (Randle, Block, Littlejohn, Putman, & Busard, 1994).

Vidalia onions are marketed as characteristically sweet onions due to Georgia's mild climate and sandy soil which allows sulfur to leach out of the root zone. Under the regulation of the USDA, as well as a state law, Vidalia onions are defined as short-day yellow granex varieties that are grown in the 13 designated counties in the state of Georgia (Vidalia Onions Grown in Georgia, 2015). It is noteworthy that the flavor in different varieties of Vidalia onion can be substantially influenced by the levels of fertilizer used in the soil (Randle, 1992).

Studies of onion flavor have been conducted in a way of correlating pyruvic acid concentration with pungency (Schwimmer & Weston, 1961; Terry, Law, Hipwood, & Bellamy, 2005; Yoo & Pike, 2001). In other studies, the level of pyruvic acid was shown to be positively related with the strength of pungency perceived by subjects of trained sensory panels (Crowther et al., 2005; Wall & Corgan, 1992). According to Yoo et al.

(2012), an onion bulb containing 3.5 μmol/mL pyruvic acid or lower was generally considered mild, while it was considered pungent with over 5 μmol/mL. On the other hand, the South Texas Onion Committee have suggested that sweet onions should have less than 4.5 μmol/mL of pyruvic acid.

In spite of all efforts, there are still several limitations in the research of sweet onion flavor. Firstly, the recommended pyruvic acid content might not be appropriate for Vidalia onions because it was specified for different types of onions. Secondly, meaningful variations in the result of pyruvic acid measurement have been reported among different laboratories, therefore making it skeptical to be used as a standardized method (Havey et al., 2002). Thirdly, pyruvic acid has no taste by itself, and has been used as an indirect measurement instead of the LF which is the major compound responsible for overall onion pungency (McCallum et al., 2005). This information indicates that pyruvic acid analysis alone cannot fully explain human perception of onion pungency. Lastly and most importantly, there have been scarce attempts to assess the desirable Vidalia onion flavor from both the instrumental and consumer sensory point of view. With respect to the sensory quality of Vidalia onions, therefore, it is important to investigate flavor profiles and control the fertilizer level considering consequential changes in the chemical composition. Nevertheless, current regulation is only focused on typological and regional terms; it is not related to the flavor characteristics such as appropriate pungency level or limitation on fertilizer level during cultivation, which could impact the overall flavor that consumers will experience.

Human perception of food comes from intricate sensory system and interpretation processes. In many cases, there is a lack of information in instrumental analyses in that they rarely capture the important perceptual process, which is described as sensory experience by human brain and following response (Lawless & Heymann, 2010). Therefore, the instrumental measurements of foods have been frequently related to the consumer hedonic results to seek the critical attributes of foods or food products from consumers' perspective (van Kleef, van Trijp, & Luning, 2006).

Partial least squares (PLS) regression has been proved to be a useful technique to relate consumer sensory structure to product/sample structure and vice versa (Martens, Bredie, & Martens, 2000). The base of PLS regression is similar to that of external preference mapping in that it aims at understanding the relationship between instrumental (X-matrix) and sensory (Y-matrix) datasets (Martens & Martens, 2001; Mitchell, Brunton, & Wilkinson, 2011). However, differences come from the way it treats data; in PLS regression, the instrumental and sensory data are used simultaneously to locate products on the map by extracting PLS components (latent variables) that represent maximized covariance between linear functions of X (instrumental measures) and Y (hedonic data) (Næs, Brockhoff, & Tomic, 2010). Indeed, it has been used in numerous works to investigate consumer preferences on various foods including olive oil and orange juice (Delgado & Guinard, 2012; di Marzo et al., 2006; Tenenhaus, Pagès, Ambroisine, & Guinot, 2005), demonstrating its efficacy in consumer studies.

To the best of our knowledge, there is no published work aimed at understanding Vidalia onion flavor through chemical composition to consumer acceptability with well-designed sensory studies. In this context, the goals of the present study were: (1) to investigate the impact of different levels of fertilizer treatment on Vidalia onion composition; (2) to establish desirable range of chemical compounds' concentration in

Vidalia onion based on consumer preferences; and (3) to gain in-depth insight into Vidalia onion consumers by relating chemical composition with consumer sensory perception and preference. Ultimately, the current study will provide useful reference to be used for quality assurance of Vidalia sweet onions.

2. Materials and Methods

2.1. Onion Samples

Six onion samples conformed by two varieties (Nunhems 1006 – sweet; Sapelo – hot) grown at three levels of fertilization (low, medium, and high) were used (Table 1). Onions were planted in December, 2014 at the UGA Vidalia Onion and Vegetable Research Center in Lyons, GA, and harvested in April, 2015.

Table 1. Description of the six onion treatments and sample codes used in the study.

Variety	Fertilizer treatment (kg/ha)			Description	Sample code
	Level	Nitrogen	Sulfur		
Nunhems 1006 (sweet)	Low	37.5	0	Sweet variety / low fertilization	SL
	Medium	134.5	59.4	Sweet variety / medium fertilization	SM
	High	190.0	118.8	Sweet variety / high fertilization	SH
Sapelo (hot)	Low	37.5	0	Hot variety / low fertilization	HL
	Medium	134.5	59.4	Hot variety /medium fertilization	HM
	High	190.0	118.8	Hot variety / high fertilization	HH

On each test day, onion samples were halved vertically through the core after removing the basal plate and stem. One half of the bulbs had its outermost and innermost layers removed, leaving the two onion leaves equidistant from the core and skin for chemical analysis. The second half of the bulbs was sealed in plastic wrap, placed in Ziploc bags, and immediately packed in Styrofoam cooler with an ice pack for transportation to the Sensory Laboratory at the University of Georgia, Griffin. All samples were evaluated by consumers within 3 hours of initial cut.

2.2. Participants

One hundred and forty-two consumers, ages ranging from 18 to 65 years, were recruited from the city of Griffin, Georgia (40 male, 102 female). Participants reported no food allergies and consuming onions and/or onion products regularly at least once a month. Consumer tests were conducted at the sensory laboratory at the University of Georgia. All evaluations were completed in sensory booths and self-administered using paper ballots. This study was reviewed and approved by the University of Georgia Institutional Review Board (IRB).

2.3. Consumer Test

2.3.1. Sample Preparation

The middle 2-3 layers of the onion bulbs were used for each test. At the time of evaluation, the onion layers were cut into 1.3 cm × 1.3 cm squares and served in a covered plastic soufflé cup (Fabri-Kal, Kalamazoo, MI) labeled with 3-digit random codes. The serving sizes were 2 pieces. Onion samples were prepared and presented within 5 min of cutting to minimize variation due to flavor changes by chemical reactions occurring after the onion cells were ruptured.

2.3.2. Consumer Test Design

The consumer test consisted of tasting six different samples in two separate sessions (3 samples/session) which were held over two consecutive days. The number of samples was determined based on the number of onions that consumers could reasonably be expected to evaluate over two sessions with no carryover effect or sensorial fatigue (Bedford 1984). The appropriateness of the number of samples was also confirmed in the previous sensory study with trained panelists (not published). A total of 7 sessions were conducted over 14 days involving 18-23 consumers each day, in which the 142 participants were distributed.

