AN ABSTRACT OF THE THESIS OF

Steven James Harper for the degree of Doctor of Philosophy in Food Science and Technology presented on May 13, 1993.

Title: Carbonation Perception: Lexicon Development and Time-Intensity Studies

Abstract approved: ______________________________
Mina R. McDaniel, Major Professor

A lexicon describing the sensory perception of carbonated water was developed. Temporal aspects and differing ingestion conditions were investigated for Bite and Burn sensation using time-intensity (T-I). Four CO_2 levels (0, 1.7, 2.8, and 4.6 volumes) at 3°C and 10°C were tested. Trained panelists used a 16-pt category scale for evaluation in the first study. One swallow (15 ml) and four continuous swallows were evaluated by trained subjects using T-I in the subsequent studies.

Lexicon included: salty, sour, bitter, cooling, astringency, bubbly, bubble size, bubble sound, gas expansion feeling, bite, burn, and numbing. Descriptor ratings, except cooling, increased as CO_2 level increased. Bubble size and bubble sound were rated higher for 10°C. Cooling, bite, burn, and numbing were rated higher for 3°C . Descriptors were divided into cooling, taste (salty, sour, bitter, astringency), trigeminal (bite, burn, and numbing), and mechanoreception descriptors (bubbly, bubble size, bubble sound, gas expansion feeling) based on PCA.

Average temporal curves for Bite and Burn demonstrated that Burn sensation (steep linear rise and long-lived exponential decay slope) was similar to previously investigated irritants while Bite (steep linear rise and decay slopes, and relatively short duration) was unlike other irritants. Sensations were qualitatively and quantitatively different. Intensity and duration of Bite and Burn were concentration dependent. Cold temperature enhanced perception. Possible psychological habituation or desensitization was observed. Most T-I parameters were correlated for both Bite and Burn. These included CO_2 level dependent and CO_2 level independent parameters. Considerable subject variability was found.

Increased exposure to CO_2 solution and increased cooling with ingestion of four continuous swallows was compared to one swallow. T-I curves for Bite (four swallows) were of higher intensity, longer duration, and developed maximum intensity plateaus. Those for Burn exhibited higher maximum intensities. At four swallows, T-I parameter correlations were strengthened, subject variability reduced and replication reproducibility improved by ease of rating afforded subjects by higher intensity sensations. Increased oral CO_2 perception with higher CO_2 levels and enhancement by cold temperature was reconfirmed. Beginnings of maximum intensity, Duration, and reaction time perceptual terminal thresholds were seen for the highest 3°C, CO_2 level. High CO_2 concentration, cold temperature, and exposure time induced these effects.

Carbonation Perception: Lexicon Development and Time-Intensity Studies

by

Steven James Harper

A THESIS

Submitted to

Oregon State University

in partial fulfillment of

the requirement for

the degree of

Doctor of Philosophy

Completed May 13, 1993

Commencement June 1993

APPROVED:

Professor of Food Science and Technology in charge of major

Head of Department of Food Science and Technology

Dean of Graduate School

Date thesis is presented: May 13, 1993

Typed by the author: Steven James Harper

ACKNOWLEDGMENT

I am grateful to my mother, Jessie Harper, for instilling in me the appreciation of the worth of a good liberal arts education. I am also appreciative of the encouragement and respect given by my father, James Harper, as I journey through my life. Their belief in me has allowed me to pursue my dreams.

Dr. Mina McDaniel has been my mentor and friend through the grueling five years I have spent as a graduate student in the Food Science Department. I thank her for her encouragement, support, and willingness to allow me flexibility in exploring the field of Food Science and Sensory Evaluation.

I am grateful to my friends, Lynne, Cida, Nancy, and John for their friendship, support and all of the encouragement they have given me during my travails. I appreciate the friendship and support received from all of the Sensory Evaluation group, and especially appreciate the advise and input from Cindy and Sonia.

I thank all of my committee members, Dr. Joe McGuire, Dr. Dan Farkas, Dr. Harry Nakaue, and Dr. Dave Thomas for their guidance through the thesis project.

I thank all of the panelists who served on my panels, enduring the pain and bubbles. I would not have been able to finish without their fine evaluations. I thank the Department of Food Science and Technology faculty and staff for giving me the opportunity to study at Oregon State University and for supporting me in my various projects. In particular, Floyd Bodyfelt has been extremely helpful with ideas, editing, and inspiration.

Of course, none of this would have been possible without the incredible love given me by my life partner, Sue Parrott. She has accompanied me on my journeys through life, has been extremely flexible and has supported me in difficult times. She has also shared in the positive experiences resulting from our adventures. This thesis is a part of her.

TABLE OF CONTENTS

Page

Page

Page

LIST OF FIGURES

Figure	Description	Page

LIST OF TABLES

CARBONATION PERCEPTION: LEXICON DEVELOPMENT AND TIME-INTENSITY STUDIES

INTRODUCTION

Carbonated beverages have become exceedingly popular with all age groups, especially among young people. The food and beverage industry routinely develops flavor formulas in order to produce a desired flavor or mouthfeel sensation. Therefore, it is quite important for the soft drink and mineral water industries to understand how flavor and mouthfeel are affected by altering the levels of CO_2 added to a given beverage, since carbonation influences these properties. Until recently, very little was known about carbonation perception. Examination of carbonation perception had been limited to relatively few studies investigating primarily the acceptability or preference of products with added CO_2.

Szczesniak (1979), surveying descriptive mouthfeel terminology for beverages, found that many carbonation related terms were suggested, with bubbly, tingly, prickly, and stinging being mentioned most often. However, more intensive research was needed as development of a complete descriptive lexicon for carbonated beverages, which includes flavor, sound, and factors associated with the trigeminal nerve system, had not been reported.

There are two primary consumption practices in the United States for carbonated beverages: The first practice is to drink the beverage at refrigerated temperature. The second is to pour the beverage over ice. It is important to understand the effect of temperatures within this range on the perceived CO_2 level of the beverages as well as understanding the effect of temperature on the sensory characteristics of the beverage.

Recently, many researchers have taken an interest in the psychophysical properties of oral CO_2 perception. Interaction between oral pungency evoked by CO_2 and basic tastants was explored by Cometto-Muñiz et al. (1987), and gender differences in perception of pungency were also examined (Cometto-Muñiz and Noriega, 1985). They found power function exponents of 1.1 for for buccal pungency. In studies by Yau and McDaniel, power functions (Yau and McDaniel,1990) and temperature effects (Yau and McDaniel, 1991) were investigated. Power function exponents for overall oral carbonation perception were determined (2.79 for swallowing and 2.65 for expectoration) and found to be more similar to electric shock than for basic tastants. They also found carbonation intensity was perceived to be higher at lower temperatures - the temperature effect being carbonation level dependent. Carbonation perception was defined as the overall perception of carbonation including stinging, burning, cooling, irritation, etc.; the feelings associated with the stimulation of free nerve endings of the trigeminal nerve. Green (1992) also found carbonation perception to be strongly temperature dependent, but concluded that enhancement experienced at cold temperatures resulted predominantly from an increase in the pungent (burning and stinging) components arising primarily from the stimulation of the nociceptive system.

Burn and Bite perception were focused on in our studies as previous studies of CO_2 and other irritants (Yau and McDaniel, 1990; Lawless, 1984) recognized these as predominant sensations for oral chemical irritation. Qualitatively, Bite and Burn represented two very different sensations.

Time-intensity (T-I) studies were first conducted by Neilson (1957). Since then, many T-I studies have been conducted focusing on basic tastes. However, no T-I studies were found with CO_2 as a stimulant. There have been a few studies which investigated temporal properties of irritants. Reaction times to ethanol application to the tongue were examined (Green, 1988) and irritation decay durations for four other irritants were determined (Lawless, 1984). It is of interest to understand the time-course of carbonation perception in order to understand its relationship with other irritants.

The goal of our research was to explore new areas of carbonation perception using sensory methodology as our tool. We were also interested in continuing the investigation into CO_2 concentration and cold interaction, as well as understanding more about the effect on carbonation perception of different ingestion conditions. Three separate studies were conducted to meet the goals of our research. The objectives of these studies included:

Study 1

Determination of the descriptors for carbonated water as developed by a trained panel.

Development of precise definitions for these descriptors and validation of the use of these descriptors.

Investigation into how the use of descriptors differ for different carbonation levels and temperatures.

Investigation of the effect of temperature and CO_2 on the intensity ratings of the descriptors.

Determination of the relationship between descriptors as temperature and CO_2 level changed.

Study 2

Determination of the time-intensity profiles for Bite and Burn perception in carbonated water.

Comparison of the perceptual relationship of Bite and Burn to each other and to other irritants which have been investigated.

Investigation of the relationship between the time-intensity parameters, the subject variability, and the CO_2-temperature relationship for each of these sensations.

Study 3

Investigation of how ingestion conditions (four swallows vs one swallow) change ratings for time-intensity parameters, average curves, CO_2 level effects, and temperature effects.

LITERATURE REVIEW

Carbonation

Reaction Mechanisms of Carbon Dioxide Dissolution

When gaseous carbon dioxide is injected into water, it is dissolved and becomes aqueous carbon dioxide. Through the chemical reaction between water and carbon dioxide, the hydration of carbon dioxide occurs. There are three major reactions of carbon dioxide in aqueous solution (Daniels et al., 1985):

(1) CO_2 (dissolved) + H_2O ------------> H_2CO_3

(2) H_2CO_3 ------------> HCO_3^- + H^+

(3) HCO_3^- -------------> CO_3^{-2} + H^+

The hydration of carbon dioxide to form carbonic acid proceeds very slowly. The value of the equilibrium constant [$K_{hydration}$ (298°K, 1 atm) = 2.6 X 10^{-3}] for the hydration indicates that the percentage of carbonic acid is very small. It is estimated at approximately 2.0% (Daniels et al., 1985). While the first reaction occurs very slowly, the carbonic acid dissociates very quickly forming bicarbonate and hydrogen ions. The third reaction also becomes a factor at very high pH values.

The reactions are dependent on the pH of the solution. At pH < 6 the primary species is CO_2, at pH 7-10 it is HCO_3^- , and at pH > 10 it is CO_3^{-2}. The primary reaction pathways at pH < 8 are:

CO_2 + H_2O -------> H_2CO_3

H_2CO_3 + OH^- ---------> HCO_3^- + H_2O

The predominant reaction pathways at pH > 10 are:

$$CO_2 + OH^- \text{------>} HCO_3^-$$

$$HCO_3^- + OH^- \text{------>} CO_3^{-2} + H_2O$$

In the range of pH 8-10 all reaction pathways are important.

The carbonated water in our studies had a pH of 3.5-3.7. Therefore we are primarily concerned with carbon dioxide in solution. However, carbon dioxide in solution is also in equilibrium with the undissolved carbon dioxide in the gaseous phase. This equilibrium is dependent on temperature and pressure.

Solubility of Carbon Dioxide

The volume of carbon dioxide in a beverage is important; it is this quantity that governs the perception of the carbonation sensation and the time of effervescence.

Theoretically, Henry's law states that the amount of a gas dissolved by a given volume of solvent at constant temperature is directly proportional to the pressure of the gas with which it is in equilibrium. Thus, solubility is dependent on temperature and partial pressure above the solution; solubility is reduced as pressure decreases or temperature increases. Carbon dioxide is unusual in that the influence of temperature on its solubility is greater than that of other gases (Zumdahl, 1986). Henry's law is obeyed very closely for carbon dioxide in aqueous systems at moderate temperatures and low pressure. At atmospheric pressure, the amount of carbon dioxide dissolved by an aqueous systems depends solely on temperature.

Inorganic compounds and compounds such as citric acid, glycerol and sucrose decrease the solubility of carbon dioxide (Jacobs, 1959). For example, a 10% reduction in the solubility of carbon dioxide is seen in a solution containing 12% sucrose.

In the beverage industry the most common measurement system for solubility of carbon dioxide in liquid is expressed as "volume". This is defined as "the amount of gas in ml that a given volume of water will absorb at 760 mm and 15.6°C" (Jacobs, 1959). This measurement is derived from the Bunsen absorption coefficient, which is defined as "the volume of gas reduced to standard conditions which at the temperature of the experiment is dissolved by one volume of solvent, the partial pressure of the gas being 760 mm" (Quinn and Jones, 1939). Gas volume test charts are often used which show the volumes at various temperatures and gage pressures. These charts are calculated based on the Boyle-Mariotte law for isothermal compression (Jacobs, 1959).

The evolution of carbon dioxide gas from liquids is of special importance to the beverage industry because of the change in psychophysical perception which occurs from this loss. A number of studies of stability of carbon dioxide gas in solution have been carried out. Findlay and King (1913) found that the rate of de-saturation was proportional to the concentration of gas in solution. In un-agitated solutions they found that the escape of gas on reduction of pressure to atmospheric was followed by a period when no gas was evolved. This situation was maintained only if the liquid remained undisturbed. After a certain time period, evolution of gas was again seen, followed by another stable period. As the concentration of carbon dioxide was reduced, these periodic evolution events occurred more infrequently. During the periods of stability noted above, any slight mechanical shock was sufficient to cause evolution.

Weyh (1975) experimented with a number of beers and water and determined that when bottle closures were carefully removed a loss of only 10% of the CO_2 occurred over a period of six hours in pale beers. He also found that 60-90% of the carbon dioxide remained after 72 hours at room temperature and that pouring reduced CO_2 content from 15-30% for various beers.

The presence of discharge nuclei, excess air in the liquid, unnecessary agitation, and improper high temperature storage all can result in rapid loss of carbonation (Jacobs, 1959). Rough, irregular-shaped particles, heavy metals, scratches on the interior bottle walls, oil droplets, and microorganisms can all accelerate lost of carbon dioxide from solution (Phillips and Woodruff, 1981).

Carbonation Procedures

Carbonation is the process of saturating a liquid with carbon dioxide. The equilibrium pressure for the gas-liquid system varies with temperature and the amounts of gas and liquid present. The time required to reach equilibrium depends on a number of factors, including temperature, pressure and relative surface area of the liquid exposed to carbon dioxide. Since liquids absorb more carbon dioxide at lower temperatures and higher pressures, carbonation systems employ low temperature, high pressure, and agitation or high surface area exposures to carbon dioxide to facilitate efficient carbonation.

Small scale pilot carbonators (Zahm and Nagel, Inc., Buffalo, NY) must be cooled and agitated in order to facilitate hydration of CO_2. Large scale commercial carbonators such as the Cem Saturator Model C (Crown Cork and Seal Co., Philadelphia, PA) use carbon dioxide pressurized containers in which the cold liquid flows in sheets down through the carbonator. The liquid becomes saturated as it flows through the carbonator.

Testing of Carbonation Level

The accurate testing of the CO_2 level for finished products is important for ensuring that proper carbon dioxide levels are present to develop the sensory qualities necessary for a desirable product.

Zahm and Nagel Co. developed a method to test carbonation which is widely used in the beverage industry. A Zahm piercing device (Zahm and Nagel Co., 1964) is used which measures pressure of the headspace as well as temperature of the product. The carbon dioxide content in the system can then be calculated.

This testing method can be biased by the amount of air in the system. When measuring bottles with an extremely small amount of head space, this error is inconsequential as practically all of the air is present in the headspace. However, there are correction methods available when an investigator suspects that a significant amount of air is in the system. Phillips and Woodruff (1981) suggested two ways of avoiding the error. First, "sniffing" can be employed. "Sniffing" is a procedure in which headspace gas is released from the container before measurements are taken. The Zahm piercing device is provided with a special valve used for "sniffing". A second method involves releasing gas into a burette filled with sodium hydroxide solution. This solution absorbs all of the CO_2, leaving only air in the burette. Repeated shaking and releasing is carried out, the air content of the burette is measured, and the total pressure value corrected.

Time consuming manometric methods have also been used as well as enzymatic and titration measurements and more recently ion analyzers. Morrison (1962) used carbonic anhydrase to catalyze the hydration of CO_2 and to provide a determination of carbon dioxide. A titrimetric method combining enzymatic and volumetric methods was developed by Caputi et al. (1970). They determined the CO_2 initially in the system using carbonic anhydrase. They titrated between pH 4 and pH 8.6 to find the amount of hydrogen ion used.

Ion analyzers have also been used in which CO_2 in the system is converted to CO_3^{-2} by sodium hydroxide and measured (Green, 1992).

Carbonation Effect on Sensory Properties

There have been relatively few studies on the effect of carbonation on sensory properties.

A number of studies have been conducted on dairy products due to interest in developing carbonated dairy products in order to increase sales of fluid milk. Lederer et al. (1991) carbonated fruit flavored milks at low levels. They found that carbonation suppressed cooked milk aroma and flavor and that sourness, astringency, chalkiness and bitterness were enhanced at the higher levels of carbonation. They used CO_2 volumes of .60, .74, and 1.42. In a study of blueberry flavored milk, overall flavor intensity, sweetness, and blueberry flavor were enhanced by carbonation (Yau et al., 1989). Carbonated yogurt was investigated by Choi and Kosikowski (1985). In this study, sweetened plain and strawberry flavored carbonated yogurt beverages were developed. A consumer acceptance test showed that 90% of respondents liked the product, while 5% disliked it. Hydrolyzed whey concentrate used as a replacement for the sucrose in the yogurt reduced acceptance.

Juice products have been another area in which carbonation seems to have potential for increasing sales and have therefore been studied. The effect of carbonating at different levels and of adjusting titratable acidity levels on acceptability of clarified pineapple juice was investigated by Baranowski and Park (1984). They found that few significant differences were found between the samples for preference, but that preference was highest for the low carbonation product (2.5 vol.). King et al. (1988) investigated carbonated muscadine grape juice. These researchers found that the carbonated juices were lighter in color (apparently caused by bleaching) and were preferred equally to non-carbonated juices. A shelf life study of carbonated soft drinks was conducted by McBride and Richardson (1983). They found evidence of deterioration with storage over the two year time period of the study, but found that all drinks were still considered satisfactory at the conclusion of the trial. A storage study using different storage temperatures and storage times was conducted on carbonated apple juice (Bright and Potter, 1979). They found that overall scores for non-carbonated and carbonated samples decreased with increased temperature and storage time but that carbonation did not affect acceptability. Response Surface methodology was used by McLellan et al. (1984) to model the sensory response to carbonated apple juice. A model including linear and quadratic effects was derived for sweetness, sourness, mouthfeel and carbonation level. Acceptability of the juice was highest for the range of typically used soluble solids and carbonation levels.

Finally, the mouthfeel of beer was evaluated; sting, bubble size, foam volume, and total CO_2 were determined to be important in describing the mouthfeel of 30 commercial beers (Langstaff et al, 1991).

Chemesthesis

Background

Taste and smell are the two primary modes of perception that come to mind when people think about food sensation. However, there is a third mode, mediated predominantly by chemosensitive fibers other than those responding to taste and smell, which has been historically referred to as the "common chemical sense" and alludes primarily to stimulation of the trigeminal system (Parker, 1912). The primary function of this sense was thought to be detection of noxious stimuli which were potentially harmful. Although there is a broad class of chemicals which can cause irritation at high concentrations, our interest lies primarily in the chemical perception of irritants (e.g. capsaicin, ethanol, piperine, CO_2, ginger oleoresin, vanillyl noneamide, etc.) relevant to food systems. This sense is now being referred to as "chemesthesis" and refers to the existence of a "chemical sensibility" rather than a specific anatomical system devoted to the detection of chemicals (Green, 1991b; Green and Lawless, 1991). The reason being, that investigators now believe that chemical sensitivity is mediated by not only what is considered to be the trigeminal nerve system, but also by chemosensitive elements of the nociceptive and thermal senses (nonspecific polymodal somatosensory fibers which are also common to cutaneous tissue).

Two interesting aspects of chemesthesis is the psychophysical power functions compared to gustatory stimulus and the loss of aversive reaction to irritants which many persons experience.

Psychophysical power functions have been found which seem compatible with its role as a warning system. Cometto-Muniz and Noriega (1985) found power functions for CO_2 oral pungency of 1.1 while power functions of greater than 2.65 were found for overall carbonation perception by Yau and McDaniel (1990). These are much greater than typical gustatory power functions which are normally <1.

Loss of aversive response seems like an unusual reaction to irritants which may be noxious in high concentrations and which are perceived by this system. However, many persons learn to like and even crave the sensations they derive from irritants such as capsaicin. No definitive explanations have been established. However, possible explanations include enhancement of salivation, desensitization, pharmacologic influences, and psychologic factors, including thrill seeking and social pressure (Rozin, 1990).

Innervation

Innervations by the trigeminal, glossopharyngeal, vagus, and chorda tympani have been found to be involved in chemesthesis. Four classes of receptors participate in this innervation: (a) mechanoreceptors (responding to mechanical or pressure stimuli); (b) thermoreceptors (responding to cold or warm stimuli); (c) nociceptors (responding to painful stimuli); and (d) propioceptors (responding to muscle or joint position). In addition chemically sensitive trigeminal free nerve endings are also thought to exist and constitute a part of this system (Silver and Finger, 1991). Two major afferents have been described which are principally responsible for conduction of impulses to the central nervous system. They are the quick-responding myelineated A-delta receptors and the slower-responding unmyelineated c-fiber receptors (Dubner and Bennett, 1983).

The trigeminal nerve (V cranial) is responsible for much of the sensory innervation of the mucous membranes of the oral cavity. The mandibular and maxillary branches of the trigeminal nerve innervate the oral cavity. Nerve branches found in the oral cavity include the lingual, which innervates the tongue and floor of the mouth; the nasopalatine nerve, which innervates portions of the hard palate; the posterior palatine nerve, which innervates the the soft palate; and the buccal nerve, which innervates the cheek mucosa. These fibers account for approx 75% of the innervation of the fungiform papillae (Farbeman and Hellekant, 1978). In the posterior portions of the mouth, the glossopharyngeal nerve carries the trigeminal nerves, as well as the gustatory fibers. A variety of other free nerve endings are also found throughout the oral mucous membrane and are especially numerous around the papillae, especially the circumvallate papillae (Finger, 1986).

The vagus nerve (X cranial) innervates the epithelium of the esophageal and oropharyngeal regions and is important to oronasal perception of irritants.

Receptors innervated by the chorda tympani (VII cranial) nerve (primarily responsible for gustatory responses) have also been found to be receptive to thermal, mechanical and chemical stimulation (Oakley, 1985).

Sensation Qualities

Chemical stimulation of receptors elicits a number of interesting physiological responses which are protective in nature. Many of the responses help to remove the source of irritation. They include salivation, local and distant vasodilation (flushing of the face, chest, and shoulders and a reddening of the conjuctiva), increased nasal secretion, tearing, and profuse sweating in the head and neck region (Lawless, 1984).

Stinging, pricking, tingling, biting, piercing, burning, itching, pungent, painful, warm, hot, and numbing, are all descriptors which have been used to describe the irritative sensations in various studies (Harper and McDaniel, 1993; Green, 1992; Cometto-Muniz et al., 1987; Stevens and Lawless, 1987).

It has been observed that 'sharper' sensations such as stinging/pricking are mediated by rapidly-conducting A-delta nociceptors, while 'duller' sensations such as burning are mediated by slower-conducting c-fiber nociceptors (Price and McHaffie, 1988; Torebjork et al., 1984).

Temporal Properties

Because chemesthesis is a system whose primary function was thought to be detection of potentially harmful stimuli one would think that the acuity of the system and the speed with which it reacts would be very rapid. In fact, it appears that stimulation is responded to rather sluggishly compared to the gustatory system and that the response is quite persistent.

Latency periods of about 5 sec. were determined for CO_2 oral pungency (Cometto-Muniz et al., 1987) while 5.9 sec. latency periods were found for application of 35% ethanol solution on the tip of the tongue (Green, 1988). However, Green also found that reaction times decreased to 1.5 sec. when 85% ethanol was used, but these times increased greatly as the application was moved toward the throat. Even 1.5 sec. is much slower than reaction times for the gustatory system. The slowness of the response seems to be due to the depth of the nerve endings, their relative scarcity, and the fact that the nerve endings are not in direct contact with saliva as the taste buds are. Only near the tongue tip and within the taste papillae are there larger concentrations (Finger, 1986).

Once the irritation is perceived the sensation increases rapidly in a manner similar to gustatory sensations. However, the decay curve for sensations is much longer than most gustatory stimulants. Lawless (1984) found that perception of a number of oral irritants (vanillyl noneamide, capsaicin, piperine and ginger oleoresin) lasted for approximately ten minutes or longer depending on the concentration.

Decay curves over time may be somewhat linear or exponential. Stevens and Lawless (1986) found that decay curves for capsaicin and piperine were different. The decay curves for capsaicin were exponential and the curves for piperine were linear. These differences are no doubt affected by the molecular properties of the each stimulus and probably is related to their degree of lipophilicity.

Taste Interactions

Irritants are seldom ingested by themselves, but instead are almost always part of a food system which includes components responsible for tastes. The interaction of taste and irritants are an integral part of our perception of food. A number of studies have been conducted investigating this interaction.

In two studies of rats, Silver et al. (1985) and Berridge (1985) discovered that trigeminal responses enhanced palatability. The first study found that trigeminal responses, but not the taste components, were desensitized when rats were injected with capsaicin, and that the rats lost part of their interest in eating. The second study discovered that when the trigeminal nerves were severed the food became less palatable for the rats.

Lawless and Stevens (1984) found that there was a diminution of taste perception after stimulation with capsaicin or piperine rinses (especially for sour and bitter tastes) They felt that the mechanism responsible for this effect was taste nerves carrying irritation information at the expense of gustatory signals. In a follow-up study investigating likers and dis-likers of chili sensation, Lawless et al. (1985) found that oral capsaicin partially masked gustatory and olfactory sensations but did not interfere with flavor identification. This effect tended to be greater in persons disliking the sensation of capsaicin. Irritation from piperine was found to be attenuated by a citric acid solution and that for capsaicin was attenuated by a sucrose solution (Stevens and Lawless 1986).

Cowart (1987) found that capsaicin did not reduce the intensity of taste stimuli when delivered in a mixed system; the earlier findings by Lawless and colleagues were attributed to the unusual presentation procedure which they used.

Two studies have investigated the interaction of tastants and CO_2. Cometto-Muniz et al. (1987) found that CO_2 interaction effects depended on the particular tastant evaluated. Sucrose sweetness and CO_2 pungency were found to have no mutual effect; sodium chloride saltiness or tartaric acid sourness and CO_2 pungency showed mutual enhancement; and quinine sulfate bitterness abated CO_2 pungency. They suggested that saltiness and sourness are qualitatively closer to oral pungency.

Yau and McDaniel (1991b) found that higher carbonation levels reduced sweetness ratings of aspartame sweetened samples, but had no effect on sucrose sweetened samples except at the 3.0 vol. CO_2 level. Carbonation was found to enhance sourness levels of citric acid and phosphoric acid at lower acid levels but not at the 3.0 vol. CO_2 level.

Temperature Interactions

The temperature of foods and beverages we ingest are rarely at the temperature of the oral cavity. Because they are usually much cooler or warmer, it is important to understand the interaction of irritants and temperature. The studies on temperature-irritant interactions have found enhancement and suppression of irritation depending on the irritant studied.

One of the first studies of temperature-irritant interaction was a study by Szolcsanyi (1977). He reported that the burning sensation of capsaicin applied to the skin could be completely eliminated by prior cooling of the skin to 28°C. This study was followed by a number of studies also indicating that irritant intensity ratings can be reduced by cooling. Sizer and Harris (1985), when testing recognition thresholds for capsaicin, found that pungency threshold was higher for 2°C samples compared to 18°C samples. Lower perceived intensity ratings were attributed to oral cooling in a number of other studies (Stevens and Lawless, 1986; Green, 1986; Green, 1990a). These investigators tested capsaicin, piperine, ethanol, sodium chloride and found that cooling significantly reduced irritation. The strength of the effect was different for each irritant (Green, 1990a). Green (1986) also found that the perception of coolness was suppressed and the perception of heat was enhanced by capsaicin solutions.

Menthol and CO_2 interact with temperature in a slightly different manner. Green (1985) discovered that the way in which menthol affected oral thermal sensations was dependent on time. When solutions were held in the mouth for 5 sec., perceptions of coolness and warmth were enhanced for water solutions below and above oral temperature. However, longer exposure to menthol attenuated the warmth for solutions above oral cavity temperature.

Two studies on oral CO_2 perception have indicated that oral irritation was increased by decreasing the temperature of the solutions. The first, by Yau and McDaniel (1991), found that carbonation intensity was perceived to be higher at lower temperatures, differences being more evident at the higher carbonation levels. This study used four temperature levels (3, 10, 16, and 22°C) and two CO_2 levels (2.4 vol. and 3.0 vol.). Green (1992) also found that cooling carbonated solutions increased ratings of oral irritation. He also found that increased CO_2 concentration increased perception of cold.

These results are consistent with three other studies which looked at different nerve systems affecting chemesthesis. Mechanoreceptor tissue from cat tongues has been shown to respond to cold with increased amplitude of response when the tissue was cooled (Henzel and Zotterman, 1951); Bryant et al. (1991) found that trigeminal receptor units in rats that were sensitive to CO_2 were also sensitive to cold stimuli; and, Oakley (1985) found that the chorda tympani nerve was sensitive to cooling.

Mechanical and Tactile Interactions

Studies of irritants now reveal that mechanical and tactile effects on the sensation of irritation, although not as important as chemical and temperature effects, definitely play a role. These studies again point to the complexity of irritation perception which involve a number of nerve systems.

Two studies (Stevens, 1982; Stevens and Hooper, 1982) investigated the effect of cooling on tactile acuity of the skin. These may not seem to be related to oral perception, however, the same nociceptor receptors are found in the skin and the oral mucosal tissue. In the first study, investigators found that tactile acuity of the skin was improved for cooling conditions relative to thermally neutral conditions. In the second study, cold objects were placed on the skin and the sensation compared to thermally neutral objects. They found that the cold objects felt heavier than the thermally neutral objects, the colder the object the heavier it felt.

Oakley (1985) discovered that the chorda tympani nerve was sensitive to mechanical stimulation, finding that when the tongue was stroked, large tactile responses were produced.

An investigation of actual tactile-irritant interaction by Green (1990a) revealed that mechanical stimulation associated with sipping and expectoration transiently suppressed the burning sensation of capsaicin irritation. He also found that pressure applied to the lingual surface heightened the ability of ethanol to stimulate nociceptors. In turn, ethanol seemed to increase the sensitivity of nociceptors to static pressure.

Chemical Sensitization

Perceptual sensitization is one of the more interesting aspects of irritation. Unlike most gustatory sensations which tend to adapt with increasing exposure, irritation has been shown to be enhanced in certain situations. Stevens and Lawless (1987) reported that when capsaicin or piperine stimuli were presented in rapid succession, the second stimulus produced a stronger sensation of irritation than the first. This effect was amplified when the second stimulus was different than the first. Green and Gelhard (1989) investigating sodium chloride irritation found that this enhancement could be extended over many stimuli and for several minutes. In a follow-up study, Green (1990a) discovered that ethanol did not exhibit this pattern, however, there was a cross-sensitizing effect on ethanol sensitivity; the intensity increasing significantly after exposure to sodium chloride.

Chemical Desensitization

Desensitization, the opposite of sensitization, is a loss of sensitivity brought about by repeated, usually intense chemical irritant stimulation. The presumed mechanism for desensitization is the depletion of Substance P (or other neuropeptides) from unmyelineated primary afferent neurons, including polymodal nociceptors. The key to obtaining desensitization (especially with lower concentration) is the insertion of a hiatus in stimulation between the desensitizing stimuli and the test stimuli. If the hiatus is too short desensitization fails to occur and sensitization may even develop (Green.

1991b). A number of studies have been done to support the notion of desensitization.

Nagy (1982) found that chronic capsaicin administration desensitized adult rats to chemically induced pain and raised thresholds for noxious stimuli. Silver et al., (1985) also found that in rats, capsaicin injections suppressed trigeminal responses. However, they did not find that it suppressed taste perception.

In humans, Lawless (1984) found that preceding a stimulus with a stronger irritant led to a short-term desensitizing effect for that stimulus. Karrer and Bartoshuk (1991a) also found that desensitization occurred on the tongue when capsaicin applications (10-1000 ppm) were used daily and in series. The desensitization lasted from one to six days depending on concentration. In another study (Karrer and Bartoshuk, 1991b) found that piperine, ginger oleoresin, ethanol, and CO_2 seltzer water all had decreased intensities after application of a single dose of capsaicin (10 ppm). However, all irritants showed complete recovery one day after testing with the exception of CO_2. Finally, Green (1991b) found that the intensity of sensations for burning and stinging were greatly reduced by repeated capsaicin exposure, but that warmth and numbness were not reduced.

Sensory Methods

Time-Intensity Literature Review

Most sensory procedures routinely used today in the sensory laboratory involve a subject providing a single response rating to an inquiry such as, "What is the intensity of sourness in this sample?", or "How well do you like this sample?" In essence, this requires the subject to "time average" their response in order to arrive at a single intensity value. This may work well in many cases, but when rating aroma, taste, texture, and sensations which dynamically change over time or which linger, it is important to use a procedure which captures the entire perception. Good examples of this include breakdown of gels upon chewing, ingestion of foods containing fat carrying flavors which change as the fat melts in the mouth, and chemesthetic responses to irritants such as CO_2 or capsaicin. These phenomena occur over time periods as short as 5-60 sec. or as long as ten minutes.

Information Provided by Time-Intensity Curves

Figure 1.1 displays a typical time-intensity curve for a single introduction of stimulus into the mouth (e.g. one swallow of carbonated water). The graph is normally marked off in horizontal units of time and vertical units of perception intensity. The time may be continuous, as in the case of a computerized recording system continuously recording time, or may be marked off in time segments as would be the case when a subject was rating a perception every 10 sec. The axis often is marked off in units of 0-100 corresponding to a scale of no intensity-extreme intensity which the subject continuously rates. A category scale of numerical intervals, or an intensity response recorded as magnitude estimation is also used and units of measure would correspond to these recording techniques.

