

## ABSTRACT

AMEERALLY, ANGELIQUE DANIELLE. Sensory and Chemical Properties of Gouda Cheese. (Under the direction of MaryAnne Drake).

Gouda cheese (G) is a Dutch, washed curd cheese that is traditionally produced from bovine milk and brined before ripening for 1-20 months. In response to domestic and international demand, U.S. production of Gouda cheese has more than doubled in recent years. An understanding of the chemical and sensory properties of G can help manufacturers to create desirable products. The objective of this study was to determine the chemical and sensory properties of Gouda cheeses. Commercial Gouda cheeses (n=36, 3 mo to 5 y, domestic and international) were obtained in duplicate lots. Volatile compounds were extracted (SPME) and analyzed by gas chromatography olfactometry (GCO) and gas chromatography mass spectrometry (GCMS). Physical analyses included pH, proximate analysis, salt content, organic acid analysis by HPLC, and color. Flavor and texture properties were determined by descriptive sensory analysis. Focus groups were conducted with cheese followed by consumer acceptance testing (n=153) with selected cheeses. Ninety aroma active compounds were detected in cheeses by SPME-GC-O. Key volatile compounds in Gouda cheeses included dimethyl sulfide, 2,3-butanedione, 2/3-methylbutanal, ethyl butyrate, acetic acid, and methional. Older cheeses had higher organic acid concentrations, higher fat and salt content, and lower moisture content than younger G. Younger cheeses were characterized by milky, whey, sour aromatic, and diacetyl flavors while older G were characterized by fruity, caramel, malty/nutty, and brothy flavors. International cheeses were differentiated by the presence of low intensities of cowy/barny and grassy flavors. Younger cheeses were characterized by higher intensities of smoothness and mouthcoating while older cheeses were

characterized by higher intensities of fracture and firmness. American consumers utilized Gouda cheeses in numerous applications and stated that packaging appeal, quality and age were more important than country of origin or nutrition when purchasing cheeses. Young and medium U.S. cheeses  $\leq 6$  mo were most liked by U.S. consumers. Three distinct consumer segments were identified with distinct preferences for cheese flavor and texture based on ripening time. Findings from this study establish key differences in Gouda cheese regarding age and origin and identify U.S. consumer desires for this cheese category.

**Key Words:**

Gouda cheese, flavor, preference mapping

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Sensory and Chemical Properties of Gouda Cheese

by

Angelique Danielle Ameerally

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## **DEDICATION**

This thesis is dedicated to my parents for their unconditional love, and to Matt for his  
laughter and support through the years.

## **BIOGRAPHY**

Angelique Danielle Ameerally was born in Georgetown, Guyana, South America to Nizam Danny Ameerally and Vivienne Gillian Ameerally. She attended Virginia Polytechnic Institute and State University and received a Bachelor's of Science degree in Food Science with a minor in Chemistry in 2009. In 2013, she pursued a Master of Science degree in Food Science under the direction of Dr. MaryAnne Drake at North Carolina State University.

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## CHAPTER 1: LITERATURE REVIEW

### I. Introduction

Dutch type cheeses are among the oldest and most important cheese varieties in terms of tons produced globally (Welthagen et al., 1998). Gouda cheese is one of the two main types of Dutch cheese, the other being Edam (Walstra et al., 1993). Both varieties are characterized as ripened firm/semi hard cheeses (Welthagen et al., 1998). The primary differences between Gouda cheese and Edam is that Gouda cheese is typically manufactured from whole milk while Edam is manufactured from partially skimmed milk and Gouda can be manufactured in weights 2.5-30 kg while Edam can be manufactured in weights 1.5-2.5 kg (Walstra, et al., 1993; Codex Alimentarius, 2013).

In The Netherlands, Gouda cheese is typically classified into one of six categories depending on age; older Gouda cheeses are more flavorful and firmer than younger Goudas (Nederlandse Zuivel Organisatie, 2013).

**Table 1. Traditional Classification of Dutch Gouda Cheese (Nederlandse Zuivel Organisatie).**

<b>Classification</b>	Jong	Jong Belegen	Belegen	Extra Belegen	Oud	Overjarig
<b>Age (months)</b>	<u>≤1</u>	2-3	4-6	7-9	10-11	<u>≥12</u>

Gouda and Edam cheeses are defined internationally by the Codex Alimentarius. Acceptable milkfat contents for Gouda and Edam cheeses ranges depending on the fat content of the dry matter used to produce the cheese (Codex Alimentarius, 2013). For both Edam and Gouda cheeses, the color may range from a near white or ivory to light yellow or yellow; the texture should be firm and suitable for cutting (Codex Alimentarius, 2013). Gas holes for Edam and Gouda cheeses may be up to 10 mm in diameter and must be distributed throughout the

interior of the cheese (Codex Alimentarius, 2013). Gouda cheese must be a flattened cylinder, a flat block, or a loaf in shape; it may be manufactured and sold with a coated or uncoated dry rind (Codex Alimentarius, 2013).

**Table 2. Primary differences between Edam and Gouda cheeses (Codex Alimentarius, 2013).**

<b>Gouda</b>		<b>Edam</b>	
Dry Matter Content Fat %	Gouda cheese % fat (dry weight)	Dry Matter Content Fat %	Edam Cheese % fat (dry weight)
30-39	48	30-39	47
40-47	52	40-44	51
48-59	55	45-49	55
>60	62	50-59	57
NA	NA	>60	62

In the U.S., Gouda cheese is defined by Code of Federal Regulations (CFR) Title 21 as having a minimum milkfat content of 46% by weight and maximum moisture content of 45% by weight (Code of Federal Regulations, 2006). With the exception of milkfat and moisture content allowances, U.S. requirements for Gouda cheese are the same as those for Edam and do not specify parameters for size, texture, color, shape and coating (Code of Federal Regulations, 2006).

**Table 3. Differences between Edam and Gouda Cheeses by Code of Federal Regulations and Codex Alimentarius Systems (Code of Federal Regulations 2006, Codex Alimentarius 2013).**

	<b>Code of Federal Regulations: Milkfat %</b>	<b>Code of Federal Regulations: Moisture %</b>	<b>Codex Alimentarius: Shape</b>	<b>Codex Alimentarius: Size (kg)</b>
<b>Gouda</b>	≥46	≤45	Flat block/flat cylinder/ loaf	2.5-30
<b>Edam</b>	≥40	≤45	Spherical cylinder/loaf	1.5-2.5

Addition of natural or artificial colorant is acceptable for U.S. production, while international standards exclusively allow the use of beta carotenes or annatto extract for Gouda cheese (Code of Federal Regulations, 2006; Codex Alimentarius, 2013). U.S. Gouda produced from

unpasteurized milk must ripen for a minimum of 60 days at a temperature of at least 2°C, but no age categories or ripening time requirements are defined by CFR (Code of Federal Regulations, 2006). Between 2010 and 2013, U.S. production of Gouda cheese increased from 19 million to 48 million pounds per year (USDA). As U.S. demand for Gouda cheese rises, characterization and relation of flavor components to consumer drivers of liking/disliking may help U.S. manufacturers to increase sales (Colonna et al., 2011; Cadwallader et al., 2009).

## **I. Production and Fermentation of Gouda Cheese**

### **Production of Gouda Cheese**

Gouda cheese is a washed curd variety traditionally produced from fresh unpasteurized cows' milk that is whole milk or at most partly skimmed (Welthagen et al., 1998, Van Den Berg, 2004). The cheese is coagulated by rennet, fermented using mixed strain starters, pressed to obtain a closed rind, and brined after pressing (Welthagen et al., 1998). The cheese should be at least partially matured after a few weeks and have undergone significant proteolysis (Welthagen et al., 1998). Cheeses produced today using this process are referred to as Goudse berenkaas (Van Den Berg, 2004).

To meet demand, the last 45 years has required a shift from artisanal to industrial scale production (Van den Berg et al., 2004). Standard commercial capacity for modern Gouda cheese plants is 30,000 tons of cheese annually (Van den Berg et al., 2004). Cow's milk is first pasteurized at 72°C for 15 seconds (Johnson et al., 1998). The milk is then placed in a cheese vat and heated to 30 °C where annatto colorant at a rate of 1 ounce: 453 kilograms of milk, microbial rennet at a ratio of 500 milliliters/4350 liters milk, and a mesophilic citrate

fermenting DL starter culture consisting of *Leuconostoc* and *Lactococcus lactis* are added (Weimer, 2007; Polzin, 2015; Johnson et al., 1998; Høier et al., 1999). Nitrates may be added at a concentration of 100 milligrams/kilogram to suppress the growth of *Clostridium* spp., but nitrate addition is less common in Europe and the U.S. where techniques such as microfiltration, lysozyme addition or milk screening can reduce gas blowouts in cheeses (Roche et al., 1983; Johnson et al., 2006; Fox et al., 2003; Borreani et al., 2008). Calcium chloride may also be added at a ratio of 296-591 milliliters/4350 liters milk (Polzin, 2015). After coagulation, the mass is cut when the curd will split cleanly into ½ inch cubes; the curd is subsequently stirred and the temperature elevated to 40 °C (Van den Berg et al., 2004; Weimer, 2007; Polzin, 2015). The curds are allowed to rest for a minimum of 15 minutes before 50% of the whey is removed and water (48°C) equal to 30% w/w milk is added to remove lactose and heat the curd (Johnson et al, 1998). The remaining 50% of the whey is removed after an additional 30-45 minutes of stirring and the curd has been allowed to firm (Johnson et al., 1998). The cheese curds are transferred to molds and gradually subjected to 25 kPa of pressure until reaching a pH of 5.4 (Polzin, 2015; Weimer, 2007). The cheese is subsequently brined by immersion of the cheese into a 1.7% salt solution for 24 h (Polzin, 2015; Messens et al., 1999). After brining, the cheese is washed in water at 50°C, dried, and stored for ripening (Weimer, 2007). Direct addition of salt to the curds may be used instead of brining, but it is more difficult to obtain a uniformly salted product by direct salting (Guinee et al., 2004). Dry salted cheeses do not undergo brining; they are instead heated after cutting to a curd temperature of 32 °C, drained, and salted at a ratio of 10 kg salt/4350 liters milk before pressing and ripening, similar to Cheddar, Colby or Jack cheeses (Polzin,

2015). Loaves are ripened in air-conditioned rooms to protect rind integrity, control evaporation, and minimize surface microbiological growth (Van den Berg et al., 2004). When produced from raw milk, Gouda cheese must be ripened for a minimum of 60 days (Code of Federal Regulations, 2006). Ripening time for Gouda cheeses ranges from 1-20 months, depending on the characteristics desired (Messens et al., 1999).

### **Microbial and Enzymatic activity in Cheese**

Traditionally, cheese fermentation was completed by bacteria naturally present in the raw milk or by addition of previously produced cheese or coagulated milk or whey as inoculant to the milk, a technique termed “backslopping” (Fox et al., 2000). Due to improved hygienic practices and the need for a consistent product, fermentation is now completed by direct addition of a starter culture to the milk (Fox et al., 2000). Dairy starter cultures are primarily composed of lactic acid bacteria (LAB), organisms capable of fermenting lactose into lactic acid (Settani et al., 2010). There are 12 genera of LAB, with *Leuconostoc*, *Lactobacillus* and *Lactococcus* comprising the majority of dairy starter cultures (Fox et al., 2000). Starter culture species can be classified as mesophilic or thermophilic, depending on their optimum temperature range; mesophilic bacteria function optimally at temperatures close to 30°C while thermophilic bacteria function optimally at temperatures close to 42°C (Fox et al., 2000). Mixed mesophilic strain cultures derived from artisanal Gouda cheese production are most commonly utilized in Northern Europe and the Netherlands; the number of strains and bacterial composition can change based on temperature, phages, and subculturing (Ayad et al., 2001). Defined mixed strain starters (DSS), consisting of at least two mesophilic and/or thermophilic bacteria, and are used to produce Gouda cheese in



countries where Cheddar cheese is manufactured (Ayad et al., 2001). DSS cultures are employed to produce Gouda cheese because of the capacity to quickly and reliably produce acid, induce eye formation, and phage resistance (Van den Berg et al., 2004). Gouda starter cultures are primarily mesophilic cultures comprised of *Lactococcus lactis* spp. *Lactis*, *Lactococcus lactis* spp. *Cremoris* and citrate positive *Leuconostoc mesenteroides* (Fox et al., 2000; Polzin, 2015). Species *Lactococcus lactis* spp. *Lactis* and *Lactococcus lactis* spp. *cremoris* are added because they are homofermentative and predominantly produce lactic acid, while heterofermentative *Leuconostoc mesenteroides* is responsible for diacetyl and eye formation resulting from CO<sub>2</sub> (Van Hoorde et al., 2008). Specifically, CO<sub>2</sub> gas produced from travels through the cheese curd until it reaches a weak point and accumulates to form an eye (McSweeney et al., 2004). Cultures are added in the form of concentrated pellets at a dosage of 500 g/1000 L with a recommended fermentation temperature of 22-34°C (Polzin, 2015). The dominant bacterial species, *Lactococcus lactis*, can comprise up to 85% of the culture (Van Hoorde et al., 2010).

### **Bacterial Cultures present in Gouda cheese**

Table 1 displays the results for microbial analysis of raw and pasteurized Gouda type cheeses (Van Hoorde et al., 2010). Cheeses analyzed by V3-16S PCR-denaturing gel electrophoresis (PCR-DGGE) analysis at varying production locations revealed the presence of several microbial species (Van Hoorde et al., 2010). Bands from species *Lactococcus lactis*, *Leuconostoc pseudomesenteroides*, *Lactobacillus curvatus*, *Penicillium pentosaceus*, and members of the *E. faecium* group were present in both pasteurized and raw Gouda cheeses (Van Hoorde et al., 2010). The presence of *Lactobacillus plantarum*, members of the

*E. faecium* group, unknown members of the genus *Enterococcus* and *Streptococcus dysgalactia*, were exclusive to raw milk Gouda cheeses (Van Hoorde et al., 2010).

In general, the microbiota of raw milk cheese is more diverse and heterogeneous than pasteurized milk cheeses; this facilitates development of more intense, stronger, and more inconsistent flavors (Kolakowski et al., 2012). Nonstarter bacteria present in Gouda cheeses include lactobacilli, enterococci and pediococci (Kolakowski et al., 2012). Higher number of bands from *Lactococcus lactis* and *Leuconostoc pseudomesenteroides*, two species present in many Gouda cheese starter cultures, were noted for pasteurized milk cheeses (Kolakowski et al., 2012). This suggests that the starter lactic acid bacteria may be better able to compete with nonstarter lactic acid bacteria in pasteurized milk Gouda cheeses (Kolakowski et al., 2012).

### **Control of Pathogenic Organisms**

The presence of pathogenic and spoilage microorganisms can affect product safety and quality (Van den Berg et al., 2004). To minimize this activity, the quality of Gouda cheese milk is regulated from the farm by temperature control and hygienic practices (Van den Berg et al., 2004). Undesired organisms include Enterobacteriaceae, coliforms, *Lactobacillus* spp. *Streptococcus thermophilus*, faecal streptococci, propionic acid bacteria and *Clostridium tyrobutyricum* (Van den Berg et al., 2004). Production steps to minimize growth include thermisation and pasteurization (Van den Berg et al., 2004). Pasteurization kills pathogens such as *Listeria monocytogenes* and Enterobacteriaceae, but does not destroy *Clostridium tyrobutyricum* spores (Van den Berg et al., 2004). *Clostridium tyrobutyricum* is of concern because it can lead to late blowing, or severe swelling in Gouda cheese by gas

production; late blowing is difficult to control because it can occur in cheese made from milk containing 5-10 spores per liter (Bester et al., 1989). Several methods exist to reduce *Clostridium tyrobutyricum* growth, including bactofugation or microfiltration of milk to reduce the concentration present, addition of nitrates to prevent butyric acid fermentation, and addition of lysozyme as a preservative (Matijasic et al., 2007). Additional manufacturing procedures, such as acid production by LAB and increased salt, also decrease pathogenic incidence in Gouda cheese (Van den Berg et al., 2004).

### **Control of Yeast Activity**

When present in sufficient concentration, yeasts may impact the ripening process and influence the flavor of the cheese in an undesirable fashion (Welthagen et al., 1998). Twenty three yeast species were isolated from a factory environment for Gouda cheese processing and ripening (Welthagen et al., 1998). *Debaryomyces hansenii* was the most abundant yeast present in the Gouda cheese; other species included *Saccharomyces cerevisiae*, *Yarrowia lipolytica*, and *Kluyveromyces marxianus* (Welthagen et al., 1998). Yeast contamination in Gouda cheese making was shown to be primarily imparted by equipment surfaces, brine bath contamination and presence of yeasts in whey (Welthagen et al., 1998).

### **Accelerated Ripening and Enzymatic Modification**

Enzyme modification of dairy products is of interest due to the potential for shorter ripening times and controlled production of flavors (Seitz et al., 1990; Pandian et al., 1990). As proteolysis is the slowest ripening step, accelerated ripening research focuses on decreasing maturation time (Fox et al., 2006). Strategies for accelerated cheese ripening

include enzyme addition, elevated ripening temperature, high pressure processing, secondary cultures and genetically modified starters (Law et al., 2000).

Increasing cheese ripening temperatures from 8°C to  $\leq 15^\circ\text{C}$  is appealing due to the potential for reduced manufacturing costs, but requires high hygienic standards and may result in off flavors due to unequal acceleration of biochemical reactions (Pandian et al., 1990; Fox et al., 2006). However, accelerated ripening of Dutch cheeses at 16°C for 56 days did not result in increased off flavors compared to cheeses ripened at 10°C, but lead to a faster decrease in cheese hardness, suggesting increased proteolysis (Pachlova et al., 2011).

Direct addition of enzymes to cheese milk during manufacture is cheap, but may result in over ripening, discoloration or inconsistent flavor, and adjunct culture use is limited in cheese due to a lack of efficacy in commercially available strains (Folkertsma et al., 1995; Seitz et al., 1990). However, direct addition of enzymes is utilized to create enzyme modified cheeses (EMC), cheese powders and cheese flavors that can be added to food products to create higher intensity cheesy flavor (Kilcawley et al., 2008).

Although starter cultures have been modified to produce lower amounts of lactic acid and accelerate maturation of full fat hard and semi hard cheeses, no commercial trials have been published (Law et al., 2000). Studies investigating the use of high pressure processing at 100-1000 MPa to accelerate cheese ripening for Cheddar, Camembert and Gouda have yielded variable results (Law et al., 2000; Malone et al., 2003). No texture or pH differences were detected in high pressure processed Gouda cheeses, but flavor differences and commercial viability have not been investigated (Messens et al., 1998; Messens et al., 2000)

## **Flavor Formation in Cheeses**

Ultimately, both the starter culture and microorganisms native to the raw milk influence flavor formation in cheeses (Marilley et al., 2004). The breakdown of lactose to lactate is a crucial first step in flavor development of a cheese (Mcsweeney et al., 2000). Lactose metabolism occurs most extensively in surface mold ripened cheeses (Mcsweeney et al., 2000). Lactate contributes to the early stages of cheese maturation and may undergo transformation by numerous other pathways to form other flavor compounds (Mcsweeney et al., 2000). Oxidation of lactate to acetate and carbon dioxide by nonstarter lactic acid bacteria is one possible reaction; acetate has been shown to contribute to the flavor of Cheddar and Dutch-type cheeses (Mcsweeney et al., 2000; Singh et al., 2003).

Though the majority of citrate native to raw milk is lost to whey, retained citrate may be further metabolized into a variety of flavor components, primarily acetate, diacetyl, acetoin, and 2,3-butanediol (Mcsweeney et al., 2000). Carbon dioxide is important to eye formation in Dutch type cheeses; gas formation results from metabolism of citrate and addition to form pyruvate and acetaldehyde (Mcsweeney et al., 2000). Diacetyl imparts a buttery flavor to younger cheeses and is typically present in low concentrations by 6 months (Urbach et al., 1997; Drake et al., 2009). Diacetyl is an important contributor to flavor of Dutch type and Cottage cheeses (Mcsweeney et al., 2000). For controlled fermentation of milk products, diacetyl and acetaldehyde are optimally present in a ratio of 4:1 (Urbach et al. 1997).

