Effects of Serving Temperature on Sensory Perception and Acceptance of Brewed Coffee

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Effects of Serving Temperature on Sensory Perception and Acceptance of Brewed Coffee

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Food Science

by

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University of Arkansas
Bachelor of Science in Food Science, 2016

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Abstract

Coffee continues to be one of the most widely consumed beverages worldwide. How an individual perceives a cup of coffee is impacted by a plethora of factors including origin, growing climate, roasting level, and consumption habits. This thesis utilized both trained descriptive panelists and untrained consumer panelists to analyze how serving/consumption temperature modulates sensory perception of brewed coffee in regards to appearance, aroma, flavor, taste, and mouthfeel. Three varieties of coffee (Colombia, Ethiopia, and Kenya) were brewed and served to panelists at four temperatures: 70, 55, 40, and 25 °C. In one study (Study 1, Chapter 3), results from descriptive analysis showed that product temperature had a larger effect in modulating sensory perception than did coffee variety. In another descriptive analysis study (Study 2, Chapter 3), trained panelists found that serving temperature had a more significant effect on perception than freshness, up to 90 minutes, of the brewed coffee sample of Ethiopian variety. Utilizing an untrained consumer panel and a Check-All-That-Apply (CATA) method to assess these same coffee samples, results showed that both serving temperature and coffee variety largely contributed to the variation in sensory perception. While these consumer panelists were more effective in differentiating between coffee varieties when assessing the samples at a lower (40 °C) temperature, liking of the sample was highest when served at hot temperatures (55 and 70 °C). This indicates that subtle attributes of brewed coffee may be easier to identify when served at lower temperatures. In a final study using CATA, additions of cream and sugar were added to the brewed coffee sample and served at four temperatures. Results showed that temperature is a significant modulator of sensory perception in enhanced coffee (i.e., brewed coffee with cream and/or sugar). The findings of this thesis show the importance of controlling temperature for the sensory evaluation of coffee products, since significant variations in both qualitative and quantitative sensory perception arise from changes in product temperature.
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Chapter 1.

General Introduction
Sensory perception is defined as “the series of events required for an organism to receive a sensory stimulus, convert it to a molecular signal, and recognize and characterize the signal” (Gene Ontology Consortium, 2018). The primary senses involved in sensory perception of humans involve five senses: vision, olfaction (smell), gustation (taste), touch, and hearing (Meilgaard et al., 2015). When evaluating food or beverage products, sensory attributes are typically perceived in order of appearance, odor/aroma, consistency and texture, and flavor (Meilgaard et al., 2015). Many of these sensory attributes are experienced simultaneously upon consumption of the sample; therefore, it may be difficult for panelists to distinguish one from another. With so many different senses being involved, many researchers have sought to explain how one sense can affect another sense. Evidence of these cross-modal interactions can be seen between many senses. For example, a sense of smell is known to be affected by the other sensory cues. In the case of vision and olfaction, congruent visual cues can aid in identification of an odor (Blackwell, 1995) and also modulate odor intensity (Zellner and Whitten, 1999). It has been found that when a solution is colored, the perceived odor intensity is higher than compared to a colorless solution (Zellner and Kautz, 1990; Kemp and Gilbert, 1997; Koza et al., 2005). One of the lesser explored interactions between senses is the association of temperature cues with other sensory cues.

The effect that temperature has on sensory perception has been studied for quite some time in a variety of products, but the actual mechanism of thermal perception is just beginning to become more understood. Perhaps the most obvious impact that temperature has on the perception of foods and beverages is how it affects liking of product. It should come to no surprise that people prefer and like to eat/drink products at the temperatures they are normally consumed (Cardello and Maller, 1982). Is this just a case of being used to something or is there
something more grand occurring to either the product or the transduction of signals from sensory neurons. Temperature has been definitively shown to be linked to taste perception, but these thermal effects have been understudied (Lemon, 2017).

Consider this scenario: Sarah, an office worker, goes to the communal coffee pot and pours herself a mug of black coffee. She sips her coffee initially as it is very hot (85 °C). After a brief moment, the coffee reaches a temperature of 70 °C. This consumption temperature is comfortable for her and she drinks a bit more of her coffee. After several more minutes, the coffee is now 50 °C. Sarah takes another drink and finds that her coffee tastes a bit differently than when she drank it earlier at 70 °C, she notices a chocolate flavor. Sarah is called over to her manager’s office where her job performance is discussed. This interaction takes 25 minutes. Sarah comes back to his office to finish her mug of coffee and finds it to be cold, almost room temperature (25 °C). She drinks the rest of her coffee and notices more subtle flavors she had not perceived before and a more pronounced sourness. She does not like the coffee as much. Sarah’s scenario illustrates the journey that a poured cup of coffee can go through and how sensory perception can be impacted.

Coffee is a very complex product containing more than 800 different volatile molecular species after being roasted (Illy, 2002). Coffee also possesses compounds leading to the perception of some basic tastes, mostly sour and bitter. Consumers typically prefer their coffee to be quite hot, often above temperatures at which burning should occur (Lee et al., 2003). However, at these hot temperatures it has been shown that roasted aromas overpower the perception of more subtle aromas, such as floral or fruity notes (Steen et al., 2017). It is understood that temperature alters aromas, flavors, and basic tastes of coffee, but both appearance and texture may be affected as well. Many previous coffee studies have only focused
on assessing products at one particular temperature, but there is clearly a need to explore how products change as a result of temperature.

This thesis aimed to determine the effects of serving temperature on the overall sensory perception of multiple varieties of coffee as perceived by both untrained consumers and trained descriptive panelists. In addition, this thesis aimed to develop a method to characterize product temperature-induced sensory variations in brewed coffee.
References


Chapter 2.

Literature Review
1. Physiology of Temperature Perception

Sensory systems have been called a link between the central nervous system and the events that occur outside of it (Feher, 2012). These systems let us perceive the world around and act accordingly. Essentially a stimulus will activate a specialized receptor and a signal will be transduced to the brain where it can be processed. This mechanism is observable in all of the major sensory systems. The sense of touch provides information regarding shape, texture, and temperature of objects (Patapoutian et al., 2003). Sensing changes in skin temperature is an important contributor to the total amount of information that is relayed and perceived from an object, or some stimuli, to an individual (Darian-Smith and Johnson, 1977). Thermal perception also can play crucial roles in habitat choice, avoidance of dangerous conditions, and behavioral thermoregulation (Breed and Moore, 2016).

Thermal sensation involves stimulation of peripheral sensory nerves that begin in the dermis which transmits electrical signals to the central nervous system through the superficial dorsal horn of the spinal cord and are terminated in the thalamus and somatosensory cortex, where some perception is made about the initial stimuli (Feng, 2014). The cutaneous sensory system contains a number of different specialized receptors ranging in functions from allowing us to feel touch, pressure, heat, and pain (Feher, 2012). The neurons of this peripheral nervous system originate in the dorsal root ganglia (Dhaka et al., 2006). In the case of temperature perception, the major receptors are generally known as thermoreceptors and include both cold and hot receptors. Thermal stimuli applied to the skin, resulting in changes in skin temperature, are perceived as either warm or cool, but adaptation of these sensations can occur quickly (Schepers and Ringkamp, 2010). Through micro-neurographic recordings, the ability of humans to discriminate cold temperatures has shown that by cooling the skin by just 1 °C, a cooling
sensation is evoked, which indicates that this is a sensitive system (Campero et al., 2001; Dhaka et al., 2006).

Feher (2012) states that cold receptors are free nerve endings with thin myelinated fibers, whereas warm receptors are free nerve endings with unmyelinated axons. The unmyelinated fibers (0.5-2 m/s) found in the warm receptors lead to a lower conduction speed compared to the myelinated fibers (12-30 m/s). Feher (2012) goes on to explain that thermoreceptors exhibit both tonic (slowly adapting) and phasic (rapid adapting) levels of activity. Hensel and Zotterman (1951) found that warm receptors continuously discharge electrical energy to constant warm temperatures. The two levels of activity allow an individual to perceive that something is hot, while also allowing them to feel warmth over a span of time.

The theory of temperature perception, otherwise known as thermoreception, has changed throughout the past few centuries. In the 19th century, studies on thermoreception were influenced by von Frey’s specificity theory of somesthesis (Green, 2004) which stated that the body has a separate sensory system for perceiving pain, just as other senses have separate systems. This theory carried over to temperature perception, which can be a contributor of pain. Boring (1942) believed that each sensory neuron of the skin was sensitive to only one form of stimulation, such as pain or temperature, and there were numerous electrophysiological studies contributing to the evidence of his beliefs. Zotterman (1935) performed experiments on cats that examined action potentials of the glossopharyngeal nerve and chorda tympani and found that applying chemical stimuli (basic tastes) to the tongue elicited small action potentials that were distinguished from touch. Zotterman (1935) also found that thermal stimuli (hot or cold water) produced action potentials similar in size and shape to those produced chemically, but concluded that these responses were separate from touch and chemesthesis. Further research by Dodt and
Zotterman (1952) led to the distinction in characteristics and behavior of warm and cold fibers that is outlined in Table 1. This research further added to the evidence of the specificity theory. However, Green (2004) stated that temperature, more specifically painful and non-painful temperature otherwise known as innocuous and noxious, is sensed by a more complex and interactive system than the one described in the specificity theory. Knowledge and elucidation of temperature perception continues into the present as researchers continue to focus on specific mechanisms.

**Table 1. Comparison of the characteristics in behavior of warm and cold receptors**

<table>
<thead>
<tr>
<th>Warm Receptor</th>
<th>Cold Receptor</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Steady discharge at constant temperatures between 20 and 50 °C</td>
<td>• Steady discharge at constant temperatures between 8-12 °C and 41 °C</td>
</tr>
<tr>
<td>• Maximum frequency between 38 and 43 °C</td>
<td>• Maximum frequency between 25 and 35 °C</td>
</tr>
<tr>
<td>• Maximum frequency of steady state discharge from a single fiber in response to a constant temperature = 3.7/s</td>
<td>• Maximum frequency of steady state discharge from a single fiber in response to a constant temperature = 9.8/s</td>
</tr>
<tr>
<td>• Aperiodic discharge of impulses in response to constant temperature</td>
<td>• Periodic discharge of impulses in response to constant temperature</td>
</tr>
<tr>
<td>• Sudden heating produces rapid volley of impulses</td>
<td>• Sudden cooling produces rapid volley of impulses</td>
</tr>
<tr>
<td>• Paradoxical discharge of phasic character in response to fall in temperature of more than 8 – 15 °C</td>
<td>• Paradoxical steady discharge in response to constant temperatures of 45 – 50 °C</td>
</tr>
</tbody>
</table>


Recent molecular biological methods have led to the discovery of the initial mechanism of temperature perception (Green, 2004). Some transient receptor potential (TRP) channel receptors are the primary transducers for temperature perception (Breed and Moore, 2016). TRPs are essentially cation channels, typically permeable for Ca$^{2+}$ and Mg$^{2+}$, that covert energy into
action potentials (Pedersen et al., 2005). Twenty-eight different TRP channels in humans have been identified and grouped into 6 families (Pedersen et al., 2005). Currently only 3 families of TRPs are associated with temperature perception: vanillloid (TRPV), melastatin (TRPM), and ankyrin (TRPA). These temperature-activated TRP ion channels are known as thermoTRPs (Schepers & Ringkamp, 2010). Within these subfamilies, 6 individual ion channels (Table 2) have been found to be thermoTRPs (Pedersen et al., 2005; Feng, 2014). These thermoTRPs are activated by distinct physiological temperature and convert thermal information into chemical and electrical signals (Patapoutian et al., 2003). Interestingly most of these thermoTRPs are also activated by non-thermal, but chemical substrates. How exactly are these ion channels activated by both temperature and chemical agonists? Three possible mechanisms have been theorized for temperature sensitivity: changes in temperature leading to production and binding of channel-activating ligands, channel proteins undergoing structural rearrangements leading to channel opening due to temperature, and that thermoTRPs sense changes in membrane tension due to lipid bilayer rearrangement resulting from temperature changes (Clapham, 2003). Voets et al. (2004) examined two thermoTRPs, cold activated TRPM8 and heat activated TRPV1, in order to address this using human embryonic kidney cells that were transiently transfected with either TRPM8 or TRPV1. These researchers suggest that thermal activation of these two TRPs arises due to basic thermodynamic principles and is due to a difference in activation energies associated with voltage-dependent opening and closing of gates. In regards to chemical agonists, Voets et al. (2004) found that gating of thermoTRPS was modified to “mimic and potentiate the thermal responses” when subjected to the chemical activators, specifically capsaicin and menthol. In contrast to these findings, utilizing similar methodology but focusing only on TRPM8, Brauchi et al. (2004) found that TRPM8 cold activation was only not voltage dependent and could be
gated separately by cold and voltage. While thermoTRP channels have been clearly shown to play a role in the perception of temperature (Dhaka et al., 2006), more research needs to be conducted in order to better understand the mechanisms involved.

Table 2. TRP channels associated with thermal perception.

<table>
<thead>
<tr>
<th>ThermoTRP Channel</th>
<th>Temperature Sensitivity</th>
<th>Non-thermal activators</th>
</tr>
</thead>
<tbody>
<tr>
<td>TRPV1</td>
<td>≥ 42 °C</td>
<td>Capsaicin, ethanol, low pH</td>
</tr>
<tr>
<td>TRPV2</td>
<td>≥ 52 °C</td>
<td>Insensitive to irritants</td>
</tr>
<tr>
<td>TRPV3</td>
<td>≥ 33 °C</td>
<td>Camphor</td>
</tr>
<tr>
<td>TRPV4</td>
<td>27 °C – 42 °C</td>
<td></td>
</tr>
<tr>
<td>TRPM8</td>
<td>&lt; 25 °C</td>
<td>Menthol, eucalyptol</td>
</tr>
<tr>
<td>TRPA1</td>
<td>&lt; 17 °C</td>
<td>Cinnamaldehyde, allicin, eugenol</td>
</tr>
</tbody>
</table>


Sime temperature perception involves transduction mechanisms sharing pathways with other sensory systems, such as taste, there should be an effect of temperature on how taste is perceived. Through functional magnetic resonance imaging Guest et al. (2007) found that oral temperature is represented in the primary taste cortex and other regions to which it connects in humans and suggests that “combination-responsive neurons may provide the basis for particular combinations of temperature, taste, texture and odor to be especially pleasant”. The growing amount of knowledge regarding temperature perception makes it a useful subject matter for research. How temperature can alter one’s sensory perception regarding food/beverages should be explored. Considerations of how temperature can affect a food/beverages’ physical attributes
or chemical composition as well as how temperature modulates an individual’s sensory systems and subsequently their perception of a product will be beneficial to the field of sensory science and consumer research.

2. Effect of Temperature on Sensory Perception

2.1 Taste

A majority of the research focusing on the effect of temperature on sensory perception has been done using basic taste solutions (salty, sweet, sour, bitter, and umami tastes). Findings typically indicate that temperature does impact perception and intensity of basic taste solutions. Results across studies do not always agree, however.