In each session, the consumers were divided into two groups (A and B) and asked to evaluate 3 uncooked Vidalia onion samples. On the first day of a session, group A evaluated 3 out of 6 samples while group B evaluated the other 3 samples (e.g. Group A – HL, SM, SH; Group B – HH, SL, HM). On the second day, each group was presented 3 samples that they did not evaluate on the first day (e.g. Group A – SL, HM, HH; Group B – SH, HL, SM). The serving sequence of each session was randomized between test days in order to balance the samples by position, thus reducing potential order effects. However, the samples were presented in the monadic sequential scheme and in the identical sequence for all participants within the same session and group. By doing this, possible changes in flavor caused by the time sensitivity of enzymatic reactions in onion cells were prevented.

Water and unsalted crackers (Saltines, The Kroger Co., Cincinnati, OH) were supplied and the consumers were instructed to rinse their mouth between tastings. There was a 10-min break before proceeding to the next sample.

2.3.3. Questionnaire

During the tasting evaluation, the consumers were asked to rate the samples for overall liking (OL) on a 9-point hedonic scale (Peryam & Pilgrim, 1957) where 1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely. The intensity of sharp/pungent/burning sensation (SPB) was scored on a 9-point scale where 1 = low, 5 = moderate, and 9 = high. In addition, willingness to buy scores (WB) were indicated on a 9-point Likert scale where 1 = extremely unlikely, 5 = moderate, and 9 = extremely likely.

After completion of the evaluation, consumers were asked to fill out a demographic questionnaire which included following details: age, gender, frequency of onion consumption, onion type(s) they consume most frequently, way of eating (cooked or raw/uncooked), aspects they care about when purchasing onions, familiarity to Vidalia onions, and quality satisfaction on Vidalia onions sold at local supermarkets.

2.4. Chemical Composition Analysis

2.4.1. Sample Extraction

The two onion layers from the half of each onion bulb were diced and placed onto the press plate of a pneumatic press similar to the one used by Randle & Bussard (1993). Solid material from the onions were collected by two stainless steel screens (4 mm and 1 mm mesh size) placed under the sample. Onion juice was flowed through the screens and collected in a beaker below the press.

2.4.2. Total Sugars

Sucrose, glucose, and fructose were quantified in onions by HPLC using a 1260 Infinity pump equipped with a refractive index detector (Agilent Technologies, Santa

Clara, CA). To prepare the samples, 1 mL of onion juice from the collection beaker was diluted into 4 mL of HPLC grade deionized H_2O. Diluted samples were filtered through a 0.45 µm nylon syringe filter (polypropylene membrane, 25 mm; Whatman plc, Maidstone, United Kingdom, Cat #: 6878-2504), and placed into target vials for HPLC analysis. A 20 µL aliquot of diluted onion juice solution was injected into the HPLC equipped with a Carbohydrate 700CH column (300 mm × 65 mm i.d., 10-µm particle size; Alltech, Deerfield, IL), along with an Alltech adsorbosphere XL SCX guard column (7.5 mm × 4.6 mm i.d.; Hamilton et al., 1997; Lee et al., 2009). Column temperature was maintained at 80 ℃, with an isocratic flow of 0.5 mL/min, using HPLC grade water as the mobile phase. Quantification was done by comparing the peak areas of the unknown samples to external standard curves for sucrose, glucose, and fructose (≥99.0%, Sigma Aldrich, Taufkirchen, Germany). A certified carbohydrates standard mix, consisting of 1000 µg/mL of a-D-glucose, fructose, lactose, maltose and sucrose, in Acetonitrile/water of 10:90 (v/v) from NSI Lab Solutions (FB-CARB, Raleigh, NC, Lot #: 042513) was used as reference, and injected along with each sequence, with an average recovery of 101.16 ± 1.7%.

2.4.3. Lachrymatory Factor (LF)

As discussed in Section 1, the LF concentration was measured as a direct indicator of pungency instead of pyruvic acid (McCallum et al., 2005). The LF content was analyzed following the procedure by Schmidt et al. (1996), with slight modifications. Thirty seconds after pressing, 4 mL of onion juice was pipetted from the juice collection beaker into a centrifuge tube containing 4 mL of 0.01% m-Xylene as internal standard (>99.0%, Acros Organics, Geel, Belgium, Legacy #: 180862500) in HPLC grade ethyl acetate (J.T. Baker, Center Valley, PA). The solution was vortexed and then centrifuged for 10 min using an HN-SII model centrifuge from Damon/ IEC Division (Needham Heights, MA). The organic layer was then extracted into 2 mL target vials and analyzed by gas chromatography with a flame ionization detector (GC-FID), within 30 min of extraction. GC analysis was conducted using a 7890A gas chromatograph (Agilent Technologies, Santa Clara, CA) with a DB-1, 30 m × 0.53 mm i.d. capillary column (J&W Scientific, Agilent Technologies, Santa Clara, CA, Part #: 125-1032), and 99.999% He carrier gas, with a flow rate of 8.5 mL/min. Injector temperature was set to 60 ℃, and oven was programed to hold at 60 ℃ for 1 min, followed by a temperature increase rate of 5 ℃/min to 200 ℃. The detector was maintained at 250 ℃. Quantification was conducted by comparing LF peak areas to the area of the internal standard (m-Xylene). The LF peak identity was confirmed by Hewlett-Packard 5890 series II GC coupled to a 5970 mass spectrometer. The chromatographic conditions used were the same as those for the GC-FID procedure, however, the column was a DB-5MS (30 m × 0.25 mm; J&W Scientific, Part #: 122-5532). The resulting chromatogram and fragmentation pattern for the LF peak were similar to those observed in previous reports (Block, Putman, & Zhao, 1992; Schmidt et al. 1996).

2.4.4. Methyl Thiosulfinates

Methyl thiosulfinates analyses were conducted by extraction of 4 mL of onion juice from the collection beaker into a centrifuge tube containing 4 mL of 0.00025% thioanisole as internal standard (>99%, Fluka Analytics, St. Louis, MO, Cat #: 88470) in HPLC grade ethyl acetate, 2 minutes after onion expression using the pneumatic press. The solution was vortexed, centrifuged for 10 min using an HN-SII model centrifuge from Damon/ IEC Division (Needham Heights, MA,). The lower organic layer was then extracted into 2 mL target vials for analysis by GC with flame photometric detection (GC-FPD) and a sulfur specific filter (Agilent Technologies, Santa Clara, CA, Part #: 1000-1437). A 7890A gas chromatograph (Agilent Technologies) with a DB-1, 5 m × 0.53 mm i.d. column (J&W Scientific, Agilent Technologies, Part #: 125-100B), and 99.999% He carrier gas, with flow maintained at 8.8 mL/min. The chromatographic conditions used were the same as those used for the LF analysis method. Quantification was conducted by summing the peak areas associated with the methyl thiosulfinate compounds and then comparing the resulting response to that of the internal standard (thioanisole). Concentrations of individual thiosulfinate were not sufficient enough to be detected using the GC-MS procedure described above; therefore compound identification was done by matching retention times and elution orders of the peaks to those observed in previous reports (Block, Naganathan, Putman, & Zhao, 1993; Schmidt et al., 1996). Use of this procedure in tandem with a sulfur specific detection system further assures accurate identification. Individual compound identification was not considered necessary due to the fact that the signal for the entire class of compounds was summed together in the chromatograms, and those compounds were eluted together within a small retention time range under the chromatographic conditions used in this procedure (Block et al., 1993; Schmidt et al., 1996).