"Zero time" is usually recorded at the time of initial sample exposure, which may be placement of the sample in the mouth, expectoration, or swallowing of the sample. There is usually a "lag time" which is then

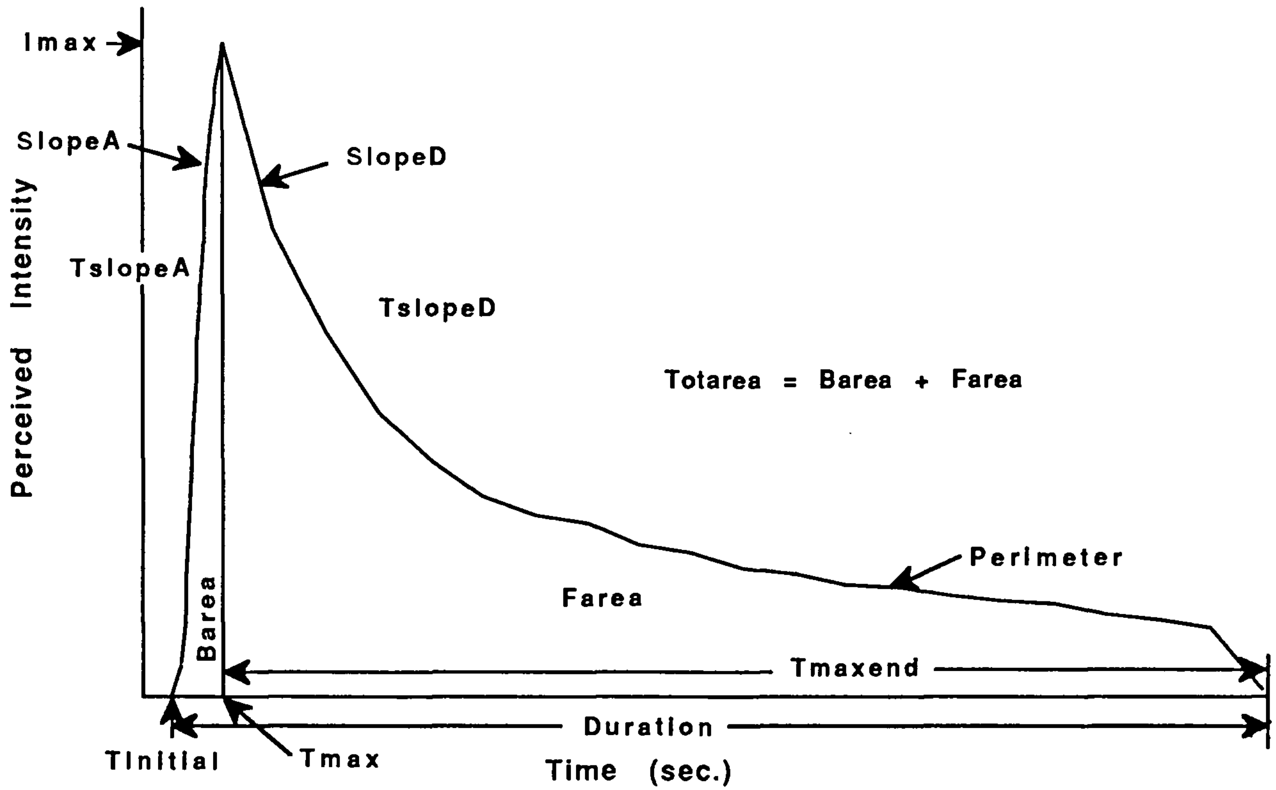


Fig. 1.1. Time-intensity parameters commonly used in studies tracking the time-course of sensations.

observed which is the time between the initial exposure and initial perception; this is referred to as Tinitial in this study. Once the response is observed the curve increases in intensity. The maximum rate of this ascending increase (rise) is called SlopeA and the time of this maximum rate onset is referred to as TslopeA. A maximum intensity is then reached (Imax) and depending on the stimulus being evaluated, a plateau of sustained maximum intensity may also be present. The time from the initial response (Tinitial) to the maximum intensity is the time to maximum intensity (Tmax). In evaluations where a plateau is present, the time when the intensity begins to decrease is noted (Tdec) and the time between Tmax and Tdec is used for evaluation (Tplateau). As the intensity drops, the maximum descending slope (decay) referred to as SlopeD and the time for SlopeD (TslopeD) are recorded. Finally the point of extinction is reached (Tend).

Other parameters which can be extracted from the time-intensity curve include: the total time of the response from Tinitial to the end of the response (Duration); the duration of time from maximum intensity to the end of the response (Tmaxend); the total area under the curve (Totarea); the area under the curve before the maximum intensity (Barea); the area under the curve after the maximum intensity (Farea); the ratio of the area before and after the maximum intensity (Ratio); and, the total perimeter of the time-intensity curve (Perimeter).

Lee and Pangborn (1986) discussed these parameters and divided them into five general categories. These categories, along with associated parameters from our studies include: (1) time-related parameters (Tinitial, Tmax, TslopeA, TslopeD, Tmaxend); (2) rate-related parameters (SlopeA, SlopeD); (3) intensity-related parameters (Imax); (4) events occurring during oral perturbations; and (5) parameters related to duration of the stimulation (Duration, Totarea, Perimeter, Barea, Farea, Ratio).

History of Time-Intensity Measurements

The first time-intensity studies were conducted and recorded using stopwatches to record the time between the introduction of the sample and the points of interest. Thus the subject may have indicated the time of initial perception, the time of maximum intensity and the time of extinction. This provided valuable information but only allowed for evaluation of approximately four points and would provide limited information for evaluation among samples.

Neilson (1957) was the first investigator to continuously record time-intensity responses allowing for the production of a time-intensity curve. In this study, subjects marked their perceived intensity on a piece of chart paper while they watched a clock. Data were gathered at a predetermined frequency. Jellinek (1964) used a ballot on which judges (while watching a clock) marked a rating from a category scale next to times which were listed on the page. The time-intensity curve was constructed from the times and intensities generated from the evaluation. Other early studies used audible cues to indicate time for evaluation (McNulty and Moskowitz, 1974). Again, time-intensity curves were constructed after each evaluation.

Larson-Powers and Pangborn (1978) utilized a moving strip chart recorder to generate continuous time-intensity curves. In this method, a chart recorder was set to advance the chart paper at a known speed. Subjects initiated the recorder with a foot pedal and moved a pen horizontally along an intensity scale indicated on a stationary paper cutter bar which overlapped the chart paper. A cardboard barrier was placed over the chart paper to prevent the subjects from viewing the evolving curve. This produced a nice time-intensity curve, but required manual evaluation of the curves before averaging or statistical analysis could be applied

An external dial potentiometer with an intensity scale indicated along its span, and linked to a chart recorder (SMURF) was used for collecting time-intensity data by Birch and Munton (1981). Schmitt et al. (1984) introduced a mechanical digitizer capable of reading individual points from chart paper curves and feeding them directly into a computer for statistical analyses.

Fairly complete computerized systems have now been introduced (Takagi and Asakura, 1984; Guinard et al., 1985; Lee, 1985). These systems utilize microprocessors to give instructions to subjects, collect data, and analyze data for programmed parameters (e.g. average curves, maximum intensity, duration, etc.). Sliding linear potentiometers with marked intensity scales, and "game paddles" moving an "X" along an intensity scale appearing on a monitor screen are the most common methods of collecting the assessments. Instructions given to the subjects via computer terminals include times to perform actions such as delivery of the sample or times for expectoration. Guinard et al. (1985) noted that "computerized time-intensity procedures gave the experimenter better control of the test conditions and reduced labor and potential human errors associated with hand-conversion of time-intensity curves into numbers for statistical analysis. The joystick facilitated highly personalized communication between the judge and the computer and precluded the conceptualization of the curve being generated by moving chart recorders."

Applications of Time-Intensity Techniques

A large portion of the studies utilizing time-intensity have been directed toward understanding the gustatory stimulants of astringency, sourness and especially bitterness and sweetness.

Time-intensity techniques have been used extensively to investigate psychophysical aspects of nutritive and non-nutritive sweeteners. These began with Birch et al. (1980) investigating evidence for a queue hypothesis in taste chemoreception and have continued to the present (Birch et al., 1982; Dubois and Lee, 1983; Naim et al., 1986; Shamil et al., 1988; Noble et al., 1991; Ott et al., 1991). Time-intensity has also been used for investigative work on model sweetener systems. These have focused on interactions with other sweeteners or interactions with fruit systems (Harrison and Bernhard, 1984; Cliff and Noble, 1990; Matysiak and Noble, 1991)

Bitterness systems have been investigated using time-intensity. (Lewis et al., 1980; Pangborn et al., 1983; Leach and Noble, 1986; Guinard et al., 1986a and 1986b; etc.).

Very few investigators have studied astringency and acids. Lee and Lawless (1991) investigated the time-course of astringent mouth characteristics for a variety of common astringents using discrete time points, rather than continuous tracking of one attribute. Straub (1992) used time-intensity to investigate the sourness and astringency in a number of common food grade acids.

Rheological studies on viscosity, firmness and melting rates of a number of gels and desserts such as ice cream and chocolate pudding have benefited greatly from the use of time-intensity techniques (Pangborn and Koyasako, 1980; Moore and Shoemaker, 1981; Muñoz et al., 1986).

Time-intensity techniques have been utilized to determine temporal responses to chemical irritants. The time-course of sensation for capsiacin, piperine, ginger and vanillyl nonamide was first investigated by Lawless (1984) and Lawless and Stevens (1986). Nasrawi (1988) also utilized time-intensity for part of her research on oral and salivary responses to capsicum. A number of other investigators have used time-intensity to determine reaction times and persistence times for irritants such as ethanol (Green, 1988).

Two unusual applications of time-intensity technique are studies by Guinard et al. (1986a; 1986b) and Taylor and Pangborn (1990). In the first study, the effects of repeated ingestion on the patterns of taste intensity for bitterness was determined for beer samples containing bitter-tasting iso-alpha-acids. In the second study chocolate milk samples with varying fat levels were evaluated by naive judges and their ratings compared to single point hedonic scaling recorded at various intervals. They found that time-intensity hedonic responses to all, but the nonfat milk sample (in a manner similar to intensity responses), varied over a 20 sec. period. The maximum and minimum time-intensity measurements also correlated significantly with results from conventional hedonic line scaling.

Variability Among Subjects' Time-Intensity Curves

Many investigators have found that there is a large degree of variability in time-intensity responses by subjects (Harrison and Bernhard, 1984; Schmitt et al, 1984; Ott et al., 1991; Guinard et al., 1986a), especially for irritants (Stevens and Lawless, 1986). Van Buuren (1992) noted that time-intensity curves appear to be determined by characteristics that are related to the subject and not the product being tested - "each curve bearing the signature of its creator." Individual differences in anatomy, oral manipulation, use of the intensity scale, and criteria for determining extinction of sensation all play a part in the variability (Noble et al., 1991) and produce large standard deviations (Swartz, 1980).

In order to address this problem of large variability and to produce time-intensity results which are more meaningful, a number of investigators have developed techniques for averaging the curves over subjects. Simple averaging of intensity ratings at common times for each individual curve to produce an overall average curve has been used by a some investigators (Leach and Noble, 1986). This method encountered the problem of multiple peaks being produced or the tail of the mean curve being dominated by the subject with the longest extinction time. Overbosch et al. (1986) developed an averaging process which addressed the problems of simple averaging. They used geometric mean normalization of the intensity ratings as well as the time ratings of all the subjects for the ascending and descending portions of the curve to produce an average overall curve. Liu and MacFie (1990) further refined this method to accommodate intensity plateaus, non-zero endpoints and non-monotonic curves, all problems left unsolved by Overbosch et al. (1986). The development of methodology for producing overall average curves which provide equal weightings for all subjects should be very beneficial to time-intensity investigations. The use of the more recent normalization procedures has proven valuable in our studies.

Descriptive Sensory Analysis

Descriptive sensory analysis is a sensory method by which the attributes of a food or product are identified and quantified using human subjects who have been specifically trained. Analysis can include all parameters of the product, but are often limited to aspects of interest (e.g. aroma, taste, appearance, aftertaste, and texture). This method is appropriate when detailed information is desired of the individual characteristics of the product. It is used for monitoring product quality, documenting sensory characteristics, correlating instrumental and chemical measurements with sensory responses, and for research guidance.

This method requires that the panel members be carefully screened, trained, and maintained by a sensory professional. Panel members must be capable of perceiving and recognizing individual sensory characteristics of a product, verbalizing their perceptions, developing a set of terms to describe the product being evaluated, rating their intensities, and reaching an agreement with fellow panel members. A collective response is required in order to statistically analyze attributes.

Generally, sessions are conducted with the panel members in which: the product is evaluated; descriptive terms are suggested by the panelists; the descriptive terms are defined; panelists agree on the terms and the definitions; physical or chemical references are suggested to help panelists perceive the attribute; and, the attributes are rated on some sort of intensity scale.

The Flavor Profile, Texture Profile, Quantitative Descriptive Analysis (QDA), Spectrum, and Free Choice Profiling methods are the most widely used descriptive analysis methods. Summaries of each method are included below. The descriptive analysis method used for investigating carbonated water perception was a hybrid of the QDA and Spectrum methods along with special procedures specific to evaluation of carbonated water.

Flavor Profile Method

The Flavor Profile method was developed by Arthur D. Little, Inc. in the late 1940's (Cairncross and Sjostrom, 1950). It is based on the concept that flavor consists of identifiable taste, odor, and chemical feeling factors plus an underlying set of sensory impressions not separately identifiable. The description of the aroma, flavor and aftertaste of a product is developed by identifying the separate characteristics contributing to the overall impression and determining their intensity. The sensory analysis includes: 1) Overall impression (amplitude). 2) Identification of perceptible aroma and flavor character notes. 3) Intensity of each character note. 4) Order in which the character notes are perceived. 5) Aftertaste.

Four to six screened and trained panelists first examine and then discuss the product of interest in an open session. A consensus agreement is reached by the panel, the product are rated on a 5-pt or 7-pt scale, and the panel leader summarizes the results in report form.

The key element in the Flavor Profile method is the panel leader and the ability of the panel to work as a team in order to reach consensus decisions. The leader is responsible for directing the conversation and providing a consensus conclusion for the test. Statistical analysis is not typically used in this method. Instead, an interpretation of the composite profile terms and intensities are provided.

Texture Profile Method

Texture Profile method was developed at the General Foods Research Center to define the textural parameters of foods (Brandt et al., 1963; Szczesniak, 1963; Szczesniak et al, 1963). Brandt and co-workers (1963) defined the texture profile as "the sensory analysis of the texture complex of a food in terms of its mechanical, geometrical, fat and moisture characteristics, the degree of each present and the order in which they appear from first bite through complete mastication." The objective of the method was to eliminate problems of subject variability, allow direct comparison of results with known material, and provide a relationship with instrumental measures (Szczesniak et al., 1963). Standard rating scales and specific reference material to represent each scale category were developed for terms which were believed to encompass rheological sensations.

Panelists, screened and selected for training, are exposed to a wide range of products from the category under investigation, and introduced to the underlying textural principles evident in the products under study. The original Texture Profile method used the 5-pt scale from the Flavor Profile method for evaluating an attribute's intensity. This was expanded to a 13-pt scale and more recently, category, line and magnitude estimation scales have been used (Meilgaard et al., 1987). Statistical analysis is conducted when line or category scales are used, otherwise actual consensus values from the panel as a whole are determined.

Concerns have been expressed regarding the methodological assigned terms and inflexible rating system employed in this method, which attempts to train inherently variant subjects to be invariant (Stone and Sidel, 1985).

Quantitative Descriptive Analysis (QDA) Method

The QDA method was developed by Stone and Sidel (Stone et al., 1974). It was developed in response to dissatisfaction among sensory analysts with the lack of statistical treatment of data obtained with the Flavor Profile method and related methods. It relies heavily on statistical analysis determination of appropriate terms, procedures, and panelists to be used for analysis of a specific product (Meilgaard, 1987). The 10 to 12 panelists are screened and selected according to their ability to discriminate differences in sensory properties among samples of the product type for which they are being trained. The panel leader acts as a facilitator, and refrains from influencing the panel. Development of consistent terminology is stressed, but the panelists are free to score as they deem appropriate using a 15 cm line scale. Panelists evaluate products one at a time in sensory booths. The results are analyzed using analysis of variance and a report of the results is written. Often "spider web" visual displays are presented comparing the attributes for each product.

Lack of leadership leading to erroneous terms, "free" approach leading to inconsistency of results, and lack of feedback to panelists on a regular basis reducing the opportunity for expanding their terminology and discrimination capabilities are criticisms which have been directed at this method (Meilgaard, 1987).

Spectrum Descriptive Analysis Method

The Spectrum descriptive analysis method was developed in the mid 1980's by Gail Vance Civille. The panel, meeting as a group, are trained on principles of whichever perceptual modalities are necessary (e.g. aroma, texture) for evaluation of the product of interest. Emphasize is placed on the qualitative and quantitative aspects of the product. Panelists are pre-screened for availability, health, product attitudes, sensory awareness and rating ability using basic taste tests, scaling tests, and modality acuity tests. The panelists are trained with the main objectives being to expose the panel to the underlying dimensions of the characteristics in the product and to provide a similar frame of reference in terminology and scaling among all participants (Muñoz and Civille, 1992). The development of attribute comparison references for development of common terminology, and anchoring intensity references for the scale being used for testing are important aspects of the Spectrum method, as they greatly reduce panelist variability. Proficient use of these references is the key to the success of determining the character profile of a product and differences between products. A 15 cm line scale is typically used and the data analysis normally uses split-plot analysis of variance to investigate panelists, the product, and the panelists by product interaction. Other multivariate data analysis techniques are also used when deemed appropriate. Final evaluation of the products is conducted in controlled environment sensory booths.

Free Choice Profile Method

The free choice profile procedure was developed relatively recently in order to compensate for panelist variability which is inevitable when working with human subjects. These include perception of different stimuli in the same product, use of different terms and scales between sessions, differing ranges of scoring between panelists, using descriptors in different ways, and panelists variance in scoring (Arnold and Williams, 1986).

This method allows panelists to choose their own vocabularies, eliminating the need for training in descriptor use. Free Choice Profile also uses a unique statistical method which is called Generalized Procrustes Analysis (GPA). The use of GPA compensates for much of the variation normally found among panelists in descriptive panels. This is done by producing a perceptual space for each panelist, and is matched with the other panelists. Three steps are required for this: first, centroids of each panelist's data are matched to eliminate the effects of usage of different parts of the scale; second, isotropic scale changes remove the variability in scoring ranges; third, the configurations are matched as closely as possible by rotation and reflection of the axes (Arnold and Williams, 1986). A consensus configuration, which is normally simplified and plotted by principal components analysis, is then calculated as the average configuration from all panelists.

One of the primary advantages of the Free Choice Profile method is that untrained panelists can be used, as well as groups of panelists who do speak the same language. Consistency by panelists is still important as well as the ability to perceive attributes and to generate sufficient terms.

CARBONATED WATER LEXICON; TEMPERATURE AND CO_2 LEVEL INFLUENCE ON DESCRIPTIVE RATINGS

Steven J. Harper and Mina R. McDaniel

Department of Food Science and Technology

Sensory Science Laboratory

Oregon State University

Corvallis, OR 97331

Oregon State University Agricultural Experiment Station

Technical Paper No. 10,016

Abstract

A lexicon for describing the sensory perception of carbonated water was developed by a trained panel. The lexicon included: salty, sour, bitter, cooling, astringency, bubbly, bubble size, bubble sound, gas expansion feeling, bite, burn, and numbing. Four CO_2 levels (non-carbonated, and 1.69, 2.75, and 4.63 volumes) and two temperature levels (3°C and 10°C) were tested. Ratings of all descriptors, except cooling, increased significantly as CO_2 level increased. Bubble size and bubble sound were rated significantly higher for 10°C samples while cooling, bite, burn, and numbing were rated higher for 3°C samples. The descriptors could be divided into four groups (cooling, taste descriptors, trigeminal descriptors, and mechanoreception descriptors) which were rated similarly based on principal component analysis.

Introduction

Carbonated beverages have become exceedingly popular with all age groups, especially among young people. In addition to the popularity of soda pop, other carbonated beverages, especially carbonated mineral water, are regularly consumed. Most of the carbonated drinks consumed are flavored, but some, such as mineral waters, are non-flavored. The food and beverage industry routinely develops flavor formulas in order to produce a desired flavor or mouthfeel sensation. It is quite important for the soft drink and mineral water industries to understand how flavor and mouthfeel are affected by altering the levels of CO_2 added to a given beverage, since carbonation influences these properties.

There are two primary consumption practices in the United States for carbonated beverages: The first practice is to drink the beverage at refrigerated temperature. The second is to pour the beverage over ice. Hence, it is important to understand the effect of temperatures within this range on the perceived CO_2 level of the beverages as well as understanding the effect of temperature on the sensory characteristics of the beverage.

Yau and McDaniel (1990) investigated the effect of increasing levels of CO_2 in water on the increase in carbonation perception. In their study, carbonation perception was defined as the overall perception of carbonation including stinging, burning, cooling, irritation, etc.; the feelings associated with the stimulation of nerve endings of the trigeminal nerve. They found power functions of 2.79 for swallowing and 2.65 for expectorating carbonated water; these functions represented a sharp increase of perceived carbonation with increasing CO_2 concentration.

Yau and McDaniel (1990) also looked at the effect of temperatures on the perception of overall carbonation intensity. They observed that carbonation intensity was perceived, by both naive and trained panels, to be greater at lower temperatures than at higher temperatures. This temperature effect was carbonation level dependent, wherein temperature differences were more evident at higher carbonation levels (Yau and McDaniel, 1991).

There have also been a few studies that investigated the effect of added CO_2 on specific attributes such as sourness and sweetness. Lederer et al. (1991) found that sourness, bitterness, and astringency were enhanced by high levels of CO_2 in flavored, carbonated milk beverages, while the sensory rating of sweetness was enhanced by carbonation of a blueberry flavored, carbonated milk beverage (Yau et al., 1989).

Development of a complete descriptive lexicon for carbonated beverages which includes flavor, sound, and factors associated with the trigeminal nerve system has not been reported. The major focus of this research was the determination of the descriptors for carbonated water as developed by the trained panel, development of precise definitions for these descriptors, and validating the use of these descriptors. In addition this study also investigated how the use of descriptors differ for different carbonation levels and temperatures, what the effect of temperature and CO_2 was on the intensity ratings of the descriptors, and what the relationship between descriptors was as temperature and CO_2 level were changed.

The information obtained from this inquiry will be valuable for understanding the descriptive profile of carbonated mineral water and should prove helpful in predicting the effect of carbonation on flavored beverages.

Materials and Methods

Samples

Commercial, bottled water (Aqua Cool, Portland, OR) was used to produce the samples. The source of the commercial, bottled water was chlorinated Cascade Mountains water. Aqua Cool filtered the water using a series of multi-media filters to remove large particles, followed by filtration through activated carbon and polymeric resins to remove color, odors, and possible chemical contaminates and chlorine. The water was then disinfected using ozone. According to Aqua Cool, the samples contained an average of 1.5 ppm calcium, .31 ppm magnesium, .7 ppm sodium and 1.0 ppm chloride.

Batches of the water were carbonated with commercial carbon dioxide (Industrial Welding, Albany, OR) in a Zahm and Nagel 18.9 liter, stainless steel carbonator (Zahm and Nagel Co., Buffalo, NY). Four carbonation levels were produced [non-carbonated, 1.69 volume (vol.) (SD=.05), 2.75 vol. (SD=.058), and 4.63 vol. (SD=.064)]. The 1.69 vol. CO_2 samples were produced using water, bottles, and CO_2 maintained at 21^{o}C. The 2.75 and 4.63 vol. CO_2 samples were produced using 21^{o}C CO_2 and 1^{o}C water, with bottles and carbonator immersed in ice. Batches of 18.9 liters for each level were produced. All samples were bottled in 828 ml, green, glass bottles and stored at 1^{o}C until evaluated.

The sample pH was 6.04 for non-carbonated, 3.71 for 1.69 vol. CO_2, 3.65 for 2.75 vol. CO_2, and 3.51 for 4.63 vol. CO_2. The pH was measured at 22^{o}C with a Corning 125 pH meter using a Sensorex epoxy-body, sealed-reference combination electrode (S200C). The pH meter was calibrated with buffers of pH 3 and 7 (Microessential Laboratory, Brooklyn, NY). Concentrations of chemical species in the carbonated water which are pertinent to the addition of CO_2 are listed in Appendix Table 5.1.

Carbonation levels were measured using a Zahm and Nagel piercing device (Zahm and Nagel Co., Buffalo, NY). Sample temperatures and headspace pressures were measured after agitation of bottles. Temperature and pressure readings were converted to "volumes CO_2 per volume water" by using a conversion table (Zahm and Nagel Co., 1964). One volume is defined as the amount of CO_2 dissolved in water at equilibrium, at 15.56°C, and at one atmosphere pressure.

Trained Panel

Training. Eight panelists (7 females and 1 male), all students or faculty of the Department of Food Science and Technology at Oregon State University, participated in training and subsequent evaluation of the carbonated samples. Panelists were consumers of carbonated beverages.

Descriptors and definitions were developed over 18 one-hour training sessions. Reference standards were used in training for taste descriptors, astringency and chalkiness (Table 2.1).

Testing. Testing was conducted in the Sensory Science Laboratory at Oregon State University, Corvallis, OR, in individual booths. Four carbonation levels (non-carbonated, and 1.69 vol., 2.75 vol., and 4.63 vol.) were tested at two temperature levels (3°C and 10°C). The 3°C samples were kept cold by packing them in ice before and during the test. The 10°C samples were tempered by placing them in a styrofoam cooler filled with

Table 2.1. Reference standards for carbonation descriptors.

Descriptors	wt/wt[a]	Source
Salty	.2%	NaCl (Morton International, Inc., Chicago, IL)
Sweet	1%	sucrose (C & H Sugar Col, Concord, CA)
Sour	.03%	citric acid (Haarman and Reimer Co., Elkhart, IN)
Bitter	.04%	caffeine (Fisher Scientific, Fairlawn, NJ)
Astringency	.08%	alum (McCormack, Baltimore, MD)
Chalky	1%	cornstarch (Best Foods, CPC International, Inc., Englewood Cliffs, NJ)

[a] All standards prepared using commercial, bottled water (Aqua Cool, Portland, OR).

10^oC water.

Samples were presented monadically in 86 ml plastic cups (Sweetheart, Maryland Cup Co., Owings Mills, MD) coded with 3 digit, random numbers. Reference standards, as described in the training, were provided and a warm-up sample of non-carbonated spring water was presented 5 minutes prior to the beginning of the test to provide similar testing conditions for the first sample as well as subsequent samples. Panelists were instructed to swallow each sample three times consecutively before rating. This procedure most closely simulated normal consumption of carbonated beverages (as evaluated in an informal study carried out by the investigator). Samples were presented at intervals of 5 minutes to facilitate accurate testing and to allow receptor recovery.

To avoid fatigue, descriptors were divided into two sets on separate ballots. The first set of descriptors included cooling, salty, sweet, sour, bitter, astringency, and chalky. The second set included bubbly, bubble size, bubble sound, gas expansion feeling, bite, burn, and numbing. The two sets of descriptors were evaluated in separate tests; each set of descriptors tested on three consecutive days with one replication being tested each day. All eight samples (carbonation by temperature treatment) were tested at each session.

Each descriptor was rated using a 16-point intensity scale (0= "none", 1= "just detectable", 3= "slight", 5= "slight to moderate", 7= "moderate", 9= "moderate to large", 11= "large", 13= "large to extreme", and 15= "extreme"). For the descriptor bubble size, the same 16-point scale was used but 1= "extremely small", 3= "small", and 15= "extremely large".

Experimental Design and Statistical Analysis

A randomized, balanced, complete block design was used with the four carbonation levels, two temperature levels, and three replications (Cochran and Cox, 1957). An assumption in this design was that panelists maintained the same sensory sensations/perceptions when evaluating the separate descriptor sets.

Analysis of variance (ANOVA), principal component analysis (PCA), and correlations were performed using version 6.03 SAS statistical package (SAS Institute, Inc., Cary, NC). Panelist, replication, temperature, CO_2 level effects, and interaction effects were tested using ANOVA. A mixed effects F-test model was used with treatment effects fixed and all other effects (including panelist) considered random (Anderson and Bancroft,1952). The data of non-perceivers (persons giving zero ratings for all temperature and CO_2 levels for a particular attribute) were not included in the data analysis, as inclusion would have diluted potential differences found by perceivers.

Panelist-by-treatment interactions were visualized by creating line graphs. Each panelist's ratings (y-axis) were plotted against each treatment (x-axis) to search for systematic inconsistencies among panelists contributing to variation. Treatment differences of descriptors with significant ($p \leq .05$) panelist-by-treatment interactions were interpreted cautiously.

Fisher's least significant difference (LSD) test ($p \leq .05$) was used to compare treatment differences. The formula and degrees of freedom used were constructed according to the formula provided by Anderson and Bancroft (1952). Using this formula, a few cases were encountered where there was a significant F-value, but differences among treatment means could not be specified because the LSD value was too large. For these cases, a t-test was used to test for significant differences among treatments.

PCA of replication mean scores for descriptors was conducted. Chalky and sweet were not analyzed because of the lack of panel agreement on their use.

Results and Discussion

Lexicon Development

Eight samples were presented to the descriptive panel for training during each of 18 panel sessions. During the first three sessions, panelists independently listed descriptors to describe the taste, mouthfeel, and sound of the samples. Twenty-two descriptors were generated from these sessions. During the following sessions, this list was narrowed as redundant terms or terms with broad, ambiguous meanings such as "fresh" or "refreshing" were eliminated. These terms were eliminated only after discussion among the panelists and panel leader, and agreement that there were terms remaining to describe the sensations represented by the eliminated terms.

An example of this elimination process were the descriptors bite and burn. Originally, pain, pricking, fizzy, tingle, sting, and bite were listed as mouthfeel terms describing a particular sensation. After several panel sessions, panelists agreed that these were all terms describing either the intensity or the location of a particular sensation which could be described by the term bite. All of the terms with the exception of bite were subsequently dropped.

The descriptor burn evolved in a similar manner. Initially, a number of terms including abrasiveness, harshness, warmth, pain, and burn were used to describe the sensation. Through discussion and training, these terms were narrowed to the term burn.

Another complex and difficult descriptor for the panelists was the term salty. Initially, mineral, baking soda, and salty were suggested as separate terms. Through discussion and training, these terms were all included as part of the term salty.

Through discussion and training the trained panel agreed upon 14 descriptors and corresponding definitions for describing the flavor, mouthfeel and sound associated with carbonated water (Table 2.2). Taste descriptors developed by the panel included salty, sweet, sour, and bitter. The list of mouthfeel descriptors was much more extensive, including cooling (physical), astringency, chalky, bubbly, bubble size, gas expansion feeling, bite (chemical), burn (chemical), and numbing. Bubbly sound was also included as a descriptor. Definitions were developed by the panel through extensive training, evaluation, and discussion. Any applicable definitions for terms presently in use as ASTM standard terminology were used (ASTM, 1991).

Even after panel agreement on terms and definitions, there were panelists who understood the definition but did not use a particular descriptor. These terms were predominantly the taste descriptors, but also included astringency and gas expansion feeling. Bitter, salty, sweet, sour, and astringency solutions were presented to the panelists in varying concentrations to make certain that panelists understood the terms and their definitions. It is well documented that panelists are occasionally confused by sour and bitter (Robinson, 1970), sour and salty (Meiselman and Dzendolet,

Table 2.2. Carbonation descriptors and definitions developed by the trained panel.

Descriptor	Definitions
Salty	the taste stimulated by NaCl in water.
Sweet	the taste stimulated by sucrose in water.
Sour	the taste stimulated by an acid such as citric acid or lactic acid in water.
Bitter	the taste stimulated by caffeine in water.
Cooling (physical)	the sensation of reduced temperature experienced as a result of exposure to thermally cold substances.
Astringency	the complex of tactile sensations due to shrinking, drawing, or puckering of the oral epithelium as a result of exposure to such substances as alums or tannins.
Chalky	the perception of particles in the mouth experienced between the tongue and the upper palate, teeth, or sides of the mouth.
Bubbly	the feeling of bubbles mechanically coming in contact with the oral epithelium including the feeling of the bubbles' movement, and/or bursting, and popping.
Bubble size	the perception of the size of the bubbles in the mouth.
Bubbly sound	the sound of bubbles bursting in the mouth.
Gas expansion feeling	the release of CO_2 from solution upon introduction into the mouth resulting in the feeling of fullness or expansion of the mouth.
Bite (chemical)	the stinging experienced primarily in the oral cavity as a result of exposure to CO_2.
Burn (chemical)	the perception of increased temperature and irritation resulting from the exposure to CO_2. The sensation lingers after the stimulus is removed.
Numbing	the perception of loss of feeling, or an anesthetized feeling within the oral cavity.

1967), and even sour and sweet (Meiselman and Dzendolet, 1967), especially at lower concentrations. These can be corrected by defining taste adjectives and providing the appropriate taste sensations (O'Mahoney et al., 1979). It was found that all panelists perceived these descriptors, but some panelists did not perceive them at the low levels found in the test samples. These were the same panelists which were classified as non-perceivers in the analysis of the testing results.

Lexicon Use

The samples were evaluated under controlled testing conditions, using the experimental design described earlier, to test which descriptors were relevant and meaningful to the descriptive profile of carbonated water.