Flavors from proteolytic products and amino acid catabolism can affect both cheese flavor and aroma; the end products are free amino acids (FAA) and peptides (Cadwallader et al.,

2009). The order in which proteolytic reactions occur is important to proper development of cheese flavor (Cadwallader et al., 2009; Mcsweeney et al., 2000). With the exception of arginine, the free amino acid concentration increases with ripening in cheese (Mcsweeney et al., 2000). Peptides can impart basic tastes to cheese and are capable of contributing to the formation of other flavors (Mcsweeney et al., 2000). Bitterness perceived in cheese primarily results from hydrophobic peptides but may be imparted by amines, acids, ketones, monoglycerides and diglycerides (Mcsweeney et al., 2000). Peptides and FAA may serve as substrates to produce flavor compounds, including transamination, dehydrogenation, decarboxylation, and reduction reactions (Marilley et al., 2004). These reactions are responsible for a wide range of compounds in cheese, including alcohols, pyruvates, aldehydes, carboxylic acids and sulfur compounds (Marilley et al., 2004).

Due to the capacity to convert into alcohols or carboxylic acids, aldehydes are not present in relatively high concentrations in cheese (Mcsweeney et al., 2000). However, they are flavor contributing due to the low parts per billion sensory thresholds of these compounds. Three Strecker degradation aldehydes formed from breakdown of amino acids have been shown to result in nutty flavor in Cheddar cheese (Avsar et al., 2004; Yvon et al., 2001). The aldehydes 2-methylbutanal, 3-methylbutanal and 2-methylpropanal were confirmed to contribute to nutty flavor by descriptive sensory analysis of cheese models spiked with these three compounds (Avsar et al., 2004).

In general, sulfur compounds are extremely volatile and are thus significant contributors to flavor of numerous cheese varieties including Cheddar, Swiss Gruyere and Camembert (Urbach et al., 1995; Preininger et al., 1993; Marilley et al., 2004; Fox et al.,

1997). Sulfur containing volatiles contributing to aroma and flavor in cheese include methanethiol, methional, dimethyl sulfide (DMS), dimethyl disulfide (DMDS), dimethyl trisulfide (DMTS) and hydrogen sulfide (Urbach et al., 1997; Cadwallader et al., 2009). These compounds result from amino acids, primarily methionine (Cadwallader et al., 2009; Mcsweeney et al., 2000). Methanethiol and DMTS impart a decomposing cabbage or decomposing odor in surface smear cheeses (Urbach et al., 1997). When present with select other compounds, methanethiol may impart odors considered typical of Cheddar flavor (Urbach et al., 1997). Hydrogen sulfide is another sulfur derived contributor to Cheddar flavor; it is formed during ripening from the breakdown of cysteine and methionine (Urbach et al., 1997). The role of sulfur containing compounds in Cheddar cheese flavor is definitive (Drake et al., 2001). A variety of sulfur flavors are documented in Cheddar cheese (Drake et al., 2001). However, the actual importance of sulfur compounds in Cheddar flavor remains controversial, in part due to the difficulty in isolation and characterization (Milo and Reineccius, 1997; Drake et al., 2010). Methional is considered characteristic to Emmental cheese due to a high odor activity value in cheeses analyzed by Aroma Extract Dilution Analysis (Preininger et al., 1993).

**Table 4. Microbial species detected by DGGE profiling of raw (R) and pasteurized (P) Goudas manufactured in two locations (Taken from Van Hoorde, 2010)**

	Curd			Day <sup>b</sup>			Wk1			Wk3			Wk5			Wk8		
	P <sup>a</sup> _BK	P_dVK	R <sup>b</sup> _dVK	P_BK	P_dVK	R_dVK	P_BK	P_dVK	R_dVK	P_BK	P_dVK	R_dVK	P_BK	P_dVK	E_BK	P_BK	P_dVK	P_BK
<i>Lc. Lactis</i>	21	21	28	34	42	34	54	52	50	48	48	40	59	63	46	53	53	38
<i>Lactococcus sp.</i>	2	2		2	3										1			3
<i>Leuc. pseudomesenteroides</i>	8	6	7	14	10	6	21	19	17	22	23	19	13	14	13			14
<i>Lb. curvatus</i>	1		2	3	3	2	4	4	4	6	3	6	4	5	6	15	19	6
<i>Lb. helveticus</i>											1		1	1	3	4	6	3
<i>Lb. plantarum</i>												4			5		1	2
<i>E. gallinarum group</i>			2		2	1		1	2		1	4		2				
<i>Lb. casei group</i>																		
<i>E. casseliflavus,</i> <i>E. malodoratus</i>											1	1		1	4			3
<i>E. faecalis</i>			6	1		5			9			6	1		6			9
<i>E. faecium group</i>	1	1	11		2	14	1	3	22	2	5	22	4	1	16	2	5	23
<i>Enterococcus sp.</i>	2		4			3	3	2	10	5	4	27	5	5	2		1	
<i>P. pentosaceus</i>			1	1		4			6			6			24	4	5	25
<i>Strep. Dysgalactiae</i>			3		1	6			10			10			4			4
<i>gamma-Proteobacteria</i>	3	1	2			4			12	2		14	2	4	8	1	2	3
Not identified			3												5	7	5	1
<b>Total</b>	38	31	69	55	63	79	83	81	142	85	86	159	89	96		86	97	134

<sup>a</sup> denotes Pasteurized cheeses and <sup>b</sup> Raw Milk cheeses



Previous work utilized steam distillation extraction (SDE) followed by gas chromatography mass spectrometry (GC-MS) to analyze volatile composition of two commercial Belgian Gouda cheeses (Van Leuven et al., 2008). Dimethyl disulfide, dimethyl trisulfide and methional were identified as the sulfur compounds present in the highest concentration (Van Leuven et al., 2008). Sulfur concentration was higher for aged Gouda cheeses and pasteurized milk Gouda cheeses, but the overall importance of sulfur derived compounds to Gouda cheese characterization and what specific sulfur compounds are detectable as odorants has not been established (Van Leuven et al., 2008).

Reactions derived from milk fat are crucial to characteristic cheese flavor (McSweeney et al., 2000). Milk fat may undergo two metabolic processes: oxidation or hydrolysis (Marilley et al., 2004). Hydrolysis of triglycerides present in milk fat is more influential than oxidation to flavor development of cheese because milk has a low oxidation-reduction potential (McSweeney et al., 2000). Short and medium chain fatty acids formed from triglycerides contribute to cheese flavor, but can be further metabolized to form additional flavor compounds such as free fatty acids, lactones, aldehydes and alcohols (McSweeney et al., 2000).

Acids detected in Cheddar cheese due to breakdown of amino acids include 2-hydroxy-3-methylbutanoic acid, 2-hydroxy-4-methylpentanoic acid, 2-hydroxy-3-methylpentanoic acid and 2-hydroxyhexanoic acid (Urbach et al., 1997). Alpha keto-acids have been detected in a variety of cheeses, including Cheddar, Gouda, Provolone, Emmenthal and Camembert (McSweeney et al., 2000). Specifically,  $\alpha$ -Keto-3-

methylbutanoic and  $\alpha$ -keto-3-methylpentanoic acids are associated with a cheesy odor (Mcsweeney et al., 2000).

Fatty acids in cheese range from C<sub>4</sub> to C<sub>20</sub> and impart flavors such as rancid, soapy, and sweaty (Cadwallader et al., 2009). Lower chain fatty acids are generally responsible for characteristic cheese flavor, but can impart rancidity at higher levels (Kanawjia et al., 1995). Odor thresholds for fat derived compounds in cheese vary depending on the fat content of the matrix; butyric acid is considered a high impact compound because it is present in high concentrations in Cheddar cheese and has a high odor activity value (Kim et al., 2011).

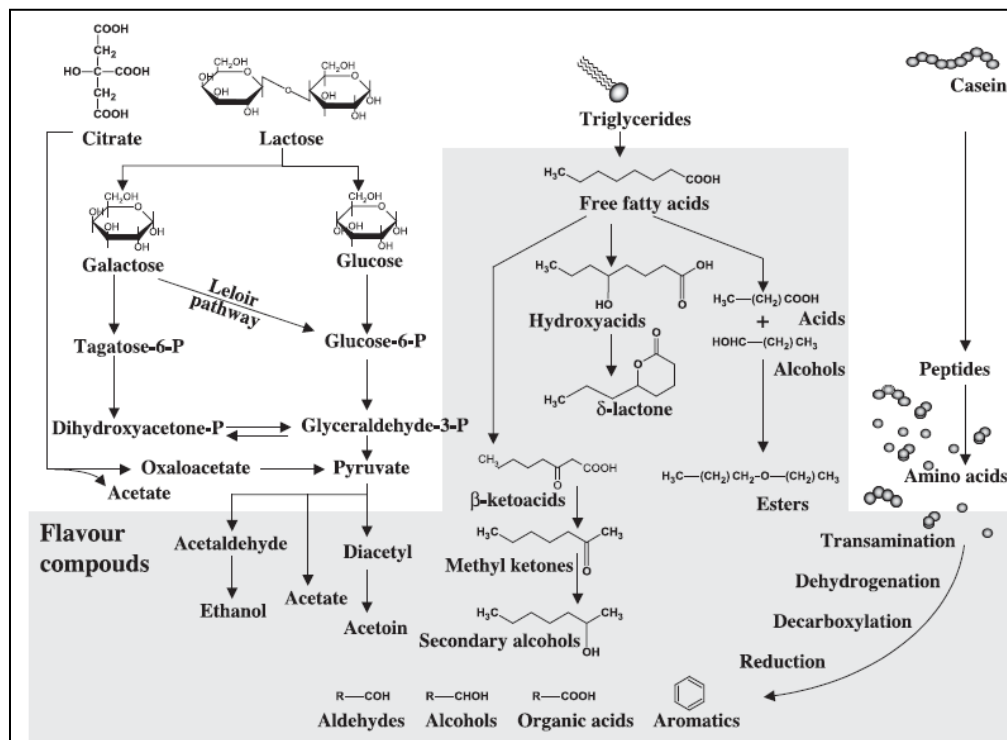


Figure 1. Three primary processes for cheese flavor development (Taken from Marilley et al., 2004).

Several factors are responsible for flavor intensity of free fatty acids, including the distribution between aqueous and fat phases, the pH of the medium, the presence of cations, and the presence of proteolytic products (Cadwallader et al., 2009). The flavor of the cheese

should be balanced in terms of fatty acid intensity versus products of glycolysis and proteolysis; moderate presence of free fatty acids in Cheddar, Dutch and Swiss type cheeses are generally regarded as rancid and are considered a flavor defect (Cadwallader et al., 2009; Mcsweeney et al., 2000). The pH of the cheese will influence the intensity of fatty acid perceived; fatty acids are overall more significant to the characteristic flavor of Italian varieties than to Cheddar, Swiss or Gouda (Mcsweeney et al., 2000). In varieties such as Romano, parmesan and provolone, lipases added to the cheese milk result in increased free carbon 4-8 fatty acids which contribute strongly to flavor (Urbach et al., 1997; Collins et al., 2003).

Due to reduced lipolytic activity, Cheddar and Gouda cheese are similar in FFA (free fatty acid) composition and contained smaller quantities of FFA than Blue type cheese (Urbach et al., 1997). However, results from Aroma Extract Dilution Analysis (AEDA) of Cheddar cheese identified seven acids as having high odor potency, including butyric acid, acetic acid and propionic acid (Christensen et al, 1995). Numerous studies have addressed FFA in Cheddar cheese; FFA composition and concentration varies between reduced fat and full fat Cheddar cheeses and between enzyme modified and control Cheddar cheeses (Nelson et al., 2004; Drake et al., 2010; Milo and Reineccius, 1997; Kilcawley et al., 2001). Acetic acid and butyric acid are generally considered to have the highest aroma impact of fatty acids in Cheddar (Drake et al., 2010; Milo and Reineccius, 1997; Kim et al., 2011). Based on sensory evaluation of full and low fat Cheddar cheese models, butyric and acetic acids was identified as a key flavor compound for both fat contents (Kim et al., 2011). Ultimately, the relationship between FFA presence and perception of cheese flavors is established for most

cheese varieties; the role of FFA in cheese as a defect or a positive aroma contributor to a variety is dependent on the concentration present and perception threshold for the compound (Collins et al., 2004). With the exception of Italian, Blue and Feta varieties, moderate FFA concentrations are considered rancid or a defect by consumers, but FFAs impart characteristic aromas and should be present in some concentration for many varieties (Collins et al., 2004; Singh et al., 2010; Dirinck et al., 1999; Singh et al., 2003; Whetstone et al., 2006).

Esters impart a fruity or coconut flavor to dairy products; this is considered more desirable in cheeses such as Parmesan and Danish Blue than varieties such as Cheddar (Urbach et al., 1997; Curioni et al., 2002; Cadwallader et al., 2009). Over 38 esters have been identified in cheese; the most common of which include ethyl butanoate, ethyl hexanoate, ethyl acetate, and ethyl octanoate (Urbach et al., 1997). Lactones are flavor compounds present in Cheddar, Gouda, Parmesan, Blue-type and other cheeses; these compounds result from the one step nonenzymatic transesterification of hydroxyl fatty acids (Alewijn et al., 2007). Over 14 types of lactones have been identified across cheese varieties (Colonna et al., 2011). Numerous delta and gamma lactones are considered influential lactones to soft varieties such as Camembert, while  $\delta$ -decalactone is important to flavor in Emmental;  $\delta$ -dodecalactone and  $\delta$ -octalactone are important to Goat cheese and Cheddar based on odor activity value (Curioni et al., 2002).

Methyl ketones form from the oxidation of saturated fatty acids into ketoacids (Urbach et al., 1997). As with fatty acids, low quantities of ketones are observed in Cheddar and Gouda compared to Italian varieties, but methyl ketones are considered characteristic to Blue cheese, varieties (Urbach et al., 1997; Patton et al., 1950; Qian et al., 2002). Heptan-2-

one is an important ketone compound to flavor in semi hard mold ripened cheese and parmesan cheeses, while 2-heptanone and 2-nonanone are crucial to Blue cheese flavor (Urbach et al., 1997; Qian et al., 2002). Nonan-2-one predominates in soft mold ripened cheese (Urbach et al., 1997). The presence of pentan-2-one and heptan-2-one in Cheddar cheese headspace is typically indicative of mold contamination (Urbach et al., 1997).

Phenols may be detected in cheese at a concentration of about 0.1 ppm (Sable et al., 1999).

Phenols impart a range of sensory flavors, including sharp, medicinal, and charred (Urbach et al., 1997). In Cheddar, phenols are considered normal in concentrations up to  $20 \mu\text{g kg}^{-1}$  (Urbach et al., 1997). Beyond this, a barny/medicine like off flavor imparted by the phenol p-cresol may be perceived (Urbach et al., 1997; Suriyaphan et al., 2001). Phenols also impart characteristic band aid and medicinal notes to smoked provolone cheese; the presence of phenols in Parmesan cheese is regarded as a desirable background characteristic (Urbach et al., 1997).

### **Autooxidation and off flavors in Dairy**

Auto oxidation of fat in milk products occurs in the polyunsaturated phospholipids fraction of the milk fat globule membrane before the main triglyceride portion (Cadwallader et al., 2009). Products differ in oxidative stability depending on whether they are suspended in water or a continuous fat phase; oxidative stability influences development of both flavor compounds and flavor defects in dairy products (Cadwallader et al., 2009). Numerous volatile compounds are produced by auto oxidation of unsaturated fatty acids; these include octanal, hexanal, 2,4-decadienal, and 2,4-heptadienal (Cadwallader et al., 2009). Thermal treatment of milk can also result in lipid degradation, thermal reactions of amino acid side

chains, Maillard reactions, and interaction of oxidative products with Maillard products (Cadwallader et al., 2009). While other thermal reactions such as peptide hydrolysis and protein dephosphorylation are possible in dairy products, they play no crucial role in dairy flavor (Cadwallader et al., 2009)

**Table 5. FFA Determination in mg kg<sup>-1</sup> for Gouda, Cheddar and Blue Cheeses (Adapted from Alewijn et al., 2005). Values in parentheses indicate the standard deviation.**

	Retention time (min)	Gouda	Cheddar	Danish Blue
Acetic acid (C2:0)	13.02	133 (7.73)	15 (0.11)	556 (267)
Propanoic acid (C3:0)	14.64	0.34 (0.08)	0.18 (0.04)	419 (397)
2-Methyl propanoic acid (C4:0i)	15.11	2.6 (0.18)		
Butanoic acid (C4:0)	16.17	12 (0.94)	3.9 (0.12)	718 (14.6)
3-Methyl butanoic acid (C5:0i)	16.92	11 (0.91)		37 (2.18)
Hexanoic acid (C6:0)	19.86	3.4 (0.19)	1.7 (0.07)	469 (14.0)
Octanoic acid (C8:0)	23.19	1.5 (0.02)	2.3 (0.03)	145 (2.41)
Decanoic acid (C10:0)	26.19	8.0 (0.19)	11 (0.11)	543 (6.80)
Dodecanoic acid (C12:0)	28.97	9.9 (0.12)	8.6 (0.03)	522 (39.8)
Tetradecanoic acid (C14:0)	31.7	27 (0.06)	27 (0.19)	1810 (163)
Pentadecanoic acid (C15:0)	33.34	1.5 (0.06)	1.6 (0.04)	94 (17.8)
Hexadecanoic acid (C16:0)	35.37	69 (0.87)	76 (0.03)	1697 (323)
Octadecanoic acid (C18:0)	40.82	2.4 (0.14)	2.1 (0.03)	59 (14.2)
Decenoic acid (C10:1)	27.05	0.80 (0.02)	1.3 (0.00)	93 (1.1)
Dodecenoic acid (C12:1)	29.55	0.89 (0.02)	0.72 (0.05)	10.0 (2.5)
Tetradecenoic acid (C14:1)	32.47	4.2 (0.16)	6.4 (0.24)	301 (47.7)
9-Hexadecenoic acid (C16:1)	36.2	4.5 (0.09)	3.9 (0.18)	434 (109)
11-Hexadecenoic acid (C16:1)	36.75	0.15 (0.15)	0.05 (0.01)	9.9 (2.6)
Oleic acid (C18:1)	42.02	36 (0.20)	43 (0.35)	5057 (1228)

## Characterization of Gouda Cheese Compounds

Volatile compounds shown to be formed during ripening of Gouda cheese include FFAs, organic acids, lactones, esters and ketones (Califano et al., 2000). Table 2 displays the FFA content for Gouda cheese along with Cheddar and Blue Cheese. Amino acid related flavor compounds and ethyl esters increased with ripening of pasteurized milk Gouda cheeses (Urbach et al., 1997). Linear aldehydes, FFAs and branched chain acids decreased with ripening. Despite the presence of fat in milk, lipolysis is negligible in Gouda type cheeses compared to other varieties (Alewijn et al., 2003). Lipases are sometimes added to dairy to accelerate ripening by increasing triglyceride hydrolysis, but soapy off flavors may result from production of medium chain fatty acids, so lipases are not typically used in modern commercial Gouda production due to the possibility of discernible flavor and texture defects (Hasan et al., 2005; Van Den Berg et al, 1993). Lipases active in Gouda cheese production are derived from the starter culture used; native milk enzymes are usually inactivated by pasteurization and lipase containing rennet paste is not used in production of Dutch type cheeses (Fox et al., 2000; Collins et al., 2003). Although *Lactococcus* is considered weakly hydrolytic compared to other lactic acid bacteria, their presence in high concentrations during ripening is responsible for FFA production (Collins et al., 2003).

Caramel imparting odorants have been identified in flavor chemistry studies of both Gouda and Emmental varieties (Preininger et al., 1996, Inagaki et al., 2015). AEDA analysis and Isotope dilution assays of Emmental were used to identify four compounds as characteristic to the variety (Preininger et al, 1993). Methional, 4-hydroxy-2,5-dimethyl-3(2H)-furanone, and 5-ethyl-4-hydroxy-2-methyl-3-(2H)-furanone were shown to be

characteristic odorants based on high odor activity value (OAV) (Preininger et al., 1993). Flavor models containing these three compounds were later generated to model Swiss like cheese flavor (Preininger et al., 1996). Thirty one odorants were identified in three Japanese Gouda cheeses (Inagaki et al., 2015). Sixteen compounds were identified as characteristic to Gouda based on an AEDA dilution factors (FD values) value greater than 4 (Inagaki et al., 2015). These compounds included two esters, three pyrazines, six acids, 3-hydroxy-4,5-dimethyl-2(5H)-furanone, 4-dodecanolide, phenylacetic acid and two unknowns (Inagaki et al., 2015). However, this study did not address potential highly volatile odorants in Gouda which may not be retained by solvent extraction.