2.5. Statistical Analysis

All of the data analyses were conducted using XLSTAT version 2015.4.01 (Addinsoft, New York, NY). The level of confidence was set at alpha equal to 0.05. Analysis of variance (ANOVA) with Fisher's least significant difference (LSD) was conducted over chemical analyses (sugars, LF and methyl thiosulfinates) and consumer data (OL, SPB, and WB) to evaluate differences among samples. Univariate analyses (canonical correlation analysis and Pearson's product-moment correlation analysis) and partial least squares (PLS) regression were performed to investigate the relationships between chemical compounds and consumer sensory scores of onion samples. PLS 2 was conducted since there were more than one variable in the data set of X-matrix (3 chemical compounds) and Y-matrix (3 consumer sensory scores). Cross-validation was applied to test the fitness of regression model.

Groups of consumers with homogeneous preferences were identified by agglomerative hierarchical clustering (AHC) using Euclidean distances and Ward's method. The consumer clusters were compared based on the socio-demographic information within each cluster. Chi-square was performed to the contingency table to test the similarity among the clusters.

3. Results and Discussion

3.1. Differences in Chemical Composition and Consumer Sensory Scores

The first step in the data analysis was to assess the source of variation in the instrumental measurement (sugars, LF, and methyl thiosulfinates) as well as in the consumer sensory scores (OL, SPB and WB) for each sample. Table 2 shows the results generated by ANOVA and differences among the samples.

Significant differences in the mean content of sugars were observed. The SH, HM and HH showed significantly higher content of sugar ($p<0.05$) while the HL had the lowest sugar content. However, there was no specific tendencies in sugar content in relation to the level of fertilizer, and no significant differences were observed in the analyzed chemical compounds between sweet (Nunhems 1006) and hot (Sapelo) variety. Concentrations of LF and methyl thiosulfinates were significantly different across the samples ($p<0.05$). Onion samples with low fertilization (SL and HL) had significantly lower content of LF and methyl thiosulfinates than those with medium and high fertilization.

Table 2 also shows the differences in mean consumer scores (OL, SPB and WB) on six Vidalia onion samples. The mean OL score showed that onions with low fertilization were preferred (SL = 7.17; HL = 6.75) to those of higher fertilization, regardless of onion variety (Nunhems 1006 or Sapelo). In terms of the OL and the SPB, consumer liking increased as the perceived pungency level decreased. WB ratings showed similar tendency of consumer preference with OL ratings.

When examining the concentration of LF and methyl thiosulfinates, OL and WB scores decreased as the concentration of those two chemical compounds increased. On the contrary, the SPB scores increased with increment of the LF and methyl thiosulfinates content in onion samples. Interestingly, the sugar content did not show consistent relationship with consumer sensory scores. The OL, SPB and WB scores fluctuated as the content of sugars increased. For example, consumers significantly preferred the SL to the SM, nonetheless these two samples were not statistically different in sugar contents ($p>0.05$). These findings demonstrated the need for examining the relative importance of chemical parameters on consumer preferences.

Table 2. Mean (± SD)[a] of instrumental parameters (n = 14) and sensory scores[b] from consumers (n = 142) for each Vidalia onion sample.[c]

	Sugars (g/100g fw)	LF (µmol/g)	Methyl thiosulfinates (nmol/g)	OL (1-9)	SPB (1-9)	WB (1-9)
SL	5.16 (± 1.51) ab	0.42 (± 0.46) b	0.24 (± 0.12) b	7.17 (± 1.82) a	2.65 (± 2.03) e	6.89 (± 2.37) a
SM	5.70 (± 1.67) ab	2.40 (± 0.79) a	0.46 (± 0.22) a	6.17 (± 2.00) cd	5.45 (± 2.25) b	5.88 (± 2.33) bc
SH	6.01 (± 1.20) a	2.46 (± 0.85) a	0.44 (± 0.21) a	6.03 (± 2.31) d	6.20 (± 1.97) a	5.54 (± 2.71) c
HL	4.66 (± 1.68) b	1.00 (± 0.90) b	0.30 (± 0.18) b	6.75 (± 1.93) ab	3.52 (± 2.25) d	6.23 (± 2.42) b
HM	6.12 (± 1.06) a	2.26 (± 0.85) a	0.48 (± 0.16) a	6.54 (± 1.85) bc	4.54 (± 2.45) c	5.99 (± 2.41) bc
HH	5.89 (± 1.36) a	2.72 (± 0.86) a	0.46 (± 0.13) a	6.25 (± 1.94) cd	5.01 (± 2.20) bc	5.87 (± 2.41) bc
Mean	5.59 (± 1.48)	1.88 (± 1.16)	0.40 (± 0.19)	6.48 (± 2.01)	4.56 (± 2.49)	6.07 (± 2.48)

[a] Mean values with the different letter within a column are significantly different ($p<0.05$).
[b] OL: overall liking (1 = dislike extremely, 5 = neither like nor dislike, 9 = like extremely); SPB: intensity of sharp/pungent/burning sensation (1 = low, 5 = moderate, 9 = high); WB: willingness to buy (1 = extremely unlikely, 5 = moderate, 9 = extremely likely).
[c] SL: sweet variety with low fertilization; SM: sweet variety with medium fertilization; SH: sweet variety with high fertilization; HL: hot variety with low fertilization; HM: hot variety with medium fertilization; HH: hot variety with high fertilization.

3.2. Correlation Analysis and Determination of Key Factors in Vidalia Onion Flavor

Canonical correlation analysis (CCA, also called canonical variate analysis) was conducted to quantitatively determine the strength of the relationship between the chemical composition (sugars, LF and methyl thiosulfinates) and consumer sensory scores (OL, SPB and WB). In this procedure, consumer scores were averaged for each onion bulb evaluated within each 14 test session, and the correlations are shown in Fig. 1.

According to the Wilk's lambda test, the first canonical dimension (Can1) was significant ($\Lambda = 0.55$; $F = 5.87$; $p<0.0001$), whereas the second and third canonical dimensions (Can2, Can3) were not ($F = 0.61$, $p = 0.66$; $F = 1.07$, $p = 0.30$, respectively). Therefore, only the Can1 was considered in further analyses. Canonical correlation coefficient (Rc) of Can1 was 0.66 and it explained 93.45% of the variance in the chemical and consumer datasets. Thus, it was clear that chemical composition and consumer sensory scores are significantly related. The results further indicate that both chemical and consumer evaluation of Vidalia onion can be used to discriminate flavor of the Vidalia onion samples investigated in this study.

As a next step, we developed separate models to predict the chemical composition (sugars, LF and methyl thiosulfinates) and consumer sensory responses (OL, SPB and WB) based on their corresponding variables. Canonical coefficients were checked to investigate the contributions of each variable to the Can1. Accordingly, the Can1 for consumer sensory responses (V_1) could be determined using the following formula:

$$V_1 = -0.669\ Y_{OL} + 0.742\ Y_{SPB} - 0.302\ Y_{WB}$$

Looking at the correlation between Can1 and each variable, all of the three variables were well explained in this canonical dimension: SPB showed the strongest correlation of 0.96

on Can1 followed by the OL (–0.83), and WB scores also showed significant negative correlation (–0.78). In addition, Pearson's correlation coefficient indicated a significant negative correlation between OL and SPB ($r = -0.65$, $p<0.05$). Thus, considering the contribution and correlation simultaneously, consumer sensory responses can be best predicted by the OL and SPB scores.