Non-perceivers. One way to validate the developed lexicon is to examine use of descriptors by the panel. During panel training some of the panelists did not use certain terms. This was also the case during testing. Salty was not detected by one panelist, astringency by two panelists, sourness by three panelists, bitterness by four panelists and two panelists did not use the term "gas expansion feeling". The results also revealed that only one panelist gave scores for chalky and two panelists used the term sweet. This was surprising because a greater number of panelists, though not always the same ones, had used these descriptors during the training. Evidence for this kind of inconsistency when working with very low concentration solutions has been discussed by other researchers (O'Mahoney et al., 1979). Chalky may have been confused with astringency by the panelists as the particulate feeling in the mouth is similar to the feeling induced by precipitation of proteins resulting from astringent compounds. Non-zero scores given by panelists for sweet were predominantly for the non-carbonated samples. Bartoshuk et al. (1964) found that the inherent taste of water is not necessarily tasteless, but after adaptation to acids, seems to taste sweet (Bartoshuk,1974). Perhaps panelists were affected by this adaptation effect, as sourness was an integral component of the descriptive profile. Chalky and sweet were dropped from subsequent analyses because of the lack of agreement. All of the other descriptors tested were utilized by the majority of the panelists in a manner consistent with their importance in describing the perception of carbonated water.

The question may be asked whether all of the descriptors are relevant to the descriptive profile of carbonated water. It is certainly possible that panelist ratings may reflect some overlapping use of the descriptors. However, a concerted effort was made during the training to clearly delineate the descriptors and their definitions to ensure that not more than one descriptor was used to describe a particular perception. This was accomplished using standards and extensive panel discussion. An examination of the definitions of the descriptors reveals that all of the definitions are distinctly different from each other (Table 2.2).

ANOVA revealed significant treatment differences by temperature level and CO_2 level for a number of the attributes (Table 2.3).

All of the descriptors were significant for panelist effect (Table 2.3). Panelist effect is commonly found in ANOVA results of trained panel data, especially when intensity standards are not used: This was the case in this study. The significant panelist effect indicated that panelists were using

Table 2.3. Significance levels[a] for main factors (temperature, CO_2 level, panelist, and replication) and all interactions for descriptors.

Source of Variance	Cooling	Salty	Sour	Bitter	Astringency	Bubbly	Bubble size	Bubbly sound	Gas expansion	Bite	Burn	Numbing
						Descriptors						
Temperature	***	ns	ns	ns	ns	ns	**	***	ns	*	*	*
CO_2 Level	ns	***	***	**	***	***	ns	***	***	***	***	***
Temp X CO_2	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Panelist	***	***	***	***	***	***	***	***	***	***	***	***
Replication	ns	ns	ns	ns	ns	ns	ns	ns	*	*	ns	ns
Pan X Rep	*	***	ns	ns	ns	**	ns	***	ns	ns	***	ns
Temp X Pan	***	ns	ns	ns	ns	ns	ns	ns	ns	ns	***	***
Temp X Rep	ns	ns	ns	ns	ns	ns	ns	ns	*	ns	ns	ns
CO_2 X Pan	ns	***	***	ns	***	**	***	***	***	***	***	***
CO_2 X Rep	ns	ns	ns	ns	ns	ns	ns	ns	*	ns	ns	ns
Temp X CO_2 X Pan	ns	ns	ns	ns	ns	ns	ns	**	*	ns	ns	ns
Temp X CO_2 X Rep	*	ns	ns	ns	ns	ns	ns	*	ns	ns	ns	ns
Temp X Pan X Rep	ns	*	ns	ns	ns	ns	ns	ns	ns	*	ns	ns
CO_2 X Pan X Rep	*	ns	*	ns	ns	*	ns	***	*	ns	ns	ns

[a] ns not significant at $p \leq .05$
*, **, *** refers to significance at $p \leq .05$, $p \leq .01$, and $p \leq .001$, respectively

different parts of the intensity scale for rating the attributes, but this did not interfere with treatment effects.

There were no significant temperature X CO_2 interactions and there was only scattered significance for all of the other interactions with the exception of panelist X CO_2 and panelist X replication (Table 2.3). Graphing the raw data using line graphs indicated minor inconsistencies throughout the data which contributed to the significant interaction effects. These inconsistencies were not systematic and were not confined to single panelists. Investigation of individual panelist standard deviations revealed only scattered cases of uncharacteristically large standard deviations, which were not confined to any one panelist.

Much of the significant panelist related variation seems to result from the sensitivity of the test to the few occurrences of panelists scoring in a slightly different manner from each other. This was most noticeable when some panelists rated both the non-carbonated and 1.69 vol. samples as zero while the other panelists rated these two samples as being different. Deletion of data from an inconsistent panelist would be expected to reduce significant interaction effects. We did not identify any panelist whom we felt was systematically inconsistent and therefore did not eliminate any panelist data from the analysis.

Another way to validate the lexicon is to examine the ratings for interrelationships among the descriptors. However, it is difficult to determine whether the panel is using descriptors independent of each other by examination of their ratings. Bubbly and gas expansion had similar ratings for CO_2 level averaged over temperature (Table 2.4). Burn and numbing had similar ratings for the samples at each carbonation level (Figure 2.1). The sensations represented by bubbly and gas expansion feeling are dissimilar

Table 2.4. Mean ratings[1], standard deviations (SD), significance levels, and LSD values for descriptors by carbonation level averaged over temperature.

Descriptor	Non-carbonated	Carbonation Level (vol.) 1.69	2.75	4.63	Sig.[2]	LSD
Bubbly	0.0[a] (0.0)	1.1[a] (1.3)	5.0[b] (2.4)	8.3[c] (2.5)	***	1.28
Bubble size	0.0[a] (0.0)	1.0[a] (1.2)	4.1[b] (2.2)	6.6[c] (2.7)	p=.06	1.43
Bubbly sound	0.0[a] (0.0)	0.2[a] (0.6)	3.8[b] (2.2)	7.0[c] (2.6)	***	1.42
Gas expansion feeling	0.0[a] (0.0)	1.3[a] (1.5)	4.8[b] (2.6)	8.5[c] (2.4)	***	1.69
Bite	0.4[a] (1.2)	3.2[b] (2.0)	6.3[c] (2.2)	9.4[d] (3.1)	***	1.79
Burn	0.4[a] (1.3)	2.0[ab] (2.1)	3.6[b] (2.3)	5.6[c] (3.0)	***	1.63
Numbing	0.7[a] (1.1)	2.1[ab] (1.6)	3.5[bc] (2.3)	4.9[c] (3.1)	***	1.66
Astringency	0.0[a] (0.0)	1.3[b] (1.2)	2.0[bc] (1.1)	2.5[c] (1.4)	***	.70
Cooling	8.4 (2.4)	8.7 (2.5)	8.9 (2.4)	8.8 (2.3)	ns	-----
Salty	0.0[a] (0.2)	2.4[b] (2.1)	2.3[b] (2.0)	2.3[b] (1.9)	***	1.22
Sour	0.2[a] (0.6)	2.5[b] (1.5)	3.4[b] (1.8)	3.7[b] (2.0)	***	1.27
Bitter	0.0[a] (0.0)	1.8[b] (1.6)	2.2[b] (1.3)	2.1[b] (1.3)	**	.95

[1] Sixteen-point intensity scale (0=none, 15=extreme)
[2] ns not significant at $p \leq .05$
*, **, *** refers to significance at $p \leq .05$, $p \leq .01$, and $p \leq .001$, respectively. Means within a row followed by the same letter are not significantly different at $p < .05$

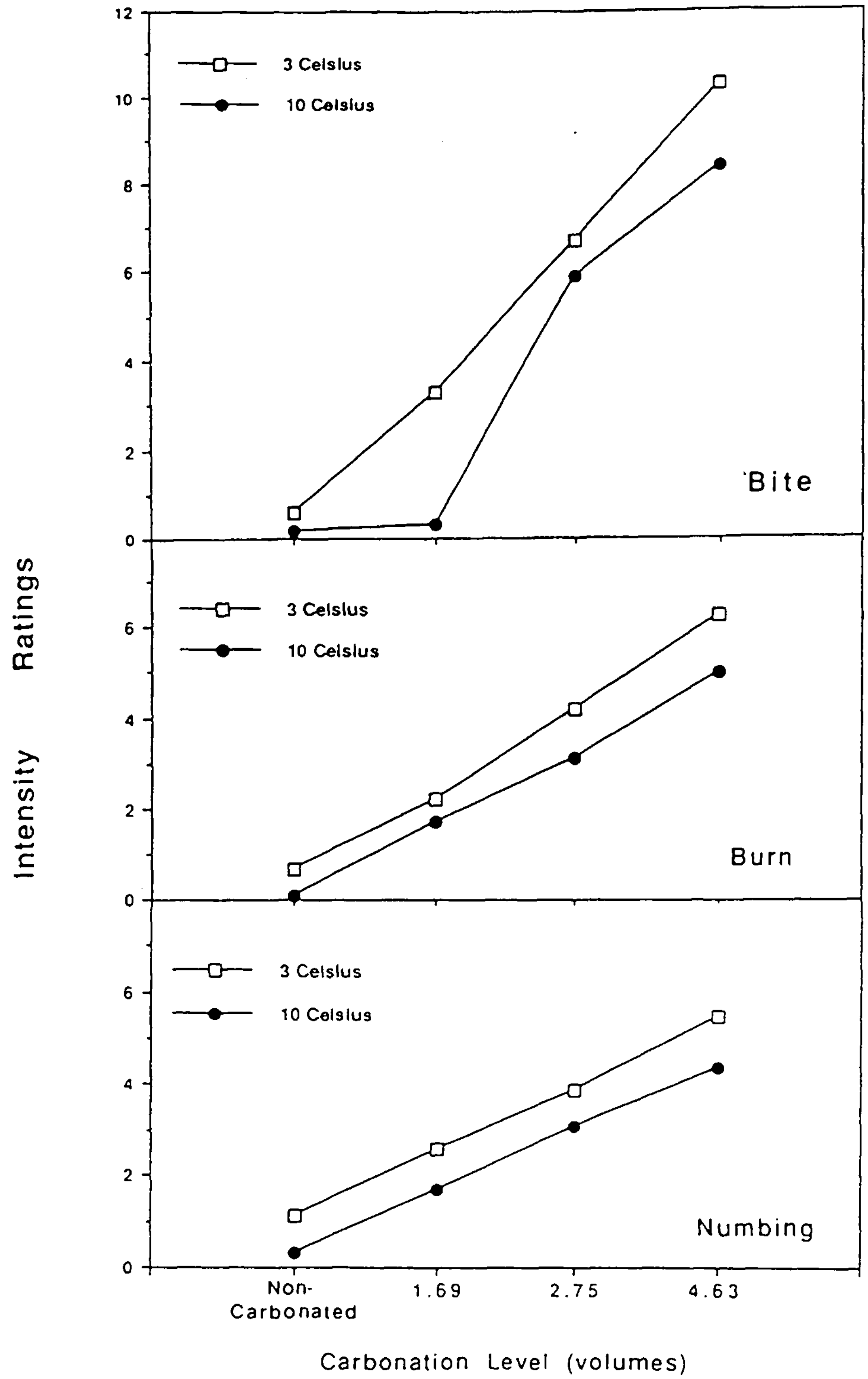


Fig. 2.1. Overall mean intensity ratings for bite, burn, and numbing by carbonation level and temperature. Scale ratings from 0=none to 16=extreme.

and would not be expected to cause confusion. However, burn and numbing may represent sensations which overlap because both their definitions pertain to a sensation which lingers, although burn is characterized by irritation and numbing is characterized by loss of feeling.

Effect of CO_2 Levels on Carbonation Perception

All of the descriptors tested, with the exception of cooling and bubble size, were significant for carbonation level averaged over temperature (Table 2.4). There were two basic patterns associated with the change in carbonation level. The first pattern was the increase in the rating of the intensity of a descriptor as the CO_2 level increased. This pattern was evident for most of the non-taste attributes, wherein each incremental level of carbonation was found to be significantly different from the preceding level. For bubbly, gas expansion feeling, and bubble sound, the non-carbonated samples and the lowest level of carbonation were not significantly different (Table 2.4). However, these levels were significantly lower compared to the middle level, and the highest level was significantly higher compared to the other levels. For bite and, to a lesser degree, for burn, numbing, and astringency, higher carbonation levels resulted in significantly higher intensity ratings at each level of carbonation (Table 2.4). This pattern is consistent with the sharp increase of perceived CO_2 magnitude experienced with increasing concentration found by Yau and McDaniel (1990), as well as the work on nasal pungency by Garcia-Medina and Cain (1982) and Cain and Murphy (1980).

The second pattern was associated with the taste descriptors of salty, bitter, and sour and was quite different compared to the non-taste descriptors.

The ratings for the three samples with CO_2 added were not significantly different than each other, however, they were significantly higher than the non-carbonated sample (Table 2.4). Sourness ratings, although not significant, did increase as the carbonation levels increased as would be expected by the decreasing pH levels recorded for the samples. Lederer et al. (1991) found that sourness, astringency and bitterness ratings were enhanced at higher carbonation levels, although the highest level of carbonation (1.42 vol.) in their study was lower than this study's lowest level of carbonation (1.69 vol.).

Effect of Temperature on Carbonation Perception

Six descriptors were found to change significantly with temperature change averaged over carbonation level: Cooling, bite, burn, and numbing were rated significantly higher in the 3°C samples, while bubble size and bubble sound were rated significantly higher in the 10°C samples (Table 2.5).

An interesting aspect of this analysis was the difference between the mouthfeel and taste components with respect to temperature. The majority of mouthfeel components were significant, conversely, none of the taste components changed significantly within the range of temperatures tested in this study. Graphing the intensity ratings for the significant descriptors demonstrates that (with the exception of cooling) they change not only with temperature but also with the change in CO_2 level (Figure 2.1 and 2.2). Bite, for example, consistently had higher intensity ratings for the carbonated samples which were at 3°C compared to 10°C. This same pattern was also evident for burn and numbing. For bubble size and bubble sound, the

Table 2.5. Mean ratings[a], standard deviations (SD), and significance levels of significant descriptors by temperature level averaged over CO_2 level.

	Cooling	Bite	Burning	Numbing	Bubble size	Bubble sound
3°C	10.4	5.2	3.3	3.3	2.6	2.5
	(1.7)	(4.3)	(3.3)	(2.8)	(3.0)	(3.2)
10°C	7.0	4.4	2.5	2.4	3.3	3.1
	(1.7)	(3.7)	(2.6)	(2.5)	(3.4)	(3.5)
Sig.[b]	***	*	*	*	**	***

[a] Sixteen-point intensity scale (0=none, 15=extreme)
[b] * $p \leq .05$, ** $p \leq .01$, *** $p \leq .001$

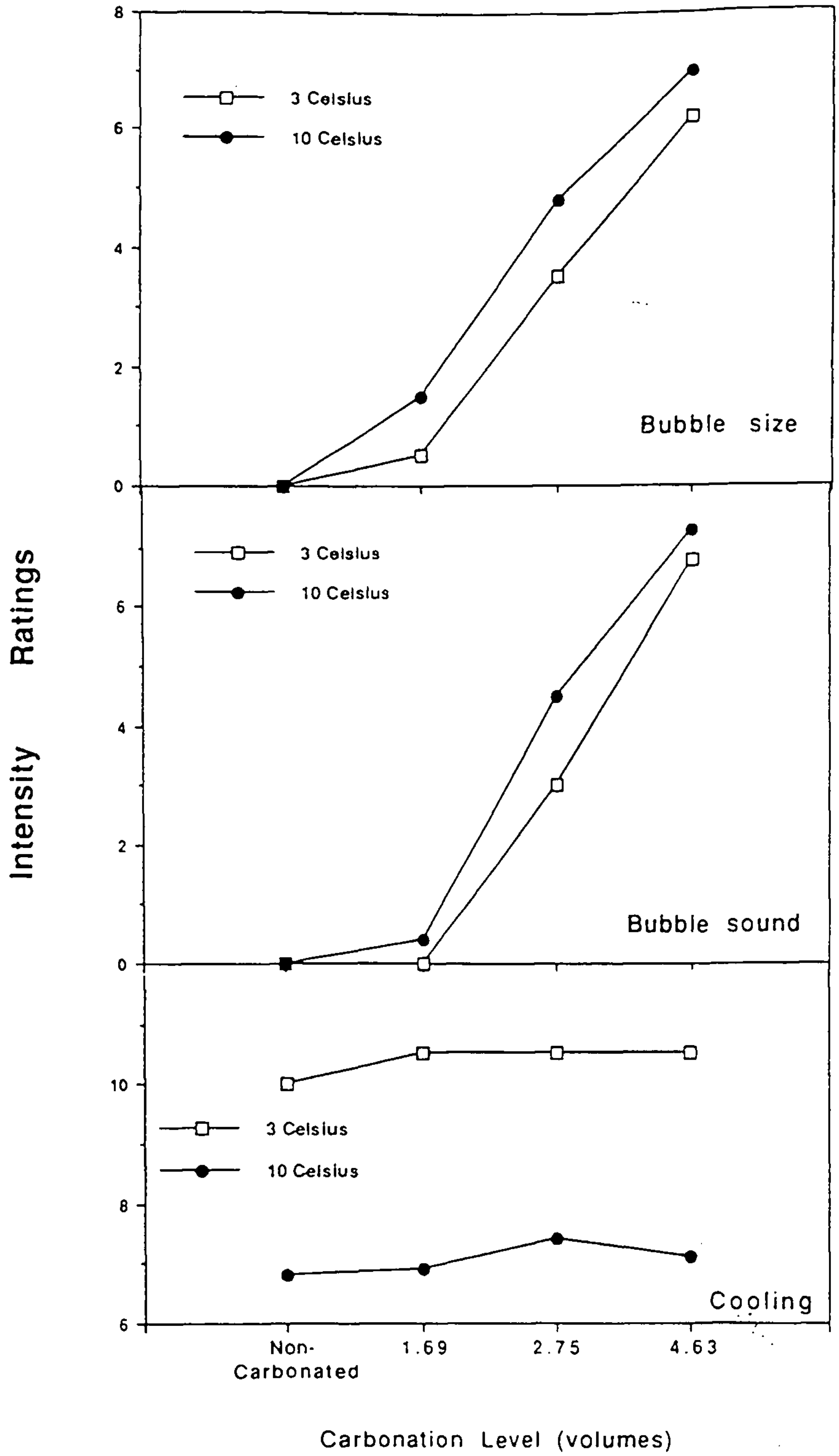


Fig. 2.2. Overall mean intensity ratings for bubble size, bubble sound, and cooling by carbonation level and temperature. Scale ratings from 0=none to 16=extreme.

pattern was the same as the carbonation level changed, but the temperature relationship was reversed as the intensity ratings were higher for the 10°C samples compared to the 3°C samples. Cooling ratings were very different for the two temperature levels with the 3°C samples rated much higher. However, these ratings were not affected by the CO_2 level.

These results are consistent with the work of Yau and McDaniel (1991), who found that carbonation intensity was perceived to be higher at lower temperatures than at higher temperatures; but contrary to the findings of Green (1990) that highlighted lowering of the ratings for "oral irritation" with decrease in temperature for capsaicin, piperine, ethanol, and NaCl. The range of temperature (24°C-46°C) in his study, however, was quite different than that used in this study.

Principal Component Analysis

The PCA model of the descriptors was explained almost entirely by the first three principal components (97%). Principal component 1 (PC1) explained 77.4%, principal component 2 (PC2) accounted for 10.7%, and principal component 3 (PC3) accounted for 8.9% of the model.

Graphs (Figures 2.3 and 2.4) of the PCA loadings give a clear picture of the goodness of the replications and how the descriptors relate to each sample, carbonation level, and temperature level. Distinct separation between each of the samples as well as the tight grouping of the three replications for each of the samples indicates that the panelists were easily able to differentiate between the samples and evaluated them in a most consistent manner. The graphs show a noticeable difference between samples based on carbonation level and temperature level. PC1 scores for

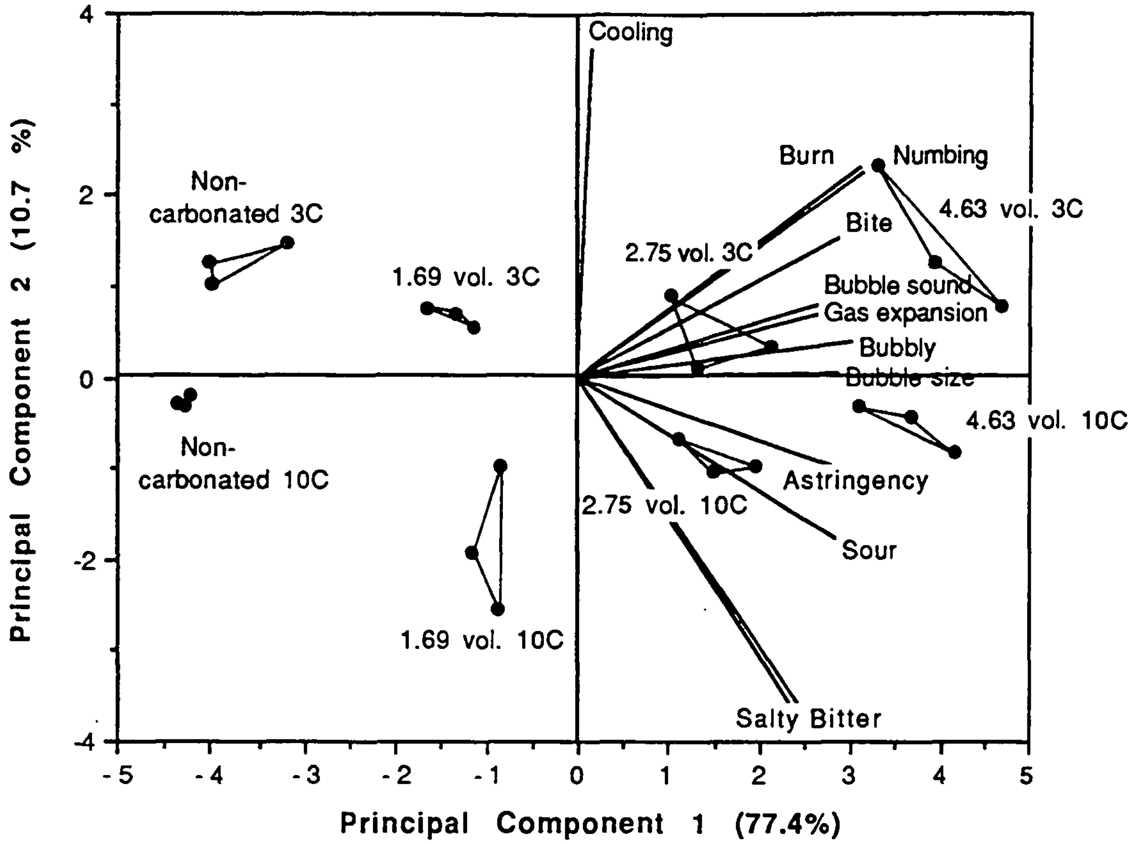


Fig. 2.3. Principal component analysis plot of intensity ratings for carbonated water descriptors for the eight samples: principal component 1 vs. 2. The three connected points for each sample represent three replications across eight panelists.

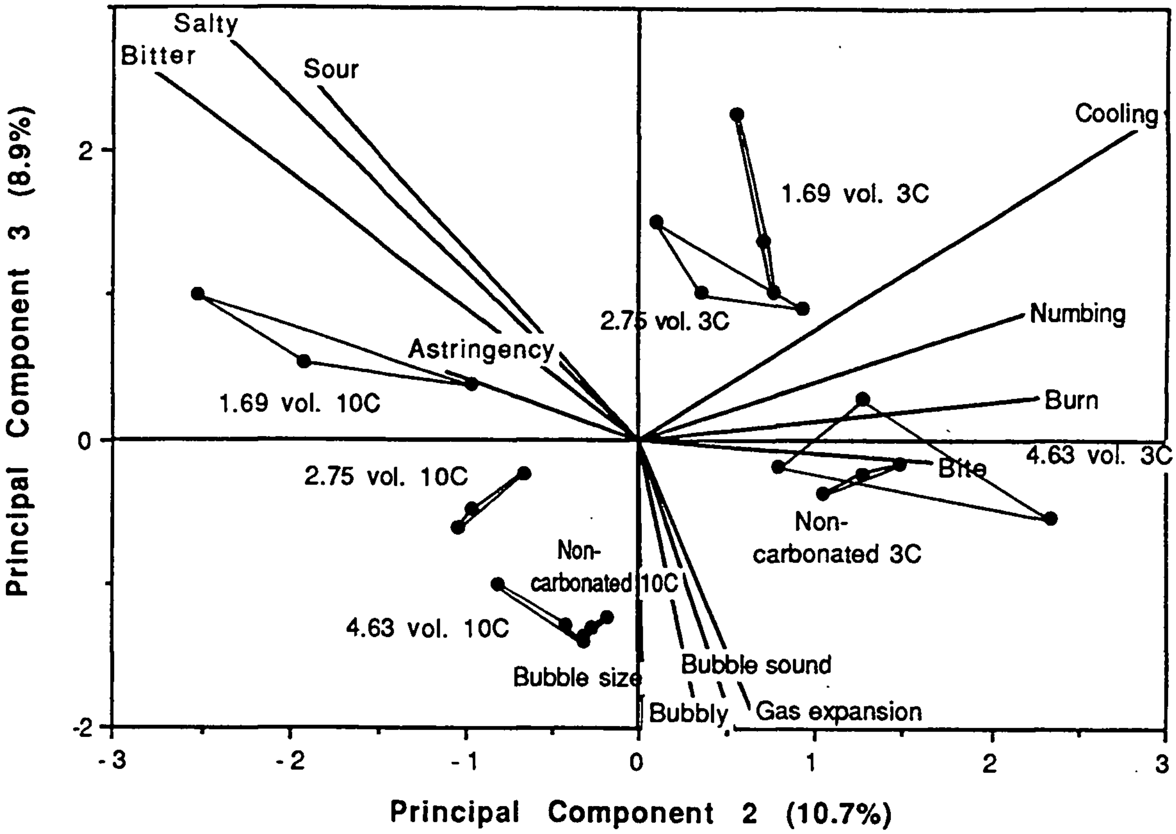


Fig. 2.4. Principal component analysis plot of intensity ratings for carbonated water descriptors for the eight samples: principal component 2 vs. 3. The three connected points for each sample represent three replications across eight panelists.

samples increased dramatically with increasing carbonation as evidenced by the spacing between carbonation levels and orientation of the plotted samples from left to right in Figure 2.3. In Figure 2.4 the sample plottings are oriented from the lower left to the upper right showing that the 3°C samples had higher scores for PC2 and PC3 compared to the 10°C samples.

PC1 can be defined as overall carbonation impact, as all terms except cooling have high loadings on PC1. PC2 can be defined as cooling, numbing, burn, and bite. This explains the major sensory differences between temperature levels after accounting for the effects of carbonation. These are primarily trigeminal responses (mouthfeel). The PC2 by PC3 plot notes a third separation in the data. The lower carbonation levels have a different perception of tastes after accounting for overall carbonation and mouthfeel.

Correlation and PCA analysis indicated that all of the descriptors, except cooling, were significantly, though not strongly, correlated. However, descriptors could be grouped to more clearly reflect their interaction with each other. The tastes saltiness, bitterness, astringency and sourness were closely related, as were bite, burn and numbing (trigeminal descriptors). Descriptors related to mechanoreception (bubbly, bubble size, bubble sound and gas expansion feeling) were very closely related, while cooling was not associated with the other attributes.

Conclusion

This research demonstrates that "overall carbonation perception" in carbonated water can be divided into a lexicon of descriptors which more accurately describe this complexity of sensations.

Additionally, the CO_2 and temperature level greatly influenced the intensity ratings for each descriptor. The intensity ratings of all descriptors, except cooling, increased significantly as CO_2 levels increased. Bubble size and bubble sound were rated significantly higher at 10°C; while cooling, bite, burn, and numbing were rated significantly higher at 3°C.

PCA revealed that all of the descriptors, except cooling, were significantly correlated and that the descriptors could be divided into four groups (cooling, taste descriptors, trigeminal descriptors, and mechanoreception descriptors) to more clearly reflect their interaction with each other.

Understanding the rating behavior for descriptors in carbonated water systems can facilitate prediction of flavor profiles of formulated, flavored, carbonated beverages. An example is understanding the interrelationship of sourness and sweetness in a beverage to which CO_2 is added.

References

Anderson, R.L. and Bancroft, R.A. 1952. *Statistical Theory in Research. Part II - Analysis of Experimental Models by Least Square,* 338, McGraw-Hill Book Co., New York.

ASTM. 1991. *1991 Annual Book of ASTM Standards. Vol. 15.07 End Use Products,* 1-3, ASTM, Philadelphia, PA.

Bartoshuk, L.M. 1974. After-dinner talk taste illusion: some demonstrations. Annals of New York Academy of Sciences. 237:279-285.

Bartoshuk, L.M., McBarney, D.H., and Pfaffmann, C. 1964. Taste of sodium chloride solutions after adaptation to sodium chloride: implications for the "water taste". Science. 143:967-968.

Cain, W.S. and Murphy, C.L. 1980. Interactions between chemoreceptive modalities of odor and irritation. Nature. 284:255.

Cochran, W.G. and Cox, G.M. 1957. *Experimental Designs,* Wiley, New York, N.Y.

Garcia-Medina, M.R. and Cain, W.S. 1982. Bilateral integration in the common chemical sense. Physiol. and Behav. 29:349.

Green, B.G. 1990. Effects of thermal, mechanical, and chemical stimulation on the perception of oral irritation. In *Chemical Senses,* Vol. ii, (B. G. Green, J. R. Mason and M. R. Kare, eds.) 172-179, Marcel Dekker Inc., New York.

Lederer, C.L. Bodyfelt, F.W., and McDaniel, M.R. 1991. The effect of carbonation level on the sensory properties of flavored milk beverages. J. Dairy Sci. 74:2100-2108.

Meiselman, H.L. and Dzendolet, E. 1967. Variability in gustatory quality identification. Perception and Psychophysics 2:496-498.

O'Mahoney, M., Goldenberg, M., Stedman, J. and Alford, J. 1979. Confusion in the use of the taste adjectives 'sour' and 'bitter'. Chemical Senses and Flavour. 4(4):301-317.

Robinson, J.O. 1970. The misuse of taste names by untrained observers. Brit. J. Psychol. 61:375-378.

Yau, N.J.N., and McDaniel, M.R. 1990. The power function of carbonation. J. Sensory Studies. 5:117-128.

Yau, N.J.N., and McDaniel, M.R. 1991. The effect of temperature on carbonation perception. Chemical Senses. 16(4):337-348.

Yau, N.J.N., McDaniel, M.R. and Bodyfelt, F.W. 1989. Sensory evaluation of sweetened flavored carbonated milk beverages. J. Dairy Sci. 72:367-377.

Zahm & Nagel Co., Inc. *Zahm Practical Testing Instruments, 15th Edition,* Zahm and Nagel Co., Inc., Buffalo, NY.

TEMPORAL ASPECTS OF CARBONATION PERCEPTION

Steven J. Harper and Mina R. McDaniel

Department of Food Science and Technology

Sensory Science Laboratory

Oregon State University

Corvallis, OR 97331

Abstract

Time-intensity of CO_2 perception (specifically Bite and Burn sensation) in carbonated water was investigated at 3°C and 10°C. CO_2 concentrations of 0, 1.7, 2.8, and 4.6 volumes were used. Results demonstrated that the time-course curve for Burn sensation (steep linear rise and long-lived exponential decay slope) was similar to previously investigated irritants while that for Bite (steep linear rise and decay slopes, and relatively short Duration) was unlike most other irritant sensations. The sensations were qualitatively and quantitatively different. Intensity and duration of Bite and Burn sensations were concentration dependent and cold temperature enhanced perception. Possible psychological habituation or desensitization was observed as intensity and duration of 10°C samples decreased with successive replications. Most time-intensity parameters were correlated with each other for both Bite and Burn and could be divided into two groups; parameters which were CO_2 level dependent and those whose ratings were independent of CO_2 level. Considerable subject variability was found as in previous time-intensity studies.

Introduction

Carbonated beverages are the most popular beverages consumed in the United States. Until recently, very little was known about carbonation perception. Examination of carbonation perception had been limited to relatively few studies investigating primarily the acceptability or preference of products with added CO_2 (Bright and Potter, 1979; Baranowski and Park, 1984; Choi and Kosikowki, 1985; King et al., 1988; McBride and Richardson, 1983; McLellan et al., 1984; Pokorny et al., 1986; Yau et al., 1989). Recently, however, many researchers have taken an interest in this area, beginning with the investigation of CO_2 as a nasal irritant. Cain and Murphy (1980) found a strong mutual interaction between pungency and odor with pungency diminishing the odors tested when CO_2 and odorants were introduced nasally. Pungency was referred to as the perception, in the respiratory tract, of the "bite" of chili pepper, "coolness" of menthol, etc. arising from the "common chemical sense" (Cain, 1981). Subjects judging the pungency of various concentrations of CO_2 presented to one or both nostrils produced ratings increasing sharply with concentration (Garcia-Medina and Cain, 1982).