Organic acids are influential to flavor and aroma compound production; lactic acid is important to quality, manufacturing and ripening in cheese (Califano et al., 2000). Production of lactic, citric, acetic and pyruvic acids in Gouda cheese is directly correlated to time and temperature (Califano et al., 2000). At the beginning of Gouda cheese production, lactic acid comprised approximately 90% of the total organic acid content; after processing and aging for duration of 70 days, lactic acid content was reduced to 73%, citric acid increased to 7.8%, propionic acid to 9.8% and acetic acid to 6.1% in Gouda samples evaluated (Califano et al., 2000). No butyric acid was detected during analysis of Gouda cheese (Califano et al., 2000).

The milk used to produce cheese is partially responsible for FFA presence in Gouda (Kanawjia et al., 1995). Commercial Edam cheeses contained relatively low concentrations of FFA (C<sub>4</sub> to C<sub>18</sub>) (Woo et al., 1984). Descriptive analysis of Edam cheese identified the attributes “smooth, mildly nutty and milky” as the top notes; this indicates FFA in Dutch



cheeses may serve as more of a background flavor component compared to other cheeses (Woo et al., 1984). It is likely FFAs in Gouda cheeses are formed from monoglycerides and diglycerides (Kanawjia et al., 1995).

Formation of methyl ketone compounds in Gouda type cheeses results from enzymatic or chemical processes of milkfat (Califano et al., 2000; Alewijn et al., 2005). Methyl ketones are not considered critical to flavor in Gouda cheese formation; this is due to their lipophilic nature and the high fat content in Gouda cheese (Califano et al., 2000). The presence of esters in Gouda is dominated by ethyl esters of long chain fatty acids (Califano et al., 2000). However, analysis of ester composition in eight commercial Gouda cheeses demonstrated that ester compounds varied among Gouda cheeses (Califano et al., 2000). Variation between esters was significantly larger in commercial raw milk cheeses than pasteurized milk cheeses (Califano et al., 2000).

Previous studies concerning Gouda cheese have shown that a variety of odor active compounds are present and possibly characteristic of Gouda cheeses (Inagaki et al., 2015; Van Leuven et al., 2008; Califano et al., 2000). Compounds identified as responsible for Gouda include those from numerous chemical classes, including esters, aldehydes, ketones, free fatty acids, pyrazines and furanones (Inagaki et al., 2015; Van Leuven et al., 2008). However, there is a need to establish characteristic Gouda flavor compounds in terms of highly volatile compounds and their relationship to analytical sensory methods.

### **Approach to Flavor Characterization**

Although flavor is ultimately defined by sensory characteristics, there is a need to understand the chemical components that contribute to flavor. Because aroma is crucial to

identification of a flavor, flavor research has historically centered on the study of volatile compounds present (Reineccius, 2005). However, flavor is now recognized as a multifaceted experience that includes chemesthesis and taste, as well as aroma (Reineccius, 2005).

Identification of aroma active components present in a food is challenging due to the complexity of the food matrix and the lower limit of instrumental analysis compared to the human nose (Reineccius, 2005).

To identify and characterize flavors present in food, descriptive sensory analysis should be utilized in conjunction with chromatographic methods such as gas chromatography mass spectrometry (GC-MS) and gas chromatography olfactometry (GC-O) (Van Leuven et al., 2008). GC-O is advantageous to the determination of key volatile compounds in dairy products because it allows only for the detection of volatiles present in concentrations higher than their thresholds, also called aroma-active or flavor-active compounds (Drake, 2004).

Relating volatile composition results from GC-MS to descriptive analysis is an effective and widely used method for volatile compound identification and characterization of key flavor compounds (Van Leuven et al., 2008). This process is accomplished by a widely accepted three step method that includes 1) selecting products containing desired flavors and profiling them by descriptive analysis 2) conducting instrumental volatile analysis to detect and identify the corresponding flavors and 3) confirming these compounds as characteristic using a combination of instrumental quantitation, threshold testing and model systems (Weimer, 2007). Descriptive analysis is used alongside instrumental detection methods because panelists are trained as instruments to provide quantitative measurements and flavor is ultimately a *sensory* perception (Plemmons et al., 1998). Numerous previous studies have

combined sensory evaluation, GC-O/GC-MS analysis and model system simulation to identify characteristic aroma active compounds of cheeses (Avsar et al., 2004; Carunchia Whetstine et al., 2003; Carunchia Whetstine et al., 2005; Christensen et al., 2005 ; Poveda et al., 2007; Suriyaphan et al., 2001). Use of SPME GCMS followed by SPME GC-O has also been effective in characterizing the aroma active compounds in sour cream (Shepard et al., 2013). Sour cream compounds were identified based on the combination of retention indices for two GC columns, comparison of odor properties and evaluating the mass spectra of the samples in comparison to literature or standards (Shepard et al., 2013; Seitz et al., 1990).

## **II. Color, Texture, and Rheological Properties of Gouda Cheese**

### **Factors affecting Color of Cheese**

Color uniformity and stability in cheese is of concern because consumer testing has shown that when the color of the cheese differs from what is expected, flavor identification decreases and the role of color on overall liking increases (Wadhvani et al., 2012). Without the addition of colorant, cheese color is the result of fat soluble carotenoids present in milkfat (Wendorff et al., 2007). Carotenoids impart a characteristic yellow color to dairy products; cheeses made from cows fed native pasture plants are higher in yellow color than those fed silage diets (Wendorff et al., 2007). Carotenoids vary in concentration based on lactation stage, breed, and feed composition; they are susceptible to breakdown by aerobic oxidation and light oxidation, although they are not degraded during thermal processing of milk (Calderon et al., 2007; Wendorff et al., 2007).

Colorants are added to cheese milk during cheese production to increase uniformity in cheese appearance (Johnson et al., 1998; Wadhvani et al., 2012). Annatto extract is

considered a natural colorant in the U.S. and is typically added to U.S. and Dutch produced Gouda and Cheddar cheeses to impart a yellow orange color (Wadhvani et al., 2012; Ebo et al., 2009; Van Middelar et al., 2011). Annatto is added to Dutch type cheeses at a concentration of 0.24 grams to 1 kg cheese (Van Middelar et al., 2011). Although annatto is capable of creating a more consistent product, it is five times more susceptible to oxidation than carotenoids (Wendorff et al., 2007). Cheeses colored by annatto may develop a pink color due to oxidation by fluorescent light exposure; increased light intensity has been correlated to increased development of the pink defect in cheese (Wendorff et al., 2007). Color fade in cheese can be minimized by optimizing packaging conditions to reduce light exposure and minimize oxygen headspace (Wendorff et al., 2007).

In a study monitoring color during ripening for Emmental and hard cheeses, regression analysis indicated that cheese body color was most affected by fat content that result from light scattering properties imparted by fat (Rohm and Jaros, 1996; Wadhvani et al., 2012 ). This is consistent with studies regarding the appearance of Mozzarella cheese, where lower fat cheeses are more translucent in color (Paulson et al., 1998). Total solids content and the cheese ripening period were also shown to have a minor effect on color (Rohm and Jaros, 1996).

### **Factors influencing Cheese Texture**

Two distinct phases related to cheese texture occur during ripening (Lawrence et al., 1987). Phase one takes place during weeks 1-2 and converts the cheese from a rubbery texture to a smoother, more homogenous one by hydrolysis of the  $\alpha$ 1 casein (Lawrence et al., 1987). Phase two occurs gradually in subsequent months and alters cheese texture by 1)

proteolysis of  $\alpha$ s1 and other caseins and 2) increased pH (Lawrence et al., 1987). Low moisture cheeses harden with age because water is consumed in the formation of ionic groups during the proteolytic process (Lawrence et al., 1987). The rate of proteolysis for cheese is primarily affected by the amount of residual rennet and milk proteinases present, the salt: moisture ratio, type of coagulant and change in pH. Changes in pH during cheese ripening influences the protein network of the curd; after a pH level below 5.5, casein submicelles will dissociate into smaller casein aggregates (Lawrence et al., 1987). As the pH approaches the isoelectric point of casein, the cheese will become shorter in texture and assume a more compact conformation (Lawrence et al., 1987). An increase in ripening temperature by 10° C will more significantly impact flavor development than texture in cheese (Lawrence et al., 1987). However, Cheddar cheeses ripened at a higher temperature increase by 20° C were shown to be more brittle and less springy in texture (Lawrence et al., 1987).

### **Texture of Gouda Cheese**

While flavor intensity across Dutch type cheeses varies, the texture is generally smooth with small holes and a semi hard to hard consistency (Welthagen et al., 1998). Citrate metabolism is responsible for the eyes in Dutch-type cheeses; this is imparted by production of CO<sub>2</sub> by lactic acid bacteria (Cadwallader et al., 2009). Crystallization of calcium D-lactate on cheese surfaces may affect texture and impart what consumers would consider a defect (Cadwallader et al., 2009).

Descriptive analysis of Gouda cheeses indicated texture differences were primarily related to age and fat content (Yates et al., 2007). Young Gouda cheeses were characterized

by higher intensities of chew-down, adhesiveness, cohesiveness, breakdown and smoothness of mouth coating; aged Goudas were higher in intensities of attributes fracturability and firmness (Yates et al., 2007). The aging of Goudas with a natural or wax rind allows moisture loss with time, ultimately resulting in increased firmness (Yates et al., 2007). Compared to regular Gouda cheese, lower fat Gouda cheeses were higher in texture attributes springiness and recovery and lower in texture attributes adhesiveness, cohesiveness and degree of breakdown; these differences can be attributed to a more intact protein matrix in lower fat Gouda Cheese (Yates et al., 2007).

### **III. Analytical means for Characterization of Cheeses**

Chromatography is the separation of compounds based their interaction with mobile and stationary phases. Column chromatography is a widely applied form of chromatography in food science flavor analysis; both gas and liquid chromatography can be used to aid in the characterization of flavor and aroma compounds.

#### **Gas Chromatography Mass Spectrometry**

In gas chromatography (GC), a gas mobile phase travels through a stationary phase containing column (Kitson et al., 1996). The analyte is carried through the column by the gas, and separation occurs based on partitioning with stationary and mobile phases (Kitson et al., 1996). Mass spectrophotometers coupled to GC (GC-MS) can be used as detectors to efficiently identify compounds based on ion fragmentation patterns (Cadawallader et al., 2009). Selected ion monitoring (SIM) is an MS mode that is advantageous because it allows for increased detection of desired ions (Seitz et al., 1990). In SIM mode, the instrument

records only the ion current for selected masses rather than the entire range of ion masses present, allowing for enhanced detection sensitivity (Kitson et al., 1996).

Several preparation methods exist for analysis of cheese by gas chromatography (GC) (Marilley et al., 2004). However, techniques such as steam (SDE- GC-MS), high vacuum and molecular distillation to extract and concentrate volatiles for GC analysis are time consuming and may result in thermal degradation (Marilley et al., 2004). These techniques are better suited for analysis of low volatile compounds such as phenols, FFA, ketones and long chain aldehydes, ketones, alcohols and esters (Marilley et al., 2004). Extraction of cheese samples with acetonitrile solvent has been shown to concentrate flavor compounds in cheese, including those from the fat (Alewijn et al, 2003). However, use of ACN as a solvent prevents detection of highly volatile compounds (Alewijn et al., 2003). SDE-GC-MS profiling using dichloromethane as a solvent resulted in identification of sixty three volatiles in Gouda-type cheeses (Van Leuven et al, 2003).

Headspace analysis is advantageous over solvent extraction due to short analysis times, increased reproducibility and reduced sample preparation (Marilley et al., 2004). However, this technique is only applicable to analysis of readily volatile and concentrated compounds (Marilley et al., 2004). Headspace analysis can be performed by capturing and injecting volatile containing air, or by immersive techniques that expose the stationary phase to the headspace and inject the stationary phase. The purge and trap method has been used to analyze headspace for Cheddar cheese, but this is method susceptible to contamination (Marilley et al., 2004). Solid phase microextraction (SPME) is an immersive headspace technique that is advantageous due to its high sensitivity and minimal sample preparation

(Marilley et al., 2004). To perform SPME, a fiber coated with a stationary phase is exposed to a sample headspace or liquid to adsorb volatiles; the loaded fiber is then desorbed onto the GC injector port (Prosen et al., 1999). SPME has been used widely as an effective method for identification of flavor volatiles in cheese (Carpino et al., 2004; Delgado et al., 2010; Di Cagno et al., 2007; Drake et al., 2010; Frank et al., 2004; Jung et al., 2013; ).

### **Gas Chromatography- Olfactometry**

Although GCMS is useful for compound identification, it cannot be used to determine whether a compound is odor active. Gas Chromatography-Olfactometry (GC-O) is an effective method for determining the identity and activity of odorants (Curioni and Bosset, 2002; Wardencki et al., 2013). During GC-O, the analyte is injected into the column and the effluent is split and sent to both a GC detection instrument and a sniffing port (Reineccius, 2005). An aromagram can be created based on detection and identification of aroma active compounds by a trained sniffer; this is valuable because it provides immediate insight into which peaks have an odor (Reineccius, 2005). Typically, more than one sniffer is used in the analysis to detect the maximum number of compounds and reduce fatigue; experienced sniffers are capable of identifying a compound based on aroma and GC retention properties (Reineccius, 2005).

GC-O has been utilized to detect hundreds of volatile compounds in cheese; carboxylic acids, sulfur related compounds, esters, alcohols, lactones and ketones are considered the most important chemical classes to cheese aroma (Thomsen et al., 2011; Curioni and Bosset, 2002). Cheese varieties previously analyzed by GC-O include, Camembert, Cheddar, Gorgonzola, Gouda, Gruyere, Parmigiano-Reggiano, Mozzarella,



Pecorino, Smear ripened, Goat's milk and Ewe's milk cheeses (Curioni and Bosset, 2002; Lecanu et al., 2002; Frank et al., 2004; Inagaki et al., 2015; Reineccius et al., 2002). GC-O sniffing of seven semi hard cheeses identified 2,3-butanedione, 3-methylbutanoic acid, (s)-2-methylbutanoic acid, methional, 1-octen-3-one, dimethyl trisulfide and methyl 2-methyl-3-furyl disulfide as the highest intensity compounds; despite not being present in all samples, these compounds were considered characteristic to semi hard cheeses because they were responsible for perceived differences between the cheeses (Thomsen et al., 2011). Japanese Gouda cheeses were analyzed using AEDA to identify potent odorants (Inagaki et al., 2015). Thirty eight aroma active compounds were identified, including esters, pyrazines, carboxylic acids, hydroxy-4,5-dimethyl-2(5H)-furanone, 4-dodecanolide and phenylacetic acids (Inagaki et al., 2015). Ten carboxylic acids, dimethyl trisulfide and methional were identified by both GCMS and GC-O (Inagaki et al., 2015). Sixteen odorants were characterized as key odorants based on increased FD factors with increasing age (Inagaki et al., 2015).

### **Liquid Chromatography**

In liquid chromatography (LC), a mobile phase travels through a stationary phase containing column and separation occurs based on compound interaction with the solid stationary and liquid mobile phase. Compounds are identified based on retention time and quantified by use of standard curves (Berger et al., 2001). LC has been used to quantify organic acids, identify starter culture composition by monitoring organic acids, determine annatto content, quantify diacetyl, and quantify acetoin in cheese (González de Llano et al., 1996; Bareth et al., 2002; Marilley et al., 2004; Zeppa et al., 2001). High Pressure Liquid Chromatography (HPLC) is a widely used and more sensitive type of liquid chromatography

that enables detection of the glycolytic and fermentative pathways and amino acid intermediates (Marilley et al., 2004). Several detectors exist for use with HPLC, including fluorescence, refractive index, electrochemical, light scattering and mass spectrometry (Berger et al., 2001). Ultraviolet-visible (UV-vis) absorbance detectors are most common detector used; they function by measuring absorbance from compounds of interest according to Beer's law (Berger et al., 2001; Swartz et al., 2010). Photo diode array (PDA) detectors function similarly to UV detectors, but can detect absorbance at multiple wavelengths to create a library and identify peaks (Swartz et al., 2010).

HPLC analysis was performed for milk samples inoculated with six different starter cultures and the organic acid composition was determined for samples, demonstrating the ability of HPLC to follow fermentation paths based on varying organic acid composition (González de Llano et al., 1996). When analyzed by HPLC for organic acids, cheeses manufactured from these six varying starter cultures were highest in orotic and citric acids; butyric acid was not detected in the samples (González de Llano et al., 1996).

Bouzas et al. (1991) proposed that HPLC could be used to simultaneously determine organic acid and sugar content during cheese ripening. Cheddar cheeses were analyzed by a cation exchange column and compounds detected using a refractive index detector connected in series to a UV-vis detector (Bouzas et al., 1991). Lactic acid and propionic acid concentrations increased during ripening, while the concentration of propionic acid decreased (Bouzas et al., 1991). Organic acid determination in Gouda cheese was performed using a cation exchange column and a photo diode array UV-Vis detector (Califano et al., 2000). Organic acid concentration in Gouda cheese tended to increase linearly with ripening time

and temperature (Califano et al., 2000). Five acids comprised 96% of the organic acid composition, with lactic acid accounting for 73% of the organic acid content (Califano et al., 2000).

#### **IV. Sensory Evaluation and Consumer Testing**

Sensory analysis is used to measure human responses resulting from use or consumption of a product, including appearance, aroma, flavor, texture, and sound (Moussaoui et al., 2010; Murray et al., 2001). Numerous sensory tests exist that can be applied to product development, quality control, and consumer liking objectives (Drake et al., 2007; Murray et al., 2001). Affective sensory testing investigates consumer acceptance and preference while analytical sensory testing is used to investigate product differences, determine thresholds, and identify and quantify product attributes (Drake et al., 2007).

##### **Descriptive Analysis**

Descriptive Analysis (DA) is an analytical sensory method for detecting and quantifying the sensory attributes of a food (Drake et al., 2007). In DA, a trained panel usually consisting of 6-12 persons evaluates samples, with individuals functioning as sensors and the group functioning as one instrument (Drake et al., 2007). DA can be utilized to investigate numerous objectives, including flavor and texture characterization, shelf life studies, and off flavor identification (Meilgaard et al., 2006). DA is advantageous because it can be related to consumer preference results using multivariate techniques to determine the optimal product formulation (Murray et al., 2001). Several procedures exist to standardize terminology and score intensities for products, including the Spectrum™ method and the Quantitative Descriptive Analysis (QDA) method (Meilgaard et al., 2006).

Descriptive analysis has been utilized to measure aging and the effect of compositional parameters on Cheddar cheese flavor formation (Drake et al., 2001; Young et al., 2004; Caspia et al., 2006; Drake et al., 2008; Drake et al., 2009; Wadhani et al., 2012). Limited work has been done to document Gouda cheese. Two commercial Gouda cheeses ripened for two months were provided by Belgian cheese makers; one cheese was manufactured using raw cow's milk and the other using milk pasteurized at an unspecified temperature. (Van Leuven et al., 2008) Cheeses were profiled by a trained panel using a lexicon adapted from Muir et al. (1995) that consisted of six aromatics and five taste attributes (Van Leuven et al., 2008). Samples scored similarly for attributes "animal, flavor intensity, taste intensity, sour, salt, bitter, pungent" (Van Leuven et al., 2008). These attributes increased in intensity with ripening, while scores for the attributes "sweet" and "creamy buttery" decreased with ripening (Van Leuven et al., 2008). Raw milk Gouda cheeses were higher in intensity for attributes "sweet, bitter, fruity/flowery, nutty, chocolate-like, animal" (Van Leuven et al., 2008). Pasteurized milk Gouda cheeses were higher in intensity for attributes "flavor intensity, sour, pungent" (Van Leuven et al., 2008).

Numerous studies regarding cheese have combined trained panelist descriptive analysis with consumer tests of liking and dissimilarity to determine consumer flavor preferences using preference mapping (Lawrence et al., 1987; Young et al., 2004; Drake, 2008; Caspia et al., 2005; Wadhani et al., 2012). Preference mapping is a multivariate technique that can be used to relate DA results to consumer acceptance data, ultimately enabling the researcher to determine specific market segments (Tenenhaus et al., 2005; Drake et al., 2004).