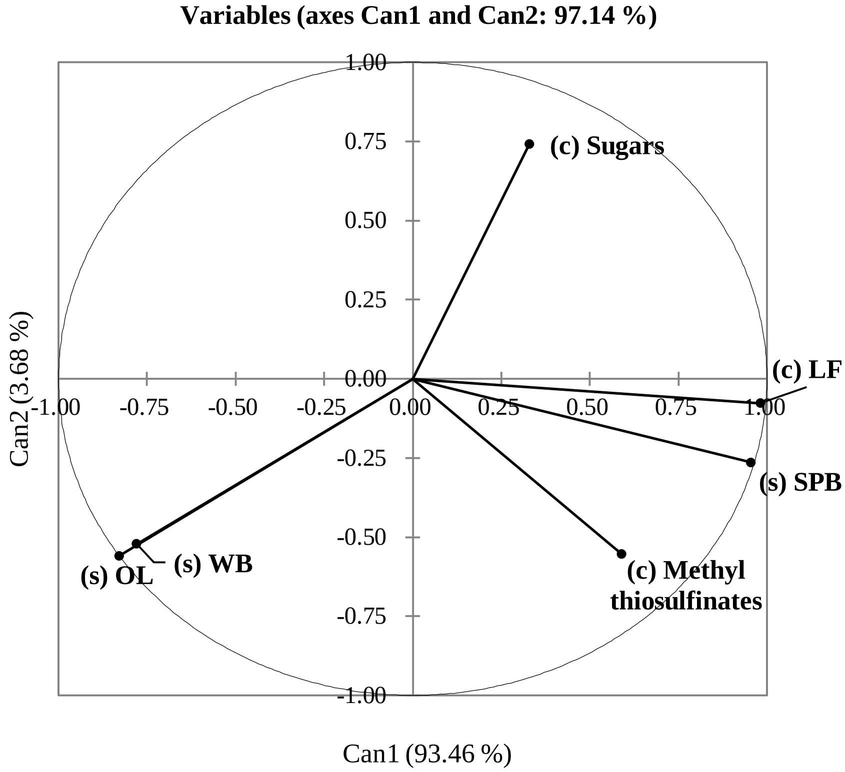

Fig. 1. Canonical correlation between chemical compounds (marked with 'c') and consumer sensory scores (marked with 's'). Consumer data (n = 142) were averaged for each onion sample within each evaluation session (n = 14). OL = overall liking, WB = willingness to buy, SPB = intensity of sharp/pungent/burning sensation.

Likewise, the formula to determine the Can1 for chemical composition (U_1) was:

$$U_1 = 0.167\ X_{sugar} + 0.903\ X_{LF} + 0.099\ X_{thiosulfinates}$$

The strongest correlation was shown in the LF (0.98) followed by methyl thiosulfinates (0.59). Sugars had weak positive correlation showing the value of 0.33. Therefore, it suggested that the LF content is the best predictor of the analyzed chemical compounds. Significant correlations with consumer OL and SPB scores ($r = -0.53$ and 0.62, respectively) also proved the appropriateness of the LF as a key factor in consumer flavor perception of Vidalia onion.

In summary, these results provided evidence that two consumer sensory scores (OL and SPB score) and LF concentration are the important factors in differentiating the flavor of Vidalia onions. Therefore, we tried to determine the consumer acceptability threshold of pungency based on the OL and SPB scores as well as the LF content.

3.3. Establishment of Desirable LF Threshold

Considering that the consumer test was performed using 9-point hedonic scale, where 5.0 was the middle point (neither liked nor disliked), overall liking score of 7.0 was chosen arbitrarily as a criterion to ensure reliable consumer preference. For each SPB score (1 to 9), the proportion of consumer acceptability (OL score ≥ 7) was investigated. Average concentration of the LF was also calculated at each SPB score. Threshold of LF content was determined by multiple regression of the % consumer acceptability on the mean LF content and SPB score, and the following equation was defined:

$$\text{Acceptability \%} = 92.12 - 0.91{*}\text{SPB} - 15.71{*}\text{LF} \quad (R^2 = 0.83)$$

As shown in Fig. 2, the LF threshold was established at 1.806 µmol/g, where 60% of the consumer acceptability was achieved and the intensity of SPB was scored between

3 and 4. Therefore, it seemed clear that Vidalia onions with LF content below 1.806 μmol/g will be not only fully acceptable to the majority of consumers, but also perceived as low pungent onions. Indeed, onions with LF below the suggested threshold were given low SPB scores and showed 69% to 72% of consumers acceptability (70% of consumer acceptability achieved = 1.336 μmol/g LF). For the LF concentrations above the threshold, however, the acceptability was presented at around 50% indicating the ambiguity of consumer liking within this range. Even the onions with the highest LF content at 2.41 and strongest SPB score at 9 were liked by nearly half of the consumers (48%). Linking to the ANOVA results (Table 2), for instance, both the HM and HL were equally liked by consumers (OL = 6.54 and 6.75, respectively; $p>0.05$). However, the LF content of the HM (2.26 μmol/g) was above the recommended threshold of 1.806 μmol/g, whereas the HL had LF content below the recommended threshold (1.00 μmol/g). This clearly indicates that the consumers reacted differently to the sensory attributes of the Vidalia onions. Accordingly, making predictions based on the averaged consumer liking may not fully capture the inter-individual differences among consumers (Guinard, Uotani, & Schlich, 2001).

Therefore, although the threshold of LF concentration at 1.806 μmol/g is recommended to be used as an indicator of consumer acceptability, it cannot tell the whole story; it neither ensures that Vidalia onions with LF content below 1.806 μmol/g would satisfy all of the consumers nor means that higher LF content above the threshold will be always unacceptable to consumers. This problem has brought the necessity of understanding individual consumer preferences on Vidalia onion in terms of multidimensional construct.