Psychophysical properties of CO_2 in solution are now being investigated. Interaction between oral pungency evoked by CO_2 and basic tastants was explored by Cometto-Muñiz et al. (1987), the researchers finding that mutual effects depended on the particular tastant employed. Gender differences in perception of pungency were also examined (Cometto-Muñiz and Noriega, 1985). They found nasal pungency power function exponents of 2.2 and 1.6, respectively, for females and males, and 1.1 for both genders for buccal pungency. In studies by Yau and McDaniel, power functions (Yau and McDaniel,1990) and temperature effects (Yau and McDaniel, 1991) were investigated. Power function exponents for overall oral carbonation perception were determined (2.79 for swallowing and 2.65 for expectoration) and found to be more similar to electric shock than for basic tastants. They also found carbonation intensity was perceived to be higher at lower temperatures - the temperature effect being carbonation level dependent with differences more evident at higher carbonation levels. Green (1992) also found carbonation perception was strongly temperature dependent, but concluded that enhancement experienced at cold temperatures resulted predominantly from an increase in the pungent (burning and stinging) components arising primarily from the stimulation of the nociceptive system. He also found that increasing levels of CO_2 increased ratings of perceived cold and that exposure continuing for several seconds led to painful sensations.

Development of descriptive terminology for carbonated water has also been investigated. Szczesniak (1979), surveying descriptive mouthfeel terminology for beverages, found that many carbonation related terms were suggested with bubbly, tingly, prickly, and stinging being mentioned most often. Biting and burning were used to describe chemical effects of beverages distinct from carbonation and along with the term stinging, all were described as having an element of quick, physical pain. Burn was considered to be classified in the "afterfeel-mouth" category involving sensations experienced primarily after the beverage is swallowed. Harper and McDaniel (1993a) developed a lexicon for describing the sensory perception of carbonated water using a trained panel. The lexicon included cooling, taste descriptors (salty, sour, bitter, astringent), trigeminal descriptors (bite, burn, numbing), and mechanoreception descriptors (bubbly, bubble size, bubble sound, gas expansion feeling). They also found that ratings of all descriptors, except cooling, increased significantly as CO_2 level increased and that cooling, bite, burn and numbing were rated higher for the 3°C sample compared to the 10°C sample.

Time-intensity (T-I) studies were first conducted by Neilson (1957) who had judges graph perceived flavor intensity over time directly on graph paper while responding to clock cues. Since then, many T-I studies have been conducted focusing predominantly on basic tastes. No T-I studies were found with CO_2 as a stimulant. Continuous tracking of a time-intensity response by computerized means allowed us to calculate a number of parameters which are inherent to the time intensity curve. Lee and Pangborn (1986) discussed these parameters and divided them into five general categories. These categories, along with associated parameters investigated in this study include: (1) time-related parameters (Tinitial, Tmax, TslopeA, TslopeD, Tmaxend); (2) rate-related parameters (SlopeA, SlopeD); (3) intensity-related parameters (Imax); (4) events occurring during oral perturbations; and (5) parameters related to duration of the stimulation (Duration, Totarea, Perimeter, Barea, Farea, Ratio). Parameters such as Tmax, Imax, Duration, and Tinitial have been used extensively by past investigators, especially for the study of sweeteners. The perimeter, slope and area parameters have been less extensively utilized.

Average curves provide valuable graphical information which aids interpretation of results when there is large variation among the subjects, as there was in this study. Overbosch et al. (1986) developed an averaging process which addressed the problems of simple averaging. He used geometric mean normalization of the intensity ratings as well as the time ratings of all subjects for the ascending and descending portions of the curve.

Liu and MacFie (1990) further refined this method to accommodate intensity plateaus, non-zero endpoints and non-monotonic curves, all problems left unsolved by Overbosch et al. (1986). The development of methodology for producing overall average curves which provide equal weightings for all subjects proved valuable to this study.

Bite and Burn were found to be the predominant and distinct descriptors in the lexicon we developed, as well as in other studies of oral irritants. With this in mind, we initiated this study to determine the T-I profiles for Bite and Burn and to compare their perceptual relationship to each other and to other irritants which have been investigated. This inquiry also gave us the opportunity to investigate the relationship between the T-I parameters, the subject variability, and the CO_2-temperature relationship for each of these sensations.

Materials and Methods

Samples

Commercial, bottled drinking water (Aqua Cool, Portland, OR) was used to produce the samples. The source of the commercial, bottled water was chlorinated Cascade Mountains water. Aqua Cool filtered the water using a series of multi-media filters to remove large particles, followed by filtration through activated carbon and polymeric resins to remove color, odors, and possible chemical contaminates and chlorine. The water was then disinfected using ozone. According to Aqua Cool, the samples contained an average of 1.5 ppm calcium, .31 ppm magnesium, .7 ppm sodium and 1.0 ppm chloride.

Batches of the water were carbonated with commercial carbon dioxide (Industrial Welding, Albany, OR) in a Zahm and Nagel 18.9 liter, stainless steel carbonator (Zahm and Nagel Co., Buffalo, NY). Four carbonation levels were produced [non-carbonated, 1.69 volume (vol.) (SD=.05), 2.75 vol. (SD=.058), and 4.63 vol. (SD=.064)]. The lowest level was chosen because it represented a level at which subjects could differentiate descriptors (Harper and McDaniel, 1993). The other levels were chosen based on prior research by Yau and McDaniel (1990) and represent levels in which a doubling of overall carbonation was perceived based on the power function for overall carbonation. The 1.69 (1.7) vol. CO_2 samples were produced using water, bottles, and CO_2 maintained at 21^{o}C. The 2.75 (2.8) and 4.63 (4.6) vol. CO_2 samples were produced using 21^{o}C CO_2 and 1^{o}C water, with bottles and carbonator immersed in ice. 18.9 liter batches of each level were produced. All samples were bottled in 828 ml, green, glass bottles and stored at 1^{o}C until evaluated.

The pH of the samples was 6.04 for non-carbonated, 3.71 for 1.7 vol. CO_2, 3.65 for 2.8 vol. CO_2, and 3.51 for 4.6 vol. CO_2. pH was measured at 22^{o}C with a Corning 125 pH meter with a Sensorex epoxy-body, sealed-reference combination electrode (S200C). The pH meter was calibrated with buffers of pH 3 and 7 (Microessential Laboratory, Brooklyn, NY).

Carbonation levels and temperatures were measured using a Zahm and Nagel piercing device (Zahm and Nagel Co., Buffalo, NY). Sample temperatures and headspace pressures were measured after agitation of bottles. Temperature and pressure readings were converted to "volumes CO_2 per volume water" using a conversion table (Zahm and Nagel Co., 1964). One volume is defined as the amount of CO_2 dissolved in water at equilibrium, 15.56^{o}C, and at one atmosphere pressure.

Training and Testing

Training. Five subjects, all students or faculty of the Department of Food Science and Technology at Oregon State University, participated in training and subsequent evaluation of the carbonated samples. Subjects were consumers of carbonated beverages and had participated in a previous panel developing and evaluating descriptors for carbonated water.

Eight one-hour training sessions were conducted to familiarize subjects with the T-I equipment and to re-familiarize the subjects with the samples and the descriptors "bite" and "burn". Bite is a sharp sensation with a quick onset experienced primarily in the oral cavity. Burn is a lingering sensation with a longer time to onset and a perception of increased temperature and irritation. Practice T-I curves for each subject were internally evaluated to make certain all subjects understood the stimulus to be evaluated and were using the T-I equipment properly.

A dilute solution of cayenne pepper (.1 g cayenne pepper (McCormick Co., Baltimore, MD)/600 g H_2O) was used as a reference standard to help subjects relate to burn perception.

Testing procedure. Testing was conducted in the Sensory Science Laboratory at Oregon State University, Corvallis, OR, in individual booths. Each individual booth was equipped with a computer monitor for providing testing instructions to the subjects, and with a data acquisition device for evaluating intensity of the samples. The data acquisition device was a 15 cm linear potentiometer with a knob which could be moved uni-directionally from left to right. A scale which was anchored with "none" and "extreme" on the ends with an unlabeled halfway marking in the middle was attached to the potentiometer. A computerized system called DASSIE (Data Acquisition System for Sensory Imput and Evaluation), developed in the Sensory Science Laboratory at Oregon State University, was used to monitor time and collect data. Data was collected every .25 seconds and was saved in a data file after being transformed to a 100-point intensity scale which corresponded to the potentiometer scale.

Eight samples were evaluated by subjects. These included samples at each carbonation level (non-carbonated, and 1.7 vol., 2.8 vol., and 4.6 vol.) at two temperatures (3°C and 10°C). The 3°C samples were packed in ice before and during the test. The 10°C samples were tempered by placing them in a styrofoam cooler filled with 10°C water. Samples were maintained in a motionless state at their respective temperatures to minimize escape of CO_2 during serving.

Samples were presented monadically in 60 ml plastic cups (Sweetheart, Maryland Cup Co., Owings Mills, MD) coded with 3 digit, random numbers. All session samples of each CO_2 level and temperature were carefully and gently poured, without agitation, into sample cups and served immediately to minimize any loss of carbonation. Each bottle was used for only one pouring. A warm-up sample of 6°C, 1.7 vol. or 6°C, 2.8 vol. water was presented 5 minutes prior to beginning of test.

Subjects were instructed to introduce the 15 ml sample into the mouth, immediately ingest the sample in one swallow, and begin to rate the sensation. Samples were presented at intervals of 5 minutes and subjects were instructed to rinse their mouths with lukewarm (approx. 25°C) water between samples.

During the testing, subjects were instructed on the procedures for the evaluation through instructions on the computer monitor in each booth. The evaluation was initiated by a countdown, with a message "to swallow" at zero time. Dassie began recording at zero time. Recording was terminated at the end of 5 minutes or by the subject pushing a button on the data acquisition device.

All eight samples were tested at each testing session.

Experimental Design and Statistical Analysis

A balanced, complete block design (Cochran and Cox, 1957) was used with four carbonation levels, two temperature levels, and three replications (reps). A separate set of evaluations was conducted for each of the sensations. Eight randomized orders were developed for each set of evaluations. One of the eight randomized orders was used for each of the sessions in which the samples were presented. It was not possible to randomize the order by panelist because of the concern with maintaining the carbonation level of the samples. Because it was not possible to evaluate all of the subjects in one sitting, many different sessions were conducted to complete each of the sets of three reps. These reps were consecutive trials for each subject using the same lot of samples. However, the subjects did not all test at the same time and, therefore, these reps should be considered to be trials and not true reps.

Principal component analysis (PCA), and correlations were performed using rep means. Panelist, rep, temperature, CO_2 level effects, and interaction effects were tested using fixed model analysis of variance (ANOVA). Fisher's least significant difference (LSD) test ($p \leq .05$) was used as the multiple comparisons test to compare differences among temperatures,

CO_2 levels, reps and panelists. All analyses were run using version 6.03 SAS statistical package (SAS Institute, Inc., Cary, NC).

A number of parameters including the typically described maximum intensity, duration, and time to maximum intensity were measured for each time-intensity curve and are defined in Table 3.1.

Average T-I curves were developed for the overall panel, reps, and individual subjects, using the normalization procedure of Liu and MacFie (1990) and a program written in C-language for the IBM computer (Yang,1993). Data was first normalized for time and intensity using average values of the parameters Imax, Tinitial, Tmax, Tdec (time at which perceptual decay accelerates), and Tend (time at which perception ends). The average curves were then produced using the normalized data sets. The overall average and subject curves were drawn using the parameters for the entire panel. Individual rep average values of these parameters were used for each set of rep average curves.

Table 3.1. Definitions of time-intensity parameters

Time-intensity parameter	Definition
Tinitial	Time to initial response.
Duration	Total time of time-intensity response from Tinitial to end of response.
Tmax	Time from initial response to time of maximum intensity.
Imax	Maximum intensity of the time-intensity response.
Totarea	Total area under the time-intensity curve.
Perimeter	Total perimeter of the time-intensity curve.
Barea	Area under the time-intensity curve before the maximum intensity.
Farea	Area under the time-intensity curve following the maximum intensity.
Ratio	Ratio of the area before the maximum intensity to the area after the maximum intensity.
SlopeA	Maximum slope of the linear portion of the ascending curve.
SlopeD	Maximum slope of the linear portion of the descending curve.
TslopeA	Total time of the linear portion of the ascending curve.
TslopeD	Total time of the linear portion of the descending curve.
Tmaxend	Duration of time from maximum intensity to end of response.

Results

Experiment One: Bite

Relationship of Time-Intensity Parameters

Previous studies have found that curve derived time-intensity parameters are often correlated and only a few parameters are needed to explain the sensation of interest. This is especially true when a large number of parameters are used. We used correlations and PCA to help determine parameters important to explaining Bite and Burn perception.

When correlations examining CO_2 level data and temperature X CO_2 level data were analyzed, significant ($p \leq .05$), strong to moderate correlations ($R^2 \geq .36$) between all parameters, with the exception of Tinitial, were found.

Principal component analysis (PCA) uses the correlation matrix to better determine not only the relationships between the parameters, but also between parameters and samples and between the samples themselves. Principal component 1 (PC1) accounted for a large part of the model, accounting for 83.4% of the variation (Figure 3.1). Principal component 2 (PC2) accounted for 10.1% of the variation while principal component 3 (PC3) was insignificant, explaining only 2.6% of the model.

PC1 was defined as being comprised of all the correlated parameters of the T-I curve. This included Slope D which is highly correlated and has a negative value by definition. PC2 was dominated by Tinitial, but also included Tmax, Duration, Tmaxend and Ratio; parameters for which higher ratings did not correspond with higher CO_2 levels. A number of distinctive groupings of parameters are apparent. Imax, Perimeter and the area

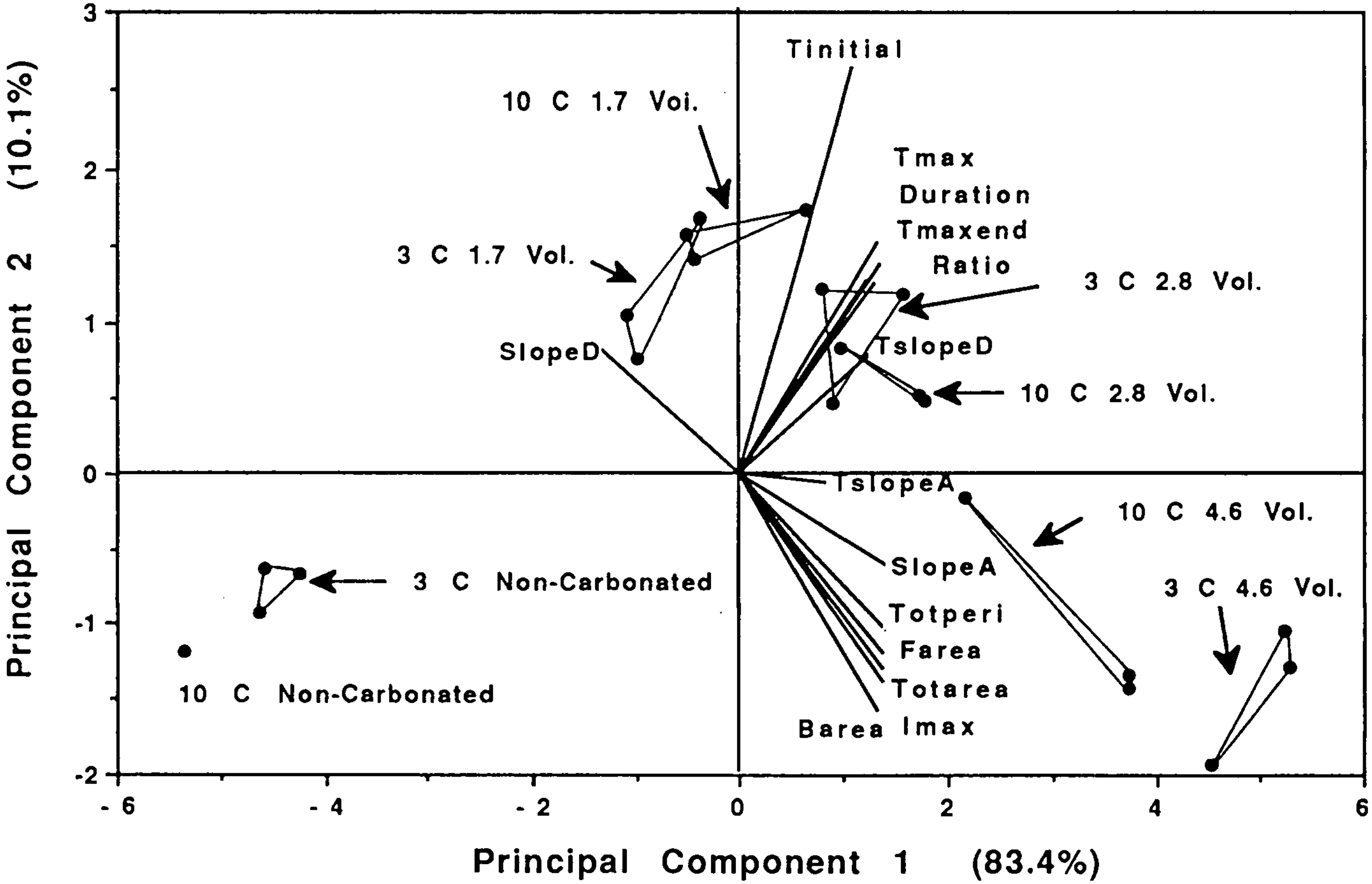


Fig. 3.1. Principal component analysis plot of ratings for Bite Parameters for the eight samples: principal component 1 vs 2. The three connected points for each sample represent three replications across five subjects.

parameters are tightly bunched as are Tmax, Duration, Tmaxend, and Ratio. Tinitial is clearly in its own space. All of the slope parameters fall between the two tightly bunched groups.

Samples for each of the CO_2 levels are clearly separated. The PC1 parameter scores are higher for higher CO_2 levels which reflects higher values for most of the actual parameter values as CO_2 level increased. Lower values of Tinitial for the non-carbonated and 4.6 vol. samples are responsible for separation of samples in PC2. The 4.6 vol. samples appear to be most influenced by parameters influenced by intensity (Imax and area parameters), while the the other carbonated samples are more related to PC2 influenced parameters (Tmax, Duration, etc.).

The non-carbonated and 4.6 vol. samples are spatially separated by temperature on the PCA graph, while the 1.7 vol. and 2.8 vol. samples don't separate by temperature. Differences between non-carbonated samples are only evident because subject #5 perceived Bite in 3°C samples while no subjects rated Bite in 10°C samples.

CO_2 Level Effects

Clear differences among the CO_2 levels for a number of parameters are evident from overall average curves (Figure 3.2). All parameters were significant ($p \leq .001$) for CO_2 level (Table 3.2). Carbonated samples were significantly different from the non-carbonated sample for all of the parameters. However, in contrast to the rest of the parameters, Ratio and Tmaxend were not significant for the different carbonated levels. Tinitial for 4.6 vol. samples occurred earlier than for other carbonated samples. Imax, along with Perimeter and the area parameters, increased with increasing

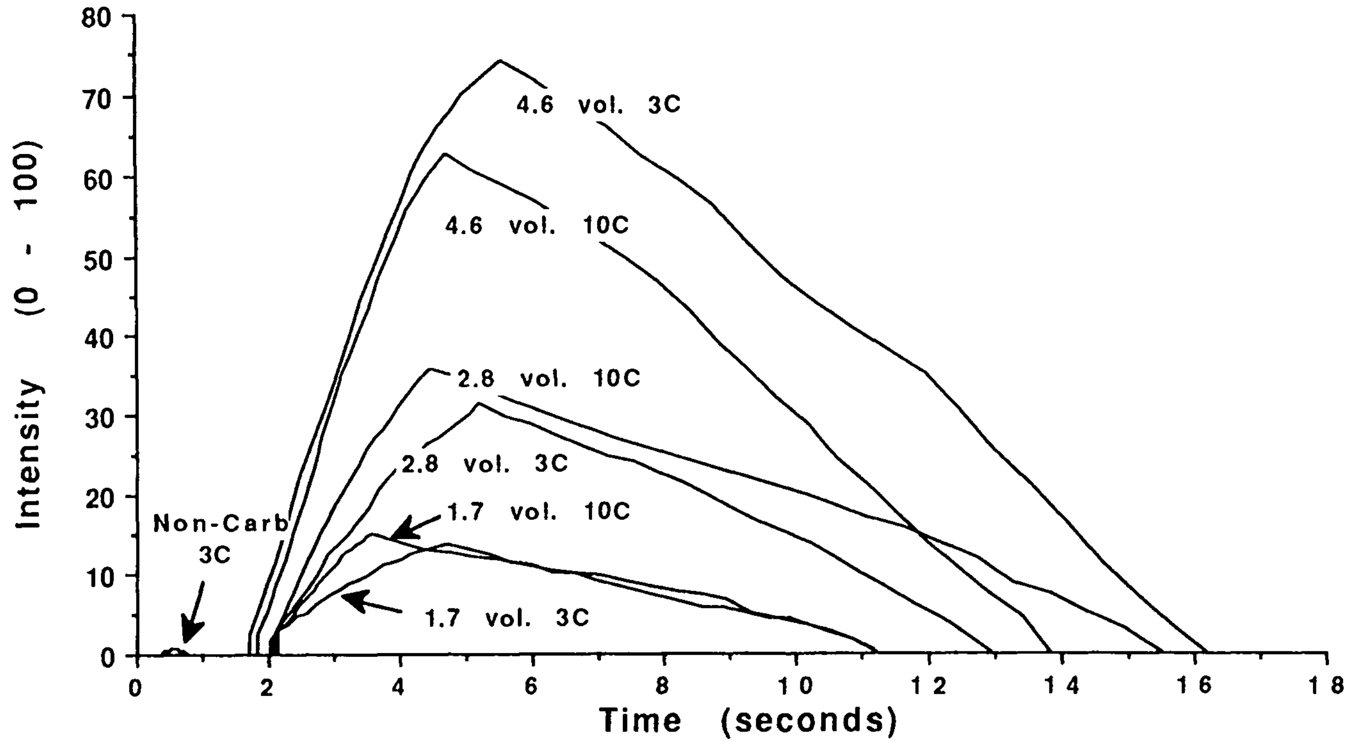


Fig. 3.2. Average time-intensity curves for all samples with the exception of the non-carbonated 10°C sample (not rated) for Bite. Curves were averaged using subject values which were normalized for time and intensity.

Table 3.2 Significance levels[a] for ANOVA main factors (Temperature, CO_2 level, Subject, and Replication) and interactions for Bite parameters.

Source of Variance	Parameters													
	Tinitial	Duration	Tmax	Imax	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
Temperature	ns	ns	**	**	*	**	ns	ns	ns	*	***	ns	ns	ns
CO_2 Level	***	***	***	***	***	***	***	***	***	***	***	***	***	***
Temp X CO_2	ns	ns	ns	***	***	***	ns	*	***	**	***	***	*	ns
Subject	***	***	***	***	***	***	***	***	**	***	***	***	***	***
Replication	ns	ns	ns	***	ns	*	*	ns	ns	ns	ns	ns	ns	ns
Subject X Rep	ns	***	ns	***	*	***	ns	*	ns	*	***	ns	ns	ns
Temp X Subject	ns	ns	ns	***	**	***	ns	ns	ns	*	**	ns	ns	ns
Temp X Rep	ns	ns	ns	***	**	**	ns	ns	ns	ns	**	*	ns	ns
CO_2 X Subject	***	***	***	***	***	***	***	***	**	***	***	***	***	***
CO_2 X Rep	ns	ns	ns	***	ns	***	ns	*	ns	ns	ns	ns	ns	ns

[a] ns not significant at $p \leq .05$

*, **, *** refers to significance at $p \leq .05$, $p \leq .01$, and $p \leq .001$, respectively

CO_2 level, each level being greater than the previous level. Comparison of the average values (17 for 1.7 vol., 33 for 2.8 vol., and 68 for the 4.6 vol.) represented an approximate "doubling" in intensity between each level. The 1.7 vol. samples clearly had a shorter Duration than for 2.8 and 4.6 vol samples, but these latter samples were similar. All slope parameters increased significantly as CO_2 level increased. And finally, Tmax occurred significantly later as CO_2 level increased.

Temperature Effects

A few temperature related differences can be determined from the overall average curve. The most obvious difference was that Tmax occurred later for all 3°C samples. Imax was greater for the 3°C 4.6 vol. sample than for the 10°C 4.6 vol. sample, but the 10°C 2.8 vol. sample Imax was greater than the 3°C 2.8 vol. sample. Imax for 1.7 vol. samples were approximately the same. The other significant parameters (Perimeter and area parameters) all had higher values for the 3°C samples.

Replications

There were no significant rep effects with the exception of lower values for Imax, Perimeter and SlopeA for rep 3 (Table 3.2). This reproducibility is exhibited in the graphing of PC1 and PC2 from PCA (Figure 3.1). Reps are clearly consistent enough to separate samples by CO_2 level. ANOVA of the two temperatures, evaluated separately (Table 3.3a and 3.3b), revealed that there were a number of parameters that were significant for 10°C samples. Imax was significant ($p \leq .001$) with four of five subjects rating the 10°C 4.6 vol.

Table 3.3 **Significance levels[a] for ANOVA main factors (CO_2 level, Subject, and Replication) and interactions for Bite parameters by temperature.**

a) 3°C

Parameters

Source of Variance	Tinitial	Duration	Tmax	Imax	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
CO_2 Level	***	***	***	***	***	***	***	***	***	***	***	***	***	***
Subject	***	***	***	***	***	***	***	***	ns	***	***	***	***	***
Replication	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Subject X Rep	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	**	ns	*	ns
CO_2 X Subject	***	***	***	***	***	***	**	**	**	***	**	***	***	*
CO_2 X Rep	ns	ns	ns	ns	ns	***	ns	ns	ns	ns	ns	ns	ns	ns

[a] ns not significant at p≤.05
*, **, *** refers to significance at p≤.05, p≤.01, and p≤.001, respectively

b) 10°C

Parameters

Source of Variance	Tinitial	Duration	Tmax	Imax	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
CO_2 Level	***	***	***	***	***	***	***	***	***	***	***	***	***	***
Subject	***	***	***	***	***	***	***	***	**	***	ns	***	***	***
Replication	ns	*	ns	***	*	*	*	ns	ns	ns	ns	*	ns	*
Subject X Rep	ns	*	*	ns	*	ns	ns	*	ns	ns	ns	ns	ns	*
CO_2 X Subject	*	***	***	***	***	***	**	***	ns	***	ns	***	***	***
CO_2 X Rep	ns	ns	ns	**	ns	**	ns	*	ns	ns	ns	**	ns	ns

[a] ns not significant at p≤.05
*, **, *** refers to significance at p≤.05, p≤.01, and p≤.001, respectively

samples lower for successive reps (especially rep three). Duration and Tmaxend also had lower values for successive reps. An interesting trend relating to the scattering in the PCA graphing and rep effects found in ANOVA was noted in the average curves for rep (Figure 3.3). For the first rep, the pattern for Imax (noted earlier) held true. But for the following reps, the values for the 10°C samples decreased substantially. Duration also decreased successively for 1.7 vol. and 4.6 vol. samples. This pattern did not occur with 3°C samples.

There was little difference between reps for 3°C samples. The only noticeable difference was that Duration increased with reps for the 2.8 vol. sample. This resulted in larger values for SlopeD, and area and perimeter parameters.

Variability Among Subjects' Time-Intensity Curves

A significant subject effect ($p \leq .001$) was found for all parameters (Table 3.2). Each subject's T-I curves were different compared to other subjects. There were significant subject X rep and subject X temperature effects for some parameters. These interactions can be predominantly explained by the change in ratings given Imax and Duration mentioned above. This, in turn, resulted in the interaction for the other parameters.

Raw data curves are difficult to use for comparing individual subjects. Average curves for each subject, which normalized the subjects to the same relative scale, were used to evaluate differences in perceptual responses. The points Imax, Tstart, Tmax, Tdec, and Tend are the same for each subject because they are based on average values for all subjects. Therefore, differences are expressed from the shapes of the curves. Two general

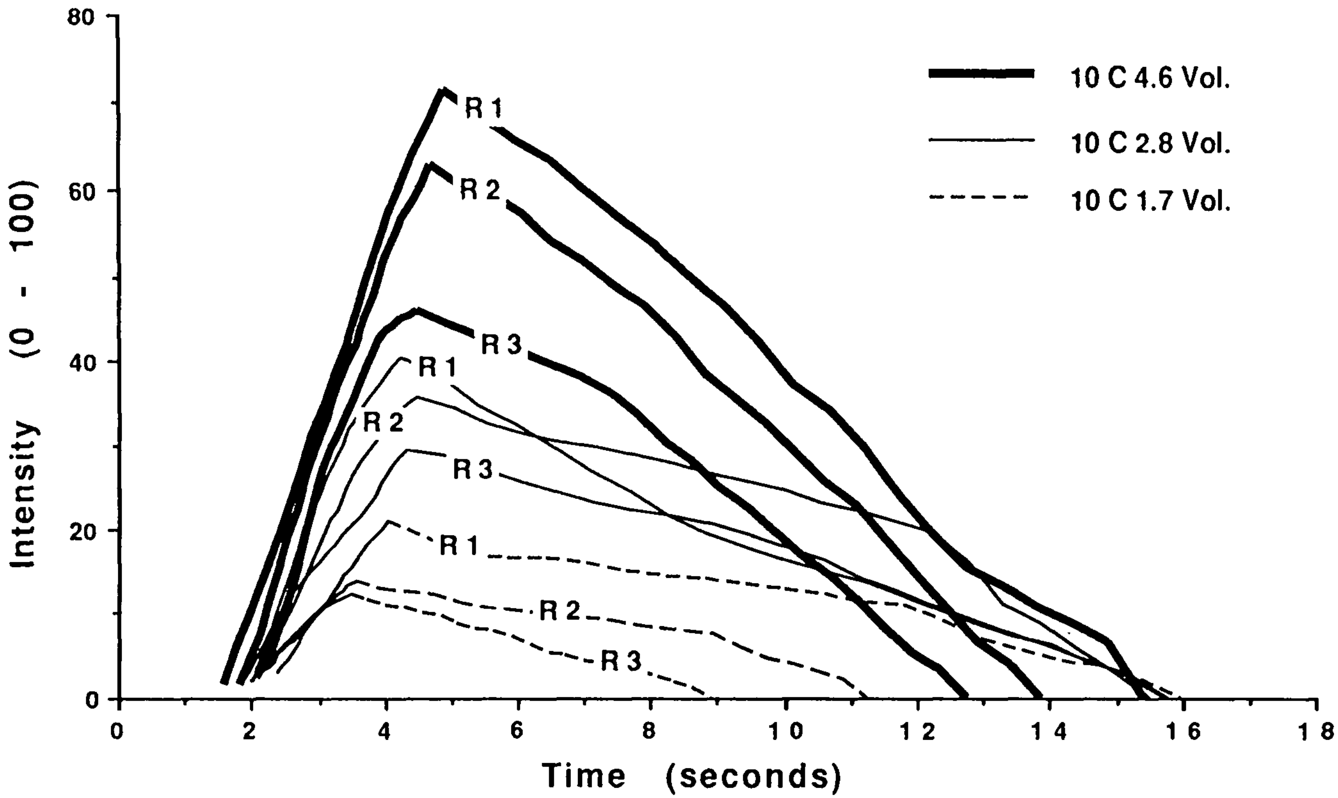


Fig. 3.3. Average replication time-intensity curves for the 10°C carbonated samples for Bite. Values of each curve for each CO_2 level decreased with each successive replication pointing to possible desensitization effects over the course of the experiment. Curves were averaged for each replication using subject values which were normalized for time and intensity.

patterns can be seen among the subjects (Figure 3.4a and 3.4b). The first pattern (exhibited by two subjects) included a steep linear SlopeA leading to Imax and a linear SlopeD which immediately fell off from Imax to the end of perception. One of the two subjects experienced a slight leveling off of sensation toward the end of perception. The second pattern (exhibited by three subjects) began with a linear SlopeA which was similar to the first pattern. However, the slope tapered off, becoming more level as it approached Imax. This was followed by a slow decrease of sensation over a period of a few seconds followed by a steep linear SlopeD to the end of the sensation. In general, this pattern exhibited a much more rounded curve pattern.

Experiment Two: Burn

Relationship of Time-Intensity Parameters

Parameters with the exception of Tinitial and Ratio were significantly correlated with each other ($R^2 \geq .31$). These correlations are reflected in the PCA in which PC1 accounted for 64.4% of the model and was comprised of all parameters with the exception of Tinitial and Ratio (Figure 3.5). PC2 accounted for 14.9% of the model and was comprised principally of Tmax, Tinitial, Ratio, TslopeA and SlopeD. PC3 was comprised of primarily Ratio and accounted for only 7.4% of the model.