## **Consumer Testing**

Consumer testing is used to measure preference and/or acceptance of a product or concept by untrained consumers (Drake et al., 2007; Meilgard et al., 2006). Consumer testing can be qualitative or quantitative; common methods of quantitative testing include acceptance testing and conjoint analysis, while common qualitative methods include focus groups and interviews (Drake et al., 2007).

Although no studies have addressed the relationship between Gouda cheese flavor and consumer acceptance, Yates et al. (2007) conducted DA and consumer testing to investigate the ideal texture of Gouda cheese. Young Gouda cheeses (<6 months) were higher in intensity for the attributes “adhesiveness”, “breakdown”, and “smoothness of mouth coating” while aged Gouda cheeses were higher in intensities of “fracturability” and “firmness” (Yates et al., 2007). Consumer testing conducted revealed a preference for younger Gouda cheeses, but flavor liking scores for the more aged Gouda did not differ from the flavor liking score for the three younger and most preferred cheeses (Yates et al., 2007). Texture characteristics of the ideal Gouda cheese include “smooth” and “creamy”, attributes characteristic of mild or young Gouda cheeses (Yates et al., 2007). These facets ultimately indicated that, while flavor plays a significant role in consumer acceptance of Gouda cheese, it is not enough to compensate for undesirable texture (Yates et al., 2007).

Specialty cheeses are generally defined as non-generic, more distinctive and individual type cheeses; they comprise a significant amount of the specialty foods market (Kupiec et al., 1998). To determine consumer attitudes towards purchase behavior of specialty cheeses and raw (RMC) versus pasteurized (PMC) cheeses, 890 specialty cheese

consumers were surveyed and tasted cheese pairs of one RMC and one PMC (Colonna et al., 2011). Consumers stated that they were most likely to purchase new specialty cheeses at “natural foods grocery stores”, “conventional grocery stores” and “farmer’s markets” (Colonna et al., 2011). Consumers who liked pungent cheeses stated that they were willing to pay more per pound of cheese; older consumers, consumers of large purchase size, and consumers who shopped in conventional supermarkets were less likely to purchase expensive cheeses (Colonna et al., 2011). Results from acceptance tasting of cheeses revealed a significant difference in preference; 426 consumers preferred RMC and 319 preferred PMC (Colonna et al., 2011). Consumers attributed their preference for RMC to be due to flavor, but results suggest that consumer perception of RMC was enhanced by labeling a cheese as from raw milk (Colonna et al., 2011).

Several studies have investigated what factors influence consumer acceptability of Cheddar cheese (CC) (Caspia et al., 2005; Drake et al., 2008; Drake et al., 2009; Roberts et al., 1994; Wadhani et al., 2012; Young et al., 2004). Previous studies by Drake et al. (2008), Caspia et al. (2005), Young et al. (2004) and Drake et al. (2009) established that consumer preferences for CC flavor and color can be related to CC age. Drake et al. (2008) used preference mapping of mild CC and revealed four distinct consumer clusters; consumer liking scores were lower for samples characterized by aged flavors and cheeses with bland flavor profiles. Sharp CC acceptability was investigated by Drake et al. (2009) and revealed five distinct consumer segments with varied preferences for flavor and CC age. Similar studies by Caspia et al. (2005) and Young et al. (2004) indicated that acceptability of CC of differing ages varied according to the consumer cluster. Both young and old CC scored high

in perceived “Cheddar cheese intensity”; this suggests that the flavors which constitute “Cheddar flavor” vary according to the consumer (Young et al., 2004).

To determine flavor preferences for Swiss cheese, consumer and descriptive data was evaluated to determine the most significant drivers of impact on liking (Liggett et al., 2008). Diacetyl was the most impactful driver of liking in Swiss cheese (Liggett et al., 2008). Additional positive drivers included whey, milk fat and umami while negative drivers of liking included cabbage, cooked and vinegar for Swiss cheese (Liggett et al., 2008). Baby Swiss was closest to the consumer ideal due to the combination of flavor and feeling factors (Liggett et al., 2008). However, additional properties not evaluated by descriptive analysis included texture and appearance (Liggett et al., 2008). Findings ultimately suggest that liking of Swiss cheese is primarily but not exclusively driven by flavor. Additional studies investigating consumer preferences for Mozzarella cheese, Finnish cheeses, and Spanish Idiazabal cheese have established preferences for these varieties based on region and the importance of flavor on acceptability (Monteleone et al., 1997; Ritvanen et al, 2005; Barcnas et al., 2001). Preference mapping with Gouda cheeses has not been conducted.

## **V. Conclusion**

Given the importance of flavor to consumer liking and increasing domestic sales of Gouda cheese, it is important to have a definitive understanding of what the key aroma active compounds in Gouda cheese are and how they influence consumer acceptance. The objective of this study was to identify the chemical and sensory properties of Gouda cheese and their relationship to drivers of liking.

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## **CHAPTER 2**

### **Sensory and Chemical Properties of Gouda Cheese**

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## ABSTRACT

AMEERALLY, ANGELIQUE DANIELLE. Sensory and Chemical Properties of Gouda Cheese. (Under the direction of MaryAnne Drake).

Gouda cheese (G) is a Dutch, washed curd cheese that is traditionally produced from bovine milk and brined before ripening for 1-20 months. In response to domestic and international demand, U.S. production of Gouda cheese has more than doubled in recent years. An understanding of the chemical and sensory properties of G can help manufacturers to create desirable products. The objective of this study was to determine the chemical and sensory properties of Gouda cheeses. Commercial Gouda cheeses (n=36, 3 mo to 5 y, domestic and international) were obtained in duplicate lots. Volatile compounds were extracted (SPME) and analyzed by gas chromatography olfactometry (GCO) and gas chromatography mass spectrometry (GCMS). Physical analyses included pH, proximate analysis, salt content, organic acid analysis by HPLC, and color. Flavor and texture properties were determined by descriptive sensory analysis. Focus groups were conducted with cheese followed by consumer acceptance testing (n=153) with selected cheeses. Ninety aroma active compounds were detected in cheeses by SPME-GC-O. Key volatile compounds in Gouda cheeses included dimethyl sulfide, 2,3-butanedione, 2/3-methylbutanal, ethyl butyrate, acetic acid, and methional. Older cheeses had higher organic acid concentrations, higher fat and salt content, and lower moisture content than younger G. Younger cheeses were characterized by milky, whey, sour aromatic, and diacetyl flavors while older G were characterized by fruity, caramel, malty/nutty, and brothy flavors. International cheeses were differentiated by the presence of low intensities of cowy/barny and grassy flavors. Younger cheeses were

characterized by higher intensities of smoothness and mouthcoating while older cheeses were characterized by higher intensities of fracture and firmness. American consumers utilized Gouda cheeses in numerous applications and stated that packaging appeal, quality and age were more important than country of origin or nutrition when purchasing cheeses. Young and medium U.S. cheeses  $\leq 6$  mo were most liked by U.S. consumers. Three distinct consumer segments were identified with distinct preferences for cheese flavor and texture based on ripening time. Findings from this study establish key differences in Gouda cheese regarding age and origin and identify U.S. consumer desires for this cheese category.

Key Words:

Gouda cheese, flavor, preference mapping

## INTRODUCTION

Gouda cheese is a washed curd Dutch cheese that is traditionally produced from bovine milk and brined before ripening for 1-20 months (Van Den Berg et al., 2004; Mellgren, 2005; Jung et al., 2013). Gouda and Edam cheeses constitute the two main types of Dutch cheese and differ internationally in their requirements for milk fat content used to produce the cheese and acceptable weights (Walstra et al., 1993; Codex Alimentarius, 2013). Gouda cheese is defined in the U.S. by the Code of Federal Regulations (CFR). CFR specifies a maximum moisture content of 45% and a minimum fat content of 46% on a dry weight basis for Gouda cheeses. Between 2010 and 2014, US production of Gouda cheese increased from 19 to 48 million pounds per year (USDA, 2014). As a result of initiatives between US manufacturers and overseas buyers, Gouda cheese export has increased dramatically since 2008 and is considered to have the most potential for cheese export (US Dairy Export Council, 2012; Cheese Market News, 2007). Understanding the sensory and chemical properties of Gouda cheese and how they influence consumer acceptance can help manufacturers to create a desirable product.

Flavor, followed by texture and appearance, are the three attributes which most influence liking of a food (Moskowitz and Krieger, 1993). Specific flavor profiles of products are documented using a trained sensory panel. Identification and characterization of key flavor compounds can then be conducted using gas chromatography mass spectrometry and gas chromatography-olfactometry (GC-O). This process has been applied to numerous foods, including sweet cream butter, berries, yogurt, milk powders, and cheeses (Drake et al., 2004; Carunchia et al., 2007, Du et al., 2011; Wright et al.,

2006; Zellner et al., 2008). Trained panel results can then be used to confirm GC-O findings and to quantitatively interpret consumer acceptance (Drake et al., 2004). Numerous studies of dairy products have correlated analytical sensory and instrumental results or analytical sensory and consumer acceptance (Childs et al., 2009; Shepard et al., 2013; Drake et al., 2008; Van Leuven et al., 2007; Murray et al., 2000; Young et al., 2004).

Previous studies with Gouda cheese have investigated fatty acid composition, the formation mechanism of lactones, and organic acid composition (Iyer et al., 1967; Alewijn et al., 2007; Califano et al., 2000). Sixty three volatiles were previously identified in two Belgian Gouda cheeses, one raw milk cheese and one pasteurized milk cheese at different ripening times, by gas chromatography mass spectrometry (GCMS). Characteristic flavor differences between the two cheeses were determined by descriptive analysis, but aroma activity was not investigated by GC-O (Van Leuven et al., 2007). Gouda cheeses previously analyzed by GCMS were differentiated from Emmental cheeses by higher intensities of concentrations of  $\delta$ -decalactone and  $\delta$ -dodecalactone and higher intensities of “buttery” notes by sensory analyses (Dirinck et al., 1999). Tungjaroenchai et al. (2004) conducted headspace analysis of Edam cheeses and identified acetic acid as the most abundant free fatty acid (FFA). Differences in FFA composition were documented between whole and reduced fat Edam cheeses (Tungjaroenchai et al., 2004).

Inagaki et al. (2015) evaluated aroma active volatile compounds in 1 young, 1 medium and 1 aged Gouda cheese. Cheeses were solvent extracted and concentrated using solvent

assisted flavor evaporation (SAFE) followed by aroma extract dilution analysis (AEDA) (Inagaki et al., 2015). Sixteen odorants increased with ripening time, but only three cheeses were evaluated (Inagaki et al., 2015). Consumer testing and descriptive analysis of Gouda cheese texture (n=21 cheeses) indicated consumer preference for a smooth/cohesive cheese texture rather than one of higher fracturability, firmness or springiness (Yates et al., 2007). This study also documented a variety of flavors and colors for commercial Gouda cheeses and the authors suggested that flavor was a key driver of liking for consumer acceptance (Yates et al., 2007). No previous study has investigated the chemical and sensory properties of a wide range of Gouda cheeses. The objective of this study was to characterize the sensory and chemical properties of Gouda cheese and to determine the drivers of liking for Gouda cheese with U.S. cheese consumers. Descriptive analysis and instrumental testing were conducted on a wide array of Gouda cheeses. Subsequently, consumer focus groups and consumer acceptance testing were conducted.

## **MATERIALS AND METHODS**

### **Gouda Cheeses**

Commercial Gouda Cheeses (n=36) were obtained in duplicate lots from five countries (USA, The Netherlands, Finland, Denmark, New Zealand). Samples ranged in age from 3 mo to 3 y and included both raw and pasteurized milk cheeses. Samples were shipped overnight and were examined for damage upon arrival. Products were stored in the dark at 4°C for descriptive analysis and consumer testing. One thousand grams of each cheese was subsampled and stored at -80°C for analytical testing and analyzed no later than two months after arrival.

## **Composition Analysis**

Proximate analysis, pH measurements, color measurements and salt content measurements were conducted in duplicate for all Gouda cheeses. Moisture content was determined by drying three grams of cheese in a vacuum oven at 110°C for 30 min and measuring the difference in mass before and after drying. Fat content was determined using a modified Mojonnier extraction method (procedure 989.05; AOAC, 2000) with 0.25 g of grated cheese. Hunter L\*a\*b\* color analysis was performed by placing a Minolta Chroma meter (CR-410; Minolta, Ramsey, NJ) directly on a 6 x 6x 3 cm block of cheese at 23° C (Dufosse et al., 2005). Cheeses were analyzed for salt content using a Salt Analyzer (TOA-DKK SAT 500; Analyticon Instruments Corp., Springfield, NJ). Salt content was determined by adding three grams of grated homogenized cheese to a 10 mL beaker and following the protocol and calculations provided by the machine manufacturer (Analyticon Instruments Corp., Springfield, NJ). Measurements for pH were conducted by placing one gram of grated cheese in a 45 mL centrifuge tube (VWR, Radnor PA) with five mL of water and vortexing the solution for 15 s to ensure mixing. The pH of the mixture was measured with a pH meter (Mettler-Toledo, GmbH, Schwerzenbach, Switzerland) by inserting the pH electrode probe (BNC; Corning Inc., Corning, NY) into the mixture (Upreti et al., 2004). Measurements were conducted on each cheese in duplicate (4 total measurements for each cheese).

## **Organic Acid Quantification**

Organic acids were extracted and analyzed according to a modified method described by Califano and Bevilacqua (2000). Five grams of cheese was grated and added to 20 mL of 0.013 N H<sub>2</sub>SO<sub>4</sub> (2.0 N; Mallinckrodt Baker Inc., Phillipsburg, NJ) in a 120 mL centrifuge

tube. Samples were shaken on high for 30 min (Barnstead Thermolyne 50800 Rotomix) and centrifuged (Sorball model RC-B5; Thermo Scientific, Waltham, MA) at 6000 x g for 10 min. The supernatant was then collected and filtered through a 0.45  $\mu\text{m}$  nylon membrane (VWR International LLC, West Chester PA). Samples were analyzed by HPLC (Waters 2996; Waters Inc., Milford, MA) using a cation exchange column (Aminex HPX-87H, 300 x 7.8 mm) with a mobile phase of 0.013 N  $\text{H}_2\text{SO}_4$  at a flow rate of 0.8 mL/min and an injection volume of 10  $\mu\text{L}$ . Cheeses were extracted in duplicate and samples were injected in duplicate for a total of four replications per sample. Separate organic acids were detected using a photodiode array detector at wavelengths 210 and 290 nm (2996; Waters Inc.) and software Empower (Waters Inc.). Organic acids were identified by comparing retention times of chemical standards and quantified by creating five point standard calibration curves for each organic acid. Organic acid standards were purchased from Sigma Aldrich (Saint Louis, MO) and VWR International LLC (West Chester, PA).

### **Gas Chromatography-Olfactometry**

Volatile aroma active compounds were extracted from cheeses by SPME and characterized by gas chromatography-olfactometry (GC-O). Five grams of grated cheese was added to a 40 mL amber screw top vial (Supelco, Bellefonte, PA) along with 17% w/w sodium chloride. Vials were heated for 25 min at 40°C using a Reacti Therm TS-18821 Heating/Stirring Module (Thermo Scientific, Waltham, MA) before a SPME fiber [divenylbenzene/Carboxen/polydimethylsiloxane (DVB/CAR/PDMS); Supelco Inc.] was exposed to each sample headspace at 2 cm for 30 min. Fibers were injected at 3 cm on an Agilent 6850 gas chromatography flame ionization detector (FID) with an olfactometry

sniffer port (Agilent Technologies Inc., Santa Clara, CA). Cheeses were evaluated in duplicate by 2 highly trained sniffers on a polar ZB-WAX (30 meter length x 0.25 mm i.d. x 0.25  $\mu$ m film thickness; Zebron; Phenomenex Inc., Torrance, CA) and a nonpolar DB-5ms column (30 meter length x 0.25 mm i.d. x 0.25  $\mu$ m film thickness; Zebron; Phenomenex Inc., Torrance, CA). The GC method used an initial temperature of 40°C for 3 min before increasing at a rate of 10°C/min to 150°C. The rate then increased by 30°C/min to 200°C and was held for 5 min. Effluent was split 1:1 between the FID and sniffer port using deactivated fused-silica capillaries (1 m length x 0.25 mm i.d.; Phenomenex Inc.). Each sniffer recorded retention time, aroma character, and perceived intensity using a 0 to 5 point intensity scale.

### **Identification of Odorants**

Tentative identifications of aroma active compounds in cheeses were based on matching odor properties and/or retention indices (RI values) calculated using an *n*-alkane series to authentic standards (Sigma Aldrich). Positive identifications were completed by comparing GC retention indices (RI) on both GC columns, mass spectra and odor properties with those of chemical standards. Chemical standards were obtained from Sigma Aldrich, St. Louis, MO and Chemstep, Martillac, France.

### **Volatile Compound Extraction and Quantification**

Volatile compounds from Gouda cheeses were extracted and quantified by SPME-GCMS. Compounds were chosen for quantification by GCMS based on frequency, GC-O results, and evaluation of previous literature (Arora et al., 1995; Preininger et al., 1996; Milo and Reineccius, 1997; Suriyaphan et al., 2001; Curioni and Bosset, 2002; Avsar et al., 2004;



Van Leuven et al., 2008). Cheeses were evaluated in triplicate using scan mode followed by select ion monitoring (SIM) mode. Three grams of grated Gouda cheese was added to a 20 mL autosampler vial containing a Teflon silicon septa face (Microliter Analytical Supplies, Suwannee, GA) along with 0.23 grams of sodium chloride. All samples were injected by CTC Analytics Combi PAL autosampler (Leap Technologies, Carrboro, NC).

Volatile fatty acids were quantified using a method adapted from Avsar et al. (2004).

Compounds were quantified using an Agilent 6890 Series GC/Agilent inert 5973 mass selective detector (MSD) equipped with a ZB-FFAP (30 m x 0.25 mm i.d. x 0.25 µm) column (Phenomenex, Torrance, CA) and analyzed using a one phase SPME fiber (PDMS; Supelco Inc.) An internal standard of 40 µl of 317 µl/l heptadecanoic acid (Sigma Aldrich, St. Louis, MO) in ether at a final concentration of 4 mg/kg was added to each vial.

Furaneol, sotolone, and homofuraneol were quantified using a method adapted from Whetstine et al. (2005) with modifications from Frank et al. (2004), Goodner et al. (1999) and Du et al. (2010). ). A 3 phase SPME fiber (DVB/CAR/PDMS; Supelco Inc.) was used to extract compounds (Frank et al., 2004). An internal standard of 80 µl of 300 mg/kg ethyl maltol (Sigma Aldrich) (Du et al., 2010) in ethanol at a final concentration of 8 mg/kg was added to each vial for these compounds. Compounds were quantified using an Agilent 7820 Series GC/Agilent 5975 MSD equipped with a ZB-5 ms (30 m x 0.25 mm i.d. x 0.25 µm) column (Goodner et al., 1999 All other compounds were quantified using a method adapted from Drake et al. (2010). These compounds were quantified using an Agilent 7820 Series GC/Agilent 5975 MSD equipped with a ZB-5 ms (30 m x 0.25 mm i.d. x 0.25 µm) column

(Phenomenex, Torrance, CA) and an internal standard of 20  $\mu$ l of 81 mg/kg 2-methyl-3-heptanone (Sigma Aldrich) in water at a final concentration of 536  $\mu$ g/kg in each vial.

Compounds analyzed on the ZB-5 column used a GC method with an initial temperature of 40°C for 3 min before increasing at a rate of 10°C/min to 90°C. The rate was then increased by 5°C/min to 200°C and 20°C/min to 250°C before being held for 5 min.

Compounds analyzed on the DB-FFAP column used a GC method with an initial temperature of 100°C for 2 min before increasing by 10°C/min to a final temperature of 245°C. For both methods, the SPME fiber was introduced into the split/splitless injector at 250°C at a pressure of 48.7 kPa, using helium as the carrier gas and a purge flow of 1,697 cm/s. A constant flow rate of 34 cm/s was maintained. The purge time was 1 min. The MS transfer line was maintained at 250°C, with the quad at 150°C and the source at 250°C.