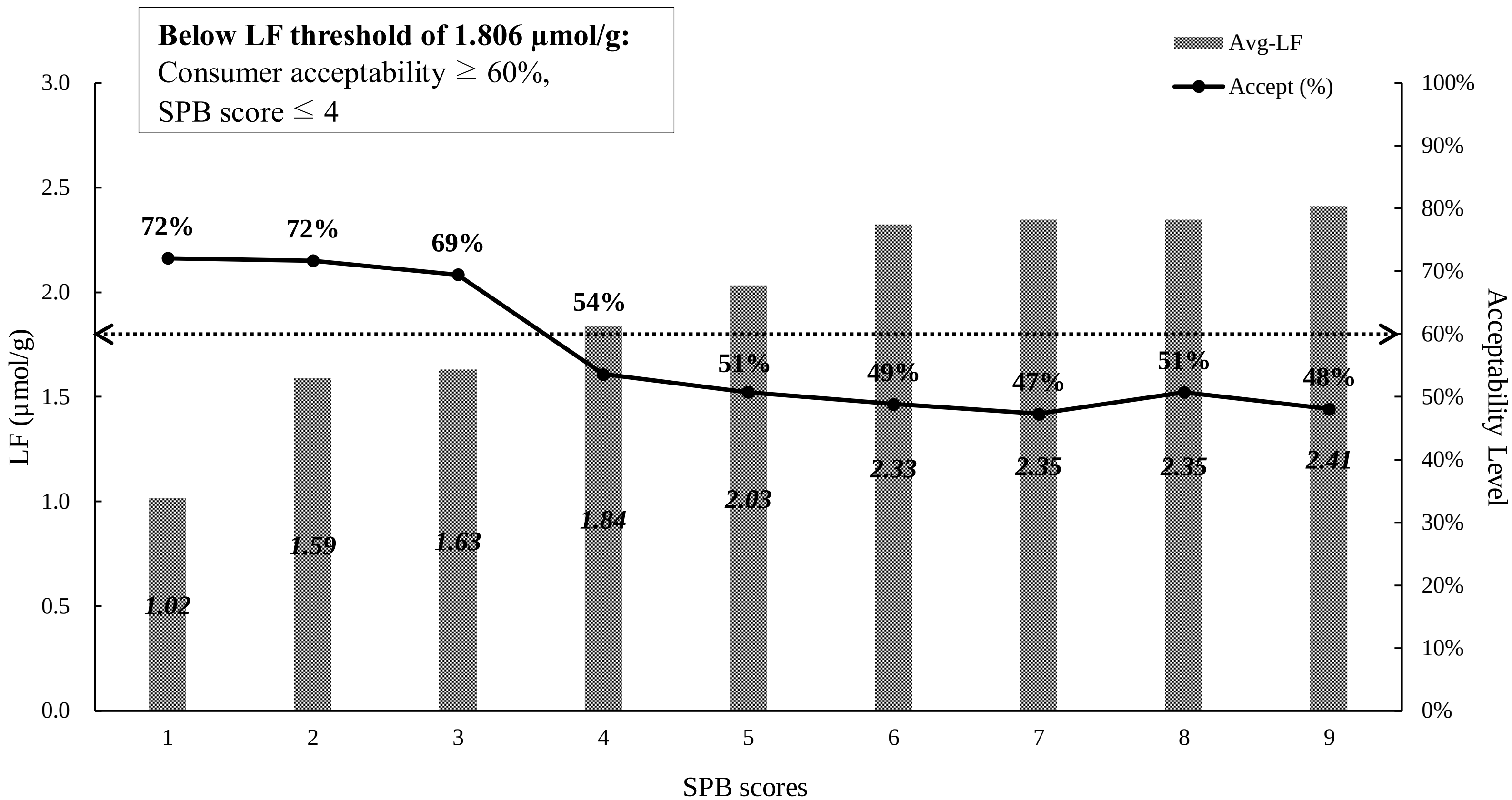

Fig. 2. Desirable LF concentration level established based on the of consumer acceptability (overall liking score ≥ 7) at each pungency score (n = 142). LF content averaged (grey bar, data labeled in italic) and proportion of consumer acceptability (solid line) within each SPB (sharp/pungent/burning sensation) score.

3.4. Consumer Preference Mapping by PLS Regression

PLS regression was performed to understand how the 6 onion samples and chemical parameters are related to the consumer responses. In the current study, chemical compounds (sugars, LF and methyl thiosulfinates) correspond to the X-matrix and the consumer scores (OL and SPB) to the Y-matrix. The variables in consumer scores which did not change for the sample effect were excluded from the PLS analysis.

Three components were automatically chosen by XLSTAT according to the cross-validation. In the present study, only the first two components (t_1, t_2) were shown since these two components were sufficient to describe the majority of consumers. In sensory science, using two components are generally adequate when there are low number of products, furthermore it allows convenience in graphical displays (Tenenhaus et al., 2005). All of the three chemical compounds were selected to be used as X variables according to their importance in the projection with respect to Y (VIP $\geq$ 0.8). A total of 99.3% of the variance of X (t_1: 92.7%; t_2: 6.6%) and 45.7% of the variance of Y (t_1: 32%; t_2: 13.7%) was explained by the first two components.

Fig. 3 shows the map of Samples $\times$ Chemical compounds $\times$ Consumer scores generated by using the first two components (t_1, t_2). The first component (t_1), opposes samples HL and SL to samples SM, HH, SH and HM. As shown, OL scores (blue dots) were largely concentrated at negative values of the t_1, revealing that the first group of onions (HL and SL) was preferred by most consumers. The result was congruous in that sample HL and SL showed the highest mean OL scores in ANOVA test (Table 2). The second group of onions (SM, HH, SH and HM) was described by consumers as more sharp, pungent and had stronger burning sensation (red dots), while less preferred than the onions in first group. For the chemical compounds, the LF and methyl thiosulfinates were highly correlated, and they were positively related to the consumer SPB scores. Thus, it clearly indicates that the OL is negatively associated with the concentration of the LF and methyl thiosulfinates.

Looking at the latent vectors, the second component (t_2) expressed the sugar content (X-loading value of –0.839). The sample HL, which had the lowest mean sugar content (Table 2), was separately placed from the other samples by the t_2 (Fig. 3., Quadrant 2). Sugar content showed some negative relationship with consumer liking (blue dots), even though there was less of a correlation ($r = -0.224$, $p<0.05$) than was true for the LF ($r = -0.553$, $p<0.0001$) and methyl thiosulfinates ($r = -0.320$, $p<0.01$).

With the common criteria, the quality of this regression was low; the cumulative percentage of explained variance for Y was equal to 45.7%, but the R^2 according to the cross-validation only amounts to 0.049 where the ideal value should be close to 1. In most preference mapping techniques including PLS regression, this low value in the quality of model fit is likely due to the diversity of the consumer perception and likings (Lawless & Heymann, 2010). Thus, this result suggested the possibility of segmentation among the consumers.