Graphing of PC1 and PC2 was useful for separating the samples, but inclusion of PC3 does not generate greater understanding as samples cannot be further characterized. PC1 scores for samples increased dramatically with increasing carbonation as evidenced by the orientation of

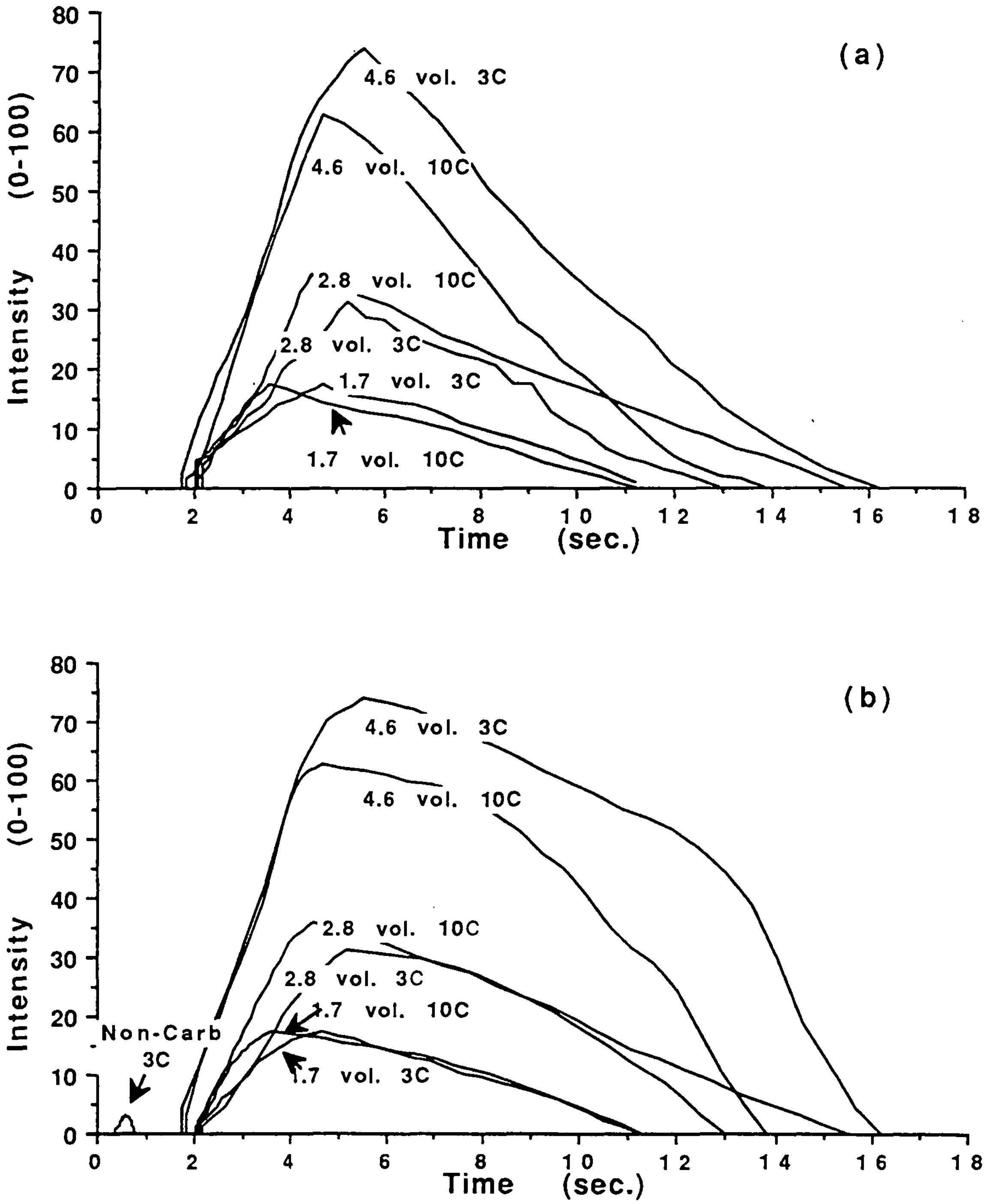


Fig. 3.4. Average curves for Bite for two subjects representing the two different patterns of evaluation by subjects. Pattern one (a) is characteristic of the scoring for three subjects, while Pattern two (b) is characteristic for two subjects.

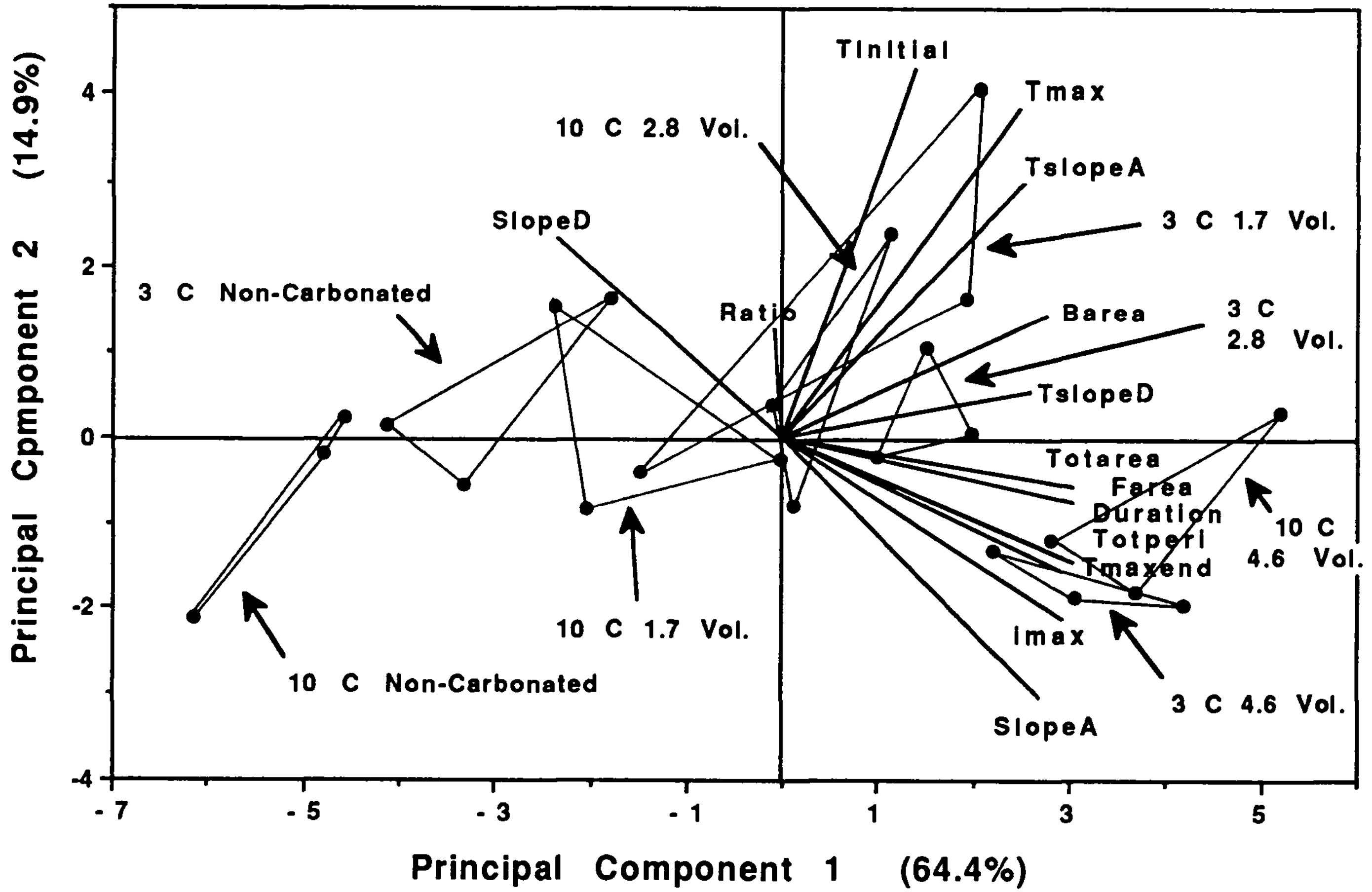


Fig. 3.5. Principal component analysis plot of ratings for Burn Parameters for the eight samples: principal component 1 vs 2. The three connected points for each sample represent three replications across five subjects.

the plotted samples from left to right in Figure 3.5. The 3°C samples generally had higher PC1 scores as they were oriented further to the right compared to 10°C samples. This is true with all levels of samples with the exception of 4.6 vol. samples.

CO_2 Level and Temperature Effects

All parameters with the exception of Ratio were significant ($p \leq .001$) for CO_2 level (Table 3.4). Carbonation levels were significantly different for the non-carbonated samples for all significant parameters. The relationship between these parameters can be visualized by the average curves (Figure 3.6). Tinitial occurred significantly sooner for 4.6 vol. samples. Values for Imax, SlopeA and SlopeD increased significantly with each increase in CO_2 level. All of the remaining parameters, with the exception of Tmax and TslopeA had similar ratings for 1.7 vol. and 2.8 vol., but had significantly higher ratings for 4.6 vol. Again, the values were higher for higher CO_2 levels. Tmax and TslopeA were not different across carbonation levels.

Comparison of the average values (22 for 1.7 vol., 35 for 2.8 vol., and 55 for the 4.6 vol.) represented an approximate 60% increase in intensity between each level.

When the data were analyzed separately for each temperature condition, results changed only slightly. The differences between samples for 10°C samples generally remained the same as for the overall ANOVA. Differences between samples for 3°C were not as clear cut, although the

Table 3.4 **Significance levels[a] for ANOVA main factors (Temperature, CO_2 level, Subject, and Replication) and all interactions for Burn Parameters.**

Source of Variance	Parameters													
	Tinitial	Duration	Tmax	Imax	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
Temperature	ns	ns	ns	*	**	ns	ns	ns	ns	ns	ns	**	ns	ns
CO_2 Level	***	***	***	***	***	***	***	***	***	***	**	***	ns	***
Temp X CO_2	ns	ns	*	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Subject	***	***	***	***	***	***	***	***	***	***	***	***	*	***
Replication	ns	*	*	ns	**	**	**	ns	**	ns	*	*	ns	*
Subject X Rep	**	**	***	ns	*	ns	*	ns	**	ns	**	*	ns	**
Temp X Subject	*	**	ns	**	***	**	ns	ns	ns	*	*	***	ns	**
Temp X Rep	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
CO_2 X Subject	***	*	**	***	**	***	***	**	ns	*	ns	**	ns	***
CO_2 X Rep	**	ns	ns	ns	ns	ns	ns	*	ns	ns	ns	ns	ns	ns

[a] ns not significant at $p \leq .05$
*, **, *** refers to significance at $p \leq .05$, $p \leq .01$, and $p \leq .001$, respectively

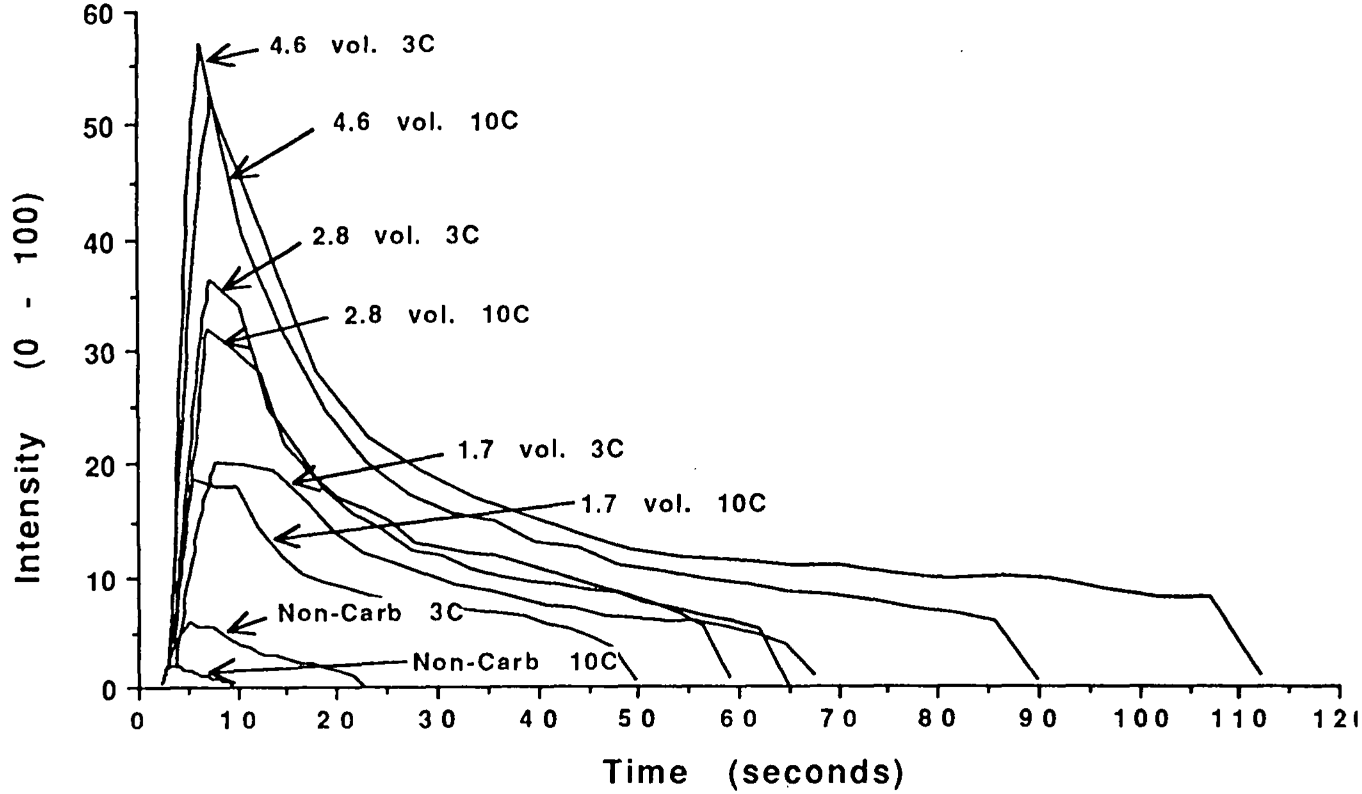


Fig. 3.6 Average time-intensity curves for all samples for Burn. Curves were averaged using subject values which were normalized for time and intensity.

overall trend of higher values for higher CO_2 levels remained. TslopeD and Barea, as well as Ratio, were not significant for 3°C samples.

Values for Imax and the other significant parameters, except Tinitial, had slightly higher values for 3°C samples. Overall, subjects found more Burn associated with 3°C samples.

Temperature X CO_2 level interactions were not significant, with the exception of Tmax. There were no significant temperature X rep interactions.

Curve Observations

The overall average curves for Burn shed light on a number of other interesting patterns (Figure 3.6). As the CO_2 level increased (not including the non-carbonated samples) a plateau which was apparent for 1.7 vol. and 2.8 vol. samples became less apparent, finally disappearing for 4.6 vol. samples. At the lower levels the initial Burn sensation was less intense, but did not fall off as rapidly.

The general shape of the average curves were very similar to each other. SlopeA was steep for all samples. After the plateau, if it was present, the sensation fell off steeply resulting in high SlopeD values. This steep descending slope did not continue to zero, but instead, became less steep, developing into a low plateau. This gradually decreasing plateau continued until the point where the subject's perception of Burn ended. At this point the slope again fell steeply to zero. The decay curve had an exponential shape. Each curve was, for the most part, oriented above each other by temperature and CO_2 level (3°C samples above 10°C samples for each CO_2 level) indicating that intensity ratings were greater for the 3°C samples compared to the 10°C samples as CO_2 levels increased.

Duration was longer for the 3°C 4.6 vol. sample than for the 10°C sample, but the opposite was true for other CO_2 levels. SlopeA and SlopeD also increased with increasing CO_2 level. Duration is characterized by a long, low intensity, residual burn plateau.

Replication Effects

There were a number of significant rep effects for the overall ANOVA (Table 3.4), but most of these effects disappeared when the data were analyzed separately for each of the temperature levels. A number of changes in the way in which the subjects rated the samples can be seen by the average curves for reps (Figures 3.7). Imax for the 4.6 vol. and 1.7 vol. 3°C samples decreased slightly for successive reps while Imax for the 4.6 vol. and 1.7 vol. 10°C samples increased with successive reps. Plateaus of Imax decreased with successive reps. Duration and SlopeA values were inconsistent among reps. The changes in ratings with each succeeding rep were sufficient to cause the rep effects.

Variability Among Subjects' Time-Intensity Curves

There were significant subject effects for all parameters indicating variability in subjects' ratings (Table 3.4). There were also subject X CO_2 level and subject X temperature interactions resulting primarily from inconsistencies in the rating of Duration.

Differences among the subjects are again evident from the average curves for subjects (Figures 3.8a and 3.8b). There were two general rating patterns for subjects. The first pattern (exhibited by three subjects) was

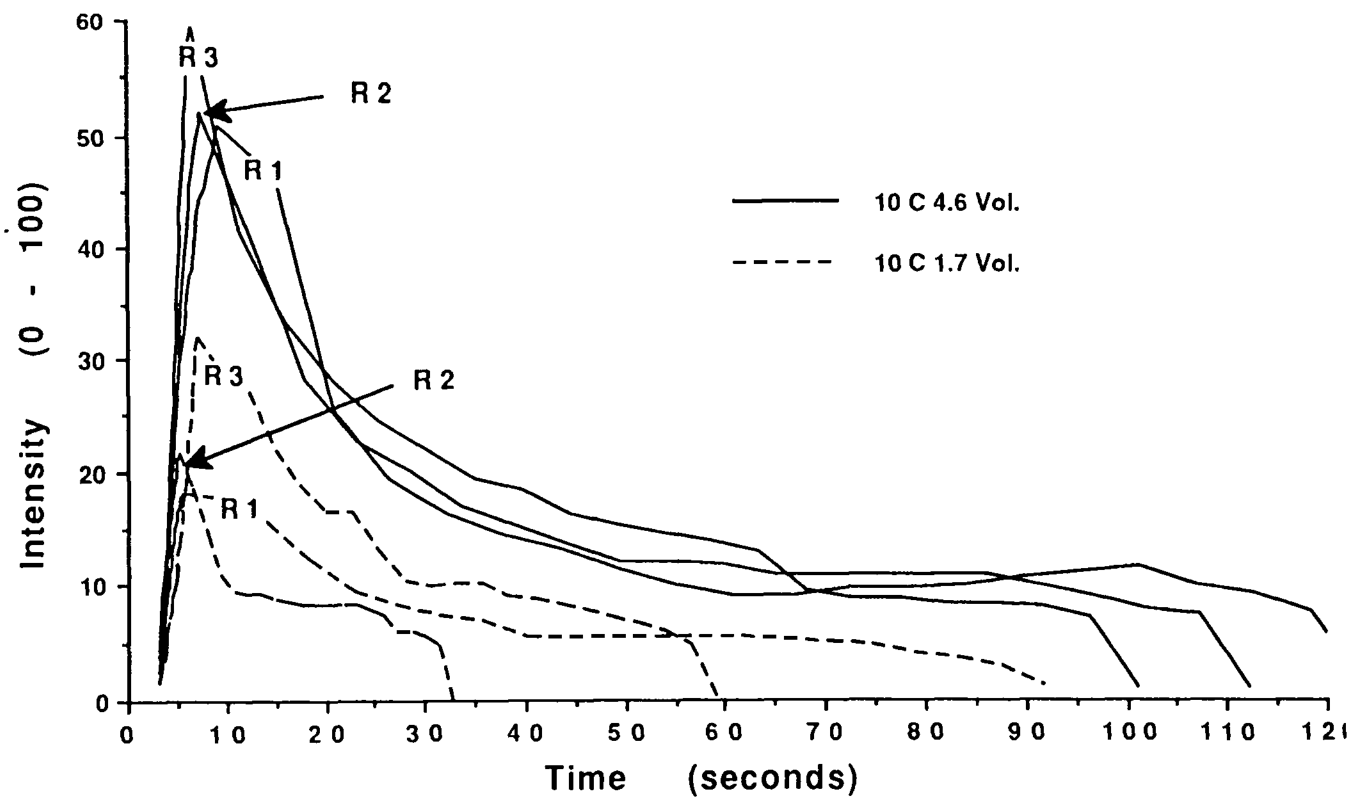


Fig. 3.7. Average replication time-intensity curves for the 1.7 vol. and 4.6 vol. 10°C samples for Burn. Values of each curve for each CO_2 level increased with each successive replication pointing to possible sensitization effects over the course of the experiment. Curves were averaged for each replication using subject values which were normalized for time and intensity.

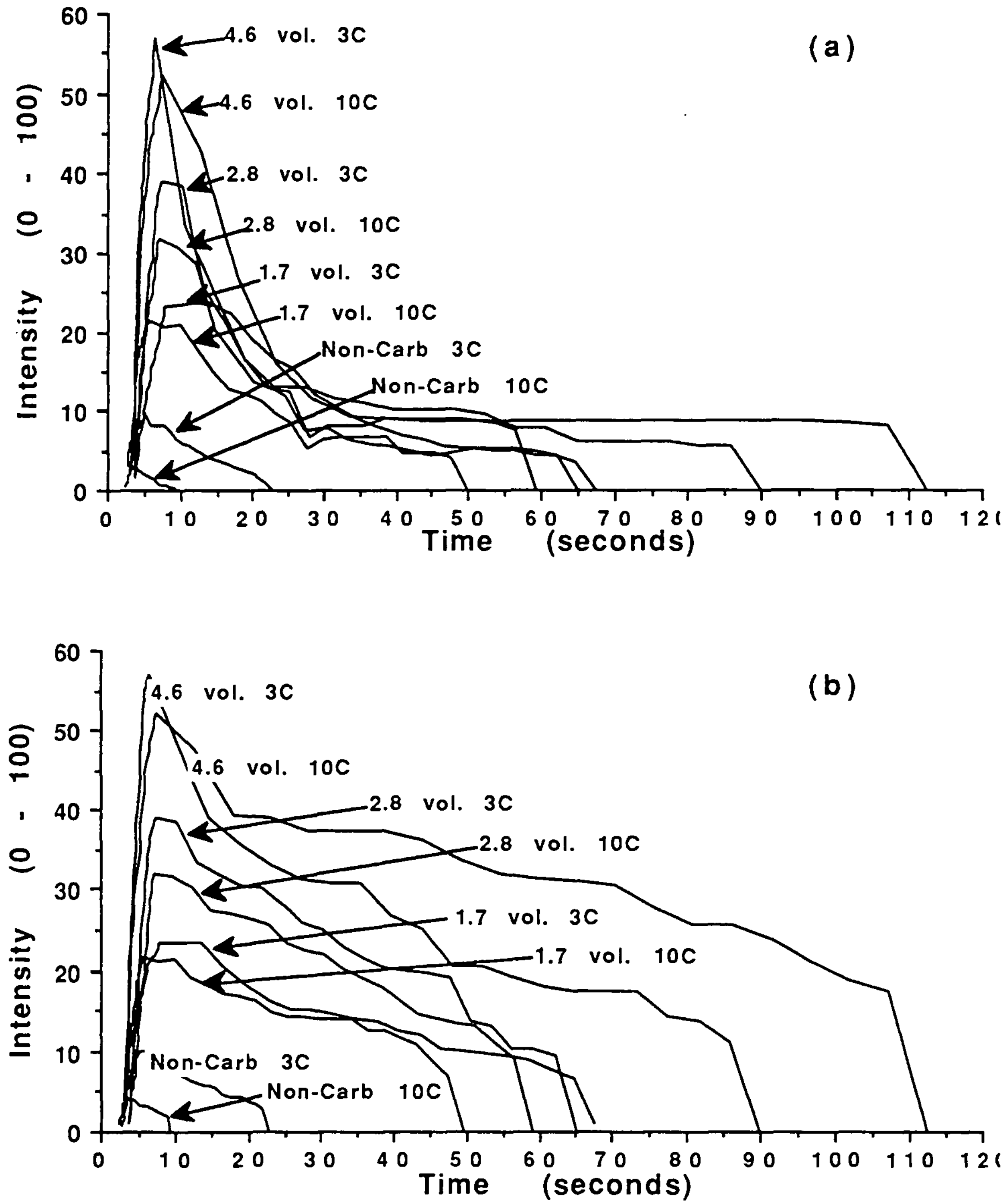


Fig. 3.8. Average curves for Burn for two of the subjects representing the two different patterns of evaluation by subjects. Pattern one (a) is characteristic of the scoring for three of the subjects, while Pattern two (b) is characteristic for two of the subjects.

characterized by a steep SlopeA leading to Imax, followed by a steep SlopeD and a long plateau of residual low intensity Burn sensation. The second pattern (exhibited by two subjects) was again characterized by a steep SlopeA, but was followed by a more gradual decrease in sensation intensity and a final steep decline of the Burn sensation. Four subjects exhibited an Imax plateau in their curve for the non-carbonated, 1.7 vol., and 2.8 vol samples.

Comparisons Between Bite and Burn

The shapes of the average curves, as well as the intensity and time parameters were different for Bite and Burn (Figures 3.2 and 3.6). Tinitial was later (~80%) and Tmax occurred later (~50%) for Burn. Duration and Tmaxend for Burn were much longer (~250%). Imax for Burn had lower ratings (~20%) and Totarea had much higher ratings for Burn (~400%). Although Totarea was much higher for Burn, Barea for Bite and Burn were comparable and all of the Totarea difference was under the decay curve (Farea). Overall, Bite and Burn had similar SlopeA but the decay curves were very different; indicative of the differences in the sensation quality, the decay curve was linear for Bite and exponential for Burn. Imax plateaus, evident in all but the highest carbonation level for Burn, were not evident for Bite. Burn also had a long low intensity residual sensation which was not present for Bite.

Only one subject perceived Bite for the non-carbonated level and only for the 3°C samples, while Burn was experienced for 3°C and 10°C non-carbonated samples by three subjects.

Discussion

Relationship of Time-Intensity Parameters

Previous time-intensity studies have primarily used a maximum intensity and a duration parameter to measure temporal response. Of the studies using parameters similar to this study, few have attempted to define the relationship between the parameters. Cliff and Noble (1990), using most of the same parameters, found that almost all of the parameters were highly correlated but that full characterization of the temporal response of fruitiness and sweetness in a model system required the use of a number of the correlated variables. The correlations and PCA results in this study showed that the parameters can be divided into highly correlated groups of parameters (evident from the PCA graphs) which would essentially describe the time-course of sensation. From these groups, Tinitial, Imax, SlopeA, Tmax, SlopeD, Duration and Ratio provided the most information and effectively described the T-I relationship for Bite and Burn. Each one of these parameters with the exception of Ratio relates to specific receptor responses which are of interest in the perception of irritants. Ratio is useful for comparing Bite and Burn.

CO_2 Level Dependent Sensation

The increasing intensity for Bite and Burn with increasing CO_2 level found in this study is consistent with the findings of past studies of oral CO_2 perception by Cometto-Muñiz and Noriega (1985), Cometto-Muñiz et al. (1987), Yau and McDaniel (1990, 1991), and Green (1992) as well as for nasal pungency perception of CO_2 (Cain and Murphy, 1980; Garcia-Medina and Cain, 1982). Cometto-Muñiz et al. and Green assumed that the sensation produced by CO_2 bubbles was primarily chemogenic and that tactile sensations were of secondary importance. We agree with that assessment but believe mechanoreceptors, responding to evolution of CO_2 from solution, certainly play a role; the bubbling sound and bubbling sensation perceived in the mouth (Harper and McDaniel, 1993) represent the presence of a pressure or mechanical stimulus. Sensitivity of the chorda tympani nerve to mechanical stimulation (Oakley, 1985) and enhancement of the perception of irritation or pain by mechanical stimulation (Green 1990a) would support this. Spatial summation has also been demonstrated by Green (1988 and 1990b) for capsaicin and ethanol stimulation of skin and labial tissue. The higher levels of CO_2 evolved as bubbles from higher CO_2 levels would increase the numbers of receptors affected, thus increasing intensity perception.

Time-Intensity Parameters

Tinitial for bite, occurring at approx 1.7 - 2.2 sec., corresponds very favorably with findings of Green (1988) that 1.5 sec. is the quickest reaction time for irritants. Tinitial for Burn of approx 3.5 sec. is quicker than the 5 sec. latency period found for oral pungency of CO_2 in humans (Cometto-Muniz et al., 1987) and the 4-9 sec latency period found for single trigeminal units in rats (Bryant et al., 1991). We noted some difficulty, as did Green (1991a), totally separating the sensations of Bite and Burn. This may have contributed to the earlier reaction times for Burn in this study. Reaction times for the 4.6 vol. samples occurring sooner for both Bite and Burn compared to the other

CO_2 levels concurs with evidence indicating that higher concentrations of irritants have quicker reaction times (Green, 1988).

Duration of the response for both Bite and Burn, which was generally higher for higher CO_2 levels, corresponds to other studies finding a positive correlation between Duration and concentration for irritants (Lawless, 1984).

The short Duration of Bite (~9 - 14 sec.) compared to Burn (~47 - 110 sec.) supports the hypothesis that a qualitative difference exists between these perceptions as well as the existence of more than one sensory pathway. It has been observed that 'sharper' sensations such as stinging/pricking (Bite) are mediated by rapidly-conducting A-delta nociceptors, while 'duller' sensations such as burning are mediated by slower-conducting c-fiber nociceptors (Price and McHaffie, 1988; Torebjork et al., 1984). The above discussion would also apply to Tmaxend which was also generally longer with increasing CO_2 levels for both Bite and Burn.

Tmax for irritants has not been investigated thoroughly. Tmax occurred later for Bite and Burn as CO_2 levels increased. Tmax also occurred significantly later for Bite in 3°C samples compared to 10°C samples. There are two possible reasons, one physiological and the other procedural. Physiologically, Cain (1981) referring to olfaction and the common chemical sense, felt that the sequestered locus of the free nerve endings of the trigeminal system retarded egress of molecules from the nerve endings, as well as retarding progress toward the endings. Higher concentration with increasing CO_2 level would tend to delay the Tmax because of buildup in the intracellular space in the epithelium. The other possibility is that physically moving the potentiometer lever required more time for higher intensity samples, as Tmax for Bite occurred relatively rapidly. Tmax occurring later for 3°C samples may be partly a function of integrative mechanisms in the central nervous system responding to temporal summation of a number of different types of receptors. This type of mechanism has been shown to exist for the onset of irritant sensation and may also prove applicable in this case (Adriaensen et al., 1980).

SlopeA and SlopeD (rates of maximum rise and decay) and their times (TslopeA and TslopeD) increased with increasing CO_2 level for both Bite and Burn. The slopes were much steeper and the times much shorter for Bite compared to Burn. This was indicative of the quality differences of these sensations; Bite is sharp and quick while Burn comes on more slowly and lasts longer.

Because Ratio was not significant for all carbonation levels for Bite and for all levels for Burn, the mechanism and receptor response affecting the perceptual rise and decay was probably the same for all CO_2 levels and both temperatures within each sensation. It was interesting to find that the subjects responded to a lessor degree, but similarly, to the non-carbonated samples for Burn.

For Bite, Imax and highly correlated area parameters, were higher for the 3°C 4.6 vol. sample compared to the 10°C sample, as expected from previous studies. But these parameters were lower for the 3°C 2.8 vol. sample compared to the 10°C sample. A number of studies have shown that mechanoreceptors, chorda tympani receptors, and receptors in the skin respond more vigorously to cold stimuli (Henzel and Zotterman, 1951; Oakley, 1985; Stevens, 1982; Stevens and Hooper, 1982; Bryant, 1991). Both 3°C and 10°C samples would evoke reaction from receptors sensitive to cold, but 3°C samples should evoke a larger response. Another consideration is concentration of the solutions. It is probable that 10°C samples evolve more CO_2 gas upon ingestion. This is evidenced by higher ratings for 10°C compared to 3°C for bubbly and bubble sound (Harper and McDaniel, 1993). Therefore, concentration of CO_2 in solution would tend to be less in 10°C samples. At the same time there may also be retronasal perception and mechanoreceptor perception of the evolved CO_2 from 10°C samples which would offset the decreased response to lower concentration. Another interesting possibility adding to the complexity of this issue is the difference in perception of colder water vs. warmer water. It is a personal observation that cold water has a rougher texture compared to warmer water. Rough was one of the preliminary descriptors in our earlier study (Harper and McDaniel, 1993a) and it may be playing a part in increasing response to colder samples. The reasons for these mixed results are complex and not altogether explainable.

For Burn, Imax ratings for the 3°C samples were higher than for the 10°C samples. This is contrary to studies of Sizer and Harris (1985) reporting that thresholds for detecting capsaicin were raised by cooling and Green (1990b) finding that cooling could completely inhibit capsaicin irritation. However, it corresponds to other findings that cooling increased ratings of irritation from CO_2 solutions (Yau and McDaniel, 1991; Green, 1992; Harper and McDaniel, 1993).

Barea ratings for Bite and Burn were similar, while Farea ratings for Burn were much higher (~400%). These findings seem to indicate that the mechanism of reception is similar for the rise in sensation, but very different for the decay for these sensations.

Desensitization and Sensitization

Desensitization was noted in the 10°C samples for Bite as ratings for both Imax and Duration decreased with successive reps. Desensitization to irritants is thought to occur with depletion of substance P in the nociceptive system thereby requiring higher concentrations of stimulant to obtain the same physiological response. Chronic capsaicin administration in rats has been shown to have a desensitization effect (Nagy, 1982; Silver et al., 1985) as has topical application of capsaicin in humans (Karrer and Bartoshuk, 1991a and 1991b). This response lasted for as long as six days depending on the concentration and frequency of capsaicin application. In the last study, the levels of response for CO_2 to a very low level of lingual topical capsaicin application did not recover in the 24 hr. time period required for the other tested irritants. Green (1991b) believes that a hiatus in stimulation between the desensitizing stimuli and the test stimuli is required for desensitization. Whether the one or two day time period between each test session in this study caused the desensitization is open to speculation. Perhaps psychological habituation, subjects not finding irritation of Bite as aversive for warmer samples, is occurring. Why this did not occur for 3°C samples is unclear. Contrary to Bite, Imax ratings for Burn for the 10°C 4.6 vol. sample was rated higher for successive reps. The reasons for this are, again, unclear.

Possible sensitization was noted in the 10°C samples for Burn, as ratings increased for successive reps. Perceptual sensitization has been demonstrated for irritants, but only over a very short time period (Stevens and Lawless, 1987; Green and Gelhard, 1989; Green, 1990a). We are unable to explain these findings.

Variability Among Subject Time-Intensity Curves

Subjects rated samples very differently compared to each other. Some subjects had very sharp, steep responses (especially for Bite) while others had more rounded or shallow responses. Many investigators have found that there is a large degree of variability in time-intensity responses by subjects (Harrison and Bernhard, 1984; Schmitt et al, 1984; Ott et al., 1991; Guinard et al., 1986), especially for irritants (Stevens and Lawless, 1986). Van Buuren (1992) noted that time-intensity curves appear to be determined by characteristics that are related to the subject and not the product being tested, "each curve bearing the signature of its creator." Individual differences in anatomy, oral manipulation, use of the intensity scale, and criteria for determining extinction of sensation all play a part in the variability (Noble et al., 1991) and produce large standard deviations (Swartz, 1980). Variability may also depend on a subject's interpretation of perception of cold sensation and how closely this interpretation relates to Bite and Burn. Green (1992) noted that subjects may associate intense sensations of burning and stinging produced by CO_2 with sensations experienced at very cold temperatures. This variability is seen in ratings for non-carbonated samples; only one subject rated Bite (3°C samples) and four subjects rated Burn in non-carbonated samples.