Literature indicates that the perception of sweet taste intensity increases in sucrose solutions as the temperature is increased. Studies have found that sweet intensity perception is impacted by both temperature and concentration and there may be some interaction between the two. In a study by Bartoshuk et al. (1982) using 7 concentrations of sucrose at 6 temperatures (4, 12, 20, 28, and 36 °C) found that sucrose solutions near body temperature were sweeter than sucrose solutions at room temp, but this was only seen in concentrations that were less than 0.52 M sucrose. When the concentration of sucrose was increased there was no statistical difference in sweetness intensity perception as a function of temperature. A similar study by Calvino (1986) using 5 concentrations at 3 temperatures (7, 37, and 50 °C) produced very similar results, with participants rating warmer samples as more sweet than colder samples. Once again, as concentration of sucrose increased, there was a smaller effect due to temperature. While sweetness perception of sucrose was shown to increase when tasted at warmer temperatures, a sweet solution of saccharin did not produce the same results (Green and Frankmann, 1988)
which indicates the effect of temperature on sweetness perception is not equal for all types of sweeteners (Frank et al., 2008).

In regards to sour tastes, there have been studies showing that temperature does impact detection and intensity perception of sour solutions, but the majority of research indicates that there is no significant effect on sour perception due to temperature (Moskowitz, 1973; Green and Frankmann, 1987). In a study using 20 participants, Lipscomb et al. (2016) found that citric acid solutions were perceived as significantly more intense at 23 °C compared to 3 and 60 °C. This result is consistent with prior findings of McBurney et al. (1973) that found that threshold detection was lowest at 32 °C for a sour solution. These results together indicate that sourness perception may be greatest at temperatures close to a normal human body temperature.

Bitterness intensity perception has been reported to be decreased with lower temperatures. In a study involving modulation of both product and tongue temperature, Green and Frankmann (1987) found that cooling the tongue and taste solution caused a reduction in perceived intensity of bitterness of a caffeine solution across different concentrations. In contrast, McBurney et al. (1973) found that bitterness threshold detection was lowest in a QSO₄ solution at 22 and 27 °C. These results confirm Paulus and Reisch's (1980) study that sought to explain the temperature effect on detection of basic taste solutions which also found that bitterness detection threshold was lowest around 20 °C.

Some more recent research reveals a unique effect of temperature on taste in that by merely changing the temperature of the tongue one can elicit taste sensations. Cruz and Green (2000) showed that warming the tongue from 20 to 35 °C can produce a sweet taste sensation while cooling the tongue from 35 to 15 °C can produce salty or sour taste sensations. It should be noted that not everyone is a “thermal taster”. Cruz and Green conclude that the gustatory system
contains thermally sensitive neurons that contribute to coding for taste even in the absence of tastants. Taste neurons and pathways seem to behave differently under different temperatures. Talavera et al. (2007) states that the Transient receptor potential cation channel subfamily M member 5 (TRPM5), which is primarily responsible for taste transduction of bitter, sweet, and umami tastes, is thermally activated by heat. This activation may be reason that changes in sweet and bitter perception are decreased at lower temperatures. Regardless of the mechanism, it is clear that temperature does affect taste perception in basic taste solutions.

2.2 Aromas

The effect of temperature on volatility is well understood. At higher temperatures, substances become more volatile, meaning they are more likely to vaporize. These volatiles can be perceived by the olfactory system as they enter the nasal cavity and reach the olfactory epithelium. It is no surprise that foods/beverages served at higher temperatures will be rated as having more intense aromas. This effect has been clearly shown in research regarding food products like cheese soup (Kahkonen et al., 1995), red and white wine (Ross and Weller, 2008), ham (Fuentes et al., 2013), and beef broth (Ventanas et al., 2010). In all of the studies mentioned, the researchers found that when samples were served at warmer temperatures, there was an increase in perceived aroma intensities. Using dynamic headspace sampling Steen et al. (2017) examined the ratio of volatiles present at 31 °C in one variety of coffee against the same coffee at warmer temperatures. Results showed that there was an accelerated release of volatile compounds above 40 °C. The effect of temperature on retronasal aromas (flavor) has not been explored as much as orthonasal aroma. However, Steen et al. (2017) also examined the perceived intensities of retronasal aromas of coffee using a trained descriptive panel. Results showed that
coffee served at lower temperatures was perceived as having more of a chocolate retronasal 
aroma when the coffee was served at lower temperatures (31, 37, and 44 °C) compared to higher 
temperatures (50, 56, and 62 °C), but the overall retronasal intensity was greatest at the higher 
temperatures. This indicates that some retronasal odors may be best perceived in samples served 
at lower temperatures. In the case of coffee, these are likely subtle flavors that may only become 
apparent or detectable when the strong roasted flavor has been diminished.

2.3 Texture

Prior studies have examined the effect that temperature has on attributes related to texture, 
such as viscosity and astringency. Smith et al. (1996) defines astringency as a dry or rough 
feeling resulting from tannins binding with salivary proteins to reduce oral lubrication. When 
examining the effect of temperature on perceived astringency intensity of cranberry juice served 
at 5 °C and 25 °C, Peleg and Noble (1999) found that samples served at 25 °C were perceived as 
more astringent than the samples that were served cooler. However, when looking at astringency 
intensity in red and white wines, Ross and Weller (2008) found no significant effect of 
temperature on astringency perception. It should be noted that Ross and Weller only examined 
wines served under a small temperature range: 4 – 18 °C for white wine and 14 – 23 °C for red 
wine. The researchers Peleg and Noble (1999) proposed that the suppression of astringency at 
lower temperatures could be the result of several mechanisms: cognitive interaction between two 
trigeminal sensations, namely cold and astringency, or an increased flow rate of saliva in 
response to cold solutions. A clearer example of the effect that temperature has on texture can be 
seen with viscosity. Increasing temperatures are typically associated with a reduction of viscosity. 
Revisiting Kahkonen’s (1995) cheese soup study, consumers found that the thickness of the soup
decreased at higher temperatures. This effect was seen regardless of the fat % that the soup was served at. More recently, Engelen et al. (2003) examined the viscosity of mayonnaise and a dessert custard at different temperatures using both consumers and instrumental analyses. Consumers reported a decrease in thickness when samples were served at higher temperatures and the instrumental analysis confirmed this result, with warmer samples having lower viscosities.

2.4 Overall Liking

In a recent review article, Lemon (2017) discussed the importance of temperature as a modulator of the intensity of gustatory, neural, and perceptual responses. Since temperature has been shown to modulate perceptual responses, there should be no surprise that a certain individual may prefer a food/beverage product at a certain temperature. This effect was clearly shown with the research of Cardello and Maller (1982), who examined this temperature effect on 13 different foods and beverages: lemonade, milk, coffee, baked ham, beef stew, pork sausage, creamed corn, hashed brown potatoes, apple pie, scrambled eggs, green beans, meatloaf, and dinner biscuits served at 5 temperatures (4.4, 12.8, 21.1, 37.8, and 57.2 °C). These researches discovered that none of the tested foods had maximum acceptability at ambient (21.1 °C) temperatures. As expected, foods that were typically served at cold temperatures, such as milk and lemonade, showed decreased acceptability with increasing temperatures, while the other foods/beverages showed an increase in acceptability as temperature increased. Coffee in particular had the least acceptability at 21.1 °C, whereas it had the greatest acceptability at 57.2 °C. In the cheese soup study by Kahkonen et al. (1995), consumers rated the soup most pleasant when it was served at the hottest serving temperature. It is possible that this increased
pleasantness can be traced back to prior experiences, where individuals likely preferred the warmer soup since that was what they typically consumed. However, they may have found the soup more pleasant because of the reduced viscosity and increased aromas. In any case, there has not yet been any research that explained if there is a direct temperature effect on the liking of a product or if the alteration of the specific attributes that are modulated by temperature play a role in overall liking.

3. Coffee

3.1 Complexity

Coffee remains to be one of the most widely consumed beverages worldwide. What exactly do people like about coffee? Is it the energizing effect that is given from ingesting caffeine, or is it the social aspect that often goes alongside having a cup of coffee? Another reason could stem from individuals liking the unique aromas and flavors that come from smelling and drinking coffee. When one drinks a cup of coffee, they typically do not imagine the long journey that the coffee has taken. The journey is composed by essentially 3 factors: predisposition (plant genetics), transformation (agronomy, climate, harvest, roasting, and brewing, etc.), and consumption (Yeretzian, 2017). Many of these factors are out of the control of consumers, but they can still alter the final cup of coffee through the brewing and consumption phase.

While brewed coffee consists mostly of water, other macromolecules such as lipids are present, as well as a multitude of other compounds. What makes coffee so unique is the sheer complexity it can contain. During the roasting process, coffee beans undergo a significant transformation due to the Maillard reaction acting on the proteins and carbohydrates present.
Roasted coffee contains more than 800 different volatile molecular species (Illy, 2002). These different volatiles are what give coffee its unique aroma. Depending on the variety of coffee, growing conditions, and roasting process a wide bouquet of aromas can be produced. The volatiles most responsible for giving coffee its unique aroma can be broken into 8 main categories as shown in Table 3. When coffee is made with using a drip brewing method, hot water passes through ground roasted coffee beans. Hot water is in contact with the grounds for roughly 4-6 min during the brewing process that allows large quantities of soluble acids caffeine, and other compounds to dissolve into the cup (Illy, 2002). The extraction of these compounds leads to different taste qualities, more specifically sour and bitter tastes, as well as astringency. The primary non-volatile components of coffee that influence the bitter taste of coffee are caffeine, trigonelline, and products produced by the thermal degradation of chlorogenic acids (Buffo and Cardelli-Freire, 2004). Non-volatile components contributing to sour taste include carboxylic acids such as citric, malic, and acetic acid. Other important non-volatiles such as polysaccharides and lipids can contribute to the viscosity of brewed coffee. In the case of astringency, the primary cause is quinic acid, which is formed from degradation products of chlorogenic acids. This nature and complexity of coffee leads it to be an ideal candidate for studies regarding the effect of temperature on taste perception and on overall sensory perception.

3.2 Effects of temperature on coffee perception

Most prior research involving brewed coffee and serving temperature has only sought to determine the optimal serving temperature in regards to consumer preference. There is not a consensus on what this temperature is, with studies showing a preferred temperature from 50-70°C (Borchgrevink et al., 1999; Lee and O’Mahony, 2002; Brown and Diller, 2008) and this
preferred temperature may be altered by the strength of coffee and additional ingredients. Lee and O’Mahony (2002) found that consumers tended to consume coffee containing sugar and creamer at a lower temperature compared to black coffee (59.0 and 61.5 °C, respectively), although there was no significant statistical difference.

While knowing the temperature at which most consumers prefer to drink their coffee is important, it still does not answer the question to how the overall sensory experience is modulated with altering the serving temperature. Few studies could be found that attempted to explain how flavor in brewed coffee is affected by serving temperature. Steen et al. (2017) used dynamic headspace sampling gas chromatography-mass spectrometry to evaluate volatile compounds of one particular coffee species, Bourbon Caturra, at different temperatures: 31, 37, 44, 50, 56, and 62 °C, and found a pronounced difference in the amount of volatiles present in regards to temperature. These researchers went on to evaluate this coffee at the same temperatures using a trained descriptive panel. Their findings were that warmer coffees were perceived as having a greater overall intensity and being perceived as more roasted, while cooler coffees were more associated with sweet and chocolate attributes. In the above study, only one variety of coffee was explored. The effect of temperature on sensory perception of one variety of coffee may not be the same in a different variety of coffee. The aforementioned researchers looked only at aroma and tastes and neglected to explore changes that may occur to appearance and textural attributes due to modulation of serving temperatures.

Yeretzian (2017) has seperated the sensory experience of coffee into four components: physical and chemical food properties, the consumption process, an individual’s neurological make-up, and an individual’s psychological and cognitive traits. Temperature can have an effect on each of these components. Starting with the physical and chemical properties of the product,
The volatile aromatic compounds are reduced when served at a lower temperature, significantly reducing the overall intensity of aroma attributes (Steen et al., 2017). Looking at the consumption process, temperature certainly alters the drinking pattern with hotter coffee likely being sipped to avoid burning the mouth while cooler coffee can be consumed at any pace. This difference in drinking pattern leads to more or less air-flow as well as frequency of swallowing, which has been shown to impact retronasal aroma perception (Buettner et al., 2002). In examining the effect of temperature on perception through an individual’s neurological make up, the effects on taste transduction as well as thermal tasting comes back into light, with some individuals perceiving tastes that may not even be present due to a change of their tongue temperature (Cruz & Green, 2000). Looking at psychology and cognitive traits, serving temperature may affect a consumer’s expectation. If a consumer expected their coffee to be hot, but it was served at a tepid temperature, the consumer would likely not enjoy the coffee as much and the perception of the product may be altered. It has been shown that emotional responses to coffee are impacted by serving temperature. Pramudya and Seo (2018) found that coffee served at ambient temperatures was more associated with negative emotional responses and low arousal compared to coffee served hot (65 °C) or cold (5 °C). The emotional response to coffee served at different temperatures may have an impact on the overall liking of the product. The effect of temperature on sensory perception of coffee should be explored more in order to gain knowledge for the ever growing coffee industry.
Table 3. Classes of volatile compounds identified in roasted coffee

<table>
<thead>
<tr>
<th>Category</th>
<th>Compounds</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sulphur compounds</td>
<td>Thiols, hydrogen sulfide, thiophenes (esters, aldehydes, ketones), thiazoles (alkyl, alcoxy and acetal derivatives)</td>
</tr>
<tr>
<td>Pyrazines</td>
<td>Pyrazine, thiol and furfural derivatives, alkyl derivatives (primary methyl and dimethyl)</td>
</tr>
<tr>
<td>Pyridines</td>
<td>Methy, ethyl, acetyl and vinyl derivatives</td>
</tr>
<tr>
<td>Pyrroles</td>
<td>Alkyl, acyl and furfural derivatives</td>
</tr>
<tr>
<td>Oxazoles</td>
<td></td>
</tr>
<tr>
<td>Furans</td>
<td>Aldehydes, ketones, esters, alcohols, acids, thiols, sulfides</td>
</tr>
<tr>
<td>Aldehydes and ketones</td>
<td>Aliphatic and aromatic species</td>
</tr>
<tr>
<td>Phenols</td>
<td></td>
</tr>
</tbody>
</table>

References


Chapter 3.

Effect of serving temperature on sensory perception of black coffee

using descriptive analysis
Abstract

Previous studies have shown the effect of temperature on human sensory perception related to aroma, taste, flavor, and mouthfeel. Thus, sensory research on foods and beverages that are served/consumed over a range of temperatures should include testing products at a range of temperatures to gauge how certain attributes are altered by temperatures. Coffee is one these products. Recent research has illustrated that serving temperature does have an impact on sensory perception of coffee, but more elaboration is needed. This study sought to determine the effect of serving temperature on sensory perception of multiple varieties of hot brewed coffee using descriptive analysis. Three varieties of coffee (Colombia, Ethiopia, Kenya) were ground, brewed, and served to six highly trained panelists at four temperatures: 70, 55, 40, and 25 °C. Results from this study found that serving temperature had an impact on 18 attributes related to appearance, aroma, taste, flavor, and mouthfeel. Using a principal component analysis, the majority of the variation in the data could be explained by PC1 (63.3%) and was mainly attributed due to differences between serving temperatures. A smaller portion of variation was explained by PC2 (21.2%) and mainly described differences between the coffee varieties. Using RV coefficients, it was shown that depending on the serving temperature, the panelists rated the products differently. A hierarchical clustering analysis further showed that the trained panel could not conclusively differentiate between the three coffee varieties. The findings of this study show the need to control product temperature across all individuals for all samples since the variation in sensory perception due to serving temperature is significant. Since the traditional technique of descriptive analysis was fairly ineffective in describing differences between the three coffee varieties in brewed coffee samples, further study is needed to increase a discrimination power of sensory testing for brewed coffee samples.
1. Introduction: Study 1

Consumers typically prefer to drink their brewed coffee at hot temperatures in the range of roughly 60-70 °C (Borchgrevink et al., 1999; Lee and O’Mahony, 2002; Brown and Diller, 2008). Because of this, it would be recommended that when performing sensory tests involving coffee, this temperature range should be kept in mind and utilized. However, the temperature at which coffee is consumed is not a static entity, it is constantly changing. In reality, consumers drink coffee at variety of temperatures typically starting very hot, as the recommended holding temperature is 80-85 °C, all the way to room temperature (25-30 °C). Consumers rarely drink their coffee at one particular temperature, but rather at a range of temperatures. This consumption of coffee at different temperatures may lead to differences in sensory perception regarding aroma and taste perception as well as overall liking. Thus, researchers should consider the effects of serving temperature on sensory perception especially on products which are consumed over a range of temperatures.