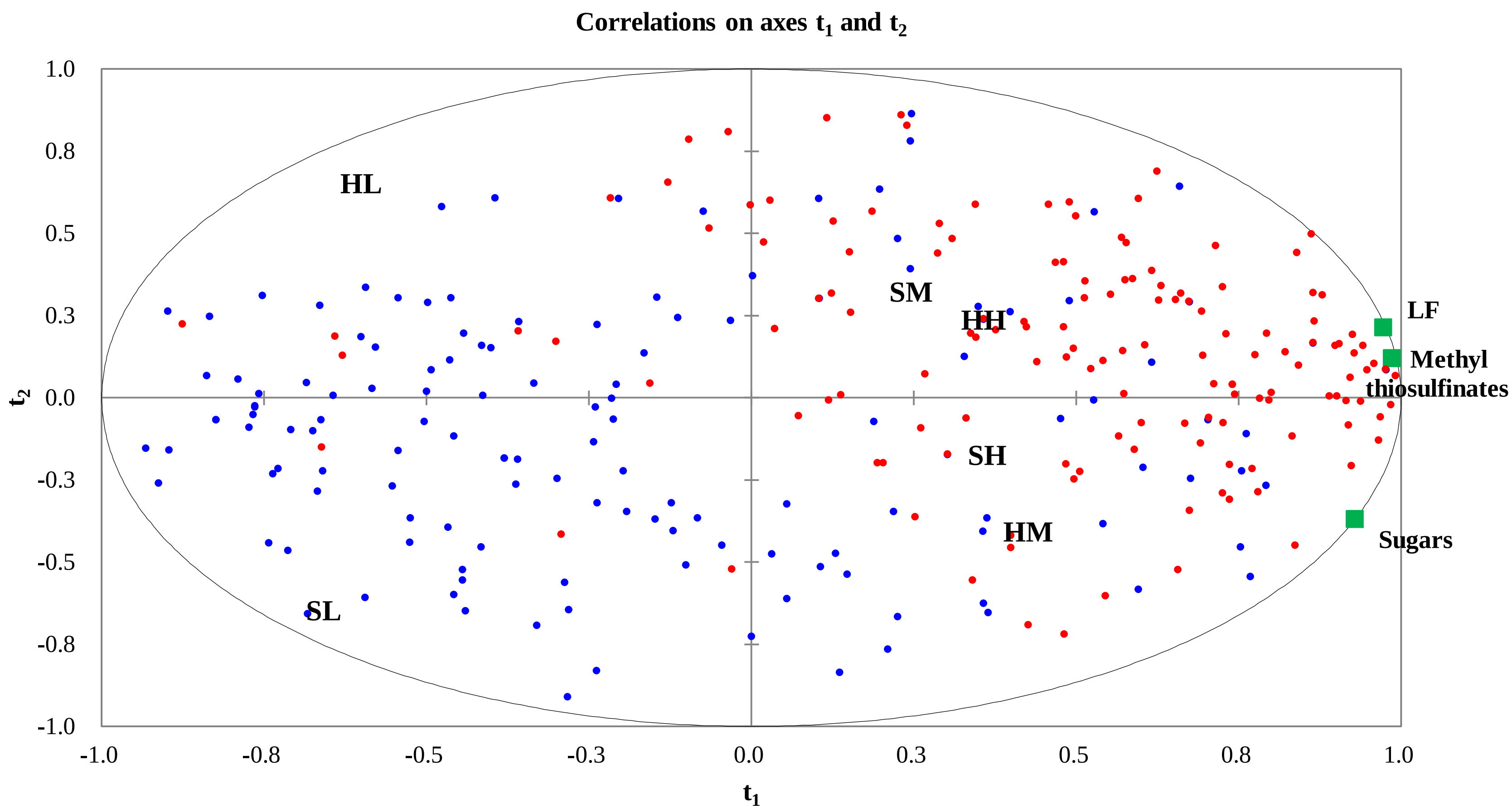

Fig. 3. PLS regression – Relationship between content of chemical compounds (sugars, LF, and methyl thiosulfinates) and consumer sensory scores (overall liking (OL) – blue dots; intensity of SPB – red dots) for 6 Vidalia onion samples. SL = sweet variety with low fertilization; SM = sweet variety with medium fertilization; SH = sweet variety with high fertilization; HL = hot variety with low fertilization; HM = hot variety with medium fertilization; HH = hot variety with high fertilization.

3.5. Clustering and Demographic Description of Consumer Groups

Agglomerative hierarchical clustering (AHC) analysis was performed on the consumer OL data using Euclidean distances and Ward's method to investigate potential consumer segments. Inspection of the dendrogram identified three clusters of consumers and there were significant differences among the clusters (ANOVA, $p<0.05$). The size of the clusters was not very balanced, with the cluster 3 being relatively small sized whereas the half of the consumers were assigned in cluster 1. Mean hedonic scores on six Vidalia onion samples given by each consumer cluster is shown in Table 3 and Fig. 4a.

Table 3. Mean hedonic scores [a] of the six samples [b] by consumer clusters. For each cluster (row), the most liked samples are in bold, while the least liked samples are in italics.

Cluster	SL	SM	SH	HL	HM	HH	Freq.
1	**7.83 a**	*6.81 a*	7.25 a	7.49 a	6.86 a	7.31 a	72
2	**7.51 a**	5.24 b	*3.94 b*	6.33 b	6.63 a	5.08 b	51
3	*3.74 b*	6.26 a	**7.00 a**	5.05 c	5.10 b	5.37 b	19
Mean	7.17	6.17	6.03	6.75	6.54	6.25	142

[a] Mean values with the different letter within a column are significantly different ($p<0.05$).

[b] SL = sweet variety with low fertilization; SM = sweet variety with medium fertilization; SH = sweet variety with high fertilization; HL = hot variety with low fertilization; HM = hot variety with medium fertilization; HH = hot variety with high fertilization.

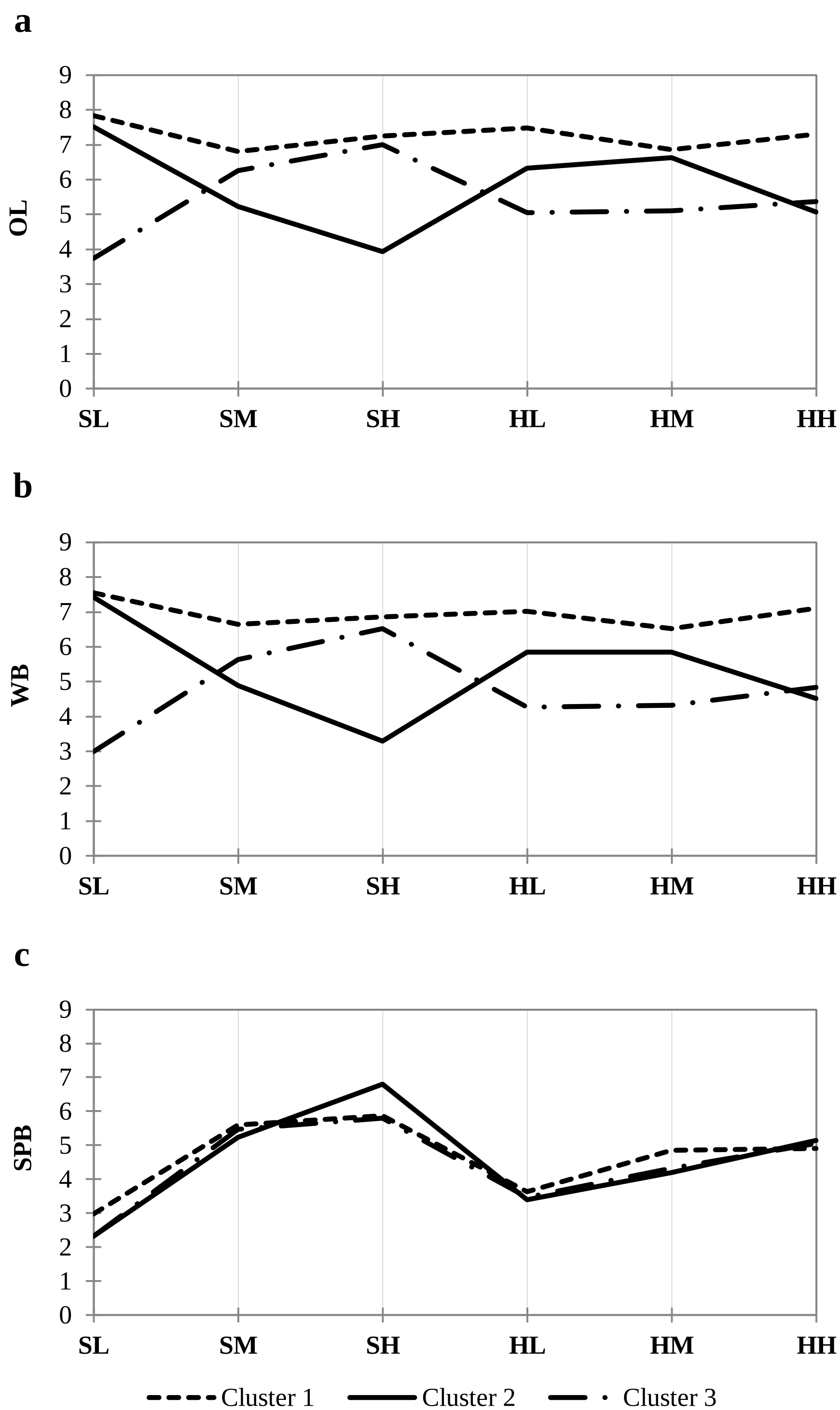

Fig. 4. Differences in averaged consumer sensory scores on six Vidalia onion samples among clusters. (a) Averaged overall liking score; (b) Averaged willingness to buy scores; (c) Averaged intensity of sharp/pungent/burning sensation scores. SL = sweet variety with low fertilization; SM = sweet variety with medium fertilization; SH = sweet variety with high fertilization; HL = hot variety with low fertilization; HM = hot variety with medium fertilization; HH = hot variety with high fertilization.