Shape of the Time-Intensity Curves

Previous investigations (Lawless, 1984) have shown that response to chemical irritation (in particular capsaicin) was characterized by a slow perceptual response and a long residual exponential decay of sensation over time. Though the time scale is different, our results proved similar for Burn. Also noticeable was a pattern of convergence of the decay to a similar level of intensity after the steep SlopeD. Although the intensities converged, the Durations were very different. In contrast to Burn, Bite had a much quicker response and a linear decay pattern which is unlike other irritants.

Imax plateaus are evident for lower CO_2 level samples for Burn. Why they are present is unclear, but they may be caused by subjects having a more difficult time feeling the exact time which perception begins to decrease in the lower CO_2 level samples.

Summary and Conclusions

The time-intensity curve for Burn sensation (steep linear rise and long-lived exponential decay slope) was similar to the irritation sensation exhibited by other irritants such as capsaicin. However, the time-course curve for Bite (steep linear rise and decay slopes, and relatively short Duration) was unlike most other irritant sensations previously investigated. The sensations are qualitatively and quantitatively different and are probably perceived by different sensory pathways. Tinitial, Tmax, Imax and Duration were very different for each sensation.

The linear increase in intensity related parameter values with higher CO_2 levels indicated a concentration dependency and suggested that receptor saturation has not yet occurred and a terminal threshold has not been reached. Cold temperature was also shown to enhance the intensity of Burn and Bite as evidenced by higher values of most time-intensity parameters for 3°C samples. This confirms results from previous studies (Yau and McDaniel, 1991; Green, 1992; Harper and McDaniel, 1993a).

Intensity and duration for 10°C samples tended to decrease with successive replications. This suggests possible psychological habituation or desensitization effects.

Most time-intensity parameters were correlated with each other for both Bite and Burn. They could be divided by PCA into two basic groups; parameters which were CO_2 level dependent and those whose ratings were independent of CO_2 level. A considerable amount of subject variability was found as in previous time-intensity studies.

References

Adriaensen, H., Gybels, J., Handwerker, H.O. and Van Hees, J. 1980. Latencies of chemically evoked discharges in human cutaneous nociceptors and of the concurrent subjective sensations. Neuroscience Letters 20:55 (cited by Green, 1988).

Baranowski, J.D. and Park, S.S. 1984. Effect of titratable acidity and carbonation level on the preference of clarified pineapple juice. J. Food Sci. 49:1220.

Bright, R.A., and Potter, N.N. 1979. Acceptability and properties of carbonated apple juice. Food Product Development April, 1979 p. 34.

Bryant, B.P., Komai, M. and Wachowiak, M. 1991. CO_2 sensitive lingual nerve neurons are differently tuned. Oral presentation at 13th annual meeting of Association for Chemoreception Sciences in Sarasota, FL.

Cain, W.S. 1981. Olfaction and the common chemical sense: similarities, differences and interactions. Chap. 6 In *Odor Quality and Chemical Structure* H.R. Moskowitz and C.B. Warren (ed.). ACS Symposium Series 148. ACS, Washington, D.C.

Cain, W.S. and Murphy, C.L. 1980. Interaction between chemoreceptive modalities of odour and irritation. Nature 284:255.

Choi, H.S. and Kosikowski, F.V. 1985. Sweetened plain and flavored carbonated yogurt beverages. J. Dairy Sci. 68:613.

Cliff, M. and Noble, A.C. 1990. Time-intensity evaluation of sweetness and fruitiness and their interaction in a model solution. J. Food Sci. 55(2):450.

Cochran, W.G. and Cox, G.M. 1957. *Experimental Designs*, Wiley, New York, N.Y.

Cometto-Muñiz, J.E. and Noriega, G. 1985. Gender differences in the perception of pungency. Physiol. & Behav. 34:385.

Cometto-Muñiz, J.E., Garcia-Medina, M.R., Calviño, A.M. and Noriega, G. 1987. Interactions between CO_2 oral pungency and taste. Perception 16(5):629.

Garcia-Medina, M.R. and Cain, W.S. 1982. Bilateral integration in the common chemical sense. Physiol. & Behav. 29:349.

Green, B.G. 1988. Spatial and temporal factors in the perception of ethanol irritation on the tongue. Percept. & Psychophys. 44(2):108.

Green, B.G. 1990a. Effects of thermal, mechanical and chemical stimulation on the perception of oral irritation. In *Chemical Senses* vol. 2 B.G. Green, J.R. Mason and M.R. Kare (ed.) Marcel Dekker, Inc., New York.

Green, B.G. 1990b. Spatial summation of chemical irritation and itch produced by topical application of capsaicin. Percept. & Psychophys. 48(1):12.

Green, B.G. 1991a. Capsaicin cross-desensitization on the tongue: psychophysical evidence that oral chemical irritation is mediated by more than one sensory pathway. Chem. Senses 16(6):675.

Green, B.G. 1992. The effects of temperature and concentration on the perceived intensity and quality of carbonation. Chem. Senses 17(4):435.

Guinard, J-X., Pangborn, R.M. and Lewis, M.J. 1986 Effects of repeated ingestion on temporal perception of bitterness in beer. Am. Soc. Brew. Chem. J. 44(1):28.

Harper, S.J. and McDaniel, M.R. 1993a. Carbonated water lexicon; temperature and CO_2 level influence on descriptive ratings. J. Food Sci. (In Press)

Harrison, S.K. and Bernhard, R.A. 1984. Time-intensity sensory characteristics of saccharin, xylitol and galactose and their effect on the sweetness of lactose. J. Food Sci. 49:780.

Hensel, H. and Zotterman, Y. 1951. The response of mechanoreceptors to thermal stimulation. J. Physiol. 115:16.

Karrer, T. and Bartoshuk, L. 1991a. Capsaicin desensitization and recovery on the human tongue. Physiol. & Behav. 49:757.

Karrer, T. and Bartoshuk, L. 1991b. Oral capsaicin desensitization and its effects on thermal, tactile and chemical stimuli. Oral presentation at 13th annual meeting of Association for Chemoreception Sciences in Sarasota, FL.

King, R.C., Sims, C.A., Moore, L.F. and Bates, R.P. 1988. Effects of maturity, skin contact and carbonation on the quality of sterile-filtered white muscadine grape juice. J. Food Sci. 53(5):1474.

Lawless, H. 1984. Oral chemical irritation: psychophysical properties. Chem. Senses 9(2):143.

Lee, W.E. and Pangborn, R.M. 1986. Time-intensity: the temporal aspects of sensory perception. Food Technol. 40(11):71.

McBride, R.L. and Richardson, K.C. 1983. Sensory assessment of the shelf life of carbonated soft drinks.

McLellan, M.R., Barnard, J. and Queale, D.T. 1984. Sensory analysis of carbonated apple juice using response surface methodology. J. Food Sci. 49:1595.

Nagy, J.I. 1982. Capsaicin: a chemical probe for sensory neuron mechanisms. In *Handbook of psychopharmacology, Vol 15* Iverson, S.D., Iverson, L.L., Snyder, S.S. (ed.) 185-235 Plenum Press, New York.

Neilson, A.J. 1957. Time-intensity studies. Drug and Cosmetic Industry 80(4):452.

Noble, A.C., Matysiak, N.L., and Bonnans, S. 1991. Factors affecting the time-intensity parameters of sweetness. Food Technol. 45(11):121.

Oakley, B. 1985. Taste responses of human chorda tympani nerve. Chem. Senses 10:469.

Ott, D.B., Edwards, C.L. and Palmer, S.J. 1991. Perceived taste intensity and duration of nutritive and non-nutritive sweeteners in water using time-intensity (T-I) evaluations. J. Food Sci. 56:535.

Pokorny, J., Davidek, Prnka, V. and Davidkova, E. 1986. Nonparametric evaluation of graphical sensory profiles for the analysis of carbonated raspberry beverages. Die Nahrung 30(2):131.

Price, D.D. and McHaffie, J.G. 1988. Effects of heterotopic conditioning stimuli on first and second pain; a psychophysical evaluation in humans. Pain 34:245.

Schmitt, D.J., Thompson, L.J., Malek, D.M. and Munroe, J.H. 1984. An improved method for evaluation of time-intensity data. J. Food Sci. 49:539.

Silver, W.L., Mason, J.R., Marshall, D.A., and Maruniak, J.A. 1985. Rat trigeminal, olfactory and taste responses after capsaicin desensitization. Brain Research 333:45.

Sizer, F. and Harris, H. 1985. The influence of common food additives and temperature on threshold perception of capsaicin. Chem. Senses 10:279.

Stevens, D.A. and Lawless, H.T. 1986. Putting out the fire: effects of tastants on oral chemical irritation. Percept. & Psychophys. 39(5)346.

Stevens, J.C. 1982. Temperature can sharpen tactile acuity. Percept. & Psychophys. 31(6):577.

Stevens, J.C. and Hooper, J.E. 1982. How skin and object temperature influence touch sensation. Percept. & Psychophys. 32(3):282.

Swartz, M. 1980. Sensory screening of synthetic sweeteners using time-intensity evaluations. J. Food Sci. 45:577.

Szczesniak, A.S. 1979. Classification of mouthfeel characteristics of beverages. In *Food Texture and Rheology* P. Sherman (ed.), Academic Press, New York.

Torebjork, H.E., Schady, W. and Ochoa, J. 1984. Sensory correlates of somatic afferent fibre activation. Human Neurobiol. 3:15.

van Buuren, S. 1992. Analyzing time-intensity responses in sensory evaluation. Food Technol. 46(2):101.

Yang, X. 1993. Personal Communication. Dept. of Agricultural Chemistry. Oregon State University, Corvallis, OR.

Yau, N.J.N. and McDaniel, M.R. 1990. The power function of carbonation. J. Sensory Studies 5:117.

Yau, N.J.N. and McDaniel, M.R. 1991. The effect of temperature on carbonation perception. Chem. Senses 16(4):337.

Yau, N.J.N., McDaniel, M.R. and Bodyfelt, F.W. 1989. Sensory evaluation of sweetened flavored carbonated milk beverages. J. Dairy Sci. 72:367.

Zahm and Nagel Co. Inc. 1964. *Zahm practical testing instruments.* Zahm and Nagel. Co. Inc., Buffalo, NY.

EFFECT OF INGESTION CONDITION ON TEMPORAL PERCEPTION OF CARBONATION

Steven J. Harper and Mina R. McDaniel

Department of Food Science and Technology
Sensory Science Laboratory
Oregon State University
Corvallis, OR 97331

Abstract

Perceptual time-intensity ratings of Bite and Burn were investigated at 3°C and 10°C using CO_2 concentrations of 0, 1.7, 2.8, and 4.6 volumes. Increased exposure to CO_2 in solution and increased cooling provided by ingestion of four continuous swallows was compared to the one swallow of a previous study. Bite and Burn sensation of carbonated water was investigated. Time-intensity average curves for Bite for four swallows were found to be of higher intensity, longer Duration, and developed maximum intensity plateaus, while those for Burn exhibited higher maximum intensities. At four swallows, time-intensity parameter correlations were strengthened, subject variability reduced and replication reproducibility improved by increased ease of rating afforded subjects by higher intensity sensations. Increased oral CO_2 perception (Bite and Burn) with higher CO_2 levels (concentrations) and enhancement by cold temperature was reconfirmed. However, beginnings of maximum intensity, Duration, and reaction time perceptual terminal threshold were seen for Bite and Burn as evidenced by drastically diminished rates of increased perception for the highest 3°C, CO_2 level. Combination of high CO_2 concentration, cold temperature, and exposure time induced these effects.

Introduction

In our previous study (Harper and McDaniel, 1993b), the time-intensity profiles of Bite and Burn sensation were determined for the ingestion of 15 ml of sample by swallowing one time. Non-carbonated and 1.7 vol., 2.7 vol., and 4.6 vol. samples of carbonated water were presented at 3°C and 10°C. The concentrations used in that study were representative of levels associated with a wide variety of carbonated beverages (e.g. strawberry soda, beer, colas, sparkling wines). The temperatures were also typical of carbonated beverages consumed on ice and out of the refrigerator. However, the ingestion condition, one swallow, was not typical of normal consumption. An informal observational study of ingestion patterns was conducted; most observed subjects ingested carbonated beverages using three or four consecutive swallows. We decided that investigating four swallows would provide information more indicative of a person's normal experience with a carbonated beverage.

Burn and Bite perception remained the focus of this study as previous studies of CO_2 (Harper and McDaniel, 1993a; Green, 1992) and other irritants (Lawless, 1984) recognized these as predominant sensations for oral chemical irritation. Qualitatively, Bite and Burn represent two very different sensations. The 'sharper' sensation of Bite (stinging/pricking) has been observed to be mediated by rapidly-conducting A-delta nociceptors, while the 'duller' sensation of Burn is mediated by slower-conducting c-fiber nociceptors (Price and McHaffie, 1988; Torebjork et al., 1984).

Increase in carbonation perception by increased CO_2 levels has been investigated by a number of researchers. A power function exponent of 1.1 was determined for the response to increasing concentrations of carbonated water (Cometto-Muñiz and Noriega, 1985). More recently, increased carbonation perception with increasing CO_2 levels has been determined in a number of studies examining various aspects of carbonation perception (Yau and McDaniel, 1990; Green, 1992; Harper and McDaniel, 1993a, 1993b). Increased perception was determined during the course of ascertaining power functions in the first two studies while similar findings were noted in the last two studies during investigations of lexicon development and time-intensity studies of carbonated water. The primary reason for this augmentation is increased concentration of CO_2 in the solution introduced into the oral cavity. Similar results have been seen for ethanol (Green, 1988) and for vanillyl nonamide, capsicum, piperine and ginger oleoresin (Lawless, 1984).

Thermal enhancement of irritation has been examined with interesting findings. For most irritants (capsaicin, piperine, ethanol, and sodium chloride) intensity ratings of perceived oral irritation were reduced by cooling (Stevens and Lawless, 1986; Green, 1986, 1990a). However, with oral CO_2 perception, intensity ratings were accentuated by introducing colder solutions into the oral cavity (Yau and McDaniel, 1991; Green, 1992; Harper and McDaniel, 1993a, 1993b).

There have only been three studies which investigated temporal properties of irritants. Reaction times to ethanol application to the tongue were examined (Green, 1988) and irritation decay durations for four other irritants were determined (Lawless, 1984). Reduced reaction times and increased duration were found to coincide with increasing concentration. The third study (Harper and McDaniel, 1993b) examined a number of temporal parameters including reaction times, duration and time to maximum intensity for various concentrations and temperatures of carbonated water.

The primary objective of this study was to investigate how the more typical condition of ingesting four swallows of carbonated water compared with ingesting one swallow (Harper and McDaniel, 1993b). Changes in time-intensity parameters and average curves, CO_2 level effects, and temperature effects were examined to understand the influence of increased exposure of the buccal cavity to CO_2 concentration and cooling.

Materials and Methods

Samples

Commercial, bottled drinking water (Aqua Cool, Portland, OR) was used to produce the samples. The source of the commercial, bottled water was chlorinated Cascade Mountains water. Aqua Cool filtered the water using a series of multi-media filters to remove large particles, followed by filtration through activated carbon and polymeric resins to remove color, odors, and possible chemical contaminates and chlorine. The water was then disinfected using ozone. According to Aqua Cool, the samples contained an average of 1.5 ppm calcium, .31 ppm magnesium, .7 ppm sodium and 1.0 ppm chloride.

Batches of the water were carbonated with commercial carbon dioxide (Industrial Welding, Albany, OR) in a Zahm and Nagel 18.9 liter, stainless steel carbonator (Zahm and Nagel Co., Buffalo, NY). Four carbonation levels were produced [non-carbonated, 1.69 volume (vol.) (SD=.05), 2.75 vol. (SD=.058), and 4.63 vol. (SD=.064)]. The lowest level was chosen because it represented a level at which subjects could differentiate descriptors (Harper and McDaniel, 1993). The other levels were chosen based on prior research by Yau and McDaniel (1990) and represent levels in which a doubling of overall carbonation was perceived based on the power function for overall carbonation. The 1.69 (1.7) vol. CO_2 samples were produced using water, bottles, and CO_2 maintained at 21°C. The 2.75 (2.8) and 4.63 (4.6) vol. CO_2 samples were produced using 21°C CO_2 and 1°C water, with bottles and carbonator immersed in ice. 18.9 liter batches of each level were produced. All samples were bottled in 828 ml, green, glass bottles and stored at 1°C until evaluated.

The pH of the samples was 6.04 for non-carbonated, 3.71 for 1.7 vol. CO_2, 3.65 for 2.8 vol. CO_2, and 3.51 for 4.6 vol. CO_2. pH was measured at 22°C with a Corning 125 pH meter with a Sensorex epoxy-body, sealed-reference combination electrode (S200C). The pH meter was calibrated with buffers of pH 3 and 7 (Microessential Laboratory, Brooklyn, NY).

Carbonation levels were measured using a Zahm and Nagel piercing device (Zahm and Nagel Co., Buffalo, NY). Sample temperatures and headspace pressures were measured after agitation of bottles. Temperature and pressure readings were converted to "volumes CO_2 per volume water" by using a conversion table (Zahm and Nagel Co., 1964). One volume is defined as the amount of CO_2 dissolved in water at equilibrium, at 15.56°C, and at one atmosphere pressure.

Training and Testing

Training. Five subjects, all students or faculty of the Department of Food Science and Technology at Oregon State University, participated in training and subsequent evaluation of the carbonated samples. Subjects were consumers of carbonated beverages and had participated in a previous panel developing and evaluating descriptors for carbonated water.

Eight one-hour training sessions were conducted to familiarize subjects with the T-I equipment and to re-familiarize the subjects with the samples and the descriptors "bite" and "burn". Bite is a sharp sensation with a quick onset experienced primarily in the oral cavity. Burn is a lingering sensation with a longer time to onset and a perception of increased temperature and irritation. Practice T-I curves for each subject were internally evaluated to make certain all subjects understood the stimulus to be evaluated and were using the T-I equipment properly.

A dilute solution of cayenne pepper (.1 g cayenne pepper (McCormick Co., Baltimore, MD)/600 g H_2O) was used as a reference standard to help subjects relate to burn perception.

Testing procedure. Testing was conducted in the Sensory Science Laboratory at Oregon State University, Corvallis, OR, in individual booths. Each individual booth was equipped with a computer monitor for providing testing instructions to the subjects, and with a data acquisition device for evaluating the intensity of the samples. The data acquisition device was a 15 cm linear potentiometer with a knob which could be moved uni-directionally from left to right. A scale which was anchored with "none" and "extreme" on the ends with an unlabeled halfway marking in the middle was attached to the potentiometer. A computerized system called DASSIE (Data Acquisition System for Sensory Imput and Evaluation), developed in the Sensory Science Laboratory at Oregon State University, was used to monitor time and collect data. Data was collected every .25 seconds and was saved in a data file after being transformed to a 100-point intensity scale which corresponded to the potentiometer scale.

Eight samples were evaluated by the subjects. These included samples for each of the carbonation levels (non-carbonated, and 1.7 vol., 2.8 vol., and 4.6 vol.) at the two temperature levels (3^oC and 10^oC). The 3^oC samples were packed in ice before and during the test. The 10^oC samples were tempered by placing them in a styrofoam cooler filled with 10^oC water. Samples were maintained in a motionless state at their respective temperatures to minimize the escape of CO_2 during serving.

Samples were presented monadically in 86 ml plastic cups (Sweetheart, Maryland Cup Co., Owings Mills, MD) coded with 3 digit, random numbers. All session samples of each CO_2 level and temperature were carefully and gently poured, without agitation, into the sample cups and served immediately to minimize any loss of carbonation. Each bottle was used for only the one pouring. A warm-up sample of 6^oC, 1.7 vol. or 6^oC, 2.8 vol. water was presented 5 minutes prior to the beginning of the test.

Subjects were instructed to immediately ingest the sample by swallowing four times continuously in a mode normal to each subject. Subjects were instructed to begin rating the sensation as soon as it was perceived. Samples were presented at intervals of 5 minutes and subjects were instructed to rinse their mouths with lukewarm (approx. 25^oC) water between each of the samples.

During the testing, subjects were instructed on the procedures for the evaluation through instructions on the computer monitor in each booth. The evaluation was initiated by a countdown, with a message "to swallow" at zero time. Dassie began recording at zero time. Recording was terminated at the end of 5 minutes or by the subject pushing a button on the data acquisition device. All eight samples were tested at each testing session.

Experimental Design and Statistical Analysis

A balanced, complete block design (Cochran and Cox, 1957) was used with four carbonation levels, two temperature levels, and three replications (reps). A separate set of evaluations was conducted for each of the sensations. Eight randomized orders were developed for each set of evaluations. One of the eight randomized orders was used for each of the sessions in which the samples were presented. It was not possible to randomize the order by panelist because of the concern with maintaining the carbonation level of the samples. Because it was not possible to evaluate all of the subjects in one sitting, many different sessions were conducted to complete each of the sets of three reps. These reps were consecutive trials for each subject using the same lot of samples. However, the subjects did not all test at the same time and, therefore, these reps should be considered to be trials and not true reps.

Principal component analysis (PCA), and correlations were performed using rep means. Panelist, rep, temperature, CO_2 level effects, and interaction effects were tested using fixed model analysis of variance (ANOVA). Fisher's least significant difference (LSD) test ($p \leq .05$) was used as the multiple comparisons test to compare differences among temperatures, CO_2 levels, reps and panelists. All analyses were run using version 6.03 SAS statistical package (SAS Institute, Inc., Cary, NC).

A number of parameters including the typically described maximum intensity, duration, and time to maximum intensity were measured for each time-intensity curve and are defined in Table 4.1.

Average T-I curves were developed for the overall panel, reps, and individual subjects, using the normalization procedure of Liu and MacFie (1990) and a program written in C-language for the IBM computer (Yang,1993). Data was first normalized for time and intensity using average values of the parameters Imax, Tinitial, Tmax, Tdec (time at which perceptual decay accelerates), and Tend (time at which perception ends). The average curves were then produced using the normalized data sets. The overall average and subject curves were drawn using the parameters for the entire panel. Individual rep average values of these parameters were used for each set of rep average curves.

Table 4.1. Definitions of time-intensity parameters

Time-intensity parameter	Definition
Tinitial	Time to initial response.
Duration	Total time of time-intensity response from Tinitial to end of response.
Tmax	Time from initial response to time of maximum intensity.
Tdec	Time at which Imax plateau ends and perceptual decay accelerates
Imax	Maximum intensity of the time-intensity response.
Totarea	Total area under the time-intensity curve.
Perimeter	Total perimeter of the time-intensity curve.
Barea	Area under the time-intensity curve before the maximum intensity.
Farea	Area under the time-intensity curve following the maximum intensity.
Ratio	Ratio of the area before the maximum intensity to the area after the maximum intensity.
SlopeA	Maximum slope of the linear portion of the ascending curve.
SlopeD	Maximum slope of the linear portion of the descending curve.
TslopeA	Total time of the linear portion of the ascending curve.
TslopeD	Total time of the linear portion of the descending curve.
Tmaxend	Duration of time from maximum intensity to end of response.

Results

Experiment One: Bite

PCA

Principal Component 1 (PC1) explained 82.7% of the model while principal component 2 (PC2) accounted for 11.3% (Figure 4.1). The remaining components did not play a significant role. All of the parameters of the time-intensity curve were equally important to PC1 with the exception of Tinitial and Ratio which were dominant for PC2 (SlopeD is included in PC1 because of its correlation and negative value by definition). The PC1 parameters all had increasing values with higher CO_2 levels, while the values of PC2 parameters did not increase with higher CO_2 values.

PC1 scores increased as CO_2 level increased. Samples were also differentiated by temperature for every CO_2 level. 3°C samples had higher scores for PC1 than comparable 10°C samples. The combined impact of all the correlated PC1 parameters resulted in scores for the 3°C samples which were comparable to 10°C samples, one CO_2 level lower. Highly carbonated samples related closely to the intensity associated parameters (Imax, area parameters, perimeter, slope parameters). The time parameters and PC2 parameters were associated with the less highly carbonated samples. The non-carbonated samples were characterized by extremely low scores for PC1.

Consistent replication reproducibility is evident from the tight grouping for each sample (Figure 4.1). Consistency was higher with higher CO_2 level.

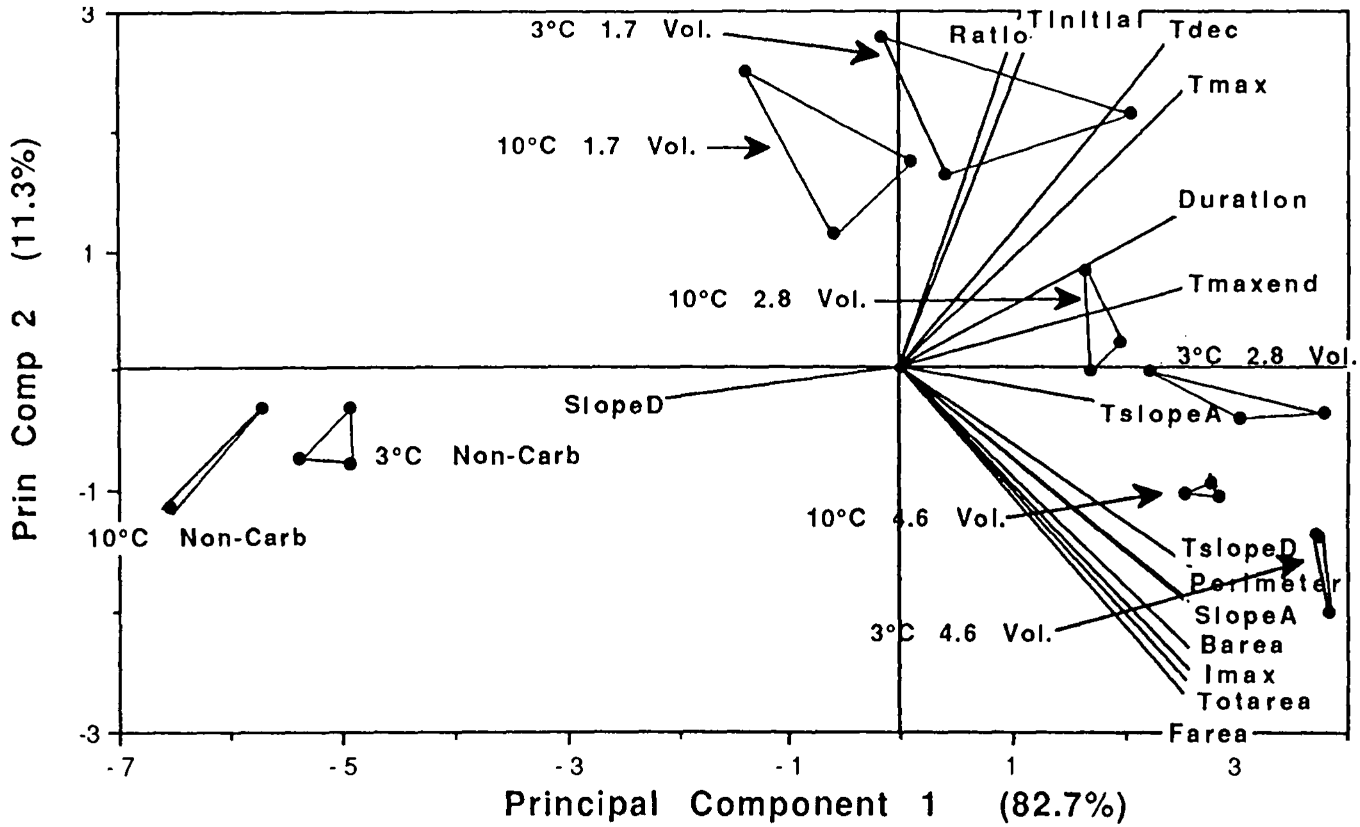


Fig. 4.1. Principal component analysis plot of ratings for Bite Parameters for the eight samples: principal component 1 vs 2. The three connected points for each sample represent three replications across five subjects.

Average Curves

The pattern of rating for the Bite sensation is evident from the shape of the overall average curves for all subjects (Figure 4.2). The initial perception occurred at ~2.5 sec. and was followed by a steep linear perceptual rise in the sensation to a maximum intensity occurring ~4.5 sec. later. A plateau of maximum intensity of ~2.25 sec. was followed by a linear perceptual decay lasting ~8.5 sec.

Four of the five subjects rated Bite in a similar fashion (Figure 4.3a). Only one of the subjects was different having a much broader, gradual curve structure with much longer plateaus (Figure 4.3b). This subject was also the only one perceiving Bite in the non-carbonated samples (3°C only).

CO_2 level and temperature differences described below are easily visualized by examining the overall average curves (Figure 4.2).

CO_2 Level Effects

Imax, area parameters, Perimeter, Duration, and slope parameters all had higher values ($p \leq .001$) at higher CO_2 levels (Table 4.2). All of these except the slope parameters and Duration had higher values for each increase in CO_2 level. The slope parameters and Duration were not significantly different for the 2.8 and 4.6 vol. samples. Tinitial was quicker the higher the carbonation level. Ratio decreased with higher CO_2 level. The plateaus were slightly shorter for the 4.6 vol. CO_2 level.

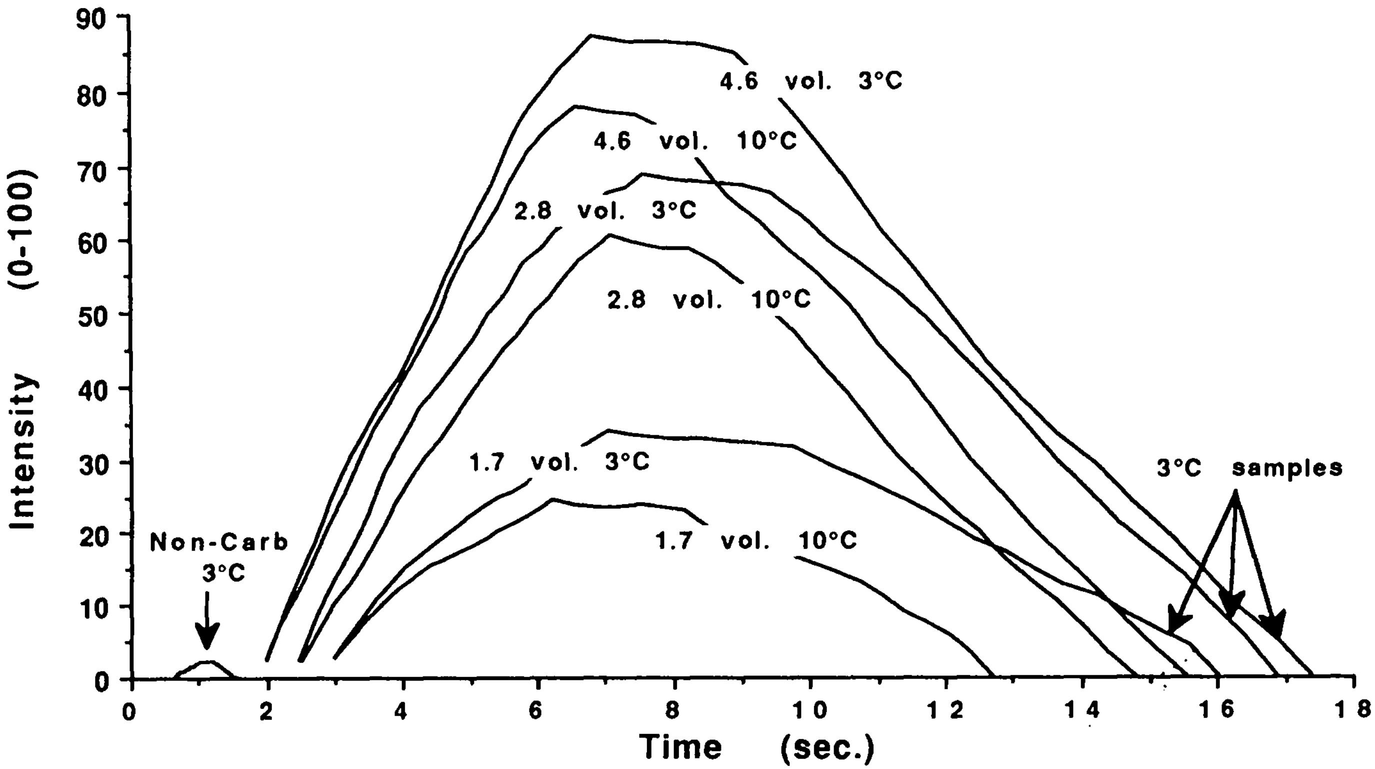
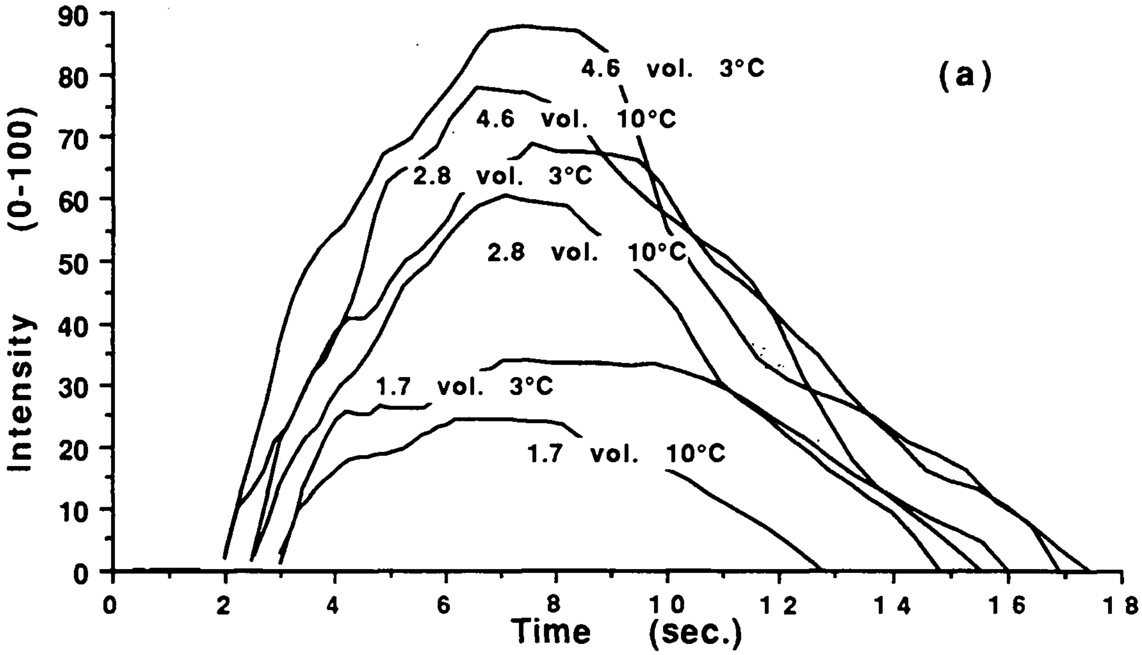


Fig. 4.2. Average time-intensity curves for all samples with the exception of the non-carbonated 10°C sample (not rated) for Bite. Curves were averaged using subject values which were normalized for time and intensity. All 3°C samples had longer Duration as indicated by arrows in lower right corner.