Research regarding the effect of serving temperature on sensory perception had been somewhat limited in only examining the overall acceptability of foods and beverages. Cardello and Maller (1982) found that foods are liked most at the temperature they are typically consumed. While these results are not surprising, they provide a basis for the thought that if overall liking is modulated by serving temperature, perhaps perceptions regarding individual attributes change as a function of serving temperature. More recently, work has been done to quantify changes in products due to serving temperature. In the case of aromas, intensities have been observed to be greater when served at warmer temperatures in products such as cheese soup, red and white wine, beef broth, and ham (Kahkonen et al., 1995; Ross and Weller, 2008; Ventanas et al., 2010; Fuentes et al., 2013). This is due to the overall volatility of aromatic compounds being increased
due to higher temperatures. With the increase in volatility, there are more aromatic compounds that enter the nasal cavity which, thereby inducting a greater perceived intensity. Since coffee is a complex product, containing more than 800 different volatile molecular species after being roasted (Illy, 2002), there should be a clear change in aroma perception depending on the temperature of the brewed coffee.

Many studies have found that modulating product or oral temperature can have an effect on how basic tastes are perceived (McBurney et al., 1973; Paulus and Reisch, 1980; Bartoshuk et al., 1982; Calvino, 1986; Green and Frankmann, 1987; Lipscomb et al., 2016). General findings are that sweetness intensity perception is increased when consumed at warmer temperatures compared to colder temperatures (Bartoshuk et al., 1982; Calvino, 1986). Green and Frankmann (1987) found that cooling the tongue to 20 °C caused a reduction in perceived bitterness intensity of a caffeine solution across different concentrations. In regards to sour taste, there is still some uncertainty on how temperature alters intensity perception with some researchers not finding any significant effect on sour perception due to temperature (Moskowitz, 1973; Green and Frankmann, 1987). However while not statistically significant, Green and Frankmann (1987) found a slight increase in perceived sourness when the tongue was cooled to 28 °C compared with 20 and 36 °C. Lipscomb et al. (2016) found that citric acid solutions were perceived as being significantly more intense at 23 °C compared to 3°C and 60 °C. This result is consistent with the prior findings of McBurney et al. (1973), who found that threshold detection was lowest, or most sensitive, at 32 °C for a sour solution compared to higher temperatures (37 and 42 °C). Thus there is some evidence to indicate that sourness perception may be increased at temperatures in the range of room temperature to body temperature. The above findings indicate
that by merely changing the temperature that a product is assessed at, human sensory perception regarding taste can be enhanced or inhibited.

Multiple methods have been previously utilized to assess coffee such as using an untrained consumer panel, a trained descriptive panel, cupping (performed by expert coffee tasters known as Q-certified coffee cuppers), and instrumental analysis. More recently multiple methods have been used in tandem in order to better describe and track changes in sensory properties and determine the overall quality of a coffee sample.

To explore how time, and ultimately temperature, influences brewed coffee, a modified descriptive analysis technique had been created called “Time scanning descriptive analysis” which aimed to provide analysis of hot foods/beverages that are consumed within a range of temperatures (Seo et al., 2009). This technique involved a time limit for the evaluation of each attribute, which in turn aided in controlling the temperature of the sample as it was being evaluated. Researchers noted that while this was an effective method in differentiating coffee samples, they did not compare if the method was more effective than conventional descriptive analysis.

Manzocco and Lagazio (2009) utilized untrained consumer data, instrumental analysis, and a trained descriptive panel in order to model shelf life acceptibility of a ready to drink coffee brew. They found that Hydrogen ion concentration along with perceived sourness from a trained descriptive panel produced a high correlation for predicting consumer rejection of a ready to drink coffee beverage. It should be noted that the samples were stored at 20 °C, but consumed at 70 °C and that both the trained panel and consumers were served only 20-mL of each sample. Due to this, there may have been variations in the overall assessment of the samples as seen with work done by Steen et al. (2017) regarding the effect of temperature on sensory perception of
brewed coffee. However, the methodology of using a trained descriptive panel along with instrumental data to predict acceptability of a product is quite ideal, as testing with consumers could be excluded resulting in both time and cost-savings.

Donfrancesco et al. (2014) sought to compare results regarding 13 coffee bean samples from Colombia, which were brewed as coffee, between a trained descriptive panel and the expert coffee graders. Once again, the temperature the coffee was tested at was 60-65 °C for the trained panel. No mention was made of the final temperature of the coffee post assessment, but the researchers may have had a temperature effect in mind as they provided panelists separate presentations in order to rate aroma and flavor. On the other hand, the cupping method that the Q-graders used involved assessing the sample at different temperatures. At 70 °C flavor and aftertaste were evaluated followed by acidity, body, and balance at 60 °C. Sweetness, uniformity, and clean cup were evaluated when the sample reached a temperature below 37 °C. While both the descriptive and cupping method was able to differentiate between many of the samples, Donfrancesco et al. (2014) noted that there was "little relationship among the individual characteristics measured by a trained sensory panel and the more broad quality characteristics measured by cuppers." One reason for this could be explained by the different valuation methodology employed by these two methods. The descriptive panel did not evaluate the coffee over a range of temperatures, while the cupping method does. Ultimately the two methods provide separate data in that cupping is used to determine the overall quality of a coffee while descriptive analysis looks at differences in specific sensory characteristics.

More recently, work has been done in order to illustrate how sensory perception of coffee changes as a function of serving temperature. A study regarding the effect of serving temperature on flavor perception of *Bourbon Caturra* coffee using descriptive and instrumental analysis was
recently performed by Steen et al. (2017). The descriptive panel rated 8 attributes (sour, bitter, and sweet basic tastes, roasted, nutty, tobacco, dark chocolate, and overall flavor intensity) of this coffee across 6 temperatures (31, 37, 44, 50, 56, and 62 °C). Dynamic headspace sampling and gas chromatography-mass spectrometry was utilized to analyze volatile compounds at different temperatures. However, these researchers encountered limitations of only analyzing samples up to 50 °C. Findings of the descriptive analysis showed significant temperature effects for overall intensity, bitter taste, sweet taste, and roasted flavor. Coffees served at higher temperatures were rated more intense in regards to overall intensity, bitter taste, and roasted flavor. Coffees served at the lowest temperature were more associated with sour tastes and a chocolate flavor. Results of the instrumental analysis revealed a clear difference in total peak area of joined total ion chromatograms based on the tested temperatures, indicating an increase in the amount of volatiles when tested at higher temperatures. However, there was a small difference in the area between the 31 and 37 °C samples which led to the conclusion that differences in coffee flavor between these two low temperatures may be very small. Interestingly they found no differences in qualitative profiles based on the temperature and that the overall ratios of the compounds stayed consistent. Unfortunately these researchers only focused on retronasal olfaction and did not ask the panelists to rate the samples orthonasally. Also, since only one particular coffee variety was analyzed, it is unsure of whether or not similar temperature effects could be seen for other varieties of coffee.

The aim of this study was to investigate the effect of serving temperature on an increased number of attributes pertaining to appearance, aroma, taste, flavor, and mouthfeel across multiple varieties of coffee using a trained descriptive panel.
2. Materials and Methods: Study 1

2.1 Sample Selection

A pilot study utilizing 10 volunteers was performed in order to find three varieties of coffee that had varying qualities in regards to sensory attributes such as appearance, aroma, and flavor. The results of this pilot study led to the selection of a Colombian, Ethiopian, and Kenyan variety.

2.2 Sample Preparation

Three types of locally roasted coffees were purchased at local markets, ground, brewed, and served to participants for testing: Ethiopian (Mama Carmen’s, Fayetteville, AR, USA), Kenyan (Onyx Coffee Lab, Fayetteville, AR, USA), and Colombian (Mountain Bird Coffee Company, Fayetteville, AR, USA). Coffee beans were ground with a coffee grinder (DBM-8, Cuisinart, East Windsor, NJ, USA) to a medium coarseness. 95 grams of coffee were place into a drip coffee maker (Model DCC-2900, Cuisinart, East Windsor, NJ, USA) along with 1.8 liters of spring water (Mountain Valley Springs Co., LLC Hot Springs, AR, USA). This led to a ratio of approximately 52.8 grams of ground coffee per liter water, which is within the range of the recommended coffee to water ratio recommended by the Specialty Coffee Association of America (SCAA, 2015). The coffees were then transferred to airpots (Model 36725.0100, Bunn, Springfield, IL, USA) where they could be dispensed into 4-ounce white Styrofoam disposable cups (Dart, Mason, MI, USA) at certain times to ensure proper temperature levels: 70, 55, 40, and 25 °C.
2.3 Descriptive sensory analysis

Descriptive sensory analysis of brewed coffee was performed at the University of Arkansas Sensory Service Center (Fayetteville, AR, USA). Six trained descriptive panelists were employed for testing the samples. Each participant had over 1,000 hours of experience evaluating food/beverage samples. Prior to testing, the panel was trained on evaluating coffee samples of 8 varieties using a modified Universal Aromatic Scale (UAS). With the help of a panel leader, a ballot was generated consisting of 32 attributes, which were separated into appearance, aroma, aromatics (flavor), basic tastes, and texture/mouthfeel (Table 1). The panel was trained using this ballot for multiple sessions until they could produce stable and reliable results within and among themselves. Each day, prior to assessment of the test samples, the panelists were given a warm up sample so that they could be calibrated against one another. Testing spanned across multiple sessions, with panelists assessing the three coffee varieties at one temperature per session. Within each session, the coffee variety serving order was randomized. Replicate measurements were obtained in a subsequent session on the same day. Panelists were given 5 minutes in between each sample and were instructed to cleanse their palate with crackers and water. 15-minute breaks were given between sessions.

2.4 Time and temperature tracking

One additional session was conducted in order to gauge exactly how much time each individual panelist took to assess each sample. The final product temperature was also measured after the assessment was complete. During this session, the panelists were unaware that they were being tracked in regards to time.
2.5 Statistical analysis

Analysis of Variance (ANOVA) was analyzed in JMP (version 13.1, SAS Institute Inc, Cary, NC, USA). Since the data produced from a descriptive panel is continuous in nature, ANOVA was performed in order to describe changes in sensory perception due to modulation of the coffee temperature and variety. Attributes that were not identified by at least 50% of the panel were removed from analysis. These included aromas of floral and skunky as well as nutty, fruity, and metallic aromatics. A three-way ANOVA model using coffee sample (S), temperature (T), and repetition (R) as main effects along with S×T, S×R, and T×R as interaction effects. Panelist was a significant source of variation for most of the sensory attributes and was included in the model as a random effect. A statistically significant difference was defined as \( P < 0.05 \). Post-hoc comparisons were performed using Tukey’s honest significant difference test.

Principal Component Analysis (PCA) on the covariance matrix, RV coefficient analysis, and Agglomerative Hierarchical Clustering (AHC) was performed using XLSTAT software (version 19.5, Addinsoft, New York, NY, USA).

3. Results and Discussion: Study 1
3.1 Effect of temperature on sensory perception of brewed coffee

The ANOVA results revealed that most sensory attributes, with the exception of green/unripe flavor and viscosity, were perceived significantly different between the 4 tested temperatures (Table 2). A significant interaction effect was seen between temperature and repetition for amount of oil, sour taste, and stale flavor (all at \( P < 0.05 \)).

When viewing temperature-dependent sensory attribute variations (Table 3.) it was found that the coffee served at 25 °C was perceived as having more visible oil and green color than the coffee samples served at 55 or 70 °C. In regards to aroma, all of the retained attributes showed a
similar trend, with the coffee evaluated at 70 °C having the greatest perceived aroma intensities compared with the coffee evaluated at 25 °C. This data coincides with previous instrumental data on volatile compounds found in the headspace of coffee samples at higher temperature (Steen et al. 2017) and follows the laws of thermodynamics. With increased volatility of aromatic compounds at high temperatures, a larger number of molecules would be present for our olfactory system to process which would lead to increased perceived intensities of aromas.

Perceived bitterness intensity was the least in samples served at 40 °C but was no different comparing samples served at 70 and 25 °C. This indicates a potential non-linear relationship between temperature and bitterness perception as proposed by McBurney et al. (1973) who found that bitterness perception involving QSO₄ was non-linear and most sensitive at 22-27 °C and least sensitive at 42 °C, which was the maximum temperature that was tested in this study. Conversely, Green and Frankmann (1987) found that bitterness intensity of a caffeine solution decreases with lower temperatures, but the methodology employed in their study involved both cooling the tongue and sample and was limited to 20, 28, and 36 °C. Direct comparisons should not be made with these past studies since temperature induces stimulus-dependent effects on bitter tastes (Lemon, 2017) and coffee contains a number of bitter compounds, such as chlorogenic acid lactones, trigonelline, and caffeine (Farah, 2012). Perception of sourness intensity increased as the sample temperature decreased with the coffee served at 25 °C being rated significantly sourer than at 70 °C. Whilst psychophysical data regarding temperature on sour perception has been quite variable (McBurney et al., 1973; Moskowitz, 1973; Green and Frankmann, 1987; Lipscomb et al., 2016; Lemon, 2017) the results from this present study show that temperature significantly impacts sour taste perception. Perceived aromatics (flavor) did not follow the same trend as aromas. Ashy flavor was most pronounced when served at 70 °C.
Cocoa/Chocolate flavor intensity was greatest at 70 °C and was diminished with cooler samples, which contradicts findings from Steen et al. (2017). Overall coffee impression and roasted flavor intensity was greatest at 70 °C and followed a linear trend in reduction as the samples were served at colder temperatures. Both stale and tobacco flavors were more pronounced in the coldest served coffee samples. One possible reason for the increase in stale flavor could be from the expectation of brewed coffee to be served hot. While a descriptive panel should not take into account how much they like or dislike a sample, there could have been a bias with the panelists disliking room temperature coffee and thus considering it to be stale, which is associated with low quality. The samples served at 25 °C were perceived as significantly more astringent than those served at 70 °C. While little research has been published regarding the effect of temperature on astringency, these results do not directly contradict those by Peleg and Noble (1999), who found that temperature had a significant effect on astringency of a cranberry juice, with 25 °C juice being rated as more astringent than juice at 5 °C. Since the present study and the one performed by Peleg and Noble (1999) do not look at similar temperatures, the results should not be directly compared.