Compounds were quantified using a standard addition method. Five point standard addition curves were constructed for each compound by adding known amounts of standards (Sigma Aldrich, St. Louis, MO; Chemstep, Martillac, France) to a model mild Gouda cheese deemed by sensory and instrumental analysis to be low in flavor related volatiles. Regression curves for each compound were calculated by subtracting the peak area found in the unspiked model cheese from the peak area for the spiked cheeses relative to the internal standard. Forty  $\mu$ l of 317  $\mu$ l/l heptadecanoic acid (Sigma Aldrich, St. Louis, MO) in ether at a final concentration of 4 mg/kg was used as an internal standard for fatty acid standard addition curves. Eighty  $\mu$ l of 300 mg/kg ethyl maltol (Sigma Aldrich) in ethanol at a final concentration of 8 mg/kg was used as an internal standard for furanone standard addition curves and 20  $\mu$ l of 81 mg/kg 2-methyl-3-heptanone (Sigma Aldrich) in water at a final

concentration of 536  $\mu\text{g}/\text{kg}$  was used as the internal standard for all other standard addition curves.

### **Descriptive Analysis**

Descriptive analysis was performed in compliance with the North Carolina State University Institutional Review Board for Human Subjects approval. A trained descriptive sensory panel (n=8, 6 females, 2 males, ages 23 to 50 y) evaluated the cheeses for flavor and a trained descriptive sensory panel (n=10, 10 females, ages 35 to 55 y) evaluated the cheeses for texture. All cheeses were served at 15°C. Panelists expectorated samples and were provided with room temperature deionized water and unsalted crackers.

For flavor evaluation Gouda cheeses were cut into 3  $\text{cm}^3$  cubes and approximately 4 cubes were placed into lidded 60 mL soufflé cups with three digit codes. Each panelist had at least 150 h of prior experience with descriptive analysis of flavor with various dairy products, including cheese and yogurt. Cheeses were evaluated for flavor in triplicate using the Spectrum™ method and an established cheese flavor lexicon (Drake et al., 2001; 2004; 2007). Panelists scored intensities using paper ballots. For texture evaluation Gouda cheeses were cut into 1  $\text{cm}^3$  cubes for texture and 16 cubes were placed into lidded 120 mL soufflé cups with three digit codes. Each panelist had approximately 100 h of prior experience with descriptive analysis of texture of dairy products, including cheese. A product specific texture scale was used to evaluate cheese texture in duplicate (Rogers et al., 2009). Data was collected using paper ballots. Results from descriptive analysis of flavor and texture were used to select representative cheeses for consumer acceptance testing.

## **Focus Groups**

Three 1.5 hour focus groups (n=28) were conducted to characterize consumer perception of Gouda cheese. Gouda cheese consumers were recruited from an online database of 8,000 individuals maintained by the Sensory Service Center (North Carolina State University). Panelists were primary shoppers with household income >\$40,000 who purchased Gouda Cheese at least twice a month and consumed cheese weekly. Focus groups were moderated by a trained guide who asked participants a series of predetermined questions in a roundtable format (Table 1). Consumers were asked questions regarding unique qualities, usage, flavor preferences, and purchase habits towards Gouda cheese. Key points from focus groups were utilized in creating the quantitative consumer panel ballot.

## **Consumer Acceptance Testing**

Consumer acceptance testing was conducted to determine consumer preferences for flavor and texture of Gouda cheeses. Testing was conducted using Compusense Cloud (Compusense Inc., Ontario, CA) with a William's randomized serving design. Representative Gouda cheeses (n=10) were selected based on market share, product mean attributes and examination of principal components biplots from descriptive analysis. Gouda cheese consumers were recruited from an online database of 8,000 individuals maintained by the Sensory Service Center (Raleigh, NC). Panelists were primary shoppers with household income >\$40,000 who purchased Gouda Cheese at least twice a month and consumed cheese weekly. Panelists were provided with consent forms, consistent with human subjects approval and an electronic ballot. Samples were prepared by cutting Gouda cheeses into 3 cm<sup>3</sup> cubes and placing into lidded 60 mL soufflé cups with three digit codes. Cheeses were

served at 8° C. Panelists were first asked to evaluate aroma, appearance and color liking for each cheese sample using a nine point hedonic scale. After consuming several bites, panelists evaluated each sample for flavor, saltiness, texture and creaminess liking using a nine point hedonic scale. Panelists used a five point anchored Just About Right scale to evaluate flavor intensity, salty taste intensity, texture and creaminess attributes. For each sample, panelists were also asked purchase intent and usage questions. Consumers were provided with spring water and unsalted crackers for palate cleansing and a 1 min delay was enforced between samples. Consumers were compensated with a \$35 gift card to a local store upon completion of the test.

### **Statistical Analysis**

Statistical analysis was performed using XLSTAT software (version 2015, Addinsoft, New York, NY). Descriptive analysis (DA), volatile compound concentration, and compositional results were analyzed using analysis of variance (ANOVA) with Fisher's least significant difference ( $p < 0.05$ ). Hierarchical agglomerative clustering was used to determine the number of consumer clusters and k means analysis was used to identify and confirm clusters. Consumer liking scores were evaluated by ANOVA with Fisher's least significant difference ( $p < 0.05$ ), just-about-right scores were evaluated by chi square analysis, and purchase intent was evaluated using a Kruskal-Wallis test. Principal component analysis was applied to descriptive analysis to determine how products were differentiated relative to one another. Partial least squares analysis was conducted and drivers of liking and disliking were determined for each cluster based on variable importance scores (VIP).

## RESULTS AND DISCUSSION

### Composition and Organic Acids

All cheeses met the moisture requirements for Code of Federal Regulations (CFR) of <45% (Table 2). Several block Gouda style cheeses (076, 158, 919, 254) did not meet CFR regulations for Gouda fat content (>46%), but these were U.S. export or international Gouda cheeses. As expected, Gouda cheeses ripened for longer periods were lower in moisture and had higher fat and salt contents than younger cheeses. Fat and moisture content were within range of previous composition analysis of Gouda cheeses by Jung et al. (2013) and Welthagen et al. (1998). Cheeses greater than 3 mo were darker in color and more yellow in color than younger cheeses, as indicated by lower L\* values and higher b\* values (Table 3). Color differences between younger and aged Gouda cheeses were likely a result of increased melanoidins responsible for brown pigmentation (Fox et al., 2000). Melanoidin formation is a nonenzymatic browning reaction that occurs in cheese and dairy products when galactose produced from lactose hydrolysis reacts with amino acids produced from proteolytic breakdown (Corzo et al., 2000; Fox et al., 1998). Color results were consistent with results for Egyptian Gouda cheeses by El-Nimr et al. (2010). All pH values for cheeses were within the pH range of 4.9-5.6 for Gouda cheeses stated by Van Den Berg et al. (2004) and the average pH value was 5.49.

Five organic acids were quantified in Gouda cheeses (Table 3). Lactic acid was present at the highest concentration for all cheeses ( $p < 0.05$ ). Overall organic acid concentration increased with ripening time and butyric acid was not detected in any of the

cheeses. These results were also consistent with organic acid determination of Gouda cheeses by Califano et al. (2000) and Skeie et al. (2001).

### **GC-O/MS Results**

Ninety aroma-active compounds were detected in cheeses by SPME-GC-O, including 6 free fatty acids, 7 sulfur derived compounds, 20 aldehydes, 10 esters, 9 nitrogen derived compounds, 3 lactones, 3 alkanes, 11 alcohols, 13 ketones, 3 furanones and 5 unknowns (Tables 4-6). Forty compounds were tentatively identified by aroma and RI. Forty five compounds were identified by aroma, RI and mass spectra. Five compounds remained unknown. The following compounds were reported for the first time as odor active in Gouda cheese by GC-O and were present in at least 30 of 34 cheeses: dimethyl sulfide, 2,3-butanedione (diacetyl), 2-methylbutanal and 3-methylbutanal. Ethyl butyrate, acetic acid, and methional were also present in at least 30 of 34 Gouda cheeses and were previously identified as significant to Gouda cheese flavor based on AEDA of solvent extracts from three cheeses by Inagaki et al (2015). Heptanal, homofuraneol and 2-methylpropanal were detected by GC-O for the first time in at least nine of 12 aged Gouda cheeses >6 mo. Sotolone, butyric and 2-isopropyl-3-methoxypyrazine were also present in the aged Goudas and were previously reported as potent odorants by Inagaki et al. (2015). Aroma active compounds listed above that were identified in at least 30 of 34 Gouda cheeses and aroma active compounds that were identified above in aged Gouda cheeses have been previously detected in Emmental, Cheddar, Blue and hard Italian cheeses by GC-O (Avsar et al., 2004; Frank et al., 2000; Pillonel et al., 2002; Qian and Reineccius, 2002).

Twenty six compounds were quantified using GC-MS, including 4 FFA, 4 sulfur derived compounds, 7 aldehydes, 3 esters, 2 pyrazines, 1 lactone, diacetyl, acetoin and 3 furanones. Nine of the compounds quantified were detected in all cheeses by GCMS. These compounds were: acetic acid, butyric acid, dimethyl sulfide, heptanal, diacetyl, ethyl butyrate, 2-methylbutanal, 3-methylbutanal and 2-methylpropanal. Sotolone, furaneol and homofuraneol were each detected in >25 Gouda cheeses by GCMS while methional and  $\delta$ -decalactone was detected in 30 Gouda cheeses by GCMS. All compounds except sotolone, homofuraneol and 2-methylpropyl acetate were previously quantified in Gouda cheeses (Inagaki et al, 2015; Van Leuven et al., 2008; Jung et al., 2008). Older cheeses were higher in concentrations of 2-methylbutanal, 3-methylbutanal, 2-isobutyl-3-methoxypyrazine, 2,5-dimethylpyrazine, and furaneol. As expected, higher concentrations of esters and lactones were detected for cheeses with higher fat contents, older cheeses, and cheeses produced from raw milk with higher numbers of microorganisms. Esters are formed from the reaction of a free fatty acid with an alcohol and lactones are formed from a one step, nonenzymatic transesterification reaction (Alewijn et al., 2005; Alewijn et al., 2007). Both esters and lactones have been shown to occur in higher concentrations in Gouda cheeses ripened for longer periods by Alewijn et al. (2005).

### **Descriptive Analysis**

Principal component analysis was applied to flavor and texture trained panel profiles of Gouda cheeses. Fifty six percent of the variability was explained on the first two principal components for flavor (Figure 1) and eighteen percent of the variability was explained on the third and fourth components (results not shown). Principal component 1 explained 43% and



was comprised of sour aromatic, whey, fruity, caramel, malty/nutty, brothy, sweet taste, salty taste and umami taste attributes. Principal component 2 explained 13% and was comprised of milkfat and cowy/barny attributes. Sour taste, diacetyl and sulfur attributes comprised third and fourth principal components (results not shown).

All Gouda cheeses had the following sensory attributes: cooked/milky, milkfat, brothy, sulfur and sour aromatic flavors, and sweet, sour, and umami tastes. Young Gouda cheeses were characterized by cooked/milky, whey, sour aromatic, and diacetyl notes while more aged cheeses were differentiated by higher intensities of sulfur, caramel, malty/nutty and fruity flavors and sour and bitter tastes (Figure 1). International cheeses 169 and 180 were differentiated by the presence of low intensities of cowy/barny, mothball and grassy flavors that were likely due to a pasture fed diet (Figure 1). Previous studies by Bendall et al. (2007), Croissant et al. (2007) and Drake et al. (2005) have documented sensory and analytical differences in U.S. versus international cheeses and milks. Bendall et al. (2007) and Croissant et al. (2007) found that flavor variability between pasture and feed based milk resulted from concentration differences for the same compounds, not from the presence of feed or breed associated compounds. Sensory differences based on country of origin were documented between Irish, U.S. and New Zealand Cheddar cheeses by Drake et al. (2005), where non U.S. cheeses were distinguished by low but distinct intensities of cowy/barny and mothball flavors.

Aged Gouda cheeses (212, 267, 324, 416, 520, 539, 608, 620, 714, 834, 995) were characterized by caramel, brothy, malty/nutty, and fruity flavors, and salty, sweet and umami tastes (Figure 1). Young et al. (2004) and Drake et al. (2001) observed similar flavor

differences in Cheddar cheese based on age. Young Cheddar cheeses with less time for flavor development were characterized by whey and diacetyl notes and older cheeses were characterized by more complex flavors and higher basic taste intensities, including sulfur, brothy, caramel, nutty, umami, sour taste, and salty taste (Young et al., 2004, Drake et al., 2001). Van Leuven et al. (2008) previously observed similar decreases in creamy and buttery flavor attributes based on ripening time in Gouda cheese. Higher intensities of sweet and bitter tastes and flowery, fruity, nutty, chocolate and animal flavors were documented in raw milk Gouda cheeses compared to pasteurized milk cheeses. There were two raw milk cheeses in the current study, (298,944, >18 mo, 4-6 mo), but these raw milk cheeses were differentiated from one another based on intensities of whey and fruity flavors. Raw milk Gouda cheeses were not distinct from pasteurized milk Goudas, possibly due to several other factors (age, make procedure, composition) that influence cheese flavor development.

Seventy five percent of the variability was explained on the first two principal components for texture (Figure 2). Principal component 1 explained 59% and was comprised of firmness, cohesion, smoothness and mouthcoating attributes. Principal component 2 explained 16% of the variability and was comprised of springiness, hand recovery, and adhesion attributes. Sixteen percent of the variability was explained on the third and fourth components for texture (results not shown). Principal component 3 explained 9% and was comprised of hand firmness and breakdown attributes. Principal component 4 explained 7% and was comprised of fracture and hand springiness attributes. Young Gouda cheeses were characterized by higher intensities of mouthcoating, smoothness of mass, and breakdown (Figure 2). Aged Dutch Gouda cheeses were characterized by higher intensities of fracture,

firmness in the mouth, and hand firmness that likely result from lower moisture content and breakdown of the protein matrix (Figure 2). Medium Gouda cheeses with higher fat contents were higher in cohesiveness and adhesiveness (Figure 2). Similar texture differences based on age in firmness, fracture, mouthcoating, smoothness and breakdown were previously detected in Gouda cheese (Yates et al., 2006).

### **Focus Group Results**

Consumers stated that the flavor of Gouda cheese was what made it unique as a variety, but were generally unable to describe the flavor profile. Most consumers expected Gouda to have a “creamy” (smooth, homogenous) texture and light yellow color, but some consumers preferred darker aged Gouda cheeses with a drier texture and crunchiness imparted by crystals. More consumers classified the variety as a specialty cheese than an everyday cheese, and used Gouda cheese in numerous applications, including entertaining, snacking, sandwiches, and cooking. Although consumers were most familiar with wedge/wheel shaped Gouda, they expressed interest in trying shredded, sliced and block format cheeses. Only two consumers were aware that Gouda cheese originated in The Netherlands and all consumers stated that they had no preference for European over American Gouda cheeses. Consumers stated that packaging appeal, quality and age were more important when shopping for a new cheese than country of origin or nutritional content.

### **Consumer Acceptance Testing**

Cheeses 495,512, 680, 919 and 944 scored highest in overall liking by all consumers. These cheeses were U.S. young or medium Gouda cheeses aged less than 6 mo. Aged Dutch Gouda cheeses 714 and 324 scored lowest in overall liking across all consumers. Based on

just about right scores, these cheeses were too high in flavor, too firm, and not creamy enough for consumers. Appearance scores were highest for cheeses with higher degrees of Hunter L\* and b\* values, indicating a preference for lighter, more yellow color. Aroma liking scores were highest for cheeses with lower concentrations of FFA and saltiness liking scores were higher for the cheeses with the least salt. Overall appearance, flavor and texture liking were consistent with consumer focus groups. Color, saltiness, firmness and creaminess liking as well as flavor intensity were correlated ( $r^2 \geq 0.95$ ) with overall liking of cheeses ( $p < 0.05$ ).

Three distinct consumer segments were identified from acceptance testing. Overall drivers of liking included sour aromatic, fruity and whey flavors, degree of breakdown and cohesion. Overall drivers of dislike included cooked/milky flavor, salty taste, and fracturability. Clusters one and two were characterized by liking of mild/medium cheeses. Cluster one consumers were driven by a liking of young cheese flavors and a dislike of high intensities of basic tastes (n=47). These consumers liked cheeses characterized by sour aromatic and whey flavors and disliked cheeses with high degrees of malty/nutty flavors, salty and umami taste. Cluster two (n=52) consumers were driven by a liking of mild/medium Gouda cheese texture and a dislike of aged cheese flavors (Figures 1, 2). These consumers liked cheeses characterized by cohesiveness, breakdown, and smoothness of mass, and disliked cheeses with high degrees of sweet, salty and umami tastes and fruity, brothy, sulfur, caramel or malty/nutty flavors. Cluster three (n=54) consumers liked firm, cohesive cheeses with fruity, caramel, brothy and malty/nutty flavors, and disliked springy texture (Figures 1, 2). Cluster one liking scores were highest for samples 512 and 919,

cheeses characterized by young flavors including diacetyl, sour aromatic and whey with higher degrees of adhesion, springy texture and high degree of recovery. Cluster two liking scores were highest for samples 495 and 944, cheeses characterized by medium flavors including sulfur and sour aromatic with a springy texture and high degree of recovery (Figures 1, 2). Cluster three consumers liked nearly all of the cheeses and were differentiated by their high liking for aged cheeses 324 and 714 (Table 4, ( $p < 0.05$ )). Cluster three liking scores were highest for cheeses 714, 324 and 512, cheeses characterized by aged flavors, including caramel, brothy, malty/nutty flavors and sweet and umami tastes with firm and fracturable textures (Figures 1, 2). These results suggest that young and medium Gouda cheeses are liked by all U.S. consumers, but the flavors and texture of aged Gouda cheeses are drivers of liking for one consumer group.

Previous studies concerning consumer acceptance of Cheddar, Edam, and Gouda cheeses have identified similar drivers of liking and consumer clusters based on flavor and texture preferences (Young et al., 2004; Murray et al., 2000; Ritvanen et al., 2005; Yates et al., 2006). Cheddar cheese consumer clusters previously identified by Young et al. (2004) and Murray et al. (2000) differed based on preference for mature cheeses with higher intensities of salty taste, flavor strength, and crumbliness versus liking of younger cheeses characterized by sweet taste and buttery flavor. Ritvanen et al. (2005) investigated Finnish consumer liking of Edam cheeses and found that appearance, mouthfeel and flavor were strongly correlated to overall liking. Edam cheeses with high overall liking were characterized by richness of flavor, salty taste, creaminess, flavor intensity and even appearance of holes, but no consumer clusters were investigated for these consumers

(Ritvanen et al., 2005). Yates et al. (2006) found that both flavor and texture were important to consumer liking of Gouda cheeses, but an undesirable texture cannot be compensated for by a liking of flavor. For the current study and the Yates et al. (2006) study, creaminess was correlated with higher overall liking for Gouda cheeses and fracturability was correlated with lower overall liking scores ( $r^2 \geq 0.93$ ).

## **CONCLUSIONS**

Compositional, sensory, and consumer acceptance differences were observed among Gouda cheeses that were primarily due to age. Based on frequency and aroma character, seven aroma active compounds can be considered characteristic to all Gouda cheeses: dimethyl sulfide, 2,3-butanedione (diacetyl), 2-methylbutanal and 3-methylbutanal, ethyl butyrate, acetic acid, and methional. Six additional compounds can be considered characteristic to aged Gouda cheeses: heptanal, homofuraneol, 2-methylpropanal, sotolone, butyric and 2-isopropyl-3-methoxypyrazine. Older cheeses were more dry, with higher concentrations of flavor compounds and organic acids. Younger Gouda cheeses were lighter in appearance, less intense in flavor and basic tastes, with a creamier and moister texture compared to older cheeses. The physical and chemical properties associated with young and medium cheeses were more appealing to U.S. consumers. Although consumers stated in focus groups that flavor was most characteristic to Gouda cheese, but drivers of liking for flavors as well as texture attributes were crucial to liking of Gouda cheeses. Generally, consumers preferred a lighter Gouda cheese color with low intensities of sulfur, sour aromatic, cooked and diacetyl flavors and low salt. In terms of texture, a Gouda cheese with a smooth texture, moderate mouthcoating and degree of breakdown was most appealing.

However, there is a consumer segment whose liking is driven by the flavors and textures common to aged Gouda cheeses, including malty/nutty, brothy, caramel, and firmness.