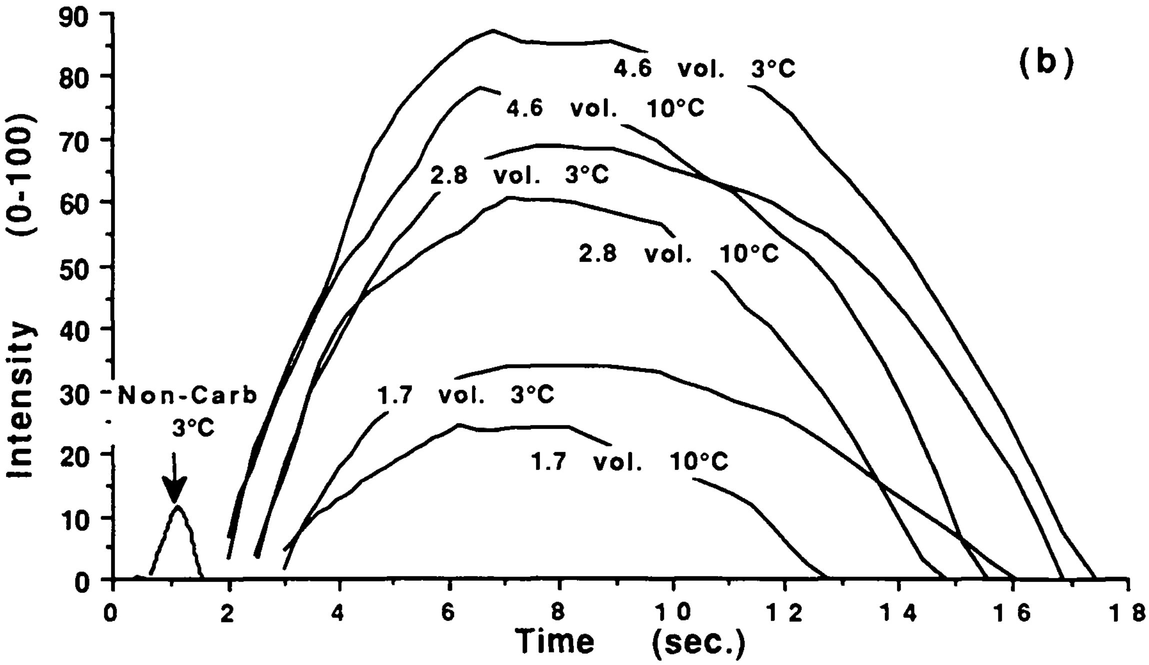


Fig. 4.3. Average curves for Bite for two of the subjects representing the two different patterns of evaluation by subjects. Pattern one (a) is characteristic of the scoring for four of the subjects, while Pattern two (b) is characteristic for one of the subjects.

Table 4.2 Mean ratings, significance, and LSD values for Bite. (a) CO_2 level (b) Temperature

(a)

CO_2 Level	TInitial	Duration	Tmax	Imax	Tdec	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
Non-Carbonated	$.50^{a}$	1.1^{a}	$.8^{a}$	6.0^{a}	$.8^{a}$	15^{a}	13^{a}	3.1^{a}	-3.0^{a}	$.3^{a}$	$.3^{a}$	8^{a}	7^{a}	$.15^{a}$	$.3^{a}$
1.7 Volumes	2.99^{d}	14.4^{a}	6.7^{b}	29.1^{b}	8.9^{b}	218^{b}	77^{b}	10.0^{b}	-9.7^{b}	2.5^{b}	2.8^{b}	73^{b}	135^{b}	$.88^{bc}$	7.7^{b}
2.8 Volumes	2.51^{c}	15.9^{b}	6.7^{b}	64.6^{c}	8.8^{b}	503^{c}	141^{c}	15.7^{c}	-11.9^{c}	4.1^{c}	5.8^{c}	184^{c}	379^{c}	$.75^{bc}$	8.5^{b}
4.6 Volumes	2.01^{b}	16.5^{b}	7.4^{c}	82.5^{d}	8.9^{b}	665^{d}	179^{d}	20.4^{d}	-13.4^{c}	4.2^{c}	6.2^{c}	217^{c}	448^{d}	$.58^{b}$	9.8^{c}
Significance	***	***	***	***	***	***	***	***	***	***	***	***	***	***	***
LSD Values	.38	1.4	.7	6.9	.8	59	18	3.5	2.1	.6	.7	38	44	.18	1.1

*, **, *** significant at .05 level, .01 level, and .001 level, respectively

abcd means within a column followed by the same letter are not significantly different at $p<.05$

(b)

Temperature	TInitial	Duration	Tmax	Imax	Tdec	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
3°C	2.03^{a}	13.0^{a}	5.7^{a}	50.2^{a}	7.2^{a}	415^{a}	113^{a}	13.5^{a}	-10.3^{a}	2.9^{a}	4.0^{a}	141^{a}	274^{a}	$.65^{a}$	7.3^{a}
10°C	1.96^{a}	11.0^{b}	5.1^{b}	40.9^{b}	6.1^{b}	286^{b}	92^{b}	11.2^{a}	-8.7^{b}	2.7^{a}	3.5^{a}	100^{b}	186^{b}	$.53^{a}$	5.8^{b}
Significance	ns	***	*	**	***	***	**	ns	*	ns	ns	**	***	ns	***
LSD Values	.27	1.0	.5	4.9	.6	42	12	2.5	1.5	.4	.5	27	31	.12	.8

ns indicates not significant at .05 level

*, **, *** significant at .05 level, .01 level, and .001 level, respectively

abcd means within a column followed by the same letter are not significantly different at $p<.05$

Temperature Effects

There was a clear separation by temperature across CO_2 levels; the 3°C samples had more Bite than the 10°C samples for every CO_2 level (Table 4.1). All parameters except for Tinitial, Ratio and the slope parameters had significantly higher values or longer times for the 3°C samples. Duration was especially temperature dependent as all 3°C samples had values higher than the 10°C samples (Figure 4.2).

When analyzed separately for each temperature results remain the same with only one exception. Ratio for the 3°C samples decreased substantially with higher CO_2 levels. This did not occur with the 10°C samples.

Experiment Two: Burn

PCA

PC1 (63.8% of the model) was comprised of all parameters associated with increasing ratings of Burn at higher CO_2 levels (Figure 4.4). These included Imax, area parameters, slope parameters, (SlopeD is a component because of its correlation and negative value by definition), Duration and Tmaxend. PC2 (12.9% of the model) was strongly dominated by loadings for Tinitial, Tmax, Tdec and Ratio (parameters whose values did not increase with higher CO_2 level). .

PC1 scores increased with higher CO_2 levels, 3°C samples having much higher scores than 10°C samples. The temperature effect was even greater for Burn than for Bite. Both 4.6 vol. and 2.8 vol. samples for 3°C had

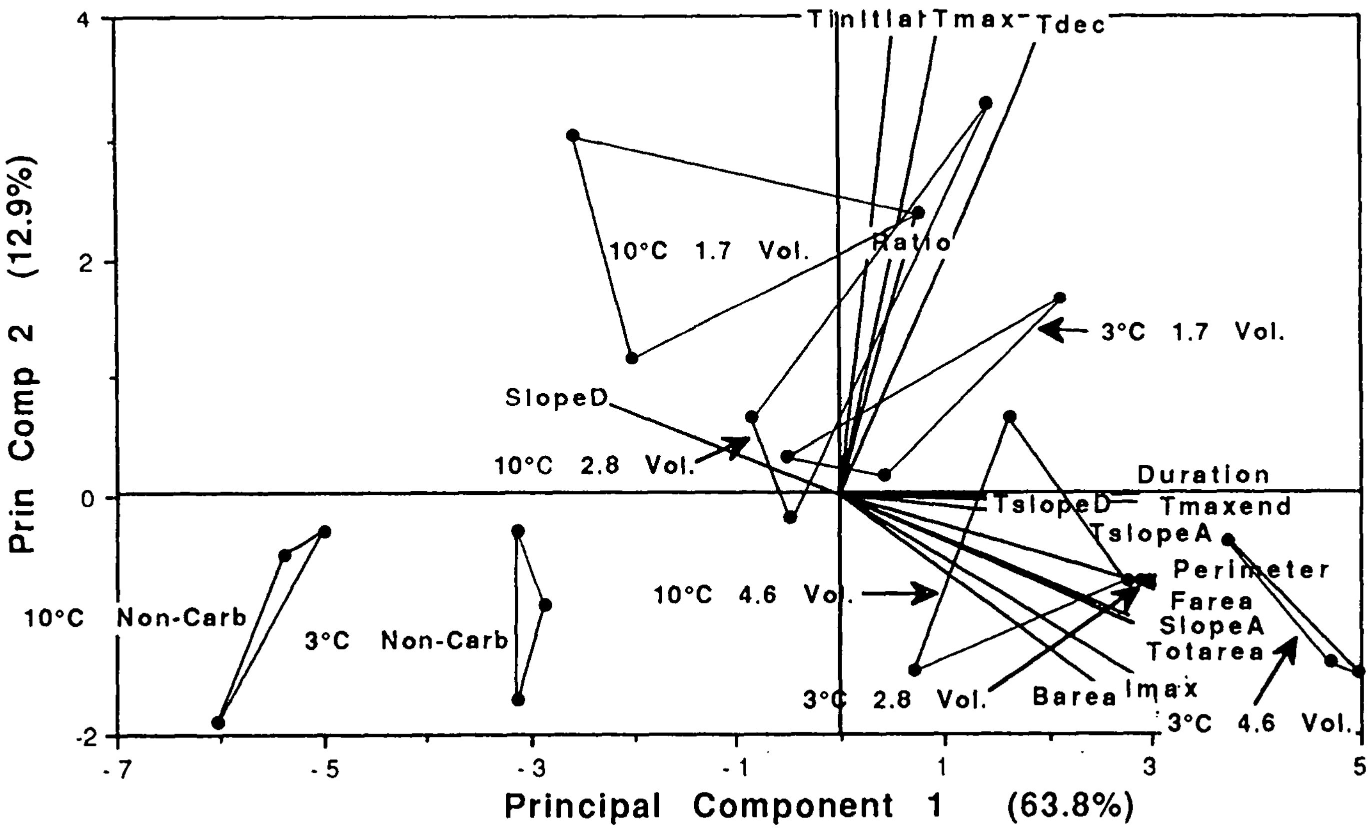


Fig. 4.4. Principal component analysis plot of ratings for Burn Parameters for the eight samples: principal component 1 vs 2. The three connected points for each sample represent three replications across five subjects.

higher PC1 scores than the 10°C 4.6 vol. sample. The 3°C 1.7 vol. sample also had higher scores than the 1.7 vol. and 2.8 vol. samples for 10°C.

The lower CO_2 level samples, as with Bite, were associated with parameters (PC2) which were not affected by increasing CO_2 level.

There was some replication variation as evidenced by the replication groupings for each sample. More variation was seen for the 1.7 vol. samples compared to others indicating potentially more difficulty rating the sample.

Principal component 3 which comprised 8.9% of the model did not provide any additional separation.

Average Curves

The pattern of rating for the Burn sensation, evident from the overall average curves for all subjects, was different compared to that for Bite (Figure 4.5). The initial perception occurred at ~5-6 sec. and was again followed by a steep linear perceptual rise in the sensation to a maximum intensity. The maximum intensity occurred ~5 sec. later followed by a plateau of maximum intensity having a duration ~2-7 sec. An exponentially shaped perceptual decay curve ensued, having a duration of approximately 30 to 97 sec., ending with a sudden drop to zero. The sudden extinction most likely resulted from uncertainty of the actual end of perception.

There were two patterns of Burn rating for subjects (Figure 4.6). Two subjects' ratings exhibited a steep perceptual slope rise, followed by a fairly steep exponentially shaped decay slope, and culminating in a long lasting, low intensity residual sensation plateau (Figure 4.6a). Two subjects had comparatively broad structures with decay curves which were much more gradual (Figure 4.6b). One subject had a curve structure which was a

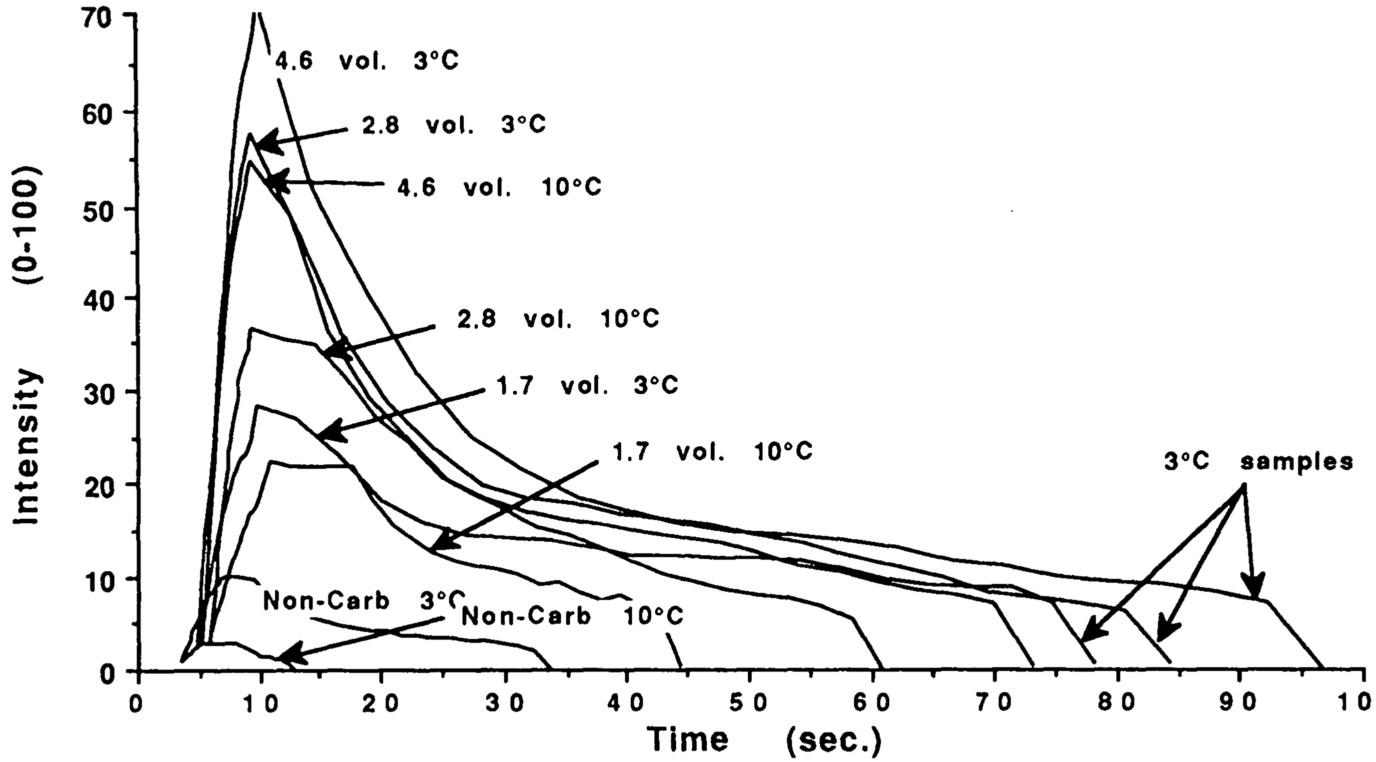


Fig. 4.5. Average time-intensity curves for all samples for Burn. Curves were averaged using subject values which were normalized for time and intensity. All 3°C samples had longer Duration as indicated by arrows in lower right corner.

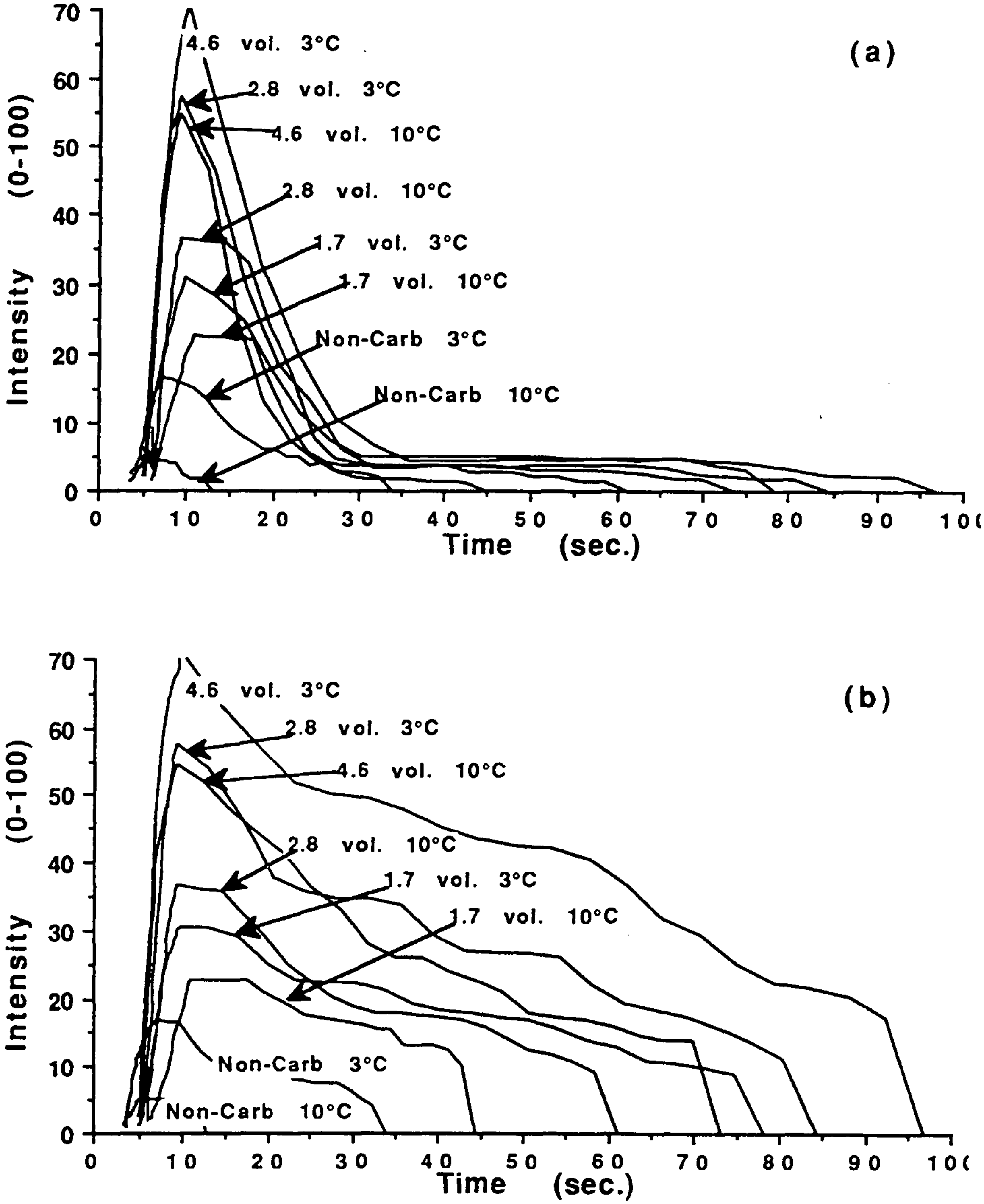


Fig. 4.6. Average curves for Burn representing the two different patterns of evaluation by subjects. Pattern one (a) is characteristic of the scoring for two of the subjects and Pattern two (b) is characteristic for two of the subjects. One of the subjects had average curves which were a combination of the two patterns.

combination of these patterns. Three of the subjects rated Burn for the non-carbonated samples.

CO_2 Level Effects

All parameter values increased with higher CO_2 level except for Tinitial, Tmax, and Tdec (Table 4.3). Tinitial was similar for all levels of carbonation. Tdec decreased with higher CO_2 levels and Tmax remained the same. This resulted in much longer Imax plateaus for the less highly carbonated samples, increasing from 1.2 sec. for 4.6 vol samples to 6.7 sec. for the 1.7 vol. samples (Figure 4.5). Imax increased between the 1.7 vol. and 2.8 vol. samples (80%) while increasing less between 2.8 vol. and 4.6 vol. (34%), indicating that we may be reaching a saturation point for receptor perception.

When analyzed separately for temperature an interesting observation is evident. First, Tinitial was not different by CO_2 level for 3°C samples, but occurred more quickly with higher CO_2 levels for 10°C samples. Secondly, Duration, Tmaxend and Totarea increased with higher CO_2 levels for the 10°C samples but not for 3°C samples. This indicates that with four swallows, combined with cold, a leveling off of perception is being approached for 3°C samples. This was not the case with 10°C samples.

Temperature Effects

Imax, area parameters, Duration, and Tmaxend all had significantly greater values for 3°C samples (Table 4.3). Duration and Tmaxend lasted much longer (~55%); each level for 3°C samples had higher values than any

Table 4.3 Mean ratings, significance, and LSD values for Burn. (a) CO_2 Level (b) Temperature

(a)

CO_2 Level	Tinitial	Duration	Tmax	Imax	Tdec	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
Non-Carbonated	3.5^{a}	22.1^{a}	6.0^{a}	10.8^{a}	9.1^{a}	373^{a}	43^{a}	3.9^{a}	-1.4^{a}	1.4^{a}	3.1^{a}	50^{a}	323^{a}	$.16^{a}$	17.4^{a}
1.7 Volumes	5.7^{b}	55.7^{b}	9.5^{b}	26.4^{b}	16.2^{c}	946^{b}	129^{b}	9.1^{b}	-3.2^{b}	2.5^{b}	5.6^{b}	73^{ab}	873^{b}	$.30^{a}$	51.0^{b}
2.8 Volumes	5.6^{b}	67.2^{bc}	9.8^{b}	47.0^{c}	14.0^{bc}	1131^{b}	190^{c}	14.9^{c}	-4.9^{c}	3.3^{c}	6.9^{bc}	109^{b}	1022^{b}	$.27^{a}$	63.2^{bc}
4.6 Volumes	5.0^{b}	80.1^{c}	10.4^{b}	62.8^{d}	11.6^{ab}	1516^{c}	232^{d}	19.8^{d}	-5.6^{c}	3.4^{c}	7.9^{c}	175^{c}	1341^{c}	$.25^{a}$	75.2^{c}
Significance	***	***	***	***	***	***	***	***	***	***	***	***	***	***	***
LSD Values	.82	16.5	1.4	4.8	3.8	263	30	3.5	1.1	.5	2.0	40	274	.17	16.9

*, **, *** significant at .05 level, .01 level, and .001 level, respectively

abcd means within a column followed by the same letter are not significantly different at $p<.05$

(b)

Temperature	Tinitial	Duration	Tmax	Imax	Tdec	Totarea	Perimeter	SlopeA	SlopeD	TslopeA	TslopeD	Barea	Farea	Ratio	Tmaxend
$3^{o}C$	4.8^{a}	68.6^{a}	9.1^{a}	43.9^{a}	12.0^{a}	1358^{a}	180^{a}	13.4^{a}	-4.2^{a}	2.9^{a}	6.5^{a}	132^{a}	1226^{a}	$.22^{a}$	64.2^{a}
$10^{o}C$	5.1^{a}	43.9^{b}	8.7^{a}	29.6^{b}	13.5^{a}	625^{b}	118^{b}	10.4^{b}	-3.3^{b}	2.4^{b}	5.3^{a}	72^{b}	553^{b}	$.25^{a}$	39.2^{b}
Significance	ns	***	ns	***	ns	**	***	*	*	**	ns	***	***	ns	***
LSD Values	.6	11.7	1.0	3.4	2.7	186	21	2.5	.8	.4	1.4	28	194	.12	12.0

ns indicates not significant at .05 level

*, **, *** significant at .05 level, .01 level, and .001 level, respectively

10°C samples. The 3°C samples had much higher levels of Imax at all CO_2 levels (~50%). This effect was accentuated for 1.7 vol. and 2.8 vol. samples; the 3°C 2.8 vol. sample was rated higher than the 4.6 vol. sample for 10°C (Figure 4.5).

Bite: Comparison of One Swallow vs Four Swallows

As noted in the introduction, one of the primary objectives of this study was to compare information we obtained to that from our previous work (Harper and McDaniel, 1993b) in which identical samples and experimental design were used but which tested ingestion of only one swallow. The comparisons which follow all relate to the previous work.

Parameter composition of PC1 and PC2 and their contribution to the model were similar. The primary difference was strengthening of the correlation of the PC1 and PC2 parameters. Separation of each CO_2 level by temperature and intensification of the relationship of increasing intensity values with higher CO_2 levels for 3°C samples resulted from four swallows. Swallowing four times vs one time increased the number of parameters which were significant for temperature and substantially heightened the effect of cold temperature and CO_2 level on Bite perception. Replication was also more consistent for four swallows, as sample grouping by replication was much tighter.

The shape of average curves were more symmetrical for the four swallow condition compared to the steep asymmetry for one swallow . Subjects rated much more consistently with each other and for each replication, resulting in less variation for four swallows. This was evidenced by the average curves for subjects and by the few replication effects for four swallows compared to one swallow.

Desensitization or sensitization effects found in the one swallow condition were not apparent in the one swallow condition.

Intensity Changes

Imax was 35-65% higher for four swallows vs one swallow depending on CO_2 level (Figure 4.7). 3°C sample Imax ratings were higher for each CO_2 level and separation between 3°C and 10°C samples increased to ~25% for four swallows compared to ~10% for one swallow. Imax plateaus were evident for the four swallow condition. This was not the case for the one swallow condition. There was a constant linear increase of Imax (~100% between each CO_2 level) for one swallow. The linear increase continued between the 1.7 vol. and 2.8 vol. for four swallows, but suddenly decreased to only ~25% from 2.8 vol. to 4.6 vol. indicating a leveling off of perception. The Imax, Totarea and Perimeter ratings of each CO_2 level for one swallow were comparable to the next higher level for four swallows. Ratio increased greatly for 3°C samples (primarily in 1.7 and 2.8 vol. samples) indicating that more of the perception occurs before Imax for four swallows.

Time Related Changes

Tinitial occurred later for four swallows but may have resulted from the need to concentrate on rating and swallowing at the same time. The quicker Tinitial response with higher CO_2 level was even more evident for four swallows. Tmax occurred later for four swallows, especially for the 1.7 vol.

samples. As a consequence, the perception time for increasing sensation was ~65% longer for four swallows.

Duration was longer for four swallows, especially for 3°C samples and 1.7 vol. samples. All 3°C carbonated samples had Duration values longer than those for 10°C samples.

TslopeA increased ~90% for four swallows indicating that perception time of increasing intensity sensation increased. However, SlopeD, TslopeD and Tmaxend were unchanged indicating that the decay perception was the same.

Burn: Comparison of One Swallow vs Four Swallows

The parameter scores for principal components were similar for both swallowing conditions but again, as with Bite, the correlation of PC1 and PC2 components were accentuated. The major difference between the swallowing conditions is a clear separation by temperature across CO_2 levels for the four swallow condition, but not for the one swallow condition. The 3°C samples have much higher PC1 scores for each carbonated and non-carbonated level for the four swallow condition while 10°C samples were comparable across conditions.

The overall curve structure was very similar for both conditions. The only differences were higher Imax and slightly higher residual sensation of decay for four swallows.

While only Imax and area parameters were significant for temperature for one swallow, all parameters except Tinitial, Tmax, Tdec, and Ratio were significant for four swallows. There were few replication effects for four swallows, while there were many for one swallow. This may indicate more difficulty in rating Burn with a lesser impact of CO_2. This was also evidenced by more variation for the 1.7 vol. level for four swallows.

Intensity Changes

Swallowing four times vs one time accentuated the effect of cold temperature and CO_2 level on the Burn perception as evidenced by increasing levels of Imax and associated parameters (especially for the 3°C samples). Imax was ~25% higher and there was a larger difference between 3°C and 10°C samples for four swallows. Most of the increase was for 3°C samples (including the non-carbonated samples). Imax was consistently higher across all CO_2 levels for four swallows. This was not true for one swallow. Totarea and Perimeter did not change for 10°C samples but were 40% higher for four swallow 3°C samples.

Time Related Changes

Tinitial occurred ~50% later for four swallows indicating possible interference from the Bite sensation. Tmax occurred ~40% later for four swallows. Even with Tinitial occurring later the perception time for increasing sensation was increased by ~30%. Tdec also occurred ~40% later resulting in plateau length remaining the same.

The average Duration for carbonated samples for both swallowing conditions was the same, although the distribution was different. Duration for 3°C samples increased while decreasing for 10°C samples for four swallows. This resulted in 3°C samples having longer Duration at each CO_2 level and all having greater values than any of the 10°C samples for the four swallow condition. Duration was very different for one swallow where there was not a difference by temperature. The greatest increase was seen for the 3°C non-carbonated sample.

Average Tmaxend was the same for both swallowing conditions, but for four swallows, the 1.7 vol. and 2.8 vol. samples for 3°C increased considerably. No increase was found for 10°C samples.

Discussion

Increase of Bite and Burn Carbonation Perception Caused by CO_2 Levels

Increased carbonation perception (specifically Bite and Burn) was again confirmed in these experiments. Our previous studies (Harper and McDaniel, 1993a; 1993b) as well as studies by Cometto-Muñiz and Noriega (1985), Cometto-Muniz et al. (1987), Yau and McDaniel (1990, 1991) and Green (1992) demonstrated this dependency. In all of these studies the intensity of sensation (exemplified by Imax in this study) was investigated.

For Bite, not only was intensity perception enhanced, but Duration of response also increased with higher CO_2 levels. Lawless (1984) found a positive correlation between Duration and concentration for a number of irritants (vanillyl noneamide, capsaicin, piperine and ginger oleoresin). In this study Duration leveled off at higher CO_2 levels; it appears that if higher CO_2 levels were investigated a limiting maximum intensity would be found. We believe that the 4.6 vol. served under the conditions used in this study came close to this limit.

The Tinitial response was also quicker with higher CO_2 levels for both Bite and Burn. This concurs with previous studies indicating that higher concentrations of irritants (ethanol and CO_2) result in quicker reaction times (Green, 1988; Harper and McDaniel, 1993b).

For Burn, Tdec decreased with increasing CO_2 level and the Imax plateaus became very short. Maximum intensity was not sustained as long at the higher CO_2 levels. This may result from possible confusion by subjects of Bite and Burn.

Enhancement of Bite and Burn Carbonation Perception by Cold Temperature

As mentioned above, there was a clear enhancement by cold of not only intensity perception, but also of the Duration of the perception. For both Bite and Burn, Imax and Duration were greatly enhanced for 3°C samples indicating that temperature affected perception. This was true at all CO_2 levels for Bite and especially accentuated for 1.7 vol. and 2.8 vol. samples for Burn. There was an additive effect of cold receptors and CO_2 receptors which has been noted in previous studies of oral CO_2 perception (Yau and McDaniel, 1991; Green, 1992; Harper and McDaniel, 1993a, 1993b). But, because the 3°C 4.6 vol sample did not see the same increase, we also believe that a terminal threshold is being approached. This already happened for Burn Duration as the average Durations were not greater than those found for one swallow in our previous study (Harper and McDaniel, 1993b).

For Burn, Tinitial occurred more quickly for higher CO_2 levels of 10°C samples but not for 3°C samples. This may be a function of concentration and receptor saturation. 10°C samples may be evolving more CO_2 gas due to warmer conditions thus reducing the concentration compared to the 3°C samples. While initial reaction time may have reached its limit with 3°C samples, the concentration limit may not have been reached in 10°C samples.

Tmax occurred later for 3°C samples. This was also found to be the case in the previous study (Harper and McDaniel, 1993b) and may be a function of integrative mechanisms in the central nervous system responding to temporal summation of responses from the different types of receptors involved with CO_2 perception and cold perception. Adriaensen et al. (1980) has noted this for onset of irritant sensation.

Increased Bite and Burn Carbonation Perception Caused by Ingestion Condition

The additional swallowing substantially accentuated the effect of cold and CO_2 level on Bite and Burn perception. This included intensity associated ratings and length of response. Although concentration of the samples and temperature of serving were not different from the previous study (Harper and McDaniel), exposure time of the oral membrane was increased from ~ 1 sec to 3-4 seconds. Increased cooling of membranes and potentially increased CO_2 exposure would result.