3.2 Effect of sample on sensory perception of brewed coffee

Significant differences between the three coffee varieties were only observed in 7 attributes (Table 2): amount of oil \( (F = 5.80, P < 0.01) \), brown color \( (F = 46.99, P < 0.001) \), green color \( (F = 13.73, P < 0.001) \), coffee impression aroma \( (F = 3.17, P < 0.05) \), roasted aroma \( (F = 3.94, P < 0.05) \), sour taste \( (F = 9.00, P < 0.001) \), and green/unripe flavor \( (F = 4.12, P < 0.05) \). Panelists generally rated intensities of the attributes consistently across repetitions \( (P \geq 0.05) \), however there was a significant effect of repetition on stale flavor \( (P < 0.05) \). There was
also significant interaction effect in regards to temperature × repetition for amount of oil, sour taste, and stale flavor (for all, \( P < 0.05 \)).

Examining differences between coffee samples (Table 3.), the Colombian variety had a significantly different appearance, namely more brown color, than the Ethiopian and Kenyan varieties. The Colombian coffee was also perceived as more intense for coffee impression and roasted aroma attributes. The Kenyan coffee was rated as most sour in addition to being perceived as having the greatest green/unripe flavor. Interestingly there were no significant effects of coffee variety × temperature, which indicates that different coffee varieties may be affected similarly by temperature.

Since the perception of a brewed cup of coffee is influenced by a plethora of factors, including genetics (Yeretzian, 2017), it should stand to reason that there would be many significant differences between the 3 varieties, however this was not the case. One potential reasoning for this could be the type of scaling that was used for the assessment of aromas and flavors. In this present study, a Universal Aromatic Scale (UAS) was used in order to more rapidly assess the samples, thus reducing the potential heat loss during the assessment. Due to this type of scaling, potential differences between varieties could have been missed since the panelists only used a small portion of the scale for many of the attributes (Muñoz and Civille, 1998). Many aromas and flavors of coffee are quite subtle, sometimes barely perceptible. Due to this subtlety, many of the panelists rated the attributes lowly (1-2) on the potential 15-point scale. In order to have the panelists use a wider portion of the scale, a product-specific scale would be suggested as it has been shown to differentiate between similar samples, such as rice, more effectively than UAS (Jarma Arroyo and Seo, 2017). Alternatively, references and scaling provided in the World Coffee Research Sensory Lexicon (2016) could be utilized in the
assessment of different coffee samples, but these are costly in terms of time as many difference references need to be given to the panelists. Also, one would find that the amount of time taken to assess many attributes of a sample of hot coffee would increase dramatically, which in turn would influence the temperature at which it was assessed. Still, the fact remains that in this present study the same scaling method (UAS) was used to assess changes in sensory perception due to temperature and variety, and that the effects due to temperature are greater than those due to variety.

3.3 Principal component analysis

Viewing the multivariate relationship of the attributes between each sample served at each temperature through PCA provides a glimpse of how temperature modulates the perception of brewed coffee. In Figure 1, approximately 84% of the variation in the data was explained by Principal Component (PC) 1 (63%) and PC2 (21%). PC1 (63%) mainly describes the difference between serving temperature as all samples tested at 70 °C are loaded negatively on PC1 while the samples served at 25 °C are loaded positively on this PC1. The coffee samples served at warmer temperatures are more associated with more intense roasted, coffee impression, and chocolate attributes, while the samples served at colder temperatures are more associated with a sour taste and stale flavor. PC2 (21%) mainly describes differences between the coffee varieties, with the Colombian coffee (positively loaded) being perceived much differently than the Ethiopian or Kenyan varieties (negatively loaded). These differences between coffee varieties resulted mostly due to appearance attributes.

PCA was also used to explore the differences between coffee varieties at each specific temperature (Figure 2). The results from this indicate that there is clear separation of each coffee
sample for each tested temperature. However, when the panelists were assessing the samples at 55 and 40 ºC, there were less defined associations between the attributes and samples which can be observed from the clumping of the attributes around the origin. This indicates that temperature affected the way that panelists rated the coffee samples and subsequently that the differences between samples may be dependent on temperature.

3.4 RV coefficients

RV coefficients were calculated based on factor score matrices of the coffee sample at each specific temperature and are displayed in Table 4. The closer an RV coefficient is to 1, the more similar the factor score matrices. A p-value of less than 0.05 indicates that the RV coefficients are significantly different from 0. The RV coefficient comparing 70 to 55 ºC was 0.99 (P < 0.001) indicating that the factor scores of the samples were similar. Similarly, the RV coefficient of 40 to 25 ºC was 1 (P < 0.001) also indicating that factor scores of the samples were similar. These results indicate that the participants rated the samples similarly between both 70-55 and 40-25 ºC. Conversely when comparing 70 to 40 or 25 ºC, the RV coefficient is 0.88 (P = 0.17) and 0.91 (P = 0.17) respectively. In the same manner the RV coefficient between 55 and 40 ºC was 0.81 (P = 0.33) which indicates that the samples were rated differently across these temperatures. Ultimately this data shows that depending on the serving temperature, the panelists rated the products differently.

3.5 Agglomerative hierarchical clustering

Using AHC, the samples could be categorized into three clusters (Figure 3). Cluster 1 contains Colombia 70 ºC, Ethiopia 70 ºC, and Kenya 70 ºC. Cluster 2 contains the Colombian
variety at 55, 40, and 25 °C. Cluster 3 is comprised of both Ethiopian and Kenyan varieties at 55, 40, and 25 °C. These results further illustrate that the panelists did not effectively differentiate between the 3 coffee varieties, particularly at 70 °C. However, these clustering results suggest that they were able to differentiate the Colombian variety at the three lowest tested temperatures from the other two samples, Kenya, and Ethiopian, which were more similar to one another.

3.6 Time and temperature change during assessment

On average, it took panelists 149 (SD = ± 32.8) seconds to assess the samples. One panelist in particular took an average of 200 seconds with one sample taking over 250 seconds. This time taken resulted in temperature changes for each target temperature (Figure 4). Samples that were served at 70 °C were nearly 60 °C when evaluations were complete. Samples served at 55 °C were approximately 50 °C after evaluation. Samples served at 40 and 25 °C mostly retained their initial temperature. Future descriptive analysis tests involving products that are served at an initial temperature that changes over times should take into consideration how long it takes for the panelists to assess a sample. Shortened ballots could also be utilized in order to reduce assessment time if it is known which key attributes contribute to the most variation between samples.

4. Introduction: Study 2

Since there were significant effects of repetition and temperature * repetition found in the above study, it is possible that the holding time may have influenced the sensory perception of the coffees as only one brew of each variety of coffee was performed on a single day. Thus, the
maximum possible time between two repetitions was approximately 75 minutes. Therefore, a separate study was performed in order to control for any effects due to holding time.

Consider drinking a freshly brewed cup of coffee compared to drinking a cup poured out of a coffee pot that has set on the warmer for a certain amount of time. Some individuals could probably tell a difference between the two, but many may not be able to. Prior researchers have sought to determine differences in sensory perception of brewed coffee due to holding duration. In a long-term scenario, Manzocco and Lagazio (2009) found that as a ready to drink coffee beverage was stored for a period of 7 days, perceived sourness increased while perceived bitterness decreased. The effect of increased sourness and decreased bitterness was seen after two days. In a more relevant study with a shorter holding duration, Feria Morales (1989), using a trained panel, found that sensory perception was effected for a brewed coffee that was left on a hot-plate for a certain amount of time (0-60 min). Acidity was increased, bitterness was decreased, but mouthfeel characteristics were not changed after 60 minutes. These findings were somewhat consistent with Pangborn (1982), who found that sourness intensity perception changed after coffee had been stored for 3 hours at 80 °C, however Pangborn did not find significant changes for bitterness or overall intensity. From these findings, along with those presented in study 1, is apparent that both serving temperature and the amount of time that brewed coffee has been held can significantly affect sensory perception. This present study sought to examine differences between coffee that had been served fresh versus coffee that had been held for a period of time in order to confirm that the findings of study 1 were due to temperature.
5. Materials and Methods: Study 2

5.1 Sample preparation

The Ethiopian Variety from the prior study was ground and brewed in drip coffee makers (Cuisinart, East Windsor, NJ, PerfecTemp® Model DCC-2900) and transferred to airpots (Bunn, Springfield, IL, Model 36725.0100) for storage until they were ready to be dispensed. The ratio of ground coffee beans to water was 95 g of ground coffee per 1.8 liters of water, which equates to 53 g coffee/liter water, which is within the recommended ratio of 50-55 g/liter described by the Specialty Coffee Association of America (SCAA, 2015). Samples were to be served under two conditions, serving temperature and freshness. Serving temperature contained four levels (70, 55, 40, and 25 °C) while freshness contained two levels (fresh and old). Fresh samples were served 15 minutes after brewing whereas the old samples were served 90 minutes after brewing. To prevent any effect due to time spent cooling, the samples that would be served at lower temperatures were poured into glass bottles, sealed using screw on lids, and subjected to water baths until reaching the desired temperature. Separate brews were performed for samples at each temperature and freshness condition for to ensure that the samples were served under the same time conditions.

5.2 Descriptive sensory analysis

Nine descriptive panelists (six from study one plus three additional panelists) took part in this study. All panelists were trained in prior sessions for assessing coffee samples. The ballot used from Study 1 was used for the evaluation of samples in this study. There were four total sessions for the panelists. Each session consisted of the assessment of four samples, one for each temperature level. Within each session, panelists were given either fresh samples or old samples.
Within each session, the order of serving temperatures was randomized. In total, the panelists assessed 16 coffee samples at 4 temperatures under 2 freshness conditions. Panelists were given 5 minutes between each sample and 15 minute breaks between sessions.

5.3 Statistical Analysis

Analysis of Variance (ANOVA) was analyzed in JMP (version 13.1, SAS Institute Inc, Cary, NC, USA) using the freshness condition as a fixed effect and serving temperature, panelist, and repetition as random effects. Principal Component Analysis (PCA) on the covariance matrix was performed using XLSTAT software (Addinsoft, New York, NY, USA).

6. Results and discussion: Study 2

6.1 Effects due to holding time

Figure 5 shows that a holding time of 90 minutes had a significant effect on green/unripe flavor, metallic flavor, astringency, and viscosity perception (for all, \( P < 0.05 \)). Pairwise comparisons using student’s t-test revealed that the Ethiopian coffee that was served fresh was perceived as having more of a green/unripe flavor while the coffee served after a 90 minute holding time was perceived as being more metallic, astringent, and viscous than the freshly served coffee. Interestingly, these results do not agree with the findings of Feria Morales (1989), who found that mouthfeel characteristics were unchanged when coffee was held at a hot temperature for an hour. While not statistically significant, both sour taste (\( P = 0.053 \)) and bitter taste (\( P = 0.062 \)) showed a trend of being more intense in the “old” coffee samples which, in the case of sour taste, is in agreement with prior research (Pangborn, 1982; Feria Morales, 1989). Had the coffee been held for a longer period of time there may have been an increased number of significantly different attributes between freshness conditions. Overall, the findings of this study
are slightly different from those in prior research but may be due to the fact that samples were evaluated with two effects, namely serving temperature and freshness conditions.

6.2 Principal component analysis

Approximately 79% of the overall variation was explained by the first two principal components (Figure 6). PC1 (67.7%) was mostly composed of high ratings of roasted and overall coffee impression attributes as well as sour taste. This first principal component explained the differences in aroma, flavor, and taste perception related to serving temperature. PC2 (11.1%) was mostly composed of skunky, cocoa/chocolate, tobacco, and metallic attributes. This second principal component primarily explained the difference in sensory perception between holding duration conditions, otherwise noted at freshness. Overall, most of the variation in the data was explained by temperature and not freshness, which indicates that the findings from Study 1 were indeed due to temperature and not a confounding factor such as holding duration.

7. Conclusion

In general, effects due to serving temperature were greater than effects due to different coffee varieties. This is a bit odd, given that discrimination between the different coffee varieties was evident during the initial pilot study. In any case, since the effects due to serving temperature on sensory perception were substantial and significant, researchers should be aware of temperature variations of products, specifically coffee, and seek to ensure that all samples are treated and evaluated similarly while considering both initial and final temperatures. Failure to treat, serve, and evaluate samples equally may result in invalid results. Another key finding from this research is that serving temperature plays a much larger role in modulating sensory perception of brewed coffee than holding duration at a hot temperature (<80-85 °C), at least up
to 90 minutes. These results may have implications for cafes and restaurants, who could possibly brew coffee less often without their customers noticing much of a difference in overall perception of the brewed coffee, as long as the coffee was still served hot. Also the findings from study 2 confirm that the significant results presented in study 1 were due to temperature and not holding duration. Since the traditional methodology of descriptive analysis was not effective in differentiating between coffee samples (Figures 2 and 3), a new methodology is needed. This method will be discussed in Chapter 4.
References


<table>
<thead>
<tr>
<th>Descriptor</th>
<th>Definition</th>
<th>Reference (Intensity)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Appearance</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amount of oil</td>
<td>Visible amount of oil on surface</td>
<td>Coffee B² (10)</td>
</tr>
<tr>
<td>Brown color</td>
<td>The amount of brown color that can be observed</td>
<td>Coffee A¹ (10)</td>
</tr>
<tr>
<td>Green color</td>
<td>The amount of green color that can be observed</td>
<td>Coffee B (12)</td>
</tr>
<tr>
<td><strong>Aroma/Flavor</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ashy (A/F)⁴</td>
<td>Associated with burnt wood materials that can be sharp, bitter, and sour</td>
<td>Ashes (UAS³)</td>
</tr>
<tr>
<td>Burnt (A/F)</td>
<td>Associated with blackened/acrid carbohydrates</td>
<td>Burnt Toast, Espresso (UAS)</td>
</tr>
<tr>
<td>Cocoa/Chocolate (A/F)</td>
<td>A brown, sweet, roasted impression</td>
<td>Powdered cocoa, chocolate bars (UAS)</td>
</tr>
<tr>
<td>Coffee impression (A/F)</td>
<td>Associated with brewed coffee</td>
<td>Brewed Coffee (UAS)</td>
</tr>
<tr>
<td>Earthy/Dirty (A/F)</td>
<td>Characteristic of dry mud, dirt or soil</td>
<td>Dry mud, dried rosemary (UAS)</td>
</tr>
<tr>
<td>Fruity (A/F)</td>
<td>Characterized by the odor produced by fruits</td>
<td>Citrus or berry fruits (UAS)</td>
</tr>
<tr>
<td>Green/Unripe (F)</td>
<td>Associated with unripe fruits characteristic of freshly cut leaves, grass, or green vegetables</td>
<td>Green legumes (UAS)</td>
</tr>
<tr>
<td>Metallic (A/F)</td>
<td>Associated with metals</td>
<td>Wet cast iron, copper pennies (UAS)</td>
</tr>
<tr>
<td>Nutty (A/F)</td>
<td>Combination of sweet, brown, woody, aromatics associated with nuts, seeds, beans, and grains</td>
<td>Wheat germ, toasted sesame seeds (UAS)</td>
</tr>
<tr>
<td>Roasted (A/F)</td>
<td>Resembling a product cooked to a high temperature, not including burnt or bitter note.</td>
<td>Roasted coffee beans (UAS)</td>
</tr>
<tr>
<td>Skunky (A/F)</td>
<td>Associated with sulfur compounds such as mercaptan, which exhibit skunk-like character</td>
<td>Skunk (UAS)</td>
</tr>
<tr>
<td>Stale (A/F)</td>
<td>Characterized by lack of freshness and early stages of oxidation</td>
<td>Cardboard (UAS)</td>
</tr>
<tr>
<td>Tobacco (A/F)</td>
<td>Associated with tobacco leaves</td>
<td>Cured tobacco (UAS)</td>
</tr>
</tbody>
</table>
Table 1. Descriptors, definitions, and references for sensory attributes of brewed coffee samples evaluated in this study

(Continued)

<table>
<thead>
<tr>
<th>Descriptor</th>
<th>Definition</th>
<th>Reference (Intensity)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bitter taste</td>
<td>Taste on tongue elicited by solutions of caffeine or quinine</td>
<td>0.05% caffeine solution (2.0) 0.08% caffeine solution (5.0) 0.15% caffeine solution (10.0)</td>
</tr>
<tr>
<td>Sour taste</td>
<td>Taste on tongue elicited by solutions of acidic compounds</td>
<td>0.05% citric acid solution (2.0) 0.08% citric acid solution (5.0) 0.15% citric acid solution (10.0)</td>
</tr>
</tbody>
</table>

**Mouth-feel**

<table>
<thead>
<tr>
<th>Descriptor</th>
<th>Definition</th>
<th>Reference (Intensity)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Astringent</td>
<td>Chemical feeling factor on the tongue described as puckering/dry</td>
<td>0.01% Alum solution (6.0)</td>
</tr>
<tr>
<td>Viscosity</td>
<td>Thickness of solution in mouth.</td>
<td>5% sucrose solution (2.0)</td>
</tr>
</tbody>
</table>

1Reference coffee A: 53 g of ground coffee beans (Sumatra variety, Mama Carmen’s, Fayetteville, AR, USA) per 1,000-mL of water were brewed using a coffee maker machine (Model: DCC-2900, Cuisinart, East Windsor, NJ, USA) and then presented in a 120-mL white Styrofoam cup (Dart, Mason, MI, USA) when its temperature was reached to room temperature (approximately 20 °C).