Although the majority of consumers consider Gouda to be a specialty cheese, they are open to trying new formats of Gouda cheese and utilize it in numerous applications. These findings can help U.S. manufacturers to understand what flavors and textures are characteristic to this variety and how to create a Gouda cheese with optimal sensory properties for U.S. consumers.

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## **APPENDICES**

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## Appendix A. Moderator's guide for Gouda cheese

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Moderator Introduction: purpose of Focus Group

Respondent introductions: Introduce yourself and share your favorite variety of cheese

Focus Area 1: Perception and use of Gouda Cheese

- Why do you buy Gouda cheese (G)? What is unique about this cheese variety that keeps you buying it?
- Where do you find G in your grocery store and what form do you buy it in?
- Who in your household (hh) consumes it and how often do you/your hh consumes it?
- In what ways do you use G and what makes it suitable for this use?
- Discuss the meaning of each of the following terms and if they are important to your G usage: Ingredient quality, Specialty product, Labels, Nutritional value, Country of origin, Artisanal produced, Raw milk G, Pasteurized milk G, Milk source i.e. cow versus sheeps milk G

Focus Area 2: The ideal Gouda Cheese

- Describe the ideal G in terms of appearance, shape and color. (show pictures and have panelist describe likes/dislikes)
- Describe the ideal G in terms of texture. What other varieties of cheese have a similar texture? (show pictures and have panelist describe likes/dislikes)
- What other factors influence your decision to purchase G?
- Do you tend to stick with G you have tried and liked or try new ones?

Focus Area 3: Gouda Cheese Purchasing

- Where do you prefer to go to purchase Gouda cheese and why?
- Describe your typical G cheese buying experience: who is with you, are there any other considerations not previously mentioned before making a G purchase
- Are there any must haves for purchasing a G? Any absolute nos?

Wrap up: Inquire as to any final thoughts and summarize the major findings

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Appendix B. Moisture, fat, solids and salt of Gouda cheeses

Sample Code	Moisture % <sup>1</sup>	Fat % <sup>1</sup>	Fat % (dry weight)	Salt % <sup>1</sup>	Age (mos)
028 <sup>4</sup>	29.8	29.8	41.2	2.28	<3
076	26.8	28.4	38.8	2.46	<3
158	29.4	30.9	43.8	1.54	<3
169 <sup>4</sup>	28.3	25.3	35.3	1.78	<3
180	39.0	31.7	51.9	2.15	<3
198 <sup>4</sup>	43.2	26.6	46.7	2.67	<3
212 <sup>4</sup>	35.6	26.6	41.3	2.62	5
247 <sup>4</sup>	25.0	38.1	50.7	2.93	9
254	27.7	30.7	42.5	2.95	<3
267 <sup>4</sup>	27.1	35.5	48.7	2.55	18
280 <sup>2</sup>	25.9	48.5	65.5	2.46	4-6
298 <sup>4</sup>	34.4	30.2	46.0	2.61	9-12
324 <sup>4</sup>	30.6	20.8	29.9	2.14	<3
342	28.1	28.5	39.6	2.11	12
373	28.4	29.4	41.1	2.92	7
386	31.2	27.5	40.0	2.78	9
416 <sup>4</sup>	23.9	39.1	51.4	2.99	9
495	28.0	31.9	44.3	2.64	<3
499 <sup>4</sup>	32.1	30.3	44.7	2.30	<3
500 <sup>4</sup>	28.3	37.4	52.2	1.73	9
520 <sup>4</sup>	17.1	40.6	49.0	3.08	12
539 <sup>4</sup>	26.0	28.4	38.4	3.27	10
608 <sup>4</sup>	22.5	34.3	44.3	3.39	18
613 <sup>4</sup>	23.7	33.2	43.6	2.61	<3
620 <sup>4</sup>	21.6	37.5	47.8	2.87	12
677 <sup>4</sup>	19.0	37.6	46.4	2.09	12
680	26.9	30.7	42.0	1.68	<3
714 <sup>4</sup>	19.1	35.2	43.6	2.61	18
788	28.6	28.6	40.1	2.20	6
834 <sup>4</sup>	17.5	33.0	40.0	1.42	5
864 <sup>4</sup>	40.0	29.6	49.3	2.72	6
919	28.2	28.5	39.6	1.28	<3
944 <sup>4</sup>	38.6	23.6	38.5	1.70	4-6
995 <sup>4</sup>	19.4	34.5	42.9	2.93	>24
<b>LSD<sup>3</sup></b>	2.40	2.38	2.72	0.15	

<sup>1</sup>Based on weight/weight <sup>2</sup>Double cream Gouda cheese made from whole milk with added cream

<sup>3</sup>Means within a column that differ by the LSD are different (p<0.05) <sup>4</sup> Internationally produced Gouda cheese

Appendix C. pH, Hunter L\*a\*b\* color, and organic acid concentrations (mg/kg) of Gouda cheeses<sup>1</sup>

Sample Code	Lactic	Citric	Acetic	Pyruvic	Propionic	L*	a*	b*	pH	Age (mos)
028	3336	56.4	123	15.7	136	85.1	-1.45	32.6	5.47	<3
076	3121	58.0	101	9.28	77.4	83.5	3.27	32	5.29	<3
158	2384	75.7	62.6	ND	106	84.1	2.57	34	5.32	<3
169	3049	27.3	111	20.0	106	83.3	-0.26	32.8	5.59	<3
180	3203	42.2	152	30.0	74.8	82.1	-0.09	34.5	5.67	<3
198	3733	59.3	111	4.89	81.8	84.5	-1.43	30.7	5.48	<3
212	3645	ND <sup>2</sup>	162	43.6	291	78.2	0.76	31.4	5.77	5
247	4343	ND	186	53.0	355	80.8	0.95	32.1	5.71	9
254	3416	ND	122	13.2	240	82.3	3.22	30	5.38	<3
267	3842	71.3	282	20.7	316	71.3	8.27	35.7	5.67	18
280	2862	37.9	71.9	17.3	74.2	72.9	4.41	33.3	5.17	4-6
298	3126	ND	118	19.8	316	76.6	7.05	33	5.48	9-12
324	5446	185	190	18.8	233	77.7	0.99	33.2	5.41	<3
342	3008	57.9	130	44.7	426	66.4	2.41	28	5.63	12
373	2595	209	ND	22.6	240	84.4	-1.99	28.7	5.55	7
386	3060	267	121	13.5	198	83.2	2.92	35.7	5.42	9
416	3576	ND	124	22.9	154	84.1	3.02	34.6	5.44	9
495	3933	123	73.9	33.4	112	80.4	-0.44	30.2	5.67	<3
499	3460	19.6	105	4.78	72.1	75.6	4.83	28.2	5.3	<3
500	3335	ND	134	26.2	382	85.8	-1.62	22.7	5.03	9
520	3124	193	27.4	32.2	92.9	81.8	-0.15	38.3	5.36	12
539	4013	ND	204	33.4	145	80.6	-0.1	31.8	5.53	10
608	1150	ND	43.5	ND	38.2	78	5.02	26.2	5.49	18
613	3616	56.4	151	18.8	134	66.1	6.71	34.9	5.39	<3
620	4969	160	188	53.2	217	69.6	5.14	35.7	5.62	12
677	4717	ND	170	49.9	475	69.9	3.71	27.9	5.48	12
680	3436	ND	147	23.9	347	82.2	3.37	33.5	5.27	<3
714	5033	ND	173	31.2	249	69.6	1.77	27.6	5.63	18
788	2762	130	ND	26.4	433	67.1	8.28	36.9	5.53	6
834	5376	ND	191	52.2	300	84.9	-2.14	25.6	5.63	5
864	3560	259	ND	28.3	158	81.7	-1.37	41.4	5.2	6
919	2968	82.9	83.1	ND	145	72.5	1.78	28.9	5.77	<3
944	3071	ND	122	13.1	230	82.7	-0.46	24.2	5.49	4-6
995	5379	ND	499	37.7	419	67.8	12.2	41.3	5.7	>24
<b>LSD<sup>3</sup></b>	598	22.3	18.5	3.95	18.3	0.95	0.21	0.45	0.17	

L\*=Lightness (L\* indicates black and L\*=100 indicates white); a\*=red/ green (negative values indicate green and positive values indicate red); b\*=blue/yellow (negative values indicate green and positive values indicate yellow) <sup>2</sup> ND=Not detected <sup>3</sup> Means within a column that differ by the LSD are different (p<0.05)



Appendix D. Consumer Acceptance results for Gouda cheese flavor and texture

		191	247	280	324	495	512	680	714	919	944
Aroma Liking		5.4fg	6.0de	6.2d	5.7ef	5.4bcd	6.6abc	7.7ab	5.2g	6.8a	6.3cd
Appearance Liking		6.6c	6.7bc	6.6c	5.6d	6.7abc	6.7bc	7.0ab	5.0e	7.1a	6.9abc
Overall Liking		5.7b	5.7b	5.7b	4.3c	6.4a	6.9a	6.5a	4.3c	6.7a	6.8a
Flavor Liking		5.3d	5.7cd	5.6d	4.3e	6.5ab	6.9a	6.2bc	4.2e	6.5ab	6.8a
Flavor	Not enough flavor	15.7%bcd	6.5%cde	19%cb	2.6%de	34.6%a	14.4%cde	18.3%bc	1.3%e	29.4%b	16.3%cd
	JAR	49.7%a	49.7%a	51.7%a	35.3%a	56.8%a	66.7%a	55.6%a	31.3%a	60.1%a	71.9%a
	Too much flavor	34.6%cd	43.8%cd	29.4%cde	62.1%ab	8.5%g	19%defg	26.1%cdef	67.3%a	10.5%fg	11.8%efg
Color liking		6.3b	6.7ab	6.6ab	5.9c	6.4b	6.4b	6.9a	5.1d	6.8a	6.6ab
Color	Too light	44.4%a	19%bc	13.1%cd	7.8%cd	43.8%a	2.6%d	7.8%cd	2.6%d	33.3%ab	34.6%ab
	JAR	34%c	55.6%bc	56.2%bc	62.7%ab	64.1%ab	66%ab	66.7%ab	73.9%ab	77.1%ab	81%a
	Too dark	0%e	3.9%de	13.1%bcd	29.4%bc	0%d	31.4%a	11.1%cd	63.4%a	0%e	1.3%de
Saltiness Liking		5.5c	5.5c	5.6c	4.8d	6.1ab	6.4a	5.8bc	4.8d	6.4a	6.4a
Saltiness	Not salty enough	13.1%a	11.1%a	11.1%a	8.5%a	17.6%a	11.8%a	17%a	5.2%a	15%a	9.8%a
	JAR	52.9%bc	57.5%bc	60.1%abc	51%c	66%abc	69.9%abc	64.1%abc	51%c	79.7%a	73.9%ab
	Too salty	34%ab	31.4%ab	28.8%ab	40.5%a	16.3%bc	18.3%bc	19%bc	43.8%a	5.2%c	16.3%bc
Texture Liking		6.3bc	5.5d	6.2c	4.3e	6.7abc	6.75ab	7.0a	4.0e	6.7abc	6.6abc
Texture	Too smooth	7.2%bc	3.9%c	30.1%a	2.6%c	6.5%bc	9.2%bc	22.2%ab	0.7%c	33.3%a	10.5%bc
	JAR	72.5%ab	51.6%bc	59.5%ab	32%cd	71.9%ab	75.2%a	77.1%a	28.8%d	66.7%ab	64.1%ab
	Too firm	20.3%f	44.4%ef	10.5%de	65.4%d	21.6%d	15.7%d	0.7%ef	70.6%a	0%ab	25.5%cd
Creaminess Liking		6.3b	5.3c	6.3b	4.0d	6.7ab	6.7ab	7.1a	3.9d	6.9a	6.6ab
Creaminess	Not creamy enough	24.2%b	48.4%a	12.4%bc	63.4%a	20.9%b	23.5%b	4.6%c	65.4%a	3.3%c	24.2%b
	JAR	69.9%a	46.4%bc	65.4%ab	32.7%c	72.5%a	71.2%a	78.4%a	31.4%c	73.2%a	68.6%ab
	Too creamy	5.9%c	5.2%c	22.2%ab	3.9%c	6.5%bc	5.2%c	17%abc	3.3%c	23.5%a	7.2%abc
Purchase Intent		2.92c	2.99bc	2.93c	2.30d	3.52a	3.63a	3.41abc	2.18d	3.50ab	3.77a

Data represents 153 consumers. Different letters in rows following means signify significant differences ( $p < 0.05$ ) Liking attributes were scored on a 9-point hedonic scale where dislike extremely =1 and like extremely =9.

JAR scales were scored on a 5-point scale where too little =1 or 2, just about right =3, and too much=4 or 5. The percentage of consumers that selected these options is presented.

Purchase intent was scored on a 5-point scale where would not buy =1 or 2, may or may not=3, and would buy=4 or 5

\*Statistical letterings were obtained from Kruskal-Wallis non-parametric test with lower scores corresponding to higher ranking.

Appendix E. Screener Questions for Gouda Cheese Consumer Testing

<b>Are you lactose intolerant?</b>
Yes
No <b>Must select</b>
<b>Please indicate your gender.</b>
Male <b>50%</b>
Female <b>50%</b>
<b>To which age group do you belong?</b>
18 years old or younger <b>Disqualify</b>
19-25 years old
26-35 years old
36-45 years old
46-55 years old
56-64 years old
65 years old and over
<b>What is your household shopping responsibility?</b>
Primary
Secondary <b>Disqualify</b>
I do not do the shopping for my household <b>Disqualify</b>
<b>Including yourself, how many people live in your household?</b>
1
2
3 – 4
5 or more
<b>Please select the category that best describes your ethnicity</b>
African american
Asian
Caucasian
Hispanic
Other
Prefer not to answer
<b>Please indicate your annual household income.</b>
Under \$25,000 per year <b>Disqualify</b>
\$25,000 to \$49,999 per year <b>Disqualify</b>
\$50,000 to \$74,999 per year
\$75,000 to \$99,999 per year
More than \$100,000 per year
<b>Please indicate your flavor preference for cheese</b>
I like mild flavored cheeses <b>50%</b>
I like more intensely flavored cheeses <b>50%</b>

Appendix F. Consumer Ballot for Gouda Cheese Consumer Testing

<p>You will now start with sample _____.</p>								
<p><b>How much do you like the AROMA of this product?</b></p>								
Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<p><b>Considering everything about this product, what is your overall impression of the APPEARANCE of this product?</b></p>								
Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<p><b>How much do you like the COLOR?</b></p>								
Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<p>Please <i>take a bite</i> of sample _____ and answer the following questions:</p>								
<p><b>Considering everything, how much do you like this sample?</b></p>								
Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<p><b>How much do you like the FLAVOR of this sample?</b></p>								
Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<p><b>How much do you like the SALTINESS of this sample?</b></p>								
Dislike Extremely	Dislike Very Much	Dislike Moderately	Dislike Slightly	Neither Like nor Dislike	Like Slightly	Like Moderately	Like Very Much	Like Extremely
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>

Appendix F cont. Consumer Ballot for Gouda Cheese Consumer Testing

<p><b>How much do you like the TEXTURE of this sample?</b></p> <p>Dislike Dislike Dislike Dislike Neither Like Like Like Like Like Extremely Very Much Moderately Slightly nor Dislike Slightly Moderately Very Much Extremely</p>									
□	□	□	□	□	□	□	□	□	□
<p><b>How much do you like the CREAMINESS of this sample?</b></p> <p>Dislike Dislike Dislike Dislike Neither Like Like Like Like Like Extremely Very Much Moderately Slightly nor Dislike Slightly Moderately Very Much Extremely</p>									
□	□	□	□	□	□	□	□	□	□
<p><b>Now please think about the flavor intensity of this cheese.</b> Would you say the flavor intensity is:</p> <p style="text-align: center;">Not nearly enough      Not enough      Just About Right      Too much      Much too much</p>									
□	□	□	□	□					
<p><b>Would you say the salty taste intensity is:</b></p> <p style="text-align: center;">Not nearly salty enough      Not salty enough      Just About Right      Too salty      Much too salty</p>									
□	□	□	□	□					
<p><b>Would you say the texture is:</b></p> <p style="text-align: center;">Not nearly soft enough      Not soft enough      Just About Right      Too soft      Much too soft</p>									
□	□	□	□	□					
<p><b>Would you say the creaminess intensity is:</b></p> <p style="text-align: center;">Not nearly creamy enough      Not creamy enough      Just About Right      Too creamy      Much too creamy</p>									
□	□	□	□	□					
<p><b>How likely would you be to purchase this CHEESE if it were available (at a reasonable price) in the store where you usually shop?</b></p> <p style="text-align: center;">Definitely would NOT buy      Probably would NOT buy      Might or might not buy      Probably would buy      Definitely would buy</p>									
□	□	□	□	□					
<p><b>Please tell us all of the ways that you would consume this cheese (CATA):</b></p> <p><input type="checkbox"/> An everyday snack</p> <p><input type="checkbox"/> Cooking</p> <p><input type="checkbox"/> Sandwich</p> <p><input type="checkbox"/> Special event/Occasion</p> <p><input type="checkbox"/> Other (please specify) _____</p>									

Appendix G. Aroma active compounds for in Gouda cheeses 028-495 (RI DB5 <600-852)

RI DB5	RI WAX	Compound	Odor	028	180	198	212	247	254	267	280	298	324	342	416	495	499
<600	693	Methanethiol	Rotten									x		x			x
<600	635	Dimethyl Sulfide	Garbage/ Pumpkin	x	x	x	x	x	x	x	x	x	x	x	x	x	x
<600	822	Butanal	Green									x	x				
<600	713	Ethanal	Solvent	x	x				x		x				x	x	
<600	912	3-methylbutanal	Malt/ Chocolate	x	x	x	x	x	x	x	x	x	x	x	x	x	x
<600	971	2,3-butanedione	Butter	x	x	x	x	x	x	x	x	x	x	x	x	x	x
603	1445	Acetic Acid	Sour Vinegar	x	x	x	x	x	x	x	x	x	x	x	x	x	x
612		Methyl Ethyl Sulfide	Garbage								x			x			
614	920	2-methylbutanal	Malt/ Almond	x	x	x	x	x	x	x	x	x	x	x	x	x	x
630	1528	Propanoic Acid	Pungent		x							x	x				
634	875	Ethyl ethanoate	Fruity						x								
661	821	2-methylpropanal	Vegetal malt	x	x	x			x	x		x	x	x	x	x	x
672	929	Ethanol	Sweet	x		x				x	x			x	x	x	x
699	1051	2,3-pentadione	Creamy	x		x		x			x	x		x			
720	990	Methyl Butanoate	Apple/Fruity		x		x	x	x	x	x	x		x		x	x
732	1282	3-hydroxybutanone	Fresh	x		x	x	x	x	x			x	x	x	x	x
749	1108	2-methyl-1-butanol	Malt						x					x	x		x
767	1115	(Z)-2-penten-1-ol	Plastic	x		x		x				x		x	x	x	
778	1106	Hexenone	Cooked/Vegetal	x		x	x				x	x	x	x	x	x	x
784	1135	4-methyl-3-penten-2-one	Medicinal			x			x			x	x	x			
788	1030	2-methylpropyl acetate	Bubblegum/ Fruity	x	x	x		x	x	x	x	x		x	x	x	
799		Octane	Alkane				x										x
800	1097	Hexanal	Green	x		x		x	x	x	x			x	x		
803	951	Ethyl Butyrate	Fruity	x	x		x	x	x	x	x	x	x	x	x	x	x
813	1160	Propyl Propionate	Fruity		x				x						x		x
819	1156	Nonenone	Baked		x				x		x					x	
823	1176	2-hydroxy-3-pentanone	Earthy														
824	1620	Butanoic Acid	Sour Cheesy	x	x	x	x	x			x	x	x	x	x	x	x
840	1190	(E)-2-hexanal	Green							x			x				
852	1063	Ethyl-3-methylbutanoate	Fruity								x			x	x	x	x

Cheeses 076,158,169,191,373, and 386 were similar in aroma composition to other cheeses and not presented in Appendices G-I

Appendix G cont. Aroma active compounds for in Gouda cheeses 499-995 (RI DB5 <600-852)