Cluster 1 was differentiated from the other clusters because the consumers in this group liked all of the samples, showing the mean OL score above 6.8 on every sample. In addition, the highest OL score for each sample was given by this cluster. The SL was most preferred (OL = 7.83) and the SM was less preferred but still liked (OL = 6.81), where the former onion contained significantly lower content of LF and methyl thiosulfinates (Table 2). The liking of about 50% of the consumers (n = 72) participated in the study was not influenced by the onion variety (Nunhems 1006 – sweet, Sapelo – hot) as well as the fertilizer level (low, medium and high). It indicates that the range of acceptability is broader for this consumers in terms of onion chemical composition.

Cluster 2 differed from the other two clusters in that the SH was disliked (OL = 3.94). The degree of liking on SM and HH were not distinctive as they were both scored close to 5.0 (anchored as 'neither liked of disliked') on the 9-point hedonic scale. The SL was given the highest OL score similar to the cluster 1 (Table 3). It appears that these consumers preferred the onions with small amount of LF and methyl thiosulfinates (Table 2). The hedonic scores decreased when these chemical compounds were present at high concentrations in the onions. This suggested that consumers in cluster 2 tended to prefer mildness and low pungency in Vidalia onions.

For cluster 3, the liking tendency was reversed from the cluster 2; the SH was most liked whereas the SL was least liked (Table 3). The SL was even scored as 'disliked (OL = 3.74)' by the consumers in this cluster despite the fact that it obtained the highest OL scores by the majority of consumers (clusters 1 and 2). It clearly indicates that there exist a consumer segment having a different point of view on onion flavor related to the preference: liked pungent onions and disliked mild onions.

In agreement with the previous analyses (CCA and PLS regression), sugar content in onions did not play an important role in differentiating consumer likings, because there was no significant differences ($p>0.05$) of sugar content between most and least liked onions in each cluster. Therefore, the drivers of likings were mainly determined by the LF and methyl thiosulfinate concentrations for all clusters.

As a summary, the means of consumer sensory scores for each cluster are also plotted in Fig. 4a-c. The WB scores (Fig. 4b) showed very similar tendency to the OL scores (Fig. 4a). Cluster 1 did not clearly distinguished six Vidalia onion samples based on the WB scores as the plotted line was all above the scores of 6.5. Again, clusters 2 and 3 showed an inverse relationship. Interestingly, the pattern of the SPB scores was similar across the three clusters (Fig. 4c) showing no statistical differences among clusters (ANOVA, $p>0.05$). With regard to the chemical compounds, the LF and methyl thiosulfinates content presented congruous patterns when plotted across the onion samples (not shown here). Even though the pattern of the SPB scores was in agreement among clusters, their likings were truly differentiated. This result implies that strong pungency and burning sensation in Vidalia onions can be a driver of either liking or disliking for the consumer segments in market, although with different size of segments.

So far, consumer clusters were explored by similarity of preference. To gain an insight whether the consumers within each cluster can be characterized in terms of socio-demographic information, its relationship to hedonic tendency was determined. Table 4 summarizes the consumer demographics, consumption and purchasing habits, and perspectives on Vidalia onion.

Table 4. Socio-demographic information summary for each cluster and the totals (percentages are in parentheses).

	p-value [a]	Cluster 1 (n = 72)	Cluster 2 (n = 51)	Cluster 3 (n = 19)	Total (n = 142)
Gender	0.228				
Female		51 (70.8%)	40 (78.4%)	11 (57.9%)	102 (71.8%)
Male		21 (29.2%)	11 (21.6%)	8 (42.1%)	40 (28.2%)
Age	0.138				
18–24		6 (8.3%)	10 (19.6%)	3 (15.8%)	19 (13.4%)
25–34		15 (20.8%)	10 (19.6%)	6 (31.6%)	31 (21.8%)
35–44		12 (16.7%)	8 (15.7%)	6 (31.6%)	26 (18.3%)
45–54		16 (22.2%)	11 (21.6%)	4 (21.0%)	31 (21.8%)
55–65		23 (31.9%)	12 (23.5%)	0 (0.0%)	35 (24.6%)
Frequency of onion consumption	0.746				
Daily		11 (15.3%)	4 (7.8%)	3 (15.8%)	18 (12.7%)
2-3 times/week		38 (52.8%)	26 (51.0%)	11 (57.9%)	75 (52.8%)
Once a week		11 (15.3%)	14 (27.5%)	3 (15.8%)	28 (19.7%)
Thrice a month		4 (5.6%)	3 (5.9%)	0 (0.0%)	7 (4.9%)
Twice a month		3 (4.2%)	2 (3.9%)	0 (0.0%)	5 (3.5%)
Once a month		3 (4.2%)	0 (0.0%)	1 (5.3%)	4 (2.8%)
Less than once a month		2 (2.8%)	2 (3.9%)	1 (5.3%)	5 (3.5%)

Table 4. Cont'd

	p-value [a]	Cluster 1 (n = 72)	Cluster 2 (n = 51)	Cluster 3 (n = 19)	Total (n = 142)
Frequently consuming onion type [b]	0.995				
Red		36 (50.0%)	23 (45.1%)	7 (36.8%)	66 (17.8%)
White		36 (50.0%)	24 (47.1%)	8 (42.1%)	68 (18.3%)
Yellow		40 (55.6%)	28 (54.9%)	12 (63.2%)	80 (21.6%)
Vidalia		58 (80.6%)	38 (74.5%)	10 (52.6%)	106 (28.6%)
Green		24 (33.3%)	13 (25.5%)	4 (21.1%)	41 (11.0%)
Shallot		5 (6.9%)	4 (7.8%)	1 (5.3%)	10 (2.7%)
Way of consumption (eating)	0.130				
Cooked		49 (68.1%)	41 (80.4%)	11 (57.9%)	101 (71.1%)
Uncooked/Raw		23 (31.9%)	10 (19.6%)	8 (42.1%)	41 (28.9%)
Important aspects in purchasing [b]	0.133				
Freshness		52 (72.2%)	39 (76.5%)	13 (68.4%)	104 (28.3%)
Shape/Size		12 (16.7%)	4 (7.8%)	2 (10.5%)	18 (4.9%)
Color of skin		16 (22.2%)	8 (15.7%)	1 (5.3%)	25 (6.8%)
Mildness		25 (34.7%)	12 (23.5%)	4 (21.1%)	41 (11.1%)
Hotness/Pungency		2 (2.8%)	1 (2.0%)	3 (15.8%)	6 (1.6%)
Sweetness		34 (47.2%)	28 (54.9%)	5 (26.3%)	67 (18.2%)
Health benefit		16 (22.2%)	7 (13.7%)	2 (10.5%)	25 (6.8%)
Package – Bag		21 (29.2%)	8 (15.7%)	1 (5.3%)	30 (8.2%)
Package – Loose		24 (33.3%)	20 (39.2%)	8 (42.1%)	52 (14.1%)