2Reference coffee B: 53 g of ground coffee beans (Rwanda variety, Westrock Coffee Co., Little Rock, AR, USA) was prepared as described above.

3Intensities for aromas and flavors were rated based on the universal aromatic scale (UAS) with some modification; soda note in Nabisco Premium Original Saltine Crackers (Mondelēz Global LLC, East Hanover, NJ, USA) = 3.0; cooked-apple note in Mott’s Natural Applesauce (Mott’s LLP, Plano, TX, USA) = 7.0.

4A and F represent aroma and flavor, respectively.
Table 2. *F*-values in the three-way analysis of variances for sensory attributes among the three coffee samples rated at four temperature-conditions: 70, 55, 40, and 25 °C

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Sample (S)</th>
<th>Temperature (T)</th>
<th>Repetition (R)</th>
<th>S x T</th>
<th>S x R</th>
<th>T x R</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Appearance</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amount of oil</td>
<td>5.80**</td>
<td>20.34***</td>
<td>1.88</td>
<td>0.82</td>
<td>0.57</td>
<td>3.33*</td>
</tr>
<tr>
<td>Brown color</td>
<td>46.99***</td>
<td>3.83*</td>
<td>2.38</td>
<td>1.75</td>
<td>0.00</td>
<td>0.66</td>
</tr>
<tr>
<td>Green color</td>
<td>13.73***</td>
<td>25.74***</td>
<td>5.43*</td>
<td>1.26</td>
<td>1.97</td>
<td>1.51</td>
</tr>
<tr>
<td><strong>Aroma</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ashy</td>
<td>1.95</td>
<td>5.56**</td>
<td>0.26</td>
<td>0.30</td>
<td>0.01</td>
<td>0.97</td>
</tr>
<tr>
<td>Burnt</td>
<td>1.12</td>
<td>4.53**</td>
<td>0.15</td>
<td>0.33</td>
<td>0.14</td>
<td>0.09</td>
</tr>
<tr>
<td>Cocoa/Chocolate</td>
<td>0.64</td>
<td>3.54*</td>
<td>0.05</td>
<td>0.52</td>
<td>0.19</td>
<td>0.49</td>
</tr>
<tr>
<td>Coffee impression</td>
<td>3.17*</td>
<td>24.90***</td>
<td>0.22</td>
<td>0.72</td>
<td>0.60</td>
<td>1.02</td>
</tr>
<tr>
<td>Earthy/Dirty</td>
<td>0.45</td>
<td>11.16***</td>
<td>0.11</td>
<td>0.27</td>
<td>0.06</td>
<td>0.25</td>
</tr>
<tr>
<td>Nutty</td>
<td>0.34</td>
<td>10.63***</td>
<td>0.11</td>
<td>0.36</td>
<td>0.14</td>
<td>0.05</td>
</tr>
<tr>
<td>Roasted</td>
<td>3.94*</td>
<td>22.19***</td>
<td>0.11</td>
<td>0.34</td>
<td>0.40</td>
<td>0.27</td>
</tr>
<tr>
<td>Tobacco</td>
<td>0.26</td>
<td>4.40**</td>
<td>0.07</td>
<td>0.26</td>
<td>0.10</td>
<td>0.87</td>
</tr>
<tr>
<td><strong>Taste</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bitter taste</td>
<td>2.16</td>
<td>4.36**</td>
<td>0.10</td>
<td>0.56</td>
<td>0.03</td>
<td>0.31</td>
</tr>
<tr>
<td>Sour taste</td>
<td>9.00***</td>
<td>33.31***</td>
<td>0.02</td>
<td>0.74</td>
<td>0.14</td>
<td>3.05*</td>
</tr>
<tr>
<td><strong>Flavor</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ashy</td>
<td>0.10</td>
<td>5.47**</td>
<td>0.32</td>
<td>0.39</td>
<td>0.14</td>
<td>0.31</td>
</tr>
<tr>
<td>Burnt</td>
<td>1.35</td>
<td>8.31***</td>
<td>0.00</td>
<td>0.27</td>
<td>1.37</td>
<td>0.38</td>
</tr>
<tr>
<td>Cocoa/Chocolate</td>
<td>1.42</td>
<td>9.67***</td>
<td>0.70</td>
<td>0.20</td>
<td>0.12</td>
<td>0.99</td>
</tr>
<tr>
<td>Coffee impression</td>
<td>1.60</td>
<td>36.98***</td>
<td>0.22</td>
<td>0.60</td>
<td>0.14</td>
<td>0.75</td>
</tr>
<tr>
<td>Earthy/Dirty</td>
<td>0.43</td>
<td>8.87***</td>
<td>0.01</td>
<td>0.25</td>
<td>0.57</td>
<td>1.14</td>
</tr>
<tr>
<td>Green/Unripe</td>
<td>4.12*</td>
<td>1.88</td>
<td>0.09</td>
<td>0.73</td>
<td>0.27</td>
<td>0.41</td>
</tr>
<tr>
<td>Roasted</td>
<td>2.29</td>
<td>61.26***</td>
<td>0.11</td>
<td>0.43</td>
<td>0.61</td>
<td>1.10</td>
</tr>
<tr>
<td>Stale</td>
<td>0.22</td>
<td>39.26***</td>
<td>4.90*</td>
<td>0.34</td>
<td>0.57</td>
<td>3.36*</td>
</tr>
<tr>
<td>Tobacco</td>
<td>0.85</td>
<td>4.52**</td>
<td>0.02</td>
<td>0.97</td>
<td>0.04</td>
<td>0.62</td>
</tr>
<tr>
<td><strong>Mouth-feel</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Astringent</td>
<td>2.67</td>
<td>2.80*</td>
<td>0.22</td>
<td>0.37</td>
<td>0.22</td>
<td>0.19</td>
</tr>
<tr>
<td>Viscosity</td>
<td>0.84</td>
<td>0.75</td>
<td>0.62</td>
<td>0.29</td>
<td>0.30</td>
<td>0.11</td>
</tr>
</tbody>
</table>

* *, **, and ***: significance at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively
Table 3. Comparisons in the mean intensity ratings (± standard deviations) for sensory attributes as a function of coffee sample and temperature condition

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Coffee sample</th>
<th>Temperature condition</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Colombia</td>
<td>Ethiopia</td>
</tr>
<tr>
<td><strong>Appearance</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amount of oil</td>
<td>4.3 (± 3.3)a</td>
<td>2.7 (± 2.8)b</td>
</tr>
<tr>
<td>Brown color</td>
<td>10.5 (± 1.8)a</td>
<td>8.4 (± 1.4)b</td>
</tr>
<tr>
<td>Green color</td>
<td>1.8 (± 1.3)b</td>
<td>2.5 (± 1.6)a</td>
</tr>
<tr>
<td><strong>Aroma</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ashy</td>
<td>0.8 (± 1.4)</td>
<td>0.5 (± 1.1)</td>
</tr>
<tr>
<td>Burnt</td>
<td>1.5 (± 1.8)</td>
<td>1.0 (± 1.5)</td>
</tr>
<tr>
<td>Cocoa/Chocolate</td>
<td>0.9 (± 1.5)</td>
<td>1.1 (± 1.4)</td>
</tr>
<tr>
<td>Coffee impression</td>
<td>5.9 (± 1.4)a</td>
<td>5.6 (± 1.2)a</td>
</tr>
<tr>
<td>Earthy/Dirty</td>
<td>0.9 (± 1.6)</td>
<td>0.8 (± 1.5)</td>
</tr>
<tr>
<td>Nutty</td>
<td>0.3 (± 1.0)</td>
<td>0.4 (± 1.1)</td>
</tr>
<tr>
<td>Roasted</td>
<td>4.3 (± 1.2)a</td>
<td>4.0 (± 0.9)ab</td>
</tr>
<tr>
<td>Tobacco</td>
<td>0.7 (± 1.3)</td>
<td>0.5 (± 1.1)</td>
</tr>
<tr>
<td><strong>Taste</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bitter taste</td>
<td>8.3 (± 1.6)</td>
<td>7.9 (± 1.5)</td>
</tr>
<tr>
<td>Sour taste</td>
<td>3.1 (± 1.6)b</td>
<td>3.1 (± 1.5)b</td>
</tr>
<tr>
<td><strong>Flavor</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ashy</td>
<td>1.0 (± 1.6)</td>
<td>0.9 (± 1.5)</td>
</tr>
<tr>
<td>Burnt</td>
<td>2.8 (± 1.6)</td>
<td>2.3 (± 1.7)</td>
</tr>
<tr>
<td>Cocoa/Chocolate</td>
<td>1.6 (± 1.7)</td>
<td>1.5 (± 1.6)</td>
</tr>
<tr>
<td>Coffee impression</td>
<td>6.9 (± 1.3)</td>
<td>6.6 (± 1.2)</td>
</tr>
<tr>
<td>Earthy/Dirty</td>
<td>1.1 (± 1.7)</td>
<td>1.2 (± 1.7)</td>
</tr>
<tr>
<td>Green/Unripe</td>
<td>0.6 (± 1.2)b</td>
<td>0.6 (± 1.3)ab</td>
</tr>
<tr>
<td>Roasted</td>
<td>5.2 (± 1.4)</td>
<td>5.0 (± 1.3)</td>
</tr>
<tr>
<td>Stale</td>
<td>1.1 (± 1.5)</td>
<td>1.2 (± 1.6)</td>
</tr>
<tr>
<td>Tobacco</td>
<td>1.2 (± 1.6)</td>
<td>1.0 (± 1.5)</td>
</tr>
<tr>
<td><strong>Mouth-feel</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Astringent</td>
<td>7.8 (± 1.0)</td>
<td>7.7 (± 1.0)</td>
</tr>
<tr>
<td>Viscosity</td>
<td>1.6 (± 0.4)</td>
<td>1.6 (± 0.4)</td>
</tr>
</tbody>
</table>

*, **, and ***: a significance at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively; N.S.: no significance at $P < 0.05$.

Mean intensity ratings with different letters in a row within either coffee products or temperature conditions for each attribute represent a significant difference at $P < 0.05$. 
Table 4. Regression vector (RV) coefficients (their corresponding $p$-values) for measuring the similarities between factor-scores of principal component analyses of the three coffee samples evaluated at four temperature-conditions: 70, 55, 40, and 25 °C

<table>
<thead>
<tr>
<th></th>
<th>70 °C</th>
<th>55 °C</th>
<th>40 °C</th>
<th>25 °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>70 °C</td>
<td>-</td>
<td>0.99 ($&lt; 0.001$)</td>
<td>0.88 (0.17)</td>
<td>0.91 (0.17)</td>
</tr>
<tr>
<td>55 °C</td>
<td>0.99 ($&lt; 0.001$)</td>
<td>-</td>
<td>0.81 (0.33)</td>
<td>0.85 (0.17)</td>
</tr>
<tr>
<td>40 °C</td>
<td>0.88 (0.17)</td>
<td>0.81 (0.33)</td>
<td>-</td>
<td>1.0 ($&lt; .001$)</td>
</tr>
<tr>
<td>25 °C</td>
<td>0.91 (0.17)</td>
<td>0.85 (0.17)</td>
<td>1.0 ($&lt; .001$)</td>
<td>-</td>
</tr>
</tbody>
</table>
Figure 1. Two plots of principal component analysis (PCA) for three coffee samples (Colombia, Ethiopia, and Kenya) evaluated at four temperature-conditions (70, 55, 40, and 25 °C). (A) represents attributes, (B) represents samples. “A” stands for aroma, “F” stands for flavor, “T” stands for taste.
Figure 2. Bi-plots of principal component analysis (PCA) for three coffee samples (Colombia, Ethiopia, and Kenya) evaluated at four temperature-conditions. (A) = 70 °C, (B) = 55 °C, (C) = 40 °C, (D) = 25 °C. “A” stands for aroma, “F” stands for flavor.
Figure 3. A dendrogram drawn by agglomerative hierarchical clustering (AHC) of three coffee samples (Colombia, Ethiopia, and Kenya) evaluated at four temperature-conditions (70, 55, 40, and 25 °C). Based on the dissimilarity of sensory attributes between coffee samples, three clusters were obtained.
Figure 4. Mean time taken to assess samples and temperature lost during descriptive analysis assessment at each target serving temperature. Error bars represent standard deviation.
Figure 5. Comparisons in mean intensity ratings for sensory attributes of coffee samples with different holding-duration. Error bars represent standard error of the means. * represents a significant difference at $P < 0.05$. “A” stands for aroma, “F” stands for flavor, “T” stands for taste, “M” stands for mouthfeel.
Figure 6. A bi-plot of principal component analysis (PCA) of an Ethiopian coffee sample evaluated at four temperature-conditions under two freshness-conditions (“Fresh” = 15 min after brewing, “Old” = 90 min after brewing). “A” stands for aroma, “F” stands for flavor, “T” stands for taste.
Chapter 4.