RI DB5	RI WAX	Compound	Odor	499	500	520	539	608	613	620	677	680	714	834	864	919	944	995
<600	693	Methanethiol	Rotten	x						x			x		x	x	x	
<600	635	Dimethyl Sulfide	Garbage/ Pumpkin	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
<600	822	Butanal	Green															
<600	713	Ethanal	Solvent									x						
<600	912	3-methylbutanal	Malt/ Chocolate		x				x				x				x	x
<600	971	2,3-butanedione	Butter	x	x	x	x	x	x	x	x	x	x	x		x	x	x
603	1445	Acetic Acid	Sour Vinegar	x														
612		Methyl Ethyl Sulfide	Garbage		x	x	x	x	x	x	x	x	x	x	x	x	x	x
614	920	2-methylbutanal	Malt/ Almond	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
630	1528	Propanoic Acid	Pungent															x
634	875	Ethyl ethanoate	Fruity		x	x	x	x	x	x	x	x	x	x	x	x	x	x
661	821	2-methylpropanal	Vegetal malt	x														
672	929	Ethanol	Sweet	x														x
699	1051	2,3-pentadione	Creamy										x					
720	990	Methyl Butanoate	Apple/Fruity	x	x	x	x	x	x		x					x	x	x
732	1282	3-hydroxybutanone	Fresh	x			x				x	x						
749	1108	2-methyl-1-butanol	Malt	x			x				x					x		
767	1115	(Z)-2-penten-1-ol	Plastic		x	x		x	x	x	x					x	x	x
778	1106	Hexenone	Cooked/Vegetal	x	x	x	x	x	x	x	x		x	x	x	x	x	x
784	1135	4-methyl-3-penten-2-one	Medicinal					x										x
788	1030	2-methylpropyl acetate	Bubblegum/ Fruity		x		x	x		x	x				x	x		x
799		Octane	Alkane			x			x			x		x				
800	1097	Hexanal	Green					x										
803	951	Ethyl Butyrate	Fruity	x	x	x	x	x	x				x	x	x	x		
813	1160	Propyl Propionate	Fruity	x														
819	1156	Nonenone	Baked						x			x						
823	1176	2-hydroxy-3-pentanone	Earthy		x		x	x		x	x		x			x	x	x
824	1620	Butanoic Acid	Sour Cheesy	x	x	x	x	x	x	x	x	x	x		x	x	x	x
840	1190	(E)-2-hexanal	Green			x	x						x				x	x
852	1063	Ethyl-3-methylbutanoate	Fruity				x	x	x				x					x

Cheeses 512 and 788 were similar in aroma composition to other cheeses and not presented in Appendices G-I

Appendix H. Aroma active compounds in Gouda cheeses 028-495 (RI DB5 852-1179)

RI DB5	RI WAX	Compound	Odor	028	180	198	212	247	254	267	280	298	324	342	416	495
854	1215	2-hexen-1-al	Cheesy	x	x		x		x		x	x			x	
897	1234	Dimethyl Pyrazine	Nutty/Brothy								x					
904	1176	Heptanal	Stale/Fatty	x	x	x	x	x	x	x	x	x	x	x	x	x
909	1463	Methional	Potato	x	x	x	x	x	x	x	x	x		x	x	x
924	1338	2-Acetyl-1-pyrroline	Popcorn				x	x	x	x	x		x	x	x	
925	1273	2-Heptanol	Mushroom	x	x		x	x			x	x	x	x	x	x
936		Unknown	Musty/Pungent												x	x
966	1315	1-octen-3-one	Earthy/Woody	x	x	x	x		x		x		x	x		
975	1382	Dimethyl Trisulfide	Sulfur/Rotten	x	x	x			x			x		x	x	
987	1345	1,5-octadien-3-one	Metallic	x		x								x	x	x
1009	1291	Octanal	Oily Stale	x		x		x	x		x	x	x	x	x	x
1010	1360	Propionylpyrrole	Popcorn	x			x	x		x			x	x	x	
1014		(Z)-3-hexenyl acetate	Pineapple		x							x		x	x	
1016	1829	Hexanoic Acid	Sour		x				x	x	x	x				
1026	1611	Acetylthiazole	Roast			x							x	x		
1038	1217	D-Limonene	Mint/Citrus			x						x				
1040	1651	1-phenylethanone	Creamy/Musty													x
1055	1421	2-methoxy-3,6-dimethylpyrazine	Earthy												x	
1072	2032	Furaneol	Sweet		x					x	x	x			x	x
1075	1558	1-octanol	Metallic/Burnt	x		x					x	x		x		
1092	1881	Guaiaacol	Potpourri	x					x							
1095		2-acetyl-2-thiazoline	Malt						x			x			x	
1104	1604	(E)-2-octenol	Plastic	x								x				x
1112	1392	Nonanal	Fatty	x						x						
1115	1601	2,4-octadienal	Vegetal								x	x	x	x	x	x
1144	2240	Sotolone	Curry	x		x	x			x		x	x	x	x	
1151	1525	E-2-nonenal	Green/Plastic							x	x	x			x	
1154	2079	Homofuraneol	Caramel/Sweet		x	x	x	x	x			x		x	x	
1167	1534	2,3-diethyl-5-methylpyrazine	Baked/Meaty		x	x	x			x	x			x		
1179	1582	2-Methylisoborneol	Earthy	x	x							x		x	x	

Cheeses 076,158,169,191,373, 386, 512 and 788 were similar in aroma composition to other cheeses and not presented in Appendices G-I.

Appendix H cont. Aroma active compounds in Gouda cheeses 499-995 (RI DB5 852-1179)

RI DB5	RI WAX	Compound	Odor	499	500	520	539	608	613	620	677	680	714	834	864	919	944	995
854	1215	2-hexen-1-al	Cheesy			x	x		x	x	x		x		x			x
897	1234	Dimethyl Pyrazine	Nutty/Brothy	x						x			x			x		
904	1176	Heptanal	Stale/Fatty	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
909	1463	Methional	Potato	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
924	1338	2-Acetyl-1-pyrroline	Popcorn	x		x	x			x			x	x				x
925	1273	2-Heptanol	Mushroom	x	x	x	x		x	x				x	x	x		
936		Unknown	Musty/Pungent	x					x				x	x				
966	1315	1-octen-3-one	Earthy/Woody	x		x		x	x	x			x			x	x	
975	1382	Dimethyl Trisulfide	Sulfur/Rotten	x		x	x	x		x	x		x	x				
987	1345	1,5-octandien-3-one	Metallic	x		x			x		x		x	x	x	x		
1009	1291	Octanal	Oily Stale	x	x	x			x	x	x	x	x	x	x	x		x
1010	1360	Propionylpyrrole	Popcorn	x				x	x				x				x	x
1014		(Z)-3-hexenyl acetate	Pineapple		x						x							
1016	1829	Hexanoic Acid	Sour	x		x	x			x			x	x				x
1026	1611	Acetylthiazole	Roast		x		x		x				x					x
1038	1217	D-Limonene	Mint/Citrus															
1040	1651	1-phenylethanone	Creamy/Musty	x	x		x						x	x	x			
1055	1421	2-methoxy-3,6-dimethylpyrazine	Earthy				x	x			x							
1072	2032	Furaneol	Sweet	x	x	x	x	x			x		x		x			
1075	1558	1-octanol	Metallic/Burnt	x	x					x	x		x					x
1092	1881	Guaiacol	Potpourri							x								
1095		2-acetyl-2-thiazoline	Malt						x	x				x				
1104	1604	(E)-2-octenol	Plastic	x			x						x					
1112	1392	Nonanal	Fatty	x		x		x	x		x	x						x
1115	1601	2,4-octadienal	Vegetal	x		x	x								x	x		
1144	2240	Sotolone	Curry			x	x	x		x	x	x			x	x	x	x
1151	1525	E-2-nonenal	Green/Plastic	x		x												
1154	2079	Homofuraneol	Caramel/Sweet		x	x	x	x		x	x				x	x		x
1167	1534	2,3-diethyl-5-methylpyrazine	Baked/Meaty	x	x	x	x		x									x
1179	1582	2-Methylisoborneol	Earthy			x									x			

Cheeses 512 and 788 were similar in aroma composition to other cheeses and not presented in Appendices G-I.



Appendix I. Aroma active compounds in Gouda cheeses (RI DB5 >1188)

RI DB5	RI WAX	Compound	Odor	028	180	198	212	247	254	267	280	298	324	342	416	495
1188	1514	2-isobutyl-3-methoxy-pyrazine	Pepper peel/Green	x	x	x				x	x	x	x	x	x	x
1192		4-acetyltoluene	Nutty												x	
1203	1243	(Z)-6-decenal	Green	x	x											
1209		Unknown	Sweet malt		x											x
1216	1472	Decanal	Cooked stale	x			x				x	x		x	x	
1230		Methyl nonanoate	Sweet									x				
1241	1611	Dihydromethyl cyclopentapyrazine	Nutty		x											
1274	2083	Octanoic Acid	Rancid	x		x	x	x	x	x	x	x	x	x	x	
1286	1633	2,4-decadienal	Green Fermented			x			x		x					
1317	1655	$\gamma$ -butyrolactone	Sweet		x										x	
1347		(E)-2-undecenal	Fatty/Green						x							
1381		Geranyl acetate	Floral													
1391	2270	Decanoic Acid	Rancid							x			x			
1392	1052	Isobutyl thiazole	Green	x												
1456	1803	2-dodecenal	Earthy/Sweet													
1512	2218	$\delta$ -decalactone	Peach/Sweet Cream					x	x		x			x	x	x
1640	2011	(E)-2-hexenoic Acid	Stale				x					x				x
1713	2359	$\gamma$ -dodecalactone	Sweet									x		x		
1749	2416	$\delta$ -dodecalactone	Sweet	x							x					
	1141	2-heptanone	Musty										x			
	1417	Unknown	Vegetal/Fermented	x			x					x				
	1479	Unknown	Ketchup/Brothy													
	1633	Phenylacetaldehyde	Honey/Sweet													
1905	2260	7-epi- $\alpha$ -eudesmol	Woody								x			x		
1949	2281	$\gamma$ -undecalactone	Sweet/toasted													
2015		(E)-isoeugenol	Sweet	x												
2060		Methyl octadecenoate	Waxy									x				

Cheeses 076,158,169,191,373 and 386 were similar in aroma composition to other cheeses and not presented in Appendices G-I.

Appendix I cont. Aroma active compounds in Gouda cheeses (RI DB5 >1188)

RI DB5	RI WAX	Compound	Odor	499	500	520	539	608	613	620	677	680	714	834	864	919	944	995
1188	1514	2-isobutyl-3-methoxy-pyrazine	Pepper peel/Green	x	x	x	x	x	x	x	x		x		x	x	x	x
1192		4-acetyl-toluene	Nutty		x		x		x		x							x
1203	1243	(Z)-6-decenal	Green		x								x					
1209		Unknown	Sweet malt					x					x					x
1216	1472	Decanal	Cooked stale	x			x	x	x						x		x	
1230		Methyl nonanoate	Sweet						x					x				
1241	1611	Dihydromethyl cyclopentapyrazine	Nutty			x	x		x	x						x		
1274	2083	Octanoic Acid	Rancid	x	x	x			x	x	x		x		x	x	x	x
1286	1633	2,4-decadienal	Green Fermented			x			x					x				
1317	1655	$\gamma$ -butyrolactone	Sweet	x	x	x												x
1347		(E)-2-undecenal	Fatty/Green						x					x				
1381		Geranyl acetate	Floral															x
1391	2270	Decanoic Acid	Rancid		x		x					x				x		
1392	1052	Isobutyl thiazole	Green				x		x									
1456	1803	2-dodecenal	Earthy/Sweet		x	x	x		x									
1512	2218	$\delta$ -decalactone	Peach/Sweet Cream	x	x					x			x		x	x		
1640	2011	(E)-2-hexenoic Acid	Stale	x		x	x		x									x
1713	2359	$\gamma$ -dodecalactone	Sweet	x	x					x								
1749	2416	$\delta$ -dodecalactone	Sweet							x								
	1141	2-heptanone	Musty						x									
	1417	Unknown	Vegetal/Fermented				x	x										
	1479	Unknown	Ketchup/Brothy	x		x				x								
	1633	Phenylacetaldehyde	Honey/Sweet			x				x								
1905	2260	7-epi- $\alpha$ -eudesmol	Woody			x												
1949	2281	$\gamma$ -undecalactone	Sweet/toasted	x											x			
2015		(E)-isoeugenol	Sweet								x							x
2060		Methyl octadecenoate	Waxy															x

Cheeses 512 and 788 were similar in aroma composition to other cheeses and not presented in Appendices G-I.

Appendix J. Concentrations of select odor active compounds in Gouda cheese (µg/kg)

Compounds	Acetic Acid	Butyric Acid	Hexanoic Acid	Octanoic Acid	Dimethyl sulfide	Dimethyl disulfide	Dimethyl trisulfide	Hexanal
028	14.6	7.44	72.1	2.74	79.1	23.5	23.1	11.6
158	72.5	6.78	22.3	1.78	47.9	22.7	22.9	3.81
180	1.27	5.98	15.1	1.73	376	29.5	30.8	40.8
212	273	43.2	87.4	5.72	250	45.1	24.9	91.3
247	27.9	54.3	54.4	5.07	89.1	24.1	22.9	68.3
254	36.4	6.99	17.9	2.31	23	24.3	22.5	3.32
267	4744	1141	939	13.3	80.2	24.3	22.8	11.9
280	15.8	6.84	16.4	1.86	23.2	22.6	22.5	4.11
298	93.3	7.88	15.4	1.76	340	51.2	25.7	109
324	248	52.1	24.4	4.66	45.8	23.5	22.6	5.16
342	78.7	8.73	16	ND <sup>2</sup>	43.8	23.8	24.3	12.6
416	45.5	6.94	22.2	2.33	29	22.9	22.9	3.27
495	14.2	6.93	15.8	ND	534	27.6	25.5	67.8
499	830	74.9	42.6	8.65	26.4	22.7	22.8	2.01
500	391	109	29.6	3.51	137	50.3	24.3	76.6
520	1358	57.7	159	6.81	187	27.8	24.9	48.2
539	1127	27.5	183	3.86	52.3	23.2	24.5	11.4
608	27.4	73.7	28.4	2.48	29.5	23.1	22.9	6.27
613	3960	342	102	7.26	93.2	26.1	36.5	75.5
620	126	19.3	33.1	3.51	45.5	31.1	28.9	53.2
677	6501	1242	1778	13.9	286	27.5	25.8	13.3
680	88.1	34.5	29.4	4.43	49.4	26.5	24.7	12.5
714	23.4	7.49	18.2	1.78	120	41.2	27.6	39.3
834	99.6	49.2	36.9	5.02	70.3	28.5	22.8	26.1
864	94.5	30.4	151	2.79	67.2	23.5	24.9	15.6
919	9.01	11.1	23.6	2.12	46.6	22.8	22.8	2.25
944	424	15.9	28.7	2.70	654	32.7	26.9	82.4
995	12.7	6.47	15.3	1.75	30	42.6	23.1	5.77
<b>LSD</b>	665	12.2	24.9	24.2	144	0.35	0.49	3.55

<sup>1</sup> Concentrations in mg/kg

<sup>2</sup> ND=Not detected in cheeses

076, 169,191,386,512 and 788 were similar in composition to other cheeses and not presented in Table 6

Appendix J cont. Concentrations of select odor active compounds in Gouda cheese (µg/kg)

Compounds	Heptanal	Octanal	Decanal	Diacetyl <sup>1</sup>	Acetoin <sup>1</sup>	Ethyl butyrate	Isobutyl Acetate	Ethyl 3-methylbutyrate	δ-decalactone
028	37.5	10.1	ND	12.5	38.8	80.6	3.69	ND2	188
158	18.4	270	18.7	2.25	8.03	20.5	ND	1.91	86.3
180	251	76.8	287	7.08	ND	345	56.3	ND	18.1
212	347	1164	361	67.2	15.1	1070	ND	ND	97.3
247	1664	2582	2641	141	76.3	490	ND	ND	43.6
254	9.16	223	ND	0.28	0.5	9.91	4.29	ND	93.2
267	60.2	1441	520	7.02	4.62	283	2.62	1.61	43.8
280	13.7	396	44.9	2.74	0.99	27.8	9.07	21.5	26.6
298	43.2	3309	287	27	115	2095	2092	1.96	229
324	37.4	58.9	26.5	5.46	1.58	54.1	0.61	ND	30.5
342	4.91	56.3	ND	1.52	ND	69	ND	5.9	78.5
416	38.4	429	26.7	1.78	0.32	34.9	31.1	ND	ND
495	29.6	826	ND	0.4	32.2	111	17	ND	72.6
499	7.81	262	21.7	1.48	ND	22.3	3.71	49.9	28.2
500	100	4858	516	12.2	25.3	654	ND	ND	56.8
520	338	5973	163	39.1	87.1	1021	289	ND	422
539	4.64	116	ND	22.4	37.6	73.3	ND	ND	26.2
608	12.6	1033	87.7	4.71	0.52	32	16.2	27.6	54.6
613	187	200	504	11.4	128	273	802	1.90	98.9
620	318	525	267	19.7	101	532	ND	ND	24.9
677	99	1313	ND	11.6	94.9	2463	ND	ND	244
680	8.83	56.1	61.3	17.7	0.55	84.1	ND	ND	374
714	139	3136	89	44.8	93.4	803	104	6.59	98.4
834	69.4	119	32.1	6.98	6.65	115	39.6	7.21	213
864	38.4	149	100	4.16	11.9	47.6	ND	22.3	92.6
919	15.4	199	37.5	1.22	ND	9.64	0.63	4.96	ND
944	450	444	649	28.8	32.9	581	ND	32.9	ND
995	21.9	15113	33.6	2.64	ND	15.9	235	16.5	97
<b>LSD</b>	3.87	28.5	47.7	8.47	16.4	7.41	30.7	1.74	62.3

<sup>1</sup> Concentrations in mg/kg

<sup>2</sup> ND=Not detected in cheeses

076, 169,191,386,512 and 788 were similar in composition to other cheeses and not presented in Table 6

Appendix J cont. Concentrations of select odor active compounds in Gouda cheese (µg/kg)

Compounds	3-methyl butanal	2-methyl butanal	2-Methyl propanal	2-isobutyl-3-methoxy pyrazine	2,5-dimethyl pyrazine <sup>1</sup>	Methional	Furaneol	Homofuraneol	Sotolone
028	0.31	0.26	0.17	10.8	ND	32.0	ND	ND	4.51
158	0.61	0.14	1.07	8.21	ND	2.69	ND	ND	1.51
180	16.2	4.49	2.03	35.4	79.2	78.2	19.9	0.71	1.03
198	0.66	0.08	0.88	2.62	ND	ND	ND	1.12	3.84
212	51.6	37.5	6.22	182	72.3	39.2	6.3	4.56	3.27
247	182	71.2	12.3	ND	62	450	78.4	10.8	2.17
254	5.23	2.34	4.08	1.65	ND	2.45	ND	0.86	ND
267	1.02	0.45	2.72	92.7	0.48	27.7	28.2	4.78	0.99
280	0.83	0.15	0.88	2.03	ND	1.84	16.5	ND	ND
298	37	2.95	5.98	110.4	ND	60	26.5	13.7	5.45
324	0.27	0.39	1.65	7.76	ND	12.1	ND	0.21	4.56
342	9.93	0.15	0.26	24.9	ND	13.1	ND	ND	3.37
416	0.25	0.11	2.34	7.4	ND	21.9	99.8	15.1	3.84
495	2.79	1.73	5.32	88.8	ND	0.09	23.9	ND	ND
499	0.11	0.03	0.98	5.38	ND	ND	23.6	ND	ND
500	11.1	1.16	13.3	60.4	186	26.2	18.7	5.75	ND
520	58.4	20.96	4.1	125	131	8.43	25.9	11.1	4.46
539	0.97	1.08	2.64	12.2	ND	98.6	47.5	24.6	3.36
608	0.41	0.61	6.14	14.8	29.9	11.2	22.4	12.3	8.55
613	6.34	2.95	1.8	39.6	ND	23.3	ND	0.32	ND
620	47.6	36.9	3.94	181	22.3	63.3	34.9	10	5.4
677	10.8	13.4	0.39	1620	413	484	36.5	9.78	3.21
680	0.09	0.18	0.13	13.1	0.34	5.23	ND	1.96	3.11
714	94.9	31.9	16.3	95.5	104	123	25.5	ND	0.83
834	0.69	0.92	4.67	26.7	0.61	94.1	ND	0.5	2.02
864	0.15	0.26	0.29	6.79	ND	49.5	281	28.4	4.95
919	ND	83.4	0.1	0.04	6.23	4.76	ND	0.43	ND
944	7.81	0.89	9.47	ND	ND	29.5	ND	0.72	10.5
995	3.98	1.79	2.62	52.1	24.7	24.7	ND	7.73	2.2
<b>LSD</b>	4.74	5.88	1.91	22.7	9.01	10.8	5.10	0.96	0.28

<sup>1</sup> Concentrations in mg/kg

<sup>2</sup> ND=Not detected in cheeses

Cheeses 076, 169,191,386,512 and 788 were similar in composition to other cheeses and not presented in Table 7.