Most time-intensity parameters were highly correlated as indicated by the PCA for both Bite and Burn. This correlation was enhanced by four swallows (especially the intensity associated parameters) compared to one swallow (Harper and McDaniel, 1993b). We believe that the increased correlation results from improved replication reproducability. This in turn was probably caused by the increased ease of rating the higher intensities resulting from four swallows. Overall, swallowing four times brought into play the full force of all of the receptors responsible for carbonation response in the posterior portion of the oral cavity.

Overall shape of the average curves changed only for Bite. The difference being the Imax plateau present in the four swallow curves (Figure 4.7).

Results from the previous study of one swallow (Harper and McDaniel, 1993b), indicated that Bite perception intensity doubled with each increasing CO_2 level. With four swallows intensity increased, but the beginning of a leveling off in perception was noted, as perception only increased 27% between 2.8 vol. and 4.6 vol. samples. There was a constant linear increase of Imax (~100% between each CO_2 level) noted in our previous study of one swallow (Harper and McDaniel, 1993b). A similar linear increase was seen in this study between the 1.7 vol. and 2.8 vol. samples, but this decreased to only ~25% from 2.8 vol. to 4.6 vol. indicating a leveling off of perception. This was accentuated for the 3°C samples. The same trend (different rate) was noted for Burn. This would seem to confirm the importance of concentration on perception, but again points to a terminal threshold being approached. The intensity accentuation for 3°C samples also points to an increase of perception affected by cold (Figure 4.7). The most probable explanation being increased response by cold receptors resulting from greater cooling by more swallows.

Bite Tinitial response was later for the four swallows, most likely resulting from interference caused by swallowing and rating at the same time. Tinitial for Burn also occurred much later. But the reason for this is probably interference from the Bite sensation. However, for both Bite and Burn, Tinitial

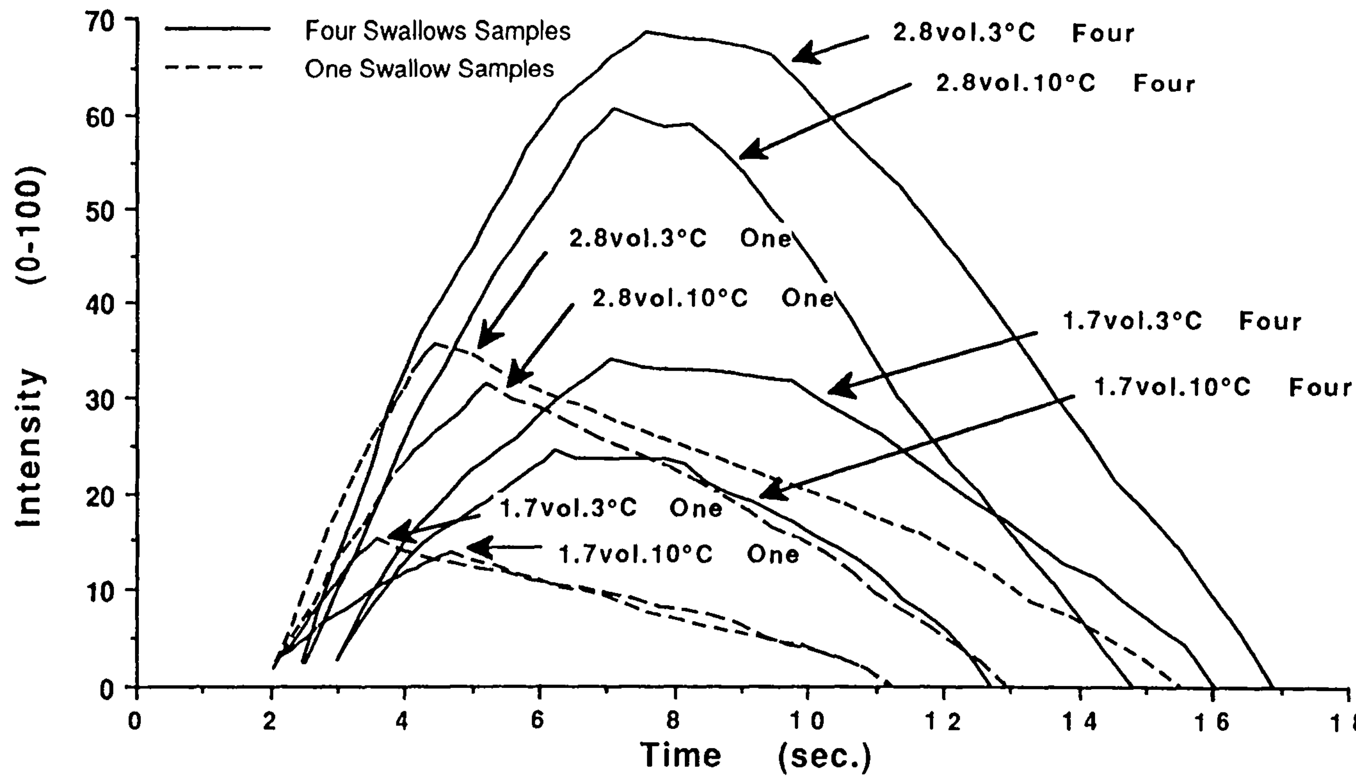


Fig. 4.7 Overall average curves of 1.7 vol. and 2.8 vol. samples for Four Swallows and One Swallow (Harper and McDaniel, 1993b).

occurring earlier with higher CO_2 level was accentuated by the four swallow condition. Green (1988) noted this quicker reaction time with increasing concentration for ethanol, and our previous study also noted this (Harper and McDaniel, 1993b). Green (1991a) and Harper and McDaniel (1993b) noted the difficulty totally separating Bite and Burn sensation especially when the initial response to Burn is occurring at a time when Bite sensation is relatively intense.

For Bite, Tmax also occurred later in the four swallow condition. Even though Tinitial and Tmax both occurred later, there was an increase in perception time of increasing intensity for four swallows.

For Bite, Tmax occurred later, TslopeA increased and Ratio increased for 3°C samples indicating that more perception sensation occurred before Imax was reached. This may indicate the role cold receptors are playing and the additive nature of CO_2 concentration and cold reception. Amount of decay perception remained unchanged.

Duration for Bite and Burn was lengthened by four swallows especially for the 3°C samples (including the non-carbonated sample). This is in contrast to one swallow where there was not a difference by temperature.

Tmax and Tdec for Burn occurred later for four swallows but the Imax plateau remained the same. In the preceeding study (Harper and McDaniel, 1993b), two possible reasons for Tmax occurring later were noted. The procedural reason, the possibility that physically moving the potentiometer lever required more time for higher intensity samples, would be ruled out by the findings of this study. Imax levels were higher, but not to the degree of affecting Tmax times. We would conclude that these results support a physiological basis such as that described by Cain (1981). He speculated that the sequestered nature of free nerve endings of the trigeminal system would retard molecule movement egressing from and progressing toward the free endings. Higher concentrations resulting from increased CO_2 exposure would cause buildup in the intracellular space in the epithelium and delay Tmax. The Imax plateau pattern remaining the same for each swallowing condition would again seem to support the assertion in the preceeding study (Harper and McDaniel, 1993b) that subjects can more easily feel the decrease in perception for Burn at high CO_2 levels. This is evidenced by the plateaus in the four swallow condition for Bite being the same at all levels. Bite, with its sharp sensation is a more upfront sensation. Burn, is a little more subtle.

Tmaxend was greatly increased for Burn for the 3°C samples but not for 10°C samples. This indicates that the cold receptors as well as receptors responsive to CO_2 interact in an additive or synergistic manner.

Summary and Conclusion

Time-intensity average curves for Bite for four swallows were found to be different compared to our previous work (Harper and McDaniel, 1993b), manifested by higher intensity, longer Duration, and development of maximum intensity plateaus reflective of the increased irritation perception. The curves for Burn were only different in that they exhibited higher maximum intensity.

Time-intensity parameter correlations were strengthened, subject variability reduced and replication reproducibility improved compared to the previous experiments with one swallow (Harper and McDaniel, 1993b). We attribute this to increased ease of rating afforded subjects by higher intensity sensations introduced in this study.

Increased oral CO_2 perception (Bite and Burn) resulting from increased CO_2 levels (concentrations) and cold temperature with ingestion of four swallows was reconfirmed. Additional time exposure of the buccal cavity to the CO_2 in solution and increased cooling for four swallows accentuated these increases. Enhancement by cold temperature was especially heightened.

Beginnings of a maximum intensity, Duration, and reaction time perceptual terminal thresholds were seen as evidenced by drastically decreased rates of increased perception seen for the highest CO_2 level samples (especially for 3°C) for both Bite and Burn. The combination of high concentration of CO_2, cold temperature, and exposure time have induced this suppression of perceptual ratings.

References

Adriaensen, H., Gybels, J., Handwerker, H.O. and Van Hees, J. 1980. Latencies of chemically evoked discharges in human cutaneous nociceptors and of the concurrent subjective sensations. Neuroscience Letters 20:55 (cited by Green, 1988).

Cain, W.S. 1981. Olfaction and the common chemical sense: similarities, differences and interactions. Chap. 6 In *Odor Quality and Chemical Structure* H.R. Moskowitz and C.B. Warren (ed.). ACS Symposium Series 148. ACS, Washington, D.C.

Cochran, W.G. and Cox, G.M. 1957. *Experimental Designs*, Wiley, New York, N.Y.

Cometto-Muñiz, J.E. and Noriega, G. 1985. Gender differences in the perception of pungency. Physiol. & Behav. 34:385.

Cometto-Muñiz, J.E., Garcia-Medina, M.R., Calviño, A.M. and Noriega, G. 1987. Interactions between CO_2 oral pungency and taste. Perception 16(5):629.

Green, B.G. 1986. Sensory interactions between capsaicin and temperature in the oral cavity. Chem. Senses 11:371.

Green, B.G. 1988. Spatial and temporal factors in the perception of ethanol irritation on the tongue. Percept. & Psychophys. 44(2):108.

Green, B.G. 1990a. Effects of thermal, mechanical and chemical stimulation on the perception of oral irritation. In *Chemical Senses* vol. 2 B.G. Green, J.R. Mason and M.R. Kare (ed.) Marcel Dekker, Inc., New York.

Green, B.G. 1991a. Capsaicin cross-desensitization on the tongue: psychophysical evidence that oral chemical irritation is mediated by more than one sensory pathway. Chem. Senses 16(6):675.

Green, B.G. 1992. The effects of temperature and concentration on the perceived intensity and quality of carbonation. Chem. Senses 17(4):435.

Harper, S.J. and McDaniel, M.R. 1993a. Carbonated water lexicon; temperature and CO_2 level influence on descriptive ratings. J. Food Sci. (In Press)

Harper, S.J. and McDaniel, M.R. 1993b. Temporal aspects of carbonation perception. Chem. Senses (Submitted).

Lawless, H. 1984. Oral chemical irritation: psychophysical properties. Chem. Senses 9(2):143.

Price, D.D. and McHaffie, J.G. 1988. Effects of heterotopic conditioning stimuli on first and second pain; a psychophysical evaluation in humans. Pain 34:245.

Stevens, D.A. and Lawless, H.T. 1986. Putting out the fire: effects of tastants on oral chemical irritation. Percept. & Psychophys. 39(5)346.

Torebjork, H.E., Schady, W. and Ochoa, J. 1984. Sensory correlates of somatic afferent fibre activation. Human Neurobiol. 3:15.

Yang, X. 1993. Personal Communication. Dept. of Agricultural Chemistry. Oregon State University, Corvallis, OR

Yau, N.J.N. and McDaniel, M.R. 1990. The power function of carbonation. J. Sensory Studies 5:117.

Yau, N.J.N. and McDaniel, M.R. 1991. The effect of temperature on carbonation perception. Chem. Senses 16(4):337.

Zahm and Nagel Co. Inc. 1964. *Zahm practical testing instruments.* Zahm and Nagel. Co. Inc., Buffalo, NY.

BIBLIOGRAPHY

Adriaensen, H., Gybels, J., Handwerker, H.O. and Van Hees, J. 1980. Latencies of chemically evoked discharges in human cutaneous nociceptors and of the concurrent subjective sensations. Neuroscience Letters 20:55 (cited by Green, 1988).

Anderson, R.L. and Bancroft, R.A. 1952. *Statistical Theory in Research. Part II - Analysis of Experimental Models by Least Square*, 338, McGraw-Hill Book Co., New York.

Arnold, G.M. and Williams, A.A. 1986. The use of generalized Procrustes techniques in sensory analysis. In *Statistical Procedures in Food Research.* Piggot, J.R. (ed.) 233-255 Elsevier Applied Science, London.

ASTM. 1991. *1991 Annual Book of ASTM Standards. Vol. 15.07 End Use Products*, 1-3, ASTM, Philadelphia, PA.

Baranowski, J.D. and Park, S.S. 1984. Effect of titratable acidity and carbonation level on the preference of clarified pineapple juice. J. Food Sci. 49:1220.

Bartoshuk, L.M. 1974. After-dinner talk taste illusion: some demonstrations. Annals of New York Academy of Sciences. 237:279-285.

Bartoshuk, L.M., McBarney, D.H., and Pfaffmann, C. 1964. Taste of sodium chloride solutions after adaptation to sodium chloride: implications for the "water taste". Science. 143:967-968.

Berridge, K.C. and Fentress, J.C. 1985. Trigeminal-taste interaction in palatability processing. Science 228:747.

Bingham, A.F., Birch, G.G., de Graaf, C, Behan, J.M. and Perring, K.D. 1990. Sensory studies with sucrose-maltol mixtures. Chem. Senses. 15(4):447.

Birch, G.G. and Munton, S.L. 1981. Use of the "SMURF" in taste analysis. Chem. Senses 6(1):45.

Birch, G.G. and Ogunmoyela, G. 1980. Taste properties of cocoa drinks with an added bitter/sweet sugar: intensity/time effects. J. Food Technol. 15:549-555.

Birch, G.G., Latymer, A., and Hollaway, M. 1980. Intensity/time relationships in sweetness: Evidence for a queue hypothesis in taste chemoreception. Chem. Senses 5(1):63-78.

Birch, G.G., O'Donnel, K., and Musgrave, R. 1982. Intensity/time studies of sweetness: psychophysical evidence for localized concentration of stimulus. Food Chem. 9:223-237.

Brandt, M.A., Skinner, E. and Coleman, J. 1963. Texture profile method. J. Food Sci. 28:404.

Bright, R.A., and Potter, N.N. 1979. Acceptability and properties of carbonated apple juice. Food Product Development April, 1979 p. 34.

Bryant, B.P., Komai, M. and Wachowiak, M. 1991. CO_2 sensitive lingual nerve neurons are differently tuned. Oral presentation at 13th annual meeting of Association for Chemoreception Sciences in Sarasota, FL.

Cain, W.S. 1981. Olfaction and the common chemical sense: similarities, differences and interactions. Chap. 6 In *Odor Quality and Chemical Structure* H.R. Moskowitz and C.B. Warren (ed.). ACS Symposium Series 148. ACS, Washington, D.C.

Cain, W.S. and Murphy, C.L. 1980. Interaction between chemoreceptive modalities of odour and irritation. Nature 284:255.

Cairncross, W.E. and Sjostrom, L.B. 1950. Flavor profile-a new approach to flavor problems. Food Technol. 4:308.

Caputi, A.Jr., Ueda, M., Walter, P. and Brown, T. 1970. Titrimetric determination of carbon dioxide in wine. Am. J. Enol. & Vitic. 21(3):140.

Choi, H.S. and Kosikowski, F.V. 1985. Sweetened plain and flavored carbonated yogurt beverages. J. Dairy Sci. 68:613.

Cliff, M. 1987. Temporal perception of sweetness and fruitiness and their interaction in model systems. M.S. thesis, Univ. of California, Davis, CA.

Cliff, M. and Noble, A.C. 1990. Time-intensity evaluation of sweetness and fruitiness and their interaction in a model solution. J. Food Sci. 55(2):450.

Cochran, W.G. and Cox, G.M. 1957. *Experimental Designs*, Wiley, New York, N.Y.

Cometto-Muñiz, J.E. and Noriega, G. 1985. Gender differences in the perception of pungency. Physiol. & Behav. 34:385.

Cometto-Muñiz, J.E., Garcia-Medina, M.R., Calviño, A.M. and Noriega, G. 1987. Interactions between CO_2 oral pungency and taste. Perception 16(5):629.

Cowart, B.J. 1987. Oral chemical irritation: does it reduce perceived taste intensity? Chem. Senses 12(3):467.

Daniels, J.A., Krishamurthi, R. and Rizui, S.H. 1985. A review of effects of carbon dioxide on microbial growth and food quality. J. Food Protect. 48(6):532.

Dubner, R. and Bennett, G.J. 1983. Spinal and trigeminal mechanisms of nociception. Ann. Rev. Neurosci. 6:381.

DuBois, G.E. and Lee, J.F. 1983. A simple technique for the evaluation of temporal taste properties. Chem. Senses. 7(3/4):237.

Farbeman, A.I. and Hellekant, G. 1978. Quantitative analysis of the fiber population in rat chorda tympani nerves and fungiform papillae. Am. J. Anat. 153:509.

Findlay, A. and King, G. 1913. Rate of evolution of gases from supersaturated solutions Part I: influence of colloids and of suspensions of charcoal on the evolution of carbon dioxide. J. Chem. Soc. (Transactions) 103:1170.

Finger, T.E. 1986. Peptide immunohistochemistry demonstrates multiple classes of perigemmal nerve fibers in the circumvallate papillae of the rat. Chem. Senses 11:135.

Garcia-Medina, M.R. and Cain, W.S. 1982. Bilateral integration in the common chemical sense. Physiol. & Behav. 29:349.

Green, B.G. 1985. Menthol modulates oral sensations of warmth and cold. Physiol. & Behav. 35:427.

Green, B.G. 1986. Sensory interactions between capsaicin and temperature in the oral cavity. Chem. Senses 11:371.

Green, B.G. 1988. Spatial and temporal factors in the perception of ethanol irritation on the tongue. Percept. Psychophys. 44:108-116.

Green, B.G. 1990a. Effects of thermal, mechanical, and chemical stimulation on the perception of oral irritation. In *Chemical Senses,* Vol. ii, (B. G. Green, J. R. Mason and M. R. Kare, eds.) 172-179, Marcel Dekker Inc., New York.

Green, B.G. 1990b. Spatial summation of chemical irritation and itch produced by topical application of capsaicin. Percept. & Psychophys. 48(1):12.

Green, B.G. 1991a. Capsaicin cross-desensitization on the tongue: psychophysical evidence that oral chemical irritation is mediated by more than one sensory pathway. Chem. Senses 16(6):675.

Green, B.G. 1991b. Oral chemesthesis: the importance of time and temperature for the perception of chemical irritants. In *Sensory Science Theory and Applications in Foods.* Lawless, H.T. and Klein, B.P. (ed.) 107-123 Marcel Dekker, New York.

Green, B.G. 1992. The effects of temperature and concentration on the perceived intensity and quality of carbonation. Chem. Senses 17(4):435.

Green, B.G. and Gelhard, B. 1989. Salt as an oral irritant. Chem. Senses 14:259.

Green, B.G. and Lawless, H.T. 1991. In *Smell and Taste in Health and Disease.* Bartoshuk, L.M., Doty, R.L., Getchell, T.V., and Snow, J.B. (ed.) 325-254 Raven Press, New York.

Guinard, J-X. 1985. Computerized time-intensity measurements of bitterness in beer and astringency in wine. M.S. thesis, Univ. of California, Davis, CA.

Guinard, J-X., Pangborn, R.M., and Lewis, M.J. 1986a. Effect of repeated ingestion on temporal perception of bitterness in beer. Am. Soc. Brew. Chem. J. 44(1):28.

Guinard, J-X., Pangborn, R.M., and Lewis, M.J. 1986b. The time-course of astringency in wine upon repeated ingestion. Enol. Vit. 37(3):184.

Guinard, J-X., Pangborn, R.M., and Shoemaker, C.F. 1985. Computerized procedure for time-intensity sensory measurements. J. Food Sci. 50:543.

Harper, S.J. and McDaniel, M.R. 1993a. Carbonated water lexicon and the influence of temperature and CO_2 levels on descriptive ratings. J. Food Sci. (In Press)

Harper, S.J. and McDaniel, M.R. 1993b. Temporal aspects of carbonation perception. Chem. Senses (Submitted).

Harrison, S.K. and Bernhard, R.A. 1984. Time-intensity sensory characteristics of saccharin, xylitol and galactose and their effect on the sweetness of lactose. J. Food Sci. 49:780.

Hensel, H. and Zotterman, Y. 1951. The response of mechanoreceptors to thermal stimulation. J. Physiol. 115:16.

Jacobs, M.B. 1959. Carbon dioxide and carbonation. In *Manufacture and Analysis of Carbonated Beverages.* Chemical Publishing Co. Inc., New York.

Jellinek, G. 1964. Introduction to and critical review of modern methods of sensory analysis (odor, taste, and flavor evaluation) with special emphasis on descriptive analysis (Flavor Profile Method). J. Nutr. Dietet. 1(3):219.

Karrer, T. and Bartoshuk, L. 1991a. Capsaicin desensitization and recovery on the human tongue. Physiol. & Behav. 49:757.

Karrer, T. and Bartoshuk, L. 1991b. Oral capsaicin desensitization and its effects on thermal, tactile and chemical stimuli. Oral presentation at 13th annual meeting of Association for Chemoreception Sciences in Sarasota, FL.

King, R.C., Sims, C.A., Moore, L.F. and Bates, R.P. 1988. Effects of maturity, skin contact and carbonation on the quality of sterile-filtered white muscadine grape juice. J. Food Sci. 53(5):1474.

Larson-Powers, N. and Pangborn, R.M. 1978. Paired comparison and time-intensity measurements of the sensory properties of beverages and gelatins containing sucrose or synthetic sweeteners. J. Food Sci. 43:41.

Lawless, H. 1984. Oral chemical irritation: psychophysical properties. Chem. Senses 9(2):143.

Lawless, H. and Stevens, D.A. 1984. Effects of oral chemical irritation on taste. Physiol. & Behav. 32:995.

Lawless, H., Rozin, P. and Shenker, J. 1985. Effects of oral capsaicin on gustatory, olfactory and irritant sensations and flavor identification in humans who regularly or rarely consume chili pepper. Chem. Senses 10(4):579.

Lawless, H.T. and Stevens, D.A. 1986. Putting out the fire: effects of tastants on oral chemical irritation. Percep. Psychophys. 5:346.

Leach, E.J. and Noble, A.C. 1986. Comparison of bitterness of caffeine and quinine by a time-intensity procedure. Chem. Senses. 11(3):339.

Lederer, C.L. Bodyfelt, F.W., and McDaniel, M.R. 1991. The effect of carbonation level on the sensory properties of flavored milk beverages. J. Dairy Sci. 74:2100.

Lee, C.B. and Lawless, H.T. 1991. Time-course of astringent sensations. Chem. Senses. 16(3):225-238.

Lee, W.E. and Pangborn, R.M. 1986. Time-intensity: the temporal aspects of sensory perception. Food Technol. 40(11):71.

Lee, W.E. III. 1985. Evaluation of time-intensity sensory responses using a personal computer. J. Food Sci. 50:1750.

Lewis, M.J., Pangborn, R.M., and Fujii-Yamashita, J. 1980. Bitterness of beer: A comparison of traditional scaling and time-intensity methods. Proc. 16th Convention Inst. of Brewing (Australia and New Zealand Sect.). 17:165.

Liu, Y.-H. and Macfie, H.J.H. 1990. Methods for averaging time-intensity curves. Chem. Senses. 15(4):471-484.

Matysiak, N.L. and Noble, A.C. 1991. Comparison of temporal perception of fruitiness in model systems sweetened with aspartame, aspartame + acesulfame K blend, or sucrose. J. Food Sci. 56(3):823.

McBride, R.L. and Richardson, K.C. 1983. Sensory assessment of the shelf life of carbonated soft drinks.

McBride, R.L. and Richardson, K.C. 1983. Sensory assessment of the shelf life of carbonated soft drinks.

McLellan, M.R., Barnard, J. and Queale, D.T. 1984. Sensory analysis of carbonated apple juice using response surface methodology. J. Food Sci. 49:1595.

McNulty, P.B. and Moskowitz, J.R. 1974. Intensity-time curves for flavored oil-in-water emulsions. J. Food Sci. 39:55.

Meilgaard, M., Civille, G.V. and Carr, B. T. 1987. Descriptive analysis techniques. In *Sensory Evaluation Techniques.* 119-127 CRC Press, Boca Raton, FL.

Meiselman, H.L. and Dzendolet, E. 1967. Variability in gustatory quality identification. Perception and Psychophysics 2:496-498.

Moore, L.J. and Shoemaker, C.F. 1981. Sensory textural properties of stabilized ice cream. J. Food Sci. 46:399.

Morrison, R.L. 1962. Enzymatic determination of carbon dioxide in lightly carbonated wines. JAOAC 45(3):627.

Muñoz, A.M. and Civille, G.V. 1992. The spectrum descriptive analysis method. In *Manual on Descriptive Analysis Testing for Sensory Evaluation.* Hootman, R.C. (ed.) 22-34 ASTM, Philadelphia, PA.

Muñoz, A.M., Pangborn, R.M., and Noble, A.C. 1986. Sensory and mechanical attributes of gel. II. Gelatin, sodium alginate and kappa-carrageenan gels. J. Texture Studies. 17:17-36.

Nagy, J.I. 1982. Capsaicin: a chemical probe for sensory neuron mechanisms. In Iverson, S.D., Iverson, L.L., Snyder, S.S. (ed.) "Handbook of psychopharmacology, Vol 15. Plenum Press, New York p. 185-235.

Naim, M., Dukan, E., Zehavi, U., and Yaron, L. 1986. The water-sweet aftertaste of neohesperidin dihydrochalcone and thaumatin as a method for determining their sweet persistence. Chem. Senses. 11(3):361.

Nasrawi, C.W. . Oral sensory responses and salivation induced by the irritation of capsaicin. Ph.D. Dissertation, University of California, Davis, 1988.

Neilson, A.J. 1957. Time-intensity studies. Drug and Cosmetic Industry 80(4):452.

Noble, A.C., Matysiak, N.L., and Bonnans, S. 1991. Factors affecting the time-intensity parameters of sweetness. Food Technol. 45(11):121.

O'Mahoney, M., Goldenberg, M., Stedman, J. and Alford, J. 1979. Confusion in the use of the taste adjectives 'sour' and 'bitter'. Chemical Senses and Flavour. 4(4):301-317.

Oakley, B. 1985. Taste responses of human chorda tympani nerve. Chem. Senses 10:469.

Ott, D.B., Edwards, C.L. and Palmer, S.J. 1991. Perceived taste intensity and duration of nutritive and non-nutritive sweeteners in water using time-intensity (T-I) evaluations. J. Food Sci. 56:535.

Overbosch, P., van den Enden, J.C. and Keur, B.M. 1986. An improved method for measuring perceived intensity/time relationships in human taste and smell. Chem. Senses. 11:331-338.

Pangborn, R.M. and Koyasako, A. 1981. Time-course of viscosity, sweetness, and flavor in chocolate desserts. J. Texture Studies. 12:141.

Pangborn, R.M., Lewis, M.J., and Yamashita, J.F. 1983. Comparison of time-intensity with category scaling of bitterness of iso-alpha-acids in model systems and in beer. J. Inst. Brew. 89:349.

Parker, G.H. 1912. The reactions of smell, taste, and the common chemical sense in vertebrates. J. Acad. Nat. Sci. Phila. 15:221.

Phillips, G. and Woodruff, J.G. 1981. Carbonating and cooling beverages. In *Beverages: Carbonated and Noncarbonated.* AVI Publishing Co., Westport, CT.

Pokorny, J., Davidek, Prnka, V. and Davidkova, E. 1986. Nonparametric evaluation of graphical sensory profiles for the analysis of carbonated raspberry beverages. Die Nahrung 30(2):131.

Price, D.D. and McHaffie, J.G. 1988. Effects of heterotopic conditioning stimuli on first and second pain; a psychophysical evaluation in humans. Pain 34:245.

Quinn, E.L. and Jones, C.L. 1936. Physical properties of carbon dioxide. In *Carbon Dioxide ACS Monograph Series.* Reinhold Publishing Co., New York.

Robinson, J.O. 1970. The misuse of taste names by untrained observers. Brit. J. Psychol. 61:375-378.

Rozin, P. 1990. Getting to like the burn of chili pepper. Biological, psychological and cultural perspectives. In *Chemical Senses. Volume 2: irritation* Green, B.G., Mason, J.R., Kare, M.R., (eds.) 231-269 Marcel Dekker, New York.

Schmitt, D.J., Thompson, L.J., Malek, D.M. and Munroe, J.H. 1984. An improved method for evaluating time-intensity data. J. Food Sci. 49:539.

Shamil, S., Birch, G.G., Jackson, A.A.S.F., and Meek, S. 1988. Use of intensity-time studies as an aid to interpreting sweet taste chemoreception. Chem. Senses. 13(4):597-605.

Silver, W.L. and Finger, T.E. 1991. The trigeminal system. In *Smell and Taste in Health and Disease.* Bartoshuk, L.M., Doty, R.L., Getchell, T.V., and Snow, J.B. (ed.) 97-108 Raven Press, New York.

Silver, W.L., Mason, J.R., Marshall, D.A., and Maruniak, J.A. 1985. Rat trigeminal, olfactory and taste responses after capsaicin desensitization. Brain Research 333:45.

Sizer, F. and Harris, H. 1985. The influence of common food additives and temperature on threshold perception of capsaicin. Chem. Senses 10:279.

Stevens, D.A. and Lawless, H.T. 1986. Putting out the fire: effects of tastants on oral chemical irritation. Percept. & Psychophys. 39(5)346.

Stevens, D.A. and Lawless, H.T. 1987. Enhancement of responses to sequential presentation of oral chemical irritants. Physiol. Behav. 39:63.

Stevens, J.C. 1982. Temperature can sharpen tactile acuity. Percept. & Psychophys. 31(6):577.

Stevens, J.C. and Hooper, J.E. 1982. How skin and object temperature influence touch sensation. Percept. & Psychophys. 32(3):282.

Stone, H. and Sidel, J.L. 1985. Descriptive analysis. In *Sensory Evaluation Practices*. 194-225 Academic Press, Inc., New York.

Stone, H., Sidel, J., Oliver, S., Woolsey, A. and Singleton, R.C. 1974. Sensory evaluation by quantitative descriptive analysis. Food Technol. 28(11):24.

Straub, A. 1992. Power function determination for sourness and time-intensity measurements of sourness and astringency for selected acids. M.S. thesis, Oregon State Univ., Corvallis, OR.

Swartz, M. 1980. Sensory screening of synthetic sweeteners using time-intensity evaluations. J. Food Sci. 45:577.

Szczesniak, A.S. 1963. Classification of textural characteristics. J. Food Sci. 28:385.

Szczesniak, A.S. 1979. Classification of mouthfeel characteristics of beverages. In *Food Texture and Rheology* P. Sherman (ed.), Academic Press, New York.

Szolcsanyi, J. 1977. A pharmacological approach to elucidation of the role of different nerve fibers and receptor endings in mediation of pain. J. Physiol. 73:251.

Takagaki, M. and Asakura, Y. 1984. Taste time-intensity measurements using a new computerized system. In Ueda, K. (ed.), Proceedings of the 18th Japanese Symposium on Taste and Smell. Japanese Association for the Study of Taste and Smell, Osaka University, Osaka, Japan, 105 (cited by Leach and Noble, 1986).

Taylor, D.E. and Pangborn, R.M. 1990. Temporal aspects of hedonic responses. J. Sensory Stud. 4:241-247.

Torebjork, H.E., Schady, W. and Ochoa, J. 1984. Sensory correlates of somatic afferent fibre activation. Human Neurobiol. 3:15.

van Buuren, S. 1992. Analyzing time-intensity responses in sensory evaluation. Food Technol. 46(2):101.

Weyh, H. 1975. Zur sensorischen beurteilung von CO_2-haltigen getrankeni. Kohlensauregehalt und rezenz. Brauwelt 115(19):638.

Yang, X. 1993. Personal Communication. Dept. of Agricultural Chemistry. Oregon State University, Corvallis, OR.

Yau, N.J.N. and McDaniel, M.R. 1990. The power function of carbonation. J. Sensory Studies 5:117.

Yau, N.J.N. and McDaniel, M.R. 1991. The effect of temperature on carbonation perception. Chem. Senses 16(4):337.

Yau, N.J.N., McDaniel, M.R. and Bodyfelt, F.W. 1989. Sensory evaluation of sweetened flavored carbonated milk beverages. J. Dairy Sci. 72:367.

Zahm & Nagel Co., Inc. 1964. *Zahm Practical Testing Instruments, 15th Edition,* Zahm and Nagel Co., Inc., Buffalo, NY.

Zumdahl, S.S. 1986. *Chemistry.* D.C. Heath and Co., Lexington MA.

APPENDIX

Table 5.1. Concentration of chemical species present in carbonated water.

Volumes of CO_2	H^+	CO_2	H_2CO_3	HCO_3^-
1.7 vol.	1.95×10^{-4}	2×10^{-2}	5.2×10^{-5}	1.2×10^{-7}
2.8 vol.	2.24×10^{-4}	7×10^{-2}	1.8×10^{-4}	3.5×10^{-7}
4.6 vol.	3.09×10^{-4}	14×10^{-2}	3.6×10^{-4}	5.1×10^{-7}

All values are expressed as moles/liter. Calculations based on Henry's Law Constants from Quinn and Jones (1936).