Effect of serving temperature on sensory perception of black coffee with untrained but frequent consumers with use of the check-all-that-apply method
Abstract

Numerous sensory testing methods have been developed and proposed to describe food and beverage sensory profiles as well as to determine differences between samples. Devoid of much training, the check-all-that-apply (CATA) method merely instructs participants to sample a product and check if they are able to perceive attributes from a provided list. This study aimed to determine the efficacy of the CATA method in describing differences between brewed coffee across multiple coffee varieties as well as describing differences in sensory perception due to modulation of the serving temperature. Three varieties of coffee were selected (Colombia, Ethiopia, Kenya) and served under four temperature-conditions (70, 55, 40, and 25 °C) to 85 consumers for assessment using the check-all-that-apply method. Results show that the CATA method was effective in describing qualitative differences between the coffee varieties while also being able to describe changes in sensory perception due to serving temperature. The findings of this study may be useful for sensory scientists seeking rapid assessment methods and to describe product temperature-dependent sensory variations in hot foods or beverages.
1. Introduction

Temperature fluctuations in foods and beverages occur often in everyday life and likely influence the overall sensory perception of consumers. When one eats a hot meal or enjoys a cold beverage, the foods and beverages tend to equilibrate with ambient temperatures. It is thought that foods and beverages consumed at different temperatures will induce different sensory perceptions. Consider eating ice cream – as it melts, it becomes sweeter. In addition, most people will report that as their beer increases in temperature, it is perceptibly more bitter compared to an ice cold beer. A study conducted by Cardello and Maller (1982) showed that serving temperatures influenced overall acceptability in 13 food and beverage items, with the highest acceptability being at the temperature the product is usually consumed. Is it the temperature itself that leads to the differing perceptions of food/beverages, or is temperature merely modulating individual attributes such as appearance, aroma, taste, and texture which in turn alters the overall acceptability?

While prior studies focusing on temperature effects on sensory perception of foods/beverages typically have used traditional consumer testing with untrained panelists or descriptive analysis with a trained panel that utilizes intensity scaling, this present research seeks to use a simple method void of intensity scaling, but rather asks consumers to indicate if they notice a particular attribute present in the sample.

Check-all-that-apply (CATA) is a method that was first used in marketing research but has been recently adopted and applied to sensory evaluation (Adams et al. 2007). CATA is a rapid method that merely asks consumers to check if a certain attribute or descriptor applies to a sample without any quantification of intensity. Due to the simplicity of the method, it is applicable to untrained panelists and has been used in product optimization (Cowden et al. 2009).
and preference mapping (Dooley et al., 2010; Meyners et al., 2013), with the addition of hedonic ratings.

Potential downsides to CATA have been outlined by Valentin et al. (2012) which involve the type of data produced, a large required number of participants, and optimization of the ballot. Nominal data, which produced by this method, typically has less power than quantitative data which is produced from descriptive analysis, thus a large sample size is necessary. The statistical analysis involved with data from CATA relies on contingency tables. With descriptive analysis five to ten subjects are typically used (Meilgaard et al., 2015), but with CATA a much larger number of subjects are needed. Results from a recent CATA method analysis study involving sample size concluded that 60-80 panelists should be used if there are wide differences between samples (Ares et al., 2014). The CATA ballot itself can also lead to bias. Ares (2013) found that the order of attributes influenced the distribution of responses for certain attributes and conclusions about differences between products. There may also be an issue with the number of descriptors on the ballot. It has been shown that with a fewer number of descriptors available, untrained panelists were able to better and more accurately characterize wine (Hughson and Boakes, 2006). Since coffee is a very complex product leading to many perceived aromas and flavors, a simple ballot may not be effective in capturing differences between products.

The objective of this study was to explore the effects that temperature had on the sensory perception of black coffee on untrained but frequent consumers of black coffee using CATA method and to compare the results to those obtained from the descriptive panel.
2. Materials and Methods

2.1 Participants

Eighty-five healthy volunteers (54 females and 31 males) ranging in age from 20 to 75 years old [mean age ± standard deviation (SD) = 41.9 ± 15.2 years] took part in this study and were recruited from the University of Arkansas Sensory Service Center’s (Fayetteville, AR) consumer database. All participants confirmed that they had no history of any major disease (e.g., diabetes, cancer or renal diseases) and no impairment to smell or taste functions. Participants were screened for being a regular consumer of coffee and reported drinking two to three cups of coffee per day (mean ± SD = 2.4 ± 1.3 cups per day). Participants were also asked how much they liked or disliked coffee in general as well as liking of black coffee a 9-point scale with 1 being dislike extremely and 9 being like extremely. Overall participants liked coffee in general very much (mean ± SD = 7.9 ± 1.3) and liked black coffee slightly (mean ± SD = 6.1 ± 2.1). In addition to the coffee hedonic questions, questions regarding liking of foods/beverages served at high and low temperatures were asked. In general, the participants liked foods served at high temperatures very much (mean ± SD = 7.6 ± 1.3) and liked foods/beverages served at low temperatures slightly (mean ± SD = 5.7 ± 2.1).

2.2 Coffee Samples and Preparation

Three types of locally roasted coffees were purchased at local markets, ground, brewed, and served to participants for testing: Ethiopian (Mama Carmen’s, Fayetteville, AR, USA), Kenyan (Onyx Coffee Lab, Fayetteville, AR, USA), and Colombian (Mountain Bird Coffee Company, Fayetteville, AR, USA). Coffee beans were ground with a coffee grinder (DBM-8, Cuisinart, East Windsor, NJ, USA) to a medium coarseness. 95 grams of coffee were place into a
drip coffee maker (Model DCC-2900, Cuisinart, East Windsor, NJ, USA) along with 1.8 liters of spring water (Mountain Valley Springs Co., LLC Hot Springs, AR, USA). This led to a ratio of approximately 53 grams of ground coffee per liter water, which is within the range of the recommended coffee to water ratio recommended by the Specialty Coffee Association of America (SCAA, 2015). The coffees were then transferred to airpots (Model 36725.0100, Bunn, Springfield, IL, USA) where they were to be dispensed into 4-ounce white Styrofoam disposable cups (Dart, Mason, MI, USA) at certain times to ensure proper temperature levels (70, 55, 40, 25 °C).

2.3 Procedure

This study was composed of 15 sessions that took place at the University of Arkansas Sensory Service Center (Fayetteville, AR). Sessions comprised of 4-10 participants and took approximately 50 minutes to complete. Participants were rewarded with a 15$ Walmart gift card for their contributions. After a brief training session with the CATA ballot and warm up sample, panelists were asked to evaluate each coffee sample using a CATA method for 49 attributes which were grouped into categories of aroma, appearance, taste, and mouthfeel. Panelists were also asked to give their hedonic rating for each sample on a 9-point scale. Panelists were given approximately 2 minutes to evaluate each sample with a minute break between samples. The shortened evaluation time allowed for the coffee to remain near its initial serving temperature as it was being tested. After each sample, panelists were instructed to cleanse their palate with an unsalted cracker and a drink of water. Samples were served in a randomized order in regards to temperature and coffee variety for each session. Due to the unequal number of participants for each session, the order of serving was not balanced and equal.
2.4 Statistical analysis

A total of 85 participants’ data was used. 12 observations (3 coffee types by 4 temperatures) were gathered from each participant resulting in 1020 total observations. Correspondence analysis (CA) and principal coordinate analysis (PCoA) were conducted using XLSTAT (Addinsoft, New York, NY, USA). Attributes selected in correspondence analysis showed a significant difference across treatments for Cochran’s Q test with a significance level of $\alpha = 0.05$. An analysis of variance model on overall liking was created using coffee variety, temperature, and their interaction as main fixed effects along with participant as a random effect. Tukey’s honest significant difference was used to determine differences in overall liking due to treatment effects with a statistically significant difference defined as $\alpha = 0.05$.

3. Results and Discussion
3.1 Overall Liking

Pooling the results of each coffee variety, there was shown to be a clear effect of temperature on liking ($F = 34.9, P < 0.001$). Overall coffee served hot (55 and 70 °C) was liked significantly more than cooler temperatures (25 and 40 °C). These results illustrate the effect that serving temperature can have on overall liking of brewed coffee and corroborate prior research which showed that the ideal consumption temperature for brewed coffee falls within the upper range of the temperatures tested here (Borchgrevink et al., 1999; Lee and O’Mahony, 2002; Brown and Diller, 2008). There was also a significant effect on overall liking due to coffee variety ($F = 31.7, P < 0.001$). These results reveal that temperature alone did not explain all of the variation in overall liking and that coffee variety can contribute in determining how much a sample is liked. There was no significant interaction between temperature and coffee variety on how much consumers liked the coffee samples ($F = 0.71, P = 0.64$). Looking separately at
overall liking for each coffee variety, a fairly linear trend can be seen for each sample (Figure 1). Regardless of coffee type, consumers liked the samples served hot significantly more than those served cold. Using a mean impact display, perceived attributes that contributed to an increase or decrease in liking could be observed (Figure 2). Sweet and roasted attributes contributed to an increase in liking for all coffee types, while burnt, bitter, and sour attributes contributed to a decrease in liking.

3.2 Correspondence analysis of all varieties and temperatures

Cochran’s Q test showed significant difference ($P < 0.05$) for 24 attributes between all samples (8 out of 21 aromas, 3 out of 3 appearance, 13 out of 22 tastes, and 0 out of 3 mouthfeel). From these significantly different attributes, a contingency table was constructed with the sum of frequencies for each sample. Chi-square analysis, which included the significantly different attributes as columns and each coffee and temperature combination as rows, revealed that the null hypothesis of there being no independence between the rows and columns should be rejected (Chi-square = 772.4, $P < 0.001$), indicating that there were detectable differences across all samples.

Correspondence analysis bi-plots were constructed using the 24 significantly different attributes. The amount of variation explained by the first two dimensions was 76.8% (Figure 3). F1 explained 51.4% of the total variation and was mainly composed of fruity aromas and tastes (loaded negatively) and bitter, burnt, and tobacco attributes (loaded positively). F1 mainly described differences between the coffee varieties. F2 explained 25.3% of the variation and was primarily composed of appearance and negative attributes (skunky and pungent tastes). F2 mainly described differences due to the serving temperatures. Ethiopian coffee was generally
rated more sweet and fruity while Kenyan was more sour and pungent. The Colombian variety was experienced as more roasted and burnt. This plot shows that there is a clear grouping of each coffee variety. As the serving temperature of the coffee was decreased, there is an observable decrease in both dimensions (F1 and F2) for each coffee variety that is nearly equal. This result suggests that regardless of coffee variety, there is a definite change in overall sensory perception due to serving temperature. As the serving temperature of the coffees were reduced, they were perceived as less roasted, less transparent, and more sour while also exhibiting more fruitiness. The results of this study corroborate recent research of a specialty coffee, were a descriptive panel found that flavor perception of coffee does indeed change with temperature (Steen et al., 2017). These researchers found that the one particular variety of coffee went from more sour and sweet at low temperatures to more bitter, roasted, and overall more intense at higher temperatures.

An additional correspondence analysis was performed to attempt to explain the change in overall sensory perception of the 3 coffee varieties pooled together. Filtering out attributes that had a significance level of $P > 0.05$, 12 attributes remained. Viewing the bi-plot, the first two dimensions explained approximately 91.7% of the variation between the four serving temperatures [Figure 4(A)]. F1 explained 78.9% of the variation and mainly described differences in sensory perception of the coffees due to serving temperature. F2 explained 12.8% of the variation and mainly described differences between the two extreme serving temperatures and the two moderate temperatures. In general coffee served hot was perceived as being roasted and transparent while cooler coffees were perceived as being more sour, pungent, and oily.
3.3 Principal coordinate analysis

PCoA was performed to identify which attributes were most associated with overall liking (Figure 5). Once again, only attributes that tested as significantly different for Cochran’s Q-test were selected for this analysis. When considering all coffee varieties pooled together, overall liking seemed to be most associated with roasted, chocolate, sweet, and fruity attributes. Looking separately at each coffee variety it can be observed that liking for the Colombian variety was associated with roasted and chocolate taste. For the Kenyan variety both roasted taste and aroma were associated with overall liking. In regards to the Ethiopian variety, nutty aroma and roasted attributes were most associated with overall liking.

3.4 Temperature effect on sensory perception of Colombian coffee

Cochran’s Q test revealed that 9 attributes showed significant difference across temperatures for Colombian coffee; cereal aroma, roasted aroma, oiliness, transparency, burnt taste, chocolate taste, papery/cardboard taste, roasted taste, and astringency. Using CA, approximately 84.7% of the variation due to serving temperature could be explained by the first two dimensions [Figure 4(B)]. An oily appearance was most noticeable at 25 °C and was significantly different from the other temperatures. Reasoning for this would be that as coffee sits for an extended period of time, and subsequently cools down, the interaction between the lipids found in coffee and water are decreased which causes the lipids, which are less dense, to rise to the surface. In regards to transparency, the 70 and 55 °C samples were viewed as more transparent than the two cooler samples. Transparency showed a correlation of -0.476 with oiliness which indicates that as coffee was perceived as oilier, it was also perceived as less transparent. Both roasted aroma and roasted taste were reported most frequently at the highest
temperature and showed a trend of decreasing as the serving temperature decreased as is expected due to there being less overall volatility for aromatic compounds at lower temperatures.

PCoA of overall liking for the Colombian coffee variety across all temperatures using the significantly different attributes determined through Cochran’s Q test was able to explain 50.8% of variation of liking using the first two dimensions (Figure 5). Overall liking was most associated with a roasted and chocolate perceived flavor while also being distant from astringent mouth feeling, burnt taste, and an oily appearance.

3.5 Temperature effect on sensory perception of Kenyan coffee

For the Kenyan variety, 8 attributes showed significant differences across temperatures (bitter aroma, roasted aroma, cloudiness, oiliness, transparency, musty/earthy taste, roasted taste, and skunky taste). CA showed a clear change in perception as the serving temperature was decreased with the first two dimensions explaining 93.4% of the variation [Figure 4(C)]. The first dimensions explained approximately 77% of the variation and mostly described the appearance change of the coffee from transparent at higher temperatures to cloudiness at lower temperatures. There is also a clear strong association of coffee served at the two highest temperatures being perceived as more roasted which is consistent with the other coffee varieties and prior literature (Steen et al., 2017).

PCoA of overall liking for the Kenyan coffee variety across all temperatures using the significantly different attributes determined through Cochran’s Q test was able to explain 47.6% of the variation contributing to liking in the first two dimensions (Figure 5). Overall liking was most associated with roasted aroma and roasted flavor. As expected, derogatory attributes such as skunky taste and cloudiness were negatively associated with liking.
3.6 Temperature effect on sensory perception of Ethiopian coffee

For the Ethiopian variety, 9 attributes showed significant differences across temperatures (berry aroma, green/vegetative aroma, nutty aroma, roasted aroma, cloudiness, oiliness, transparency, roasted taste, and sour taste). CA analysis showed a clear change in perception as serving temperature was decreased and the first two dimensions explained 93% of the overall variation between serving temperatures [Figure 4(D)]. At higher serving temperatures, the Ethiopian coffee was perceived as more nutty, roasted, and transparent while at lower serving it was perceived more frequently with a berry aroma, sour taste, and oily and cloudy appearance.

PCA of overall hedonic liking for the Kenyan coffee variety across all temperatures using the significantly different attributes determined through Cochran’s Q test was able to explain 40% of the variation of overall liking (Figure 5). Increased overall liking was tightly grouped with roasted aroma, nutty aroma, and roasted taste. Perception of sour taste, green/vegetative aroma, and cloudiness are not associated with an ideal product of this variety of coffee.