Appendix K. Trained panel profiling of Gouda cheese flavor (n=36)

Sample Code	Cooked	Sour Aromatic	Whey	Diacetyl	Milkfat	Fruity	Caramel	Malty/Nutty	Brothy	Sulfur	Bitter	Cowy/Barny	Grassy
028	4.4	0.75	2.4	ND	4.0	ND	2.5	ND	2.0	1.3	ND	1.3	1.3
076	3.6	1.0	3.0	1.3	3.3	ND	0.67	ND	1.9	1.8	0.93	ND	ND
158	3.3	ND	2.0	ND	3.7	ND	2.5	1.7	2.3	1.8	ND	ND	ND
169	3.3	1.4	2.2	ND	3.0	1.5	1.3	1.3	2.4	1.3	0.50	2.8	ND
180	3.5	1.2	2.3	ND	3.3	1.2	0.75	0.75	2.5	3.0	1.5	1.5	1.4
191	4.0	1.8	2.6	0.83	3.0	ND	0.50	ND	2.1	2.4	1.0	ND	ND
198	3.4	2.3	2.7	ND	3.3	ND	1.0	ND	2.1	3.3	ND	ND	ND
212	4.5	ND	0.50	ND	4.5	1.7	4.3	4.0	3.7	1.3	0.75	ND	ND
247	3.2	ND	ND	ND	3.7	1.9	4.3	3.6	3.5	1.1	ND	ND	ND
254	3.0	0.90	2.0	1.0	3.3	ND	1.7	0.67	2.0	2.4	0.50	ND	ND
267	4.0	0.75	ND	ND	4.0	1.4	4.7	3.9	3.7	1.0	1.4	ND	ND
280	3.0	1.3	1.5	ND	4.5	ND	ND	ND	2.2	2.3	0.50	ND	ND
298	2.5	0.50	1.0	ND	3.8	2.7	2.7	2.0	3.0	1.4	0.83	1.6	ND
324	3.0	0.82	2.5	ND	3.4	ND	1.0	1.0	2.0	2.5	1.0	ND	ND
342	3.5	ND	ND	ND	3.5	1.3	3.3	2.3	3.2	2.4	ND	0.75	ND
373	4.0	1.5	2.1	0.50	4.0	ND	ND	1.8	2.0	1.5	0.67	ND	ND
386	3.0	0.50	2.5	0.50	4.0	ND	1.0	0.77	1.5	1.5	1.5	ND	ND
416	3.0	0.83	2.6	ND	3.1	ND	1.5	ND	2.1	2.1	0.83	ND	ND
495	3.6	0.75	1.0	ND	4.0	ND	2.0	ND	4.1	2.7	ND	ND	ND
499	3.0	2.0	1.0	ND	3.0	ND	ND	ND	ND	ND	1.0	ND	ND
500	3.0	ND	1.0	ND	3.3	3.6	3.5	2.9	2.6	1.4	0.83	ND	ND
512	4.0	ND	2.2	ND	4.0	ND	1.5	ND	2.5	1.0	0.50	ND	ND
520	3.0	0.50	ND	ND	3.1	1.0	4.7	4.3	4.8	1.1	0.65	ND	ND
539	3.5	1.5	2.8	ND	3.5	2.3	1.6	1.0	2.5	1.0	0.50	0.50	1.0
608	3.0	ND	ND	ND	3.6	1.0	4.0	1.8	2.8	1.4	0.50	ND	ND
613	3.6	1.0	2.2	1.5	3.8	ND	1.7	1.2	2.0	2.5	0.75	ND	ND
620	3.2	ND	2.1	ND	3.7	2.9	3.0	2.5	1.4	1.3	0.75	ND	ND
677	3.0	ND	2.4	ND	3.5	3.5	2.0	2.3	2.5	1.0	0.50	3.1	1.0
680	4.0	1.0	2.7	1.4	4.0	ND	1.7	ND	1.5	1.8	0.50	ND	ND
714	3.9	ND	ND	ND	3.8	1.4	3.5	3.3	2.8	1.6	ND	ND	ND
788	3.3	0.57	2.5	ND	3.0	ND	1.7	ND	2.8	1.8	0.83	ND	ND
834	3.0	ND	ND	ND	4.2	1.4	4.1	3.4	3.1	1.4	ND	ND	ND
864	3.0	0.90	2.6	ND	3.0	ND	1.3	0.83	2.0	2.7	1.0	1.0	ND
919	3.0	1.5	2.3	ND	3.0	ND	ND	ND	2.0	3.4	1.2	ND	ND
944	3.5	1.5	2.7	1.0	3.7	ND	1.3	ND	1.9	1.9	1.6	ND	ND
995	3.6	ND	ND	ND	3.7	1.2	4.7	2.5	4.1	1.3	0.50	ND	ND

Appendix K cont. Trained panel profiling of Gouda cheese basic tastes (n=36)

Sample Code	Sour	Sweet	Salty	Umami	Bitter
028	2.8	2.5	3.80	3.2	ND
076	2.8	1.9	2.3	2.1	0.93
158	2.4	3.4	3.8	3.5	ND
169	3.0	3.6	3.2	3.4	0.50
180	3.4	2.6	3.2	2.8	1.5
191	3.7	2.5	3.7	3.2	1.0
198	3.3	2.8	3.6	2.3	ND
212	2.6	4.2	5.5	4.5	0.75
247	2.3	4.1	4.3	3.7	ND
254	2.8	2.3	4.8	3.5	0.50
267	3.0	3.7	5.9	4.6	1.4
280	2.8	2.4	4.7	3.4	0.50
298	2.8	3.8	3.0	2.8	0.83
324	2.7	2.0	4.1	3.0	1.0
342	2.7	4.2	4.1	4.0	ND
373	2.7	2.7	3.8	3.5	0.67
386	2.5	2.5	4.3	3.4	1.5
416	2.8	2.4	4.0	2.7	0.83
495	2.1	3.0	5.0	3.9	ND
499	3.8	2.0	4.7	3.5	1.0
500	3.2	3.9	3.6	3.1	0.83
512	2.5	3.0	3.8	4.0	0.50
520	2.9	4.0	5.4	4.6	0.65
539	2.0	3.6	5.0	4.2	0.50
608	2.8	3.1	5.1	4.0	0.50
613	3.0	3.6	2.5	3.5	0.75
620	3.4	3.7	3.9	3.1	0.75
677	2.0	3.6	4.9	4.2	0.50
680	3.0	2.2	3.5	3.0	0.50
714	2.7	3.2	4.3	3.9	ND
788	2.8	2.6	4.4	3.3	0.83
834	2.8	3.6	4.4	3.8	ND
864	3.4	2.5	3.6	3.0	1.0
919	3.0	2.0	3.2	2.6	1.2
944	3.8	2.5	3.5	2.7	1.6
995	2.7	3.6	4.8	4.0	0.50

Appendix L . Trained panel profiling of Gouda cheese texture (n=36)

Sample Code	Hand Firmness	Hand Springiness	Hand Recovery	Firmness	Fracture	Breakdown
028	8.4	7.3	6.3	6.9	4.3	5.3
076	13.0	14.0	14.0	11.0	8.7	2.4
158	10.0	14.0	13.0	8.9	6.7	4.7
169	11.0	14.0	14.0	9.5	5.9	8.1
180	12.0	14.0	13.0	9.7	6.2	7.7
187	6.3	13.0	13.0	6.6	4.7	9.7
191	9.5	13.0	13.0	8.7	6.4	9.1
198	8.9	13.0	13.0	7.1	4.8	10.0
212	13.0	1.5	1.9	10.0	7.1	6.9
235	14.0	ND	ND	11.0	7.8	6.9
254	8.2	13.0	12.0	7.3	5.2	11.0
267	14.0	ND	ND	11.0	8.4	6.8
298	10.0	2.4	2.2	8.3	6.1	10.0
318	9.5	13.0	13.0	8.0	5.5	8.6
342	6.0	13.0	12.0	5.7	4.8	12.0
373	7.1	11.0	11.0	7.1	5.2	9.0
386	6.3	12.0	11.0	5.0	4.1	12.0
416	11.0	11.0	12.0	7.0	5.0	11.0
499	12.0	ND	ND	9.2	5.9	6.7
500	16.0	2.0	1.8	8.6	5.6	11.0
512	9.5	7.8	6.8	9.0	5.8	7.2
520	12.0	ND	ND	10.0	9.0	5.8
539	9.3	12.0	10.3	7.8	4.3	9.9
608	8.1	7.3	5.7	6.5	4.0	7.0
612	12.0	5.0	7.0	11.0	8.0	4.3
613	9.2	9.3	8.9	7.8	6.9	7.9
620	12.0	0.5	0.5	10.0	6.5	3.0
629	4.1	14.0	12.0	3.8	2.0	7.5
677	14.0	ND	ND	13.0	11.0	3.6
707	9.5	13.0	13.0	8.7	6.4	9.1
788	5.3	12.0	12.0	5.8	4.7	8.1
834	14.0	ND	ND	13.0	10.0	2.0
847	1.9	5.4	10.3	14.0	14.0	8.3
864	9.4	12.0	12.0	8.0	5.9	8.8
904	9.5	11.0	9.5	4.8	3.5	9.8
995	14.0	ND	ND	11.0	8.6	6.8



Appendix L cont. Trained panel profiling of Gouda cheese texture (n=36)

Sample Code	Cohesiveness	Adhesiveness	Mass Smoothness	Mouthcoating
028	6.5	6.3	7.3	7.5
076	2.7	3.5	3.5	4.7
158	4.5	5.0	5.8	6.8
169	8.9	8.2	8.8	9.3
180	8.8	8.2	8.2	8.8
187	10.2	9.2	10.6	10.5
191	9.67	9.3	8.7	8.7
198	10.3	9.4	10.5	10.7
212	7.4	8.3	5.8	5.6
235	7.1	8.4	5.4	5.5
254	11.0	9.5	11.3	11.3
267	6.8	8.4	4.8	4.6
298	10.0	9.7	9.3	9.1
318	9.2	8.5	9.6	9.4
342	11.0	10.3	11.7	11.2
373	9.7	8.6	9.4	10.1
386	12.0	10.6	11.9	11.6
416	12.0	10.0	12.0	12.0
499	8.0	9.0	11.0	11.5
500	11.0	10.0	10.3	9.8
512	7.0	6.0	6.9	7.15
520	7.0	4.0	2.3	4.0
539	8.5	9.0	12.5	13.0
608	8.1	8.4	11.8	12.0
612	5.5	5.8	3.0	3.5
613	8.4	8.3	8.4	8.8
620	4.7	5.0	2.0	2.0
629	8.1	7.7	12.5	13.0
677	7.0	7.8	6.3	6.0
707	9.7	9.3	8.7	8.7
788	11.0	9.3	11.4	11.0
834	4.8	5.3	4.8	4.7
847	5.6	8.1	8.3	8.0
864	9.1	8.6	8.2	8.3
904	10.0	8.5	10.0	11.0
995	6.9	8.7	4.9	3.9



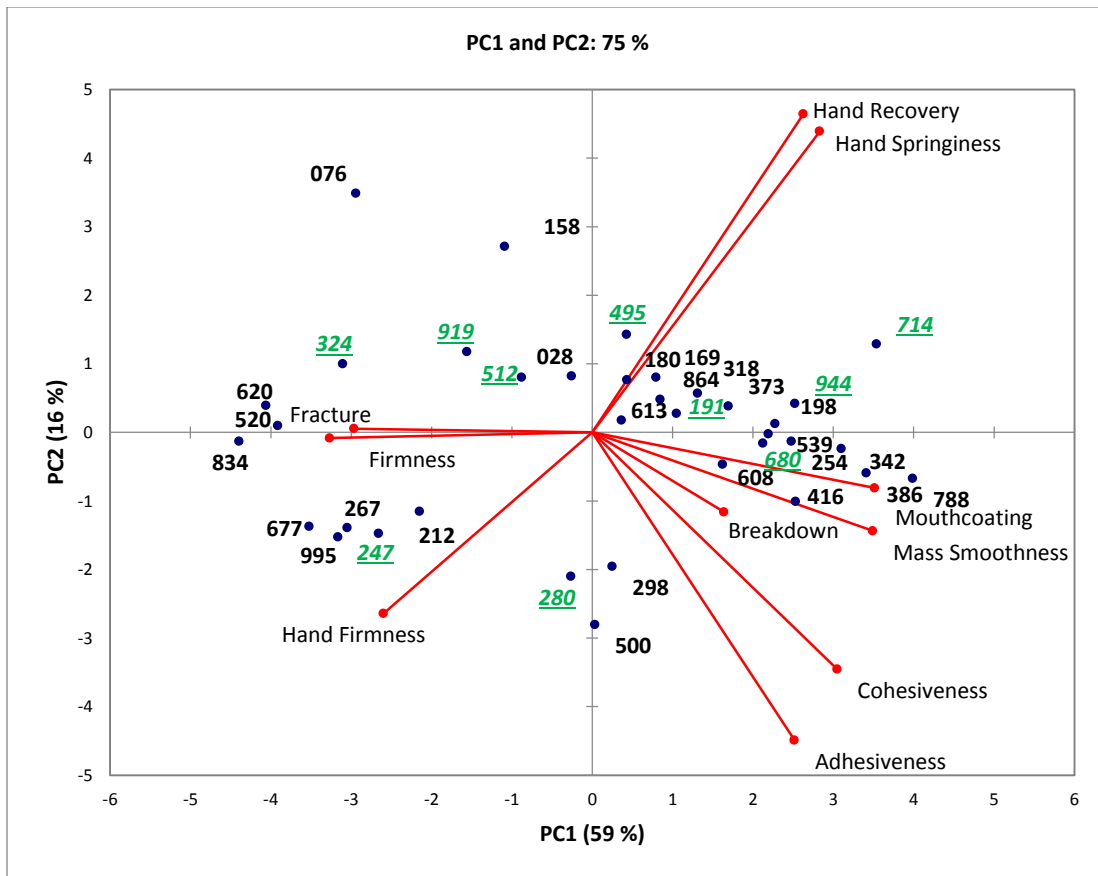


Figure 2. Principal components analysis biplot of flavor texture for Gouda cheeses. Underlined cheeses were selected for consumer testing.

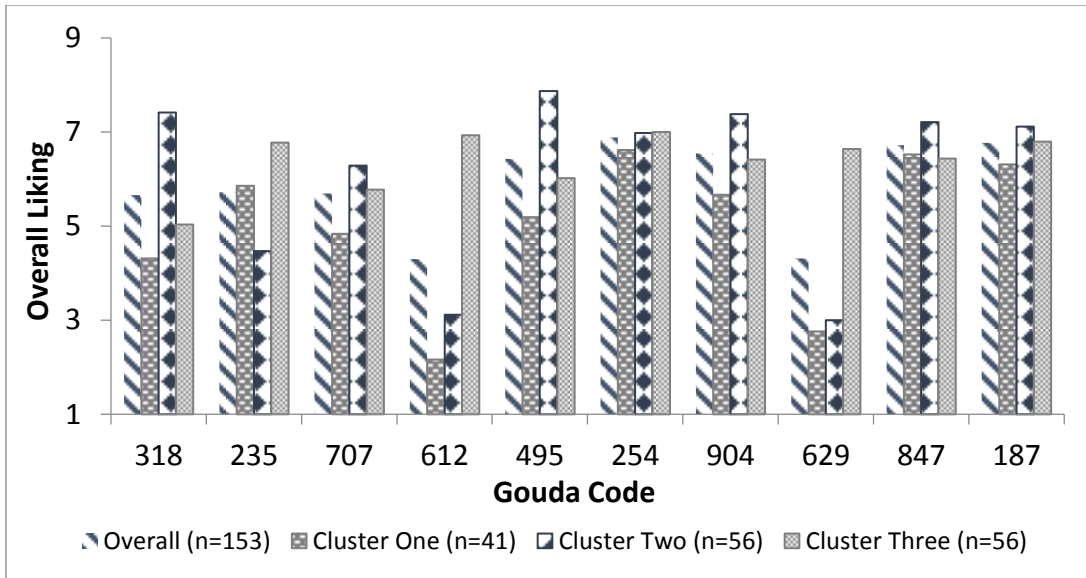


Figure 3. Overall consumer liking scores for Gouda cheeses

Liking was scored on a 9 point hedonic scale, where 1 was “dislike extremely” and 9 was “like extremely”.

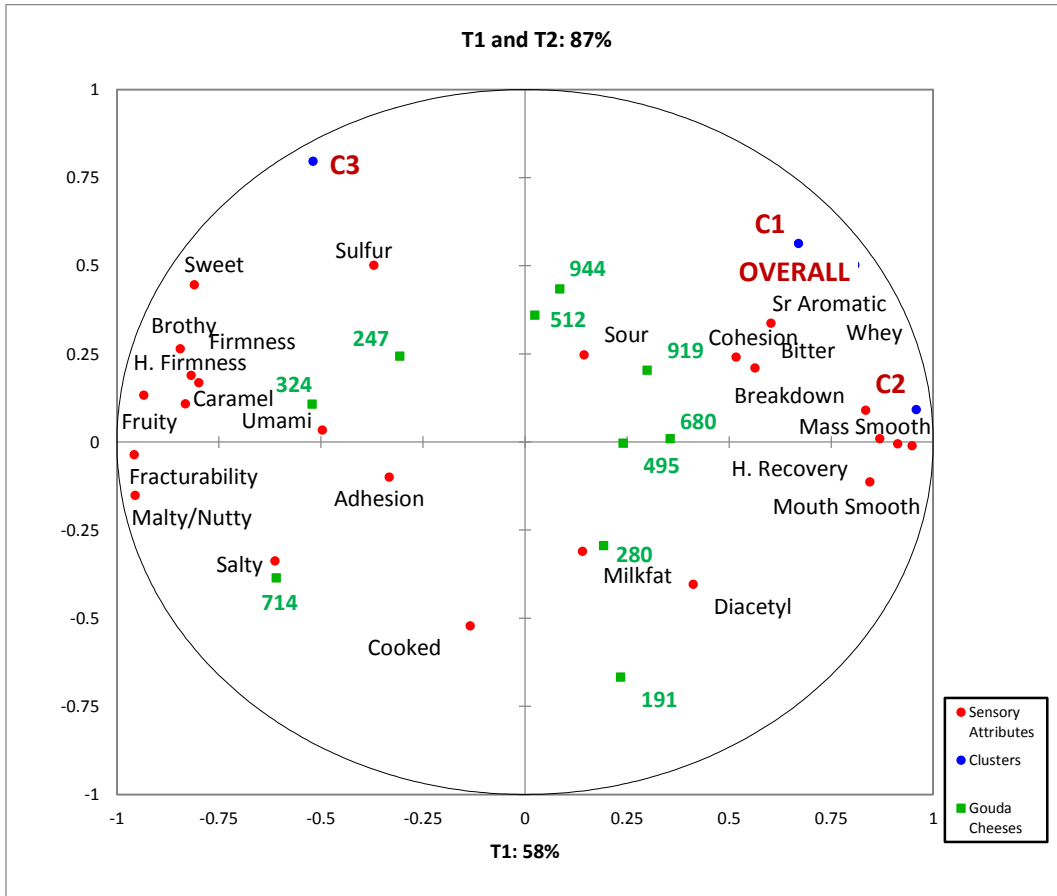


Figure 4. Partial least squares (PLS) correlation biplot of trained panel and consumer liking scores

Flavor and texture attributes are included in this biplot. Numbers represent cheeses that were included in consumer testing. C1 represents cluster 1 (n=47), C2 represents cluster 2 (n=52) and C3 represents cluster 3 (n=54)