Table 4. Cont'd

	p-value [a]	Cluster 1 (n = 72)	Cluster 2 (n = 51)	Cluster 3 (n = 19)	Total (n = 142)
Familiarity with Vidalia onion	0.633				
Yes		61 (84.7%)	41 (80.3%)	15 (78.9%)	117 (82.4%)
Neutral		4 (5.6%)	1 (2.0%)	1 (5.3%)	19 (13.4%)
No		7 (9.7%)	9 (17.7%)	3 (15.8%)	6 (4.2%)
Quality satisfaction on Vidalia onion [c]	0.243				
Yes – satisfied		48 (76.2%)	26 (61.9%)	10 (62.5%)	84 (69.4%)
Neutral		13 (20.6%)	12 (28.6%)	6 (37.5%)	31 (25.6%)
No – not satisfied		2 (3.2%)	4 (9.5%)	0 (0.0%)	6 (5.0%)

[a] Obtained from chi-square test on the contingency table. The p-value > 0.05 indicates the independence of two categorical variables (i.e. female, male (rows) vs. clusters 1, 2 and 3 (columns)). In this case, there was no association found among clusters and socio-demographic variables.

[b] Multiple answers were allowed for these questions, thus percentages add up to over 100%.

[c] Asked only for those who answered "Yes" or "Neutral" for Vidalia onion familiarity question.

Consumers were not differentiated by the frequency of onion consumption ($p > 0.05$). A large number of consumers consumed onions more than once a week, regardless of their different preference tendency. In all clusters, the female proportions were higher than male proportions since the participants in this study were mostly female. In terms of onion cooking habits, most consumers eat cooked onions instead of fresh/raw onions. Majority of the consumers were familiar with Vidalia onions and satisfied with its quality sold at local market.

In cluster 1 (liked all onions at any intensity of pungency), the largest age group (31.9%) was between 55 and 65, followed by 45-54 (22.2%). They consumed Vidalia onions most frequently showing the highest percentage among three clusters. For the other onion types, it showed balanced distribution; half of the consumers in this group often consumed red and white onions, while one-third of them consumed green onions as well. Generally, these onions are relatively more pungent and hot compared to the sweet onions (e.g. Vidalia® or Walla Walla). It suggested that these consumers enjoy different onion types with various pungency level, although Vidalia onion is their favorite one. This finding was in agreement with their liking tendency on the six samples. They were very concerned about freshness when purchasing onions. Sweetness was the second most important aspects, followed by mildness and package – loose.

Cluster 2 (preferred less pungent onions) had a balanced distribution of ages with slight bias towards 45-65 years. Vidalia was also the most frequently consumed onions, and yellow onion was the second most. Unlike the cluster 1, other types of onions were not very frequently consumed by these consumers (< 50%). This is likely because Vidalia onions, as a sweet onion type, are known to be sweeter and less pungent than other types of onions. They had similar consideration with cluster 1 on onions when deciding purchase; freshness, sweetness and package – loose.

The consumers in cluster 3 (preferred pungent onions and disliked mild onions) were relatively balanced in gender, but younger than the other clusters. About 60% of them were aged between 25 and 44, while the consumers in the other two clusters were mostly older than 45 years old. Yellow onions were more frequently consumed than Vidalia sweet onions for these consumers. They didn't care much about sweetness when buying onions, whereas it was the second most important aspects for the other clusters. Instead, hotness/pungency was considered more importantly for the consumers in this cluster; on the contrary, it was the least important aspects for the cluster 1 and 2. Larger proportion of consumers in this cluster enjoyed uncooked, raw onions. It is worth to mention that onions become sweeter when cooked (caramelized) and their preferences for pungent onion might have been reflected in their consumption habit. These findings suggest that younger people have higher acceptability on the hotness or pungency in onions, in some part, moreover they tend to look for stronger flavors rather than mildness.

As the current study was conducted with consumers recruited in the city of Griffin, it is assumed that the findings can broadly stand for the state of Georgia. Further study is required to include wide selection of consumers from different areas in the U. S., which will reveal more extensive answers to which factors drive consumer liking in Vidalia onion. Since the focus in this paper was on the chemical composition and consumer preference, inclusion of descriptive sensory analysis and extra information from consumers such as ethnicity, income, importance of price or motivation of purchase, will allow acquiring much thorough knowledge.

4. Conclusion

This study revealed that chemical composition of Vidalia onion is correlated with consumer sensory scores. Onion samples with low fertilization contained lower content of LF and methyl thiosulfinates than samples with medium and high fertilization, resulting in higher average hedonic and willingness to buy scores from consumers. Consumer liking tended to decrease when they perceived higher intensity of pungency in the samples. Content of sugars was not influential both on chemical composition and consumer scores.

It was recommended that maintaining the content of the LF below 1.806 μmol/g in Vidalia onions is desirable to render low pungency and higher acceptability to the majority of consumers. While this threshold could be used by the Vidalia onion committee as an internal criterion in the field of cultivation to assure the quality as sweet onions, it is also important to have in-depth understanding of potential market segments with different consumer liking tendency.

From the results of this study on six Vidalia onion samples, it was apparent that there exist three different groups of consumers. Cluster 1 was made up of consumers with a liking in all onions at any intensity of pungency. Cluster 2 represented preference to less pungent onions showing considerably higher consumption frequency in Vidalia onions. Cluster 3, the most unique segment, was the group of consumers who liked pungent onions whereas they disliked mild ones. They were mostly younger consumers and did not care much about sweetness when purchasing onions.

This study could prove to be a valuable methodology of assessing the influence that onion variety and sulfur fertilizer level have on the flavor of Vidalia onions and may ultimately allow a more guided approach to ensure the good quality in terms of consumer preferences.

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SUMMARY AND CONCLUSION

The objective of the study was to investigate the relationship between onion chemical composition (sugars, LF and methyl thiosulfinates) and the actual flavor perception in two Vidalia varieties, Nunhems 1006 (sweet) and Sapelo (hot), grown at different levels of fertilizer.

The first study showed that the content of LF and methyl thiosulfinates in Vidalia onions were highly correlated to the burning sensation and aftertaste perceived by sensory panels. However, there was no significant correlation between sugar content and sweetness. Given these results, we can conclude that the LF and methyl thiosulfinates content play important roles in the sensory characterization of onion flavor.

The second study revealed that chemical composition of Vidalia onion is correlated with consumer sensory perception and preference. Consumer preferences tended to decrease when they perceived higher intensity of pungency and burning sensation in onions. Conforming to the findings of the first study, sugar content was not influential both on chemical composition and consumer scores. It was recommended that the LF content below 1.806 µmol/g in Vidalia onions would be desirable to render low pungency and higher acceptability to the majority (60%) of consumers. On the other hand, three consumer segments that have different liking tendency were found in the PLS regression and agglomerative hierarchical clustering (AHC) analysis. Cluster 1 was made up of consumers who liked onions at any intensity of pungency. Cluster 2 represented preference to less pungent onions. Interestingly, consumers in cluster 3 liked pungent onions whereas they disliked mild ones.

This study could prove to be a valuable methodology of assessing the influence that onion variety and fertilizer level have on the flavor of Vidalia onions and may ultimately allow a more guided approach to ensure the good quality in terms of consumer preferences.