3.7 Product effect on sensory perception at all temperatures

Overall at all temperatures, the Ethiopian variety was perceived as more fruity smelling and tasting as well as being sweeter. The Kenyan coffee was generally perceived as more sour and pungent regardless of temperature. The Colombian coffee was more strongly associated with bitter, burnt, and roasted attributes compared to the other varieties at all serving temperatures. There appears to be much tighter groupings of attributes for each coffee as the serving temperature is reduced, with the 25 and 40 °C serving temperatures appearing to most definitively separate each coffee variety with little overlapping of attributes (Figure 6). This
finding also indicates that as temperature is reduced, there are greater detectable differences in overall sensory perception between the coffee varieties.

Cochran’s Q test conducted on the three coffee varieties at each serving temperature revealed that 9 attributes were significantly different among the three coffee varieties at 70 °C, 16 attributes were significantly different at 55 °C, 19 attributes were significantly different at 40 °C, and 15 attributes were significantly different at 25 °C (Table 1). This seems to indicate that there is less ability for consumers to differentiate differences between coffee varieties served at a high temperature. Thus, it would be recommended to serve coffee at a reduced temperature (40–55 °C range) in order to best describe differences between coffee samples.

4. Conclusion

Product temperatures influence sensory perception and overall liking of brewed coffee. Regardless of coffee variety, consumers liked the coffees served at the warmer temperatures (70 and 55 °C) compared to the colder temperatures (40 and 25 °C). As the serving temperature of coffee decreased, sensory perception of sour taste and fruity attributes increased, appearance transitioned to more oily, and perception of non-ideal attributes such as pungent and skunky were reported more frequently. The check-all-that-apply method was able to rapidly differentiate between varieties of coffee and describe changes in sensory perception due to temperature. To best characterize sensory attributes of brewed, it is recommended to evaluate coffee samples in a wide range of temperature.
References


Table 1. List of significantly different attributes according to Cochran’s Q test between the three coffee varieties at four temperatures (70°C, 55°C, 40°C, and 25°C)

<table>
<thead>
<tr>
<th></th>
<th>70 °C</th>
<th>55 °C</th>
<th>40 °C</th>
<th>25 °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>*Berry A</td>
<td>*Berry A</td>
<td>*Berry A</td>
<td>*Berry A</td>
<td></td>
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<tr>
<td>*Berry F</td>
<td>*Berry F</td>
<td>*Berry F</td>
<td>*Berry F</td>
<td></td>
</tr>
<tr>
<td>*Fruity A</td>
<td>*Fruity A</td>
<td>*Fruity A</td>
<td>*Fruity A</td>
<td></td>
</tr>
<tr>
<td>*Fruity F</td>
<td>*Fruity F</td>
<td>*Fruity F</td>
<td>*Fruity F</td>
<td></td>
</tr>
<tr>
<td>*Pungent F</td>
<td>*Pungent F</td>
<td>*Pungent F</td>
<td>*Pungent F</td>
<td></td>
</tr>
<tr>
<td>*Sour T</td>
<td>*Sour T</td>
<td>*Sour T</td>
<td>*Sour T</td>
<td></td>
</tr>
<tr>
<td>*Sweet T</td>
<td>*Sweet T</td>
<td>*Sweet T</td>
<td>*Sweet T</td>
<td></td>
</tr>
<tr>
<td>Bitter T</td>
<td>Bitter A</td>
<td>Bitter T</td>
<td>Ashy A</td>
<td></td>
</tr>
<tr>
<td>Burnt F</td>
<td>Burnt A</td>
<td>Brown sugar F</td>
<td>Bitter A</td>
<td></td>
</tr>
<tr>
<td>Metallic F</td>
<td>Burnt A</td>
<td>Bitter T</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cloudy</td>
<td>Burnt F</td>
<td>Cloudy</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cereal F</td>
<td>Chemical F</td>
<td>Metallic F</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chocolate F</td>
<td>Cloudy</td>
<td>Oily</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Oily</td>
<td>Green/Veg F</td>
<td>Sweet A</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sweet A</td>
<td>Roasted F</td>
<td>Transparent</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Transparent</td>
<td>Skunky F</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Sweet A</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Tobacco A</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Transparent</td>
<td></td>
<td></td>
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</tbody>
</table>

“A” stands for aroma, “T” stands for taste, “F” stands for flavor. * indicates a significant difference at all temperatures for an attribute.
Figure 1. Mean overall liking for 3 coffee varieties (Colombian, Kenyan, and Ethiopian) served at 4 temperatures (70, 55, 40, and 25 °C). Mean ratings with different letters are significantly different within each variety ($P < 0.05$). Error bars represent the standard error of the mean. *** represents a significant difference at $P < 0.001$. 
Figure 2. Mean overall liking impact of 3 coffee varieties (Colombian, Kenyan, and Ethiopian) served at 4 temperatures (A = 70, B = 55, C = 40, and D = 25 °C). While bars to the right of the 0 on the x-axis significantly increased liking, bars to the left of the 0 on the x-axis significantly decreased liking ($P < 0.05$).
Figure 3. Correspondence analysis bi-plot of 3 coffee varieties (C = Colombian, K = Kenyan, and E = Ethiopian) at 4 temperatures (70, 55, 40, and 25 °C). Attributes are denoted by a circle while products are denoted with a square. Attributes displayed were determined as significantly different across coffee varieties according to Cochran's Q test with a significance of 0.05. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Figure 4. Correspondence analysis bi-plot of 4 temperatures (70, 55, 40, and 25 °C) for 3 coffee varieties (Colombian, Kenyan, and Ethiopian). Attributes displayed on each plot were determined as significantly different across serving temperatures according to Cochran’s Q test with a significance 0.05. (A) = all varieties pooled together. (B) = Colombian variety. (C) = Kenyan variety. (D) = Ethiopian variety. “A” stands for aroma, “T” stands for taste, “F” stands for flavor.
Figure 5. Principal coordinate analysis of 3 coffee varieties (Colombian, Kenyan, and Ethiopian) and all varieties combined with 4 pooled temperatures (70, 55, 40, and 25 °C). Attributes displayed on each separate plot were determined as significantly different across temperatures according to Cochran’s Q test with a significance of 0.05. (A) = all varieties pooled together. (B) = Colombia variety. (C) = Kenya. (D) = Ethiopia. “A” stands for aroma, “T” stands for taste.
Figure 6. Correspondence analysis bi-plots of 3 coffee varieties (Colombian, Kenyan, and Ethiopian) at each temperature (70, 55, 40, and 25 °C). Attributes displayed on each separate bi-plot were determined as significantly different across coffee varieties according to Cochran’s Q test with a significance of 0.05. (A) corresponds to 70 °C. (B) corresponds to 55 °C. (C) corresponds to 40 °C. (D) corresponds to 25 °C.
Chapter 5.

Effect of serving temperature on sensory perception of enhanced coffee

using check-all-that-apply
Abstract

A large portion of coffee consumers do not drink their coffee black. Generally, consumers will add some sort of creamer, sugar, or both. It is also unlikely that an individual will drink their coffee beverage at one specific temperature but will rather consume it over a range of temperatures. The objective of this study was to determine the effects of additions (cream and/or sugar) on sensory perception of coffee as well as to determine the effect of serving temperature for each serving condition. An Ethiopian coffee variety was selected, brewed, and served to 75 consumers under 4 enhancement levels (black, with cream, with sugar, with cream and sugar) for evaluation using a check-all-that-apply format. Coffees served with enhancements were liked more than those served without enhancements. Temperature was found to have a significant impact on how much each sample was liked overall, but there was no interaction found between enhancements and serving temperature for how much a sample was liked. Serving temperature altered the reported qualities of each enhancement type in a significant manner. The findings suggest that the addition of cream and sugar mask perception of negative attributes which increases liking.
1. Introduction

Many people do not really enjoy their coffee served black and they will add cream, sugar, or other flavors to their hot brewed coffee. A survey conducted earlier this year for screening purposes found that more than 70% of participants (258 out of 364) preferred to drink their coffee with some sort of enhancement. However, there is generally no standard amount of these enhancements added and obviously vary widely from one individual to another. How these enhancements alter the sensory perception of coffee beverages have been studied, but as shown in the previous chapters, the effect of serving temperature on coffee perception is quite significant. Thus, it is pertinent to explore potential effects of serving temperature on coffee that has been enhanced in some form. Recent studies by Green and Nachtigal (2012, 2015) have shown that somatosensory stimuli may modulate flavor and taste perception and that sweetness perception adaptation is increased when given colder solutions. These findings indicate that taste/flavor perception is influenced by temperature and that other somatosensory cues, such as texture, may have an effect. By adding a different textural component (creamer) and a sweetener (sucrose), it stands to reason that overall sensory perception can be modulated. By exploring the effect of temperature on the sensory perception of each coffee beverage, it may be better understood what role temperature has on modulating overall sensory perception for enhanced coffee.

Research by Bücking and Steinhart (2002) which utilized High-Resolution Gas Chromatography/Mass Spectrometry and Gas Chromatography/Olfactometry as well as trained sensory panelists found that a clear shift of odor profiles after the addition of a milk or vegetable product creamer to a brewed coffee. Generally, after addition of a creamer, there was a decreased intensity for “roasted” aroma but an increase in milk-like attributes. Interestingly the sensory
panel could not differentiate between a black coffee beverage and a sucrose enhanced coffee beverage (0.045 g sucrose/ mL coffee), however most attributes showed a lower perceived aroma intensity in the sample enhanced with sucrose. The attributes impacted the most were bitter and sour. Finally, Bücking and Steinhart (2002) concluded that both sucrose and creamer had an impact on flavor release in coffee beverages. Similarly, to many previous researchers, the samples were served at only one particular temperature, 55 °C, with no regard to the effect that serving temperature may have on aroma, taste, and flavor perception.

In a more recent study, Charles et al. (2015) used a single variety of coffee under two roasting levels (light and dark roast) and two levels of sugar concentration (0 and 100 mg/mL) that was evaluated using Temporal Dominance of Sensations (Pineau et al., 2003). Results implied that the addition of sugar suppressed bitterness, sourness, roasted and burnt attributes whilst also improving dominant perception of attributes such as caramel, nutty and overall flavor regardless of the roasting level. Simply by adding sugar to the brewed coffee flavor and smell perception were modulated.

Using descriptive analysis Adhikari (2017) evaluated the impact of additions (milk and sugar) on the sensory properties of hot brewed coffee. Coffee with milk and sugar showed significantly higher intensities for sweetness along with lower intensities for roasted and ashy attributes. Black coffee was rated significantly more intense for bitterness. The addition of sugar was found to have a greater impact than milk in modulating sensory perception. Once again, as with prior studies, this research on enhanced coffee beverages failed to consider the modulating effect of serving temperature.
This present study was designed in order to utilize the rapid check-all-that-apply method to describe the effect of serving temperature on sensory perception of brewed coffee that has had an addition of creamer, sugar, or creamer plus sugar.

2. Materials and Methods

2.1 Amount of Cream/Sugar to be added

An initial study utilizing 40 healthy volunteers (25 females and 15 males) ranging in age from 21 to 72 years old [mean age ± standard deviation (SD) = 42.9 ± 15.0 years] took part in this study and were recruited from the University of Arkansas Sensory Service Center’s (Fayetteville, AR) consumer database. All participants confirmed that they had no history of any major disease (e.g., diabetes, cancer or renal diseases) and no impairment to smell or taste functions. Participants were screened for being a regular consumer of coffee and reported drinking two to three cups of coffee per day (mean ± SD = 2.9 ± 1.4 cups per day). Participants were also asked how much they liked or disliked coffee in general as well as liking of black coffee and enhanced coffee a 9-point scale with 1 being dislike extremely and 9 being like extremely. Overall participants liked coffee in general very much (mean ± SD = 8.4 ± 0.8), liked black coffee slightly to moderately (mean ± SD = 6.9 ± 2.1), and liked enhanced coffee moderately to very much (mean ± SD = 6.9 ± 2.1). These participants were given a black coffee sample (approximately 50 mL) along with pre-weighed containers of creamer (Original Coffee Powdered Creamer, Coffee-mate, Nestle, Glendale, CA, USA) and sucrose (Great Value, Bentonville, AR, USA) and asked to slowly add as much cream and sugar until it reached their desired amount. The remaining amount of sugar and creamer was subtracted from the initial amount to determine the amount used. The average amount of creamer used was 2.3 g ± 1.9 g
which equals 0.046 g per mL. The average amount of sugar used was 2.3 g ± 2.2 g. Based on these results the amount of both cream and sugar to be added to the samples was 40 g per L brewed coffee.

2.2 Participants

Seventy-five healthy volunteers (47 females and 28 males) ranging in age from 20 to 76 years old [mean age ± standard deviation (SD) = 45.2 ± 15.8 years] took part and completed this study. Panelists were recruited from the University of Arkansas Sensory Service Center’s (Fayetteville, AR) consumer database. All participants confirmed that they had no history of any major disease (e.g., diabetes, cancer or renal diseases) and no impairment to smell or taste functions. Participants were screened for being a regular consumer of coffee and reported drinking three to four cups of coffee per day (mean ± SD = 3.4 ± 1.2 cups per day). Participants were also asked how much they liked or disliked coffee in general as well as liking of black coffee a 9-point scale with 1 being dislike extremely and 9 being like extremely. Overall participants liked coffee in general very much (mean ± SD = 8.2 ± 0.8), liked black very much (mean ± SD = 7.1 ± 1.5), and liked enhanced coffee moderately to very much (mean ± SD = 7.8± 1.4). In addition to the coffee hedonic questions, questions regarding liking of foods/beverages served at high and low temperatures were asked. In general, the participants liked foods served at high temperatures very much (mean ± SD = 7.6 ± 1.3) and liked foods/beverages served at low temperatures slightly (mean ± SD = 6.7 ± 1.9).
2.3 Sample Preparation

One Ethiopian variety of coffee (Mama Carmen’s, Fayetteville, AR, USA) was used for this study. Coffee beans were ground with a coffee grinder (DBM-8, Cuisinart, East Windsor, NJ, USA) to a medium coarseness. 95 grams of coffee were place into a drip coffee maker (Model DCC-2900, Cuisinart, East Windsor, NJ, USA) along with 1.8 liters of spring water (Mountain Valley Springs Co., LLC Hot Springs, AR, USA). This led to a ratio of approximately 53 grams of ground coffee per liter water, which is within the range of the recommended coffee to water ratio recommended by the Specialty Coffee Association of America (SCAA, 2015). The coffee was then transferred to airpots (Model 36725.0100, Bunn, Springfield, IL, USA) where it was infused with the enhancements (cream, sugar, or both) in the proper ratio 40 grams per liter. Samples that were to be served at a low temperature were dispensed into glass bottles which were placed into water baths to more rapidly reach the desired temperature. Samples were then poured into 4-ounce white Styrofoam disposable cups (Dart, Mason, MI, USA) and served at the appropriate temperature.

2.4 Procedure

This study was composed of 20 sessions (each panelist attended two sessions separated by one week) that took place at the University of Arkansas Sensory Service Center (Fayetteville, AR). Sessions comprised of 4-10 participants and took approximately 40 minutes to complete. On day one participants received samples under two category conditions (black, with sugar, with cream, with cream and sugar) over four temperatures (70, 55, 40, and 25 °C) in a randomized order by both serving conditions for each session. On the second day participants were given samples from the other two category conditions. After a brief training session with a slightly
modified CATA ballot and warm up sample, panelists were asked to evaluate each coffee sample using a CATA method for 55 attributes which were grouped into categories of aroma, appearance, taste, and mouthfeel. Panelists were also asked to give their hedonic rating for each sample on a 9-point scale. Panelists were given approximately 2 minutes to evaluate each sample with a minute break between samples. After each sample, panelists were instructed to cleanse their palate with an unsalted cracker and a drink of water. Participants were rewarded with a 30$ Walmart gift card for their completion of both days.

2.5 Statistical analysis

Correspondence analysis (CA) was conducted using XLSTAT (Addinsoft, New York, NY, USA). Attributes selected in correspondence analysis showed a significant difference across treatments for Cochran’s Q test with a significance level of $\alpha = 0.1$ for each individual enhancement type and $\alpha = 0.01$ between every sample. An analysis of variance model on overall liking was created using enhancement, temperature, and their interaction as main fixed effects along with participant as a random effect. Tukey’s honest significant difference was used to determine differences in overall liking due to treatment effects with a statistically significant difference defined as $\alpha = 0.05$.

3. Results and Discussion

3.1 Overall Liking

There was shown to be a clear main effect of temperature on liking within each enhancement level; black ($F = 7.42, P < 0.001$), cream ($F = 5.9, P < 0.001$), sugar ($F = 13.9, P < 0.001$), and cream + sugar ($F = 6.2, P < 0.001$). Within every enhancement type, the samples
served at the two warmest temperatures (70 and 55 °C) were liked significantly more than the coldest temperature (25 °C). There was also found to be no interaction effect between enhancement and serving temperature \((F = 0.59, P = 0.80)\) indicating that overall liking is not impacted differently by serving temperature depending on which temperature it is served at. It can, however, be observed that in the cream + sugar samples there was maximum liking in the samples served at 55 °C (Figure 1), which is in agreement with Lee and O’Mahony (2002) who found that consumers preferred drinking coffee enhanced with cream and/or sweetener around 60 °C.

When viewing effects due to enhancement type, significant differences were seen between cream + sugar and the other categories \((F = 94.78, P < 0.001)\). As shown in Figure 1, coffees served with cream + sugar were liked significantly more than coffee served black and coffee served with only cream or sugar. Likewise, coffee served black was liked significantly less than coffee served with cream or sugar.

### 3.2 Correspondence analysis of all enhancements and temperatures

Using Correspondence Analysis, the effects of both enhancements and temperature could be viewed using a bi-plot. Cochran’s Q test showed significant differences \((P < 0.01)\) between enhancements and temperature for 43 of the 55 tested attributes. As shown in Figure 2, the first two dimensions explained approximately 87% of the variation between the samples. F1 explained 61.4% of the total variation and was mainly composed of dairy, cream, and appearance attributes. F1 mainly described differences between samples containing cream or no cream. F2 explained 26.0% of the variation and was primarily composed of sweet attributes. F2 mainly described differences due to samples having sugar added or not. There was no clear trend of
temperature shown in Figure 2 which indicates that differences between serving temperatures are less significant than differences due to the addition of enhancements in brewed coffee. Based on the amount of variation explained in F1 compared to F2 it would appear that cream alters the overall sensory experience of coffee more than sugar. However, based on the distance of the cream + sugar cluster compared to the black coffee cluster it is evident that both the addition of cream and sugar significantly impact perception of brewed coffee more than either cream or sugar alone which confirms findings by Adhikari (2017). In general, samples served black were most associated with astringency, sour taste, bitter taste, and negative attributes such as chemical, pungent, metallic, and ashy flavors. Samples served with sugar added were more associated with sweetness and fruity flavors. This may be due to individuals associating sweetness with fruits or that the sugar may have increased perception of the fruitiness that was inherently present in the Ethiopian coffee. Samples served with cream were perceived as having a lighter color and possessing dairy attributes. Samples with cream and sugar added were perceived more frequently as having a chocolate flavor, which makes sense since chocolate is typically associated with both cream and sugar.

Since much of the variation explained was due to appearance. A separate correspondence analysis was run which only examined differences between samples in regards to aroma, taste, and flavor. Cochran’s Q test showed significant differences ($P < 0.01$) for 33 out of 45 attributes. Illustrated in Figure 3, 86% of the variation is still explained in the first two dimensions, but now the variation is more evenly explained over the first two dimensions (55% and 31% compared to 61% and 26%). The first dimension primarily describes the differences between enhanced coffee and black coffee. The second dimension primarily describes differences in sensory perception between coffee served with cream versus served with sugar. Once again, no clear trends of
serving temperature can be seen. Overall, associations of each enhancement type and attributes remained relatively unchanged. The major difference between Figures 2 and 3 lies in the difference in sensory perception when comparing coffee served black to coffee served with either cream or sugar. Based on the proximity of both cream or sugar enhanced samples to the samples served black, it can be inferred that adding sugar only and creamer only modulated aroma, taste, and flavor of this particular brewed coffee relatively equally. By combining creamer and sugar, the overall sensory experience of a cup of coffee is massively changed.

3.3 Temperature effect on sensory perception of brewed black coffee

Among the four tested temperatures, 10 attributes showed significant differences between the brewed black coffee samples of Ethiopian variety for Cochran’s Q test: cloudiness, color (light and dark), burnt, cocoa, and tobacco aromas, earthy and roasted flavors, sour taste, and mouth coating. Figure 4 shows the correspondence analysis bi-plot that was constructed using these 10 attributes. 87.5% of the variation in these attributes could be explained using two dimensions (Figure 4). The first dimension explains 55.7% of the variation and appears to describe changes in sensory perception due to serving temperature with the sample served at 25 °C being perceived as cloudier, having a lighter color, and being more sour. These findings are consistent with the results from the previous two chapters which also found an increase in sourness perception, increase in cloudiness, and change in color with the Ethiopian coffee served black at low temperatures. The second dimension explains 32% of the variation and describes differences from the between the hottest served sample (70 °C) and the others with the 70 °C being perceived more frequently as having a burnt aroma.
3.4 Temperature effect on sensory perception of brewed coffee enhanced with cream

Cochran’s Q test revealed that 14 attributes showed significant difference across temperatures for the brewed coffee samples that were served with an addition of creamer: bitter, brown sugar, burnt, chemical, earthy, and roasted aromas, cloudy and oily appearance, earthy, nutty, roasted taste, as well as mouth coating and thickness. Using CA on these samples and 14 attributes, approximately 83.7% of the variation was explained using the first two dimensions (Figure 5). The first dimension (64.7%) described differences between the sample served at 25 °C and the samples served at warmer temperatures (40, 55, and 70 °C). Surprisingly, panelists more frequently reported that the 25 °C samples were thin which contradicts the aforementioned findings that samples are typically perceived as more viscous as they cool (Kahkonen et al., 1995). The second dimension mainly described differences between the three warmer temperatures (40, 55, and 70 °C) with 70 °C being more associated with bitter and roasted attributes. No significant change in reported sourness frequency was observed which may indicate that cream inhibits sour taste perception in coffee. Adhikar (2017) also reported a decrease in sourness in samples that had milk or milk + sugar added.

3.5 Temperature effect on sensory perception of brewed coffee enhanced with sugar

When viewing the differences between the brewed coffee samples served with sugar added, 12 attributes showed significant difference for Cochran’s Q test: berry, bitter, green, and pungent aromas, oily and transparent appearance, brown sugar and skunky flavor, sour and sweet taste, as well as thickness. The CA bi-plot constructed using these 4 samples and 12 attributes is presented in Figure 6 and shows that the first two dimensions explain 91.7% of the variation in the data. The first dimension explains 64% of the variation and mainly describes differences
between the four serving temperatures with the warmer samples (55 and 70 °C) being perceived more frequently for a bitter aroma and sweet taste. This increase in sweet taste perception at warmer temperatures has been previously discussed and shows consistency with previous research (Bartoshuk et al., 1982; Calvino, 1986). Once again it can be observed that the colder sample is associated with thin-ness and sour taste, as well as skunky flavor.

3.6 Temperature effect on sensory perception of brewed coffee enhanced with cream and sugar

Cochran’s Q test revealed that 12 attributes showed significant difference across temperatures for the brewed Ethiopian coffee that had an addition of both creamer and sugar: bitter, dairy, earthy, pungent, roasted, and sweet aromas, cloudiness, color and oily appearance, tobacco flavor, and thickness. Overall, 89.2% of the variation in these attributes and samples were explained by the first two dimensions using CA. Once again, the primary source of variation was mainly described due to serving temperature which is illustrated in Figure 7 by the first dimension which explained 64% of the variation. The three samples served at the warmer temperatures were more associated with roasted and earthy aromas while the coldest sample was more associated with sweet, dairy aromas and a thin mouthfeel. Previous literature shows that both sucrose and creamy solutions should decrease in viscosity as temperatures increase (Swindells et al., 1958), but the participants reported the opposite in that the warmer samples were actually perceived as more thick. These results could be due to panelists simply not liking the colder samples as much and supplying attributes that they perceive to be more associated with a poor-quality coffee beverage. Typically high quality coffees are known for having a better “body” otherwise known as a thicker or more viscous mouthfeel (Hoffman, 2014). The second
dimension explained 25% of the variation and most described differences from the samples served at 70 °C and the samples served at 55 °C. The samples served at 55 °C with cream and sugar added were most associated with a thick mouthfeel and were liked the most, which strengthens the thought of panelists selecting negative attributes to describe the samples that were not liked as much.

4. Conclusion

To summarize, the addition of cream, sugar, or cream + sugar has an impact on how brewed coffee is perceived in regards to appearance, aroma, taste, flavor, mouthfeel, and overall liking. Samples served with cream and sugar were liked significantly more than samples served with either sugar or cream or served black. Serving temperature was found to have a significant impact on the overall sensory perception of “enhanced” brewed coffee. Regardless of which enhancements were added, temperature affected how much participants liked the samples overall, with the samples served at 70 and 55 °C being liked significantly more than samples served at 25 °C. Perception of potential negative attributes were masked in samples served with enhancements.
References

Adhikari, J. (2017). Impact of consumption temperature and additions (milk and/or sugar) on sensory properties of hot brewed coffee.


Figure 1. Mean overall liking of brewed coffee of Ethiopian variety for 4 enhancement levels (Black, Cream, Sugar, and Cream + Sugar) served at 4 temperatures (70, 55, 40, and 25 °C). Error bars represent the standard error of the mean. Post-hoc testing performed with Tukey’s HSD. Different lowercase letters indicate a significant difference in means within each enhancement type. Different capital letters indicate a significant difference in means between enhancement types.
Figure 2. Correspondence analysis bi-plot of brewed Ethiopian coffee served under four enhancement levels (B = Black, S = Sugar, C = Cream, and C + S = Cream and Sugar) at four temperatures (70, 55, 40, and 25 °C). Attributes displayed were determined as significantly different across samples according to Cochran’s Q test with a significance of 0.01. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Figure 3. Correspondence analysis bi-plot of only aroma, flavor, and taste attributes of a brewed Ethiopian coffee served under four enhancement levels (B = Black, S = Sugar, C = Cream, and C + S = Cream and Sugar) at four temperatures (70, 55, 40, and 25 °C). Attributes displayed were determined as significantly different across samples according to Cochran’s Q test with a significance of 0.01. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Figure 4. Correspondence analysis bi-plot of brewed Ethiopian coffee served black at four temperatures (70, 55, 40, and 25 °C). Attributes displayed were determined as significantly different across samples according to Cochran’s Q test with a significance of 0.1. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Figure 5. Correspondence analysis bi-plot of brewed Ethiopian coffee served with creamer at four temperatures (70, 55, 40, and 25 °C). Attributes displayed were determined as significantly different across samples according to Cochran’s Q test with a significance of 0.1. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Figure 6. Correspondence analysis bi-plot of brewed Ethiopian coffee served with sugar at four temperatures (70, 55, 40, and 25 °C). Attributes displayed were determined as significantly different across samples according to Cochran’s Q test with a significance of 0.1. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Figure 7. Correspondence analysis bi-plot of brewed Ethiopian coffee served with cream and sugar at four temperatures (70, 55, 40, and 25 °C). Attributes displayed were determined as significantly different across samples according to Cochran’s Q test with a significance of 0.1. “A”, “F”, and “T” represent aroma, flavor, and taste, respectively.
Chapter 6.
Overall Conclusion
Variations in product temperature have exhibited an effect on how attributes of food and beverages are perceived. This thesis demonstrates that serving temperature modulates the overall sensory experience of a coffee beverage. These findings were shown using trained panelists with descriptive analysis and untrained panelists through the CATA method. One key difference between the two methods that were examined here was that the descriptive panelists took a much longer time to assess the products, leading to potentially confounding results. Untrained panelists using CATA were able to rapidly assess each sample ensuring that the product was tested close to its original serving temperature. Since one limitation of CATA is that only nominal data is produced, researchers cannot determine the intensity differences between samples. Despite this limitation, descriptive analysis was largely ineffective at describing difference between coffee varieties compared to the CATA method.

Based on this research, to best describe differences between coffee beverage samples, it is recommended to serve samples at a temperature in the 40 – 55 °C range. However, if the goal of a researcher is to describe qualities of a particular coffee beverage, he/she should evaluate the product at a range of temperatures. The research presented in this thesis shows the importance of maintaining product temperatures across all samples for every individual during sensory testing. If this is neglected, significant differences regarding sensory perception and overall liking may be lost.

In order to increase consumer appreciation of coffee beverages, coffee roasters and baristas should seek to optimize roasting, brewing, and serving conditions to maintain sensory quality over a wide range of temperatures.
Appendix 1.

Research compliance protocol letters
February 9, 2017

MEMORANDUM

TO: Han-Seok Seo
Tonya Tokar
Ragita Pramudya
Matt Chapko
Sara Jama Arroyo

FROM: Ro Windwalker
IRB Coordinator

RE: New Protocol Approval

IRB Protocol #: 17-01.415

Protocol Title: The Effect of Serving Condition on Sensory Characteristics of Food and Beverages

Review Type: ☑ EXEMPT ☐ EXPEDITED ☐ FULL IRB

Approved Project Period: Start Date: 02/06/2017, Expiration Date: 02/05/2018

Your protocol has been approved by the IRB. Protocols are approved for a maximum period of one year. If you wish to continue the project past the approved project period (see above), you must submit a request, using the form Continuing Review for IRB Approved Projects, prior to the expiration date. This form is available from the IRB Coordinator or on the Research Compliance website (https://vprpred.uark.edu/units/rscp/index.php). As a courtesy, you will be sent a reminder, two months in advance of that date. However, failure to receive a reminder does not negate your obligation to make the request in sufficient time for review and approval. Federal regulations prohibit retroactive approval of continuation. Failure to receive approval to continue the project prior to the expiration date will result in Termination of the protocol approval. The IRB Coordinator can give you guidance on submission times.

This protocol has been approved for 700 participants. If you wish to make any modifications in the approved protocol, including enrolling more than this number, you must seek approval prior to implementing those changes. All modifications should be requested in writing (email is acceptable) and must provide sufficient detail to assess the impact of the change.

If you have questions or need any assistance from the IRB, please contact me at 109 MLKG Building, 5-2208, or irb@uark.